

# Assessment of morphological and chlorophyll content traits in Bambara groundnut (*Vigna subterrannea* [verdc L.]



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**Background:** Bambara groundnut has promising economic potential due to its various uses such as food, medicinal and agronomic benefits. To date, there are no significant breeding efforts aimed at cultivar development and the crop remains underutilised.

**Aim:** To characterise and identify Bambara groundnut landraces using agro-morphological and chlorophyll traits to identify breeding lines for cultivar development.

**Setting:** The experiment was conducted in Limpopo and North West provinces (Loskop and Molelwane research farms).

**Methods:** This study used randomised complete block design (RCBD) with three replications.

**Results:** Analysis of variance revealed significant variations among most of the landraces used. Highly significant positive correlations were observed for most of the traits recorded. The principal component analysis (PCA) indicated that the first six principal components contributed 78.34% of the total variability among the test genotypes that were being evaluated. The biplot and dendrogram further grouped the landraces according to traits associated with them. Shannon Weaver's diversity index showed a wide variability among the accessions in qualitative morphological traits.

**Conclusion:** The agro-morphological traits and chlorophyll content characterisation showed significant variability within the landraces tested. These findings would be beneficial for breeders to choose the desirable traits associated with the Bambara groundnut landraces in the breeding programme for the development of new breeding population.

**Contribution:** The identified superior landraces will be utilised by the small-scale and large-scale commercial farmers for direct cultivation and marketing, and can be used as parents in breeding programmes.

**Keywords:** Bambara groundnut; agronomic traits; characterisation; genetic coefficient variance; phenotypic coefficient variance.

## Introduction

Bambara groundnut (*Vigna subterranea* [L.] Verdc.) is an African legume crop that has been cultivated for many years even before the groundnut (*Arachis hypogaea* L.) (Hillocks, Bennet & Mponda 2012). It is an indigenous and underutilised grain legume food crop, which is mainly grown by female subsistence farmers in semi-arid parts of Africa (Ntundu et al. 2006). This legume has numerous names which differ according to ethnic groups in South Africa and include Phonda (Venda), Ditloo-marapo (Sepedi), and Tindhluwa (Tsonga) (Swanevelder 1998).

This legume has been termed a complete food and is grown mainly for its seed, which constitutes about 63% carbohydrates, 19% protein and 6.5% fats (Abedije et al. 2018; Anhwange & Atoo 2015; Bamishaiye, Adegbola & Bamishaiye 2011; Mbuma et al. 2022), and has the potential to alleviate risk of nutritional insecurity (Siwale et al. 2023). It also has promising economic potential due to its various uses such as food, medicinal, and agronomic benefits, including the plant parts that can be used for animal feed. Freshly harvested and dry Bambara groundnuts are consumed in many ways after processing for human consumption. In Eastern Africa, Bambara groundnuts are roasted and milled, and the flour is used to make soup, a relish, and a substitute for coffee (Mubaiwa et al. 2018). Moreover, the crop can improve the fertility of the soil through nitrogen

**Note:** This article was republished with updated naming convention of the graphical material: Figure 1 changed to Table 3 and Figure 2 changed to Table 4, consequently renumbering all the remaining tables and figures accordingly. This correction does not alter the study's findings of significance or overall interpretation of the study's results. The publisher apologises for any inconvenience caused.

fixing bacteria (Hillocks et al. 2012; Mayes et al. 2019). Although this crop has advantages for food security, Bambara groundnut accessions are merely landraces (Unigwe et al. 2016) with low yield potential; hence, identification of potential breeding lines based on the agronomic traits and release of improved cultivars for small-scale and large-scale cultivation is critical in the breeding programme. Therefore, the study was conducted to provide a better understanding of phenotypic trait- and physiological variations among 20 landraces and their relationship between plant traits and yield components. Also, the study explored phenotypic trait variations using agro-morphological traits for potential contribution to selecting breeding lines for high yields and related traits this crop offers. The study also evaluated the phenotypic variability and the relationship among accessions and to estimate the frequency distribution of the qualitative traits assessed.

## Research methods and design

### Description of experimental sites

The experiment was conducted in two extreme ecological sites. The first site was located in Limpopo Province (Loskop research station [25.1773°S, 29.3936°E with average temperatures between 16°C and 31°C]) during 2019–2020 cropping season. The second site was established in North West province (Molelwane research farm [25°48'00"S, 25°38'21"E with the average temperatures ranging from 22°C to 34°C]) in 2020–2021 cropping season. The soil type at both locations was classified as the Hutton series according to the South African soil classification system (Kasirivu, Mterechera & Dire 2011) and is reddish with a sandy loam texture. The total rainfall during the growing seasons at Molelwane trial site was 590 mm in 2019–2020 and 561 mm in 2020–2021, while at Loskop it was 429 mm in 2019–2020 and 442 mm during the 2020–2021 growing season.

### Planting materials

Twenty Bambara groundnut landraces were obtained from the Agricultural Research Council (ARC) gene bank, Pretoria, South Africa. A randomised complete block design (RCBD) with three replications was used. Each plot was 2.7 cm × 60 cm with inter- and intra-row spacing of 60 cm × 30 cm. Each plot had 2 rows, with each row consisting of 10 plants. Sowing was done on a flat bed, with one seed sown at each station. Weeds were controlled by hoeing. No fertiliser was applied at both sites (Gerrano et al. 2015).

### Data collection

Table 1 shows the quantitative data that were collected (on three randomly selected plants averaged per plot) during the pre-harvest and post-harvest growth stages of the crop. Qualitative morphological data were collected using Bambara groundnut descriptors (IPGRI/IITA/BAMNET 2000) on the traits presented in Table 2.

**TABLE 1:** Quantitative traits that were collected and the methods or procedures used.

| Quantitative traits                   | The method and procedures used for collecting data                                                                                                                                                                                                                                                                                                                                  |
|---------------------------------------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| Chlorophyll content                   | Measured using chlorophyll meter.                                                                                                                                                                                                                                                                                                                                                   |
| Terminal leaf length and width        | Recorded 10 weeks after planting; average length and width of three leaves from each plant at the fourth node of 3 healthy plants.                                                                                                                                                                                                                                                  |
| Leaf area: area (mm)                  | Measured (estimated) from 3 leaves per plant at 10 weeks from sowing. Leaf area was then estimated based on the central leaflet length and width using the method of (Cornelissen et al. 2003) in the following equation: $A = \sigma * L * W * \pi / 4$ (2), where: L = Length of the leaflet (cm) W = Width of the leaflet (cm) $\pi = 3.1416$ $\sigma = 0.95$ correction factor. |
| Days to 50% flowering                 | Determined visually by counting the number of days when 50% of the plants in a subplot had opened flowers.                                                                                                                                                                                                                                                                          |
| Number of nodes per stem              | Number of nodes counted at harvest on randomly selected three plants.                                                                                                                                                                                                                                                                                                               |
| Number of branches per plant          | Number of branches counted at randomly selected three plants.                                                                                                                                                                                                                                                                                                                       |
| Plant height (mm)                     | Measured from the ground level (at the base of the plant) to the tip of the highest point, including the terminal leaflet. Recorded 10 weeks after planting; the average height of 3 plants.                                                                                                                                                                                        |
| Days to maturity                      | This parameter was taken when about 80% of the pods on a sample plant were matured. 1–2 plants were uprooted in each subplot, and the number of matured pods was indicated by the dark markings of the internal shell wall.                                                                                                                                                         |
| Pod length and width (mm)             | Digital Vernier Calliper was used to measure the greatest length and width of 10 randomly selected dried pods per plant (at 12% moisture content).                                                                                                                                                                                                                                  |
| Seed length and width (mm)            | Digital Vernier Calliper was used to measure the greatest length and width of 10 randomly selected dried seeds per plant (at 12% moisture content)                                                                                                                                                                                                                                  |
| Number of pods per plant              | The numbers of pods were taken from three plants after harvesting                                                                                                                                                                                                                                                                                                                   |
| One hundred seeds weight per plot (g) | One hundred seeds were counted and averaged after shelling in a plot base                                                                                                                                                                                                                                                                                                           |
| Yield per plant (g)                   | The seed yield per plants was weighed on an electric scale                                                                                                                                                                                                                                                                                                                          |

mm, millimeter; g, gram; cm, centimeter;  $\pi$ , pi;  $\sigma$ , sigma.

**TABLE 2:** Descriptors and codes used for the characterisation of Bambara groundnut landraces.

| Qualitative markers                           | Descriptor and code                                        |
|-----------------------------------------------|------------------------------------------------------------|
| Growth habit                                  | 1 Bunch; 2 Semi-bunch; 3 Spreading                         |
| Flower colour                                 | 1 Yellow; 2 White                                          |
| Terminal leaflet shape                        | 1 Round; 2 Oval; 3 Elliptic; 4 Lanceolate                  |
| Stem hairiness                                | 0 Absent 3 Sparse 7 Dense                                  |
| Seed colour                                   | Visual technique and Munsell colour chart used             |
| Seed shape                                    | 1 Round; 2 Oval; 3 Ovate; 4 Spherical                      |
| Dark pigmentation on the terminal leaflet     | 0 Absent 1 Present                                         |
| Colour of the fully expanded terminal leaflet | 1 Green, 2 Red, 3 Purple                                   |
| Pod colour                                    | 1 Yellow; 2 Brown; 3 Reddish-brown; 4 Purple               |
| Pod texture                                   | 1 Smooth; 2 Little grooves; 3 Much grooves; 4 Much grooves |

### Data analysis

Statistical Analysis System (SAS 2021) version 9.4 was used to analyse the data. Data subjected to analysis of variance (ANOVA) to explore differences among test landraces. Mean separation was carried out using Fisher's protected least significant difference (LSD) at the 5% significance level of probability. Correlations between yield and yield-related contributing traits and principal

component analysis (PCA) were also performed to reveal variability among characters. Cluster and biplot analyses were performed to study similarities and dissimilarities between the tested landraces based on the traits recorded. For qualitative morphological data, the phenotypic frequency distributions of the phenotypic characters were computed. Shannon Weaver diversity index was calculated from the phenotypic frequencies to analyse and interpret the phenotypic diversity for each trait (Zavinon et al. 2019):

$$H' = 1 - \sum_{i=1}^n (\log_e) P_i \quad [\text{Eqn 1}],$$

where  $P_i$  is the proportion of accessions in the  $i$ th class of an  $n$ -class descriptor and  $n$  is a phenotypic class for a descriptor.

### Estimation of variance components

The phenotypic, genotypic, and environmental variances, and coefficient of variation were calculated according to the formula described by Khan et al. (2020):

Environmental variance:

$$\delta^2_e = \text{MSE} \quad [\text{Eqn 2}]$$

Genotypic variance:

$$\delta^2_p = \frac{MSG - MSE}{rs} \quad [\text{Eqn 3}]$$

Phenotypic variance:

$$\delta^2_p = \sigma^2_g + \sigma^2_e \quad [\text{Eqn 4}],$$

where mean square of genotypes (MSG) is the mean square due to genotype, MSE is the mean square of error (environmental variance), and  $r$  is the number of replications and  $s$  the number of sites.

Phenotypic coefficient of variance:

$$\text{PCV} = \sqrt{\frac{(\delta^2_g)}{GM}} * 100 \quad [\text{Eqn 5}]$$

Genotypic coefficient variance:

$$\text{GCV} = \sqrt{\delta^2_g} / GM * 100 \quad [\text{Eqn 6}],$$

where:

$\delta^2_p$  = phenotypic variance

$\delta^2_g$  = genotypic variance

GM = grand mean of the character studied

Estimation of heritability in a broad sense: broad sense heritability ( $h^2$ ), expressed as the percentage of the ratio of genotypic variance ( $\delta^2_g$ ) to the phenotypic variance ( $\delta^2_p$ ) was calculated using the following formula (Owusu et al. 2021):

$$h^2 = \frac{\delta^2_g}{\delta^2_p} \times 100 \quad [\text{Eqn 7}]$$

**TABLE 3:** Combined mean values across sites showing variation in vegetative traits of Bambara groundnut.

| Genotype | DTF     | NNS      | NBS      | DM        | CHL      | TLL    | TLW   | LArea   | PH       | SC     |
|----------|---------|----------|----------|-----------|----------|--------|-------|---------|----------|--------|
| Bamb1    | 77.33   | 20.12b-e | 10.17ed  | 167.67    | 35.33b-d | 13.82  | 6.97  | 74.22   | 20.95def | 15     |
| Bamb2    | 77.00   | 18.17c-f | 12.5b-e  | 173.83a-c | 53.57a   | 12.58  | 5.65  | 53      | 21.13c-f | 16.33  |
| Bamb3    | 84.17   | 20.33a-e | 13.83a-c | 172.17a-f | 38.45b-d | 14.05  | 6.433 | 67.02   | 21.4c-f  | 16.33  |
| Bamb4    | 77.67   | 25a      | 14.5a-c  | 170.5a-g  | 44.95a-c | 13.78  | 7.02  | 74.93   | 21.72cde | 19.83  |
| Bamb5    | 75.67   | 21.5a-d  | 16ab     | 173.67abc | 42.07a-d | 13.02  | 6.7   | 72.1    | 24.75b-e | 18.5   |
| Bamb6    | 75.00   | 17.67def | 14.17a-c | 170.5a-f  | 35.42b-d | 14.97  | 6.73  | 75.68   | 24.75a   | 14.833 |
| Bamb7    | 77.33   | 20.5a-d  | 14.67a-c | 164.67g   | 53.03a   | 14.9   | 6.53  | 76.25   | 23.22a-d | 18.17  |
| Bamb8    | 75.00   | 19.83b-e | 16ab     | 166.83efg | 49.7ab   | 14.22  | 6.63  | 75.9    | 20.93d-f | 18.67  |
| Bamb9    | 77.17   | 15.67ef  | 14.17a-c | 171.83a-f | 36.77b-d | 13.65  | 6.6   | 67.52   | 21.65c-e | 18.83  |
| Bamb10   | 75.83   | 19.17b-f | 13.33a-d | 173.33a-d | 44.25a-c | 14.5   | 5.75  | 60.93   | 19.27f   | 16.83  |
| Bamb11   | 77.50   | 19.83b-e | 13.17a-e | 165.83g   | 46.92a-c | 14.78  | 7.3   | 83.13   | 22.93a-d | 18.67  |
| Bamb12   | 77.33   | 18.17c-f | 12.67b-e | 171.67a-f | 29.7d    | 13.63  | 6.55  | 68.05   | 20.5ef   | 17     |
| Bamb13   | 80.83   | 19.83b-e | 16.33a   | 168.83b-g | 41.6a-d  | 14.52  | 7.43  | 81.48   | 22.33b-e | 14.33  |
| Bamb14   | 77.17   | 18.67c-f | 11.33c-e | 172.67a-d | 36.45b-d | 15.55  | 6.23  | 73.63   | 20.05ab  | 16.33  |
| Bamb15   | 75.17   | 20.5a-d  | 12.5b-e  | 170.5a-g  | 34.05d   | 12.95  | 6.23  | 61.12   | 21.8c-e  | 17.83  |
| Bamb16   | 79.67   | 14.83f   | 9.67e    | 172.83a-d | 36.48b-d | 13.62  | 6.13  | 63.98   | 20.3ef   | 17.17  |
| Bamb17   | 78.17   | 19.83b-c | 12.83a-e | 167d-g    | 37.72b-d | 13.9   | 7.05  | 79.3    | 20.87d-f | 19.5   |
| Bamb18   | 77.50   | 23.5ab   | 13a-e    | 173a-d    | 48.7ab   | 15.47  | 6.816 | 81.23   | 22.88a-d | 20.33  |
| Bamb19   | 75.17   | 22.83a-c | 13.17a-e | 174.17ab  | 40.18a-d | 14.75  | 6.95  | 83.25   | 22.8a-d  | 17.5   |
| Bamb20   | 78.00   | 20.17b-e | 13.17a-e | 175.83a   | 28.75d   | 15.55  | 7.57  | 86.85   | 21.4c-f  | 17     |
| <i>p</i> | ns      | *        | *        | *         | *        | ns     | ns    | ns      | ***      | ns     |
| LSD      | 0.2     | 0.02     | 0.03     | 0.03      | 0.02     | 0.1    | 0.5   | 0.1     | 0.08     | 0.2    |
| CV       | 6.05    | 20.99    | 23.72    | 3.28      | 30.9     | 12.8   | 19.46 | 25.75   | 8.89     | 21.26  |
| GM       | 1185.02 | 67.81    | 31.59    | 76.4      | 254.64   | 145.35 | 3.67  | 5611.52 | 189.41   | 471.95 |

Note: The letters just shows the existence of the differences and not same letters showing their similarity. Means within the same column followed by different letters are significantly different at  $p < 0.05$ .

CV, coefficient of variation; LSD, least significant difference; DTF, Days to 50% flowering; NNS, Number of nodes per stem; NBS, Number of branches per plant; DM, Days to maturity; CHL, Chlorophyll content; TLL, Terminal length; TLW, Terminal leaflet width; LArea, leaflet area; PH, Plant height; SC, Stand count; GM, grand mean.

\*\*\*, highly significant at  $p \leq 0.001$ ; \*, significant at  $p \leq 0.05$ .

**TABLE 4:** Combined mean values showing variation in yield and related traits of Bambara groundnut across test sites.

| Genotypes      | PodL      | PodW      | SL       | SW        | HSW       | YPP         | NPP       |
|----------------|-----------|-----------|----------|-----------|-----------|-------------|-----------|
| Bamb1          | 19.64cde  | 12.2abc   | 12.32cd  | 9.78bcd   | 43.08c-g  | 40.4bc      | 88.5      |
| Bamb2          | 19.55cde  | 12.37ab   | 14.34a   | 10.17bc   | 44.13c-g  | 45.34bc     | 94.83     |
| Bamb3          | 21.32ab   | 12.55ab   | 10.9gh   | 9.93bcd   | 38.76g    | 22.8c       | 117.17    |
| Bamb4          | 21.65a    | 12abc     | 10.25h   | 10.01bcd  | 45.09c-g  | 25.33c      | 105.83    |
| Bamb5          | 20a-d     | 11.77abc  | 12.62c   | 10.07bcd  | 43.86c-g  | 31.95 bc    | 89.83     |
| Bamb6          | 19.57cde  | 10.03dc   | 10.9gh   | 9.23d     | 59.31a    | 43.25 bc    | 64        |
| Bamb7          | 19.9b-e   | 11.85abc  | 13.65ab  | 11.85a    | 48.21c-g  | 41.28 bc    | 138.33    |
| Bamb8          | 20.52a-d  | 11.93abc  | 12.75bc  | 10.18bc   | 48.83b-f  | 71.64a      | 141.3     |
| Bamb9          | 20.15a-d  | 12.15abc  | 11.23fgh | 10.53b    | 51.86abc  | 45.6 bc     | 79        |
| Bamb10         | 20.45a-d  | 12.35ab   | 11.27e-h | 9.9bcd    | 51.25a-d  | 41.67 bc    | 95.5      |
| Bamb11         | 21.2abc   | 12.75a    | 12.28cde | 10.07bcd  | 43.94c-g  | 44.12 bc    | 83.17     |
| Bamb12         | 18.27e    | 11.9abc   | 12.38c   | 9.82bcd   | 43.8c-g   | 37.98 bc    | 67.33     |
| Bamb13         | 20.42a-e  | 11.77abc  | 11.93c-f | 9.65bcd   | 49.75a-f  | 45.56 bc    | 78.33     |
| Bamb14         | 19.23de   | 12.63a    | 11.88c-g | 10.07bcd  | 44.87c-g  | 48.97ab     | 69.5      |
| Bamb15         | 19.95bcd  | 12.57ab   | 12.25c-f | 10.38bc   | 50.45a-e  | 32.1 bc     | 49.67     |
| Bamb16         | 20.3a-d   | 12.42ab   | 12.67bc  | 10.22bc   | 40.65     | 33.79 bc    | 66.33     |
| Bamb17         | 19.88b-e  | 10.15d    | 12.12c-f | 10.07bcd  | 58.35b    | 36.49 bc    | 95.33     |
| Bamb18         | 19.17a-d  | 12.28abc  | 11.75c-g | 9.57cd    | 43.14c-g  | 36.69 bc    | 99        |
| Bamb19         | 18.97de   | 11.32bcd  | 11.93c-f | 10.25bc   | 39.87     | 30.95bc     | 63.5      |
| Bamb20         | 20.38a-d  | 12.33ab   | 11.3d-g  | 9.63bcd   | 41.27     | 31.88bc     | 44.83     |
| <b>P value</b> | <b>**</b> | <b>**</b> | <b>*</b> | <b>**</b> | <b>**</b> | <b>****</b> | <b>ns</b> |
| LSD            | 0.02      | 0.02      | 0.01     | 0.01      | 0.02      | 0.07        | 0.3       |
| CV             | 7.2       | 9.15      | 7.35     | 7.79      | 18.77     | 50.84       | 69.81     |
| GM             | 16.4      | 7.65      | 20.59    | 23.11     | 279.779   | 840.7       | 3737.18   |

Note: The different letters shows the existence of variation.

CV, coefficient of variation; LSD, least significant difference; PodL\_mm, Pod length; PodW\_mm, Pod width; SL, Seed length; SW, Seed width; HSW, 100 Seed weight; YPP, mass of seeds per plant; NPP, number of pods per plant.

\*\*\*, highly significant at  $p \leq 0.001$ ; \*\*, highly significant at  $p \leq 0.05$ ; \*, significant at  $p \leq 0.05$ .

Genetic advance (GA) was estimated as per the formula given by Khan et al. (2020):

$$GA = \frac{\sqrt{\delta^2 g \times \delta^2 p}}{\delta^2 p} \quad [\text{Eqn 8}],$$

where:

GA-expected genetic advance

$\delta^2 p$  = phenotypic variance

$\delta^2 g$  = genotypic variance

$K$  = the constant selection differential at 5% selection intensity ( $k = 2.063$ )

## Ethical considerations

This article followed all ethical standards for research without direct contact with human or animal subjects.

## Results

### Variation in vegetative traits and some yield related traits across test sites of Bambara groundnut

The ANOVA showed that there was significant variation among the tested Bambara groundnut landraces for traits recorded (Table 3 and Table 4). Results on vegetative traits revealed that Bamb3 had the highest number of days to 50% flowering at 84.2, whereas Bamb6 had the shortest number of days to 50% flowering at 75 (Table 5). Bamb4 had the highest number of nodes per stems at 25, whereas Bamb16 had the least at 14.8. Bamb13 had the highest number of

branches at 16.3, whereas Bamb16 had the lowest at 9.7. Bamb20 had the longest days to reach maturity at 175.8, whereas Bamb11 had the shortest at 165.8. Bamb2 had the highest chlorophyll content of 53.6 g/mL, whereas Bamb20 had the least chlorophyll content at 28.8 g/mL. Bamb20 also recorded the longest leaflets at 15.6 mm. Bamb2 had the shortest leaflet at 12.3 mm, value of Bamb6 had the tallest plant with a value of 24.6 mm, whereas Bamb11 had the shortest plant at 14.3 mm. Variation was also observed in yield and yield-related traits (Table 4), where Bamb4 recorded the tallest pod length at 21.7 mm whereas Bamb12 recorded 18.3 mm. Bamb14 had the widest pod width at 12.6 mm, while Bamb17 had a pod width of 10.2 mm. Bamb6 had the longest seed length at 14.3 mm, whereas Bamb4 had the shortest at 10.3 mm. Bamb7 had the biggest seed width at 11.8 mm, while Bamb11 had the smallest at 10.5 mm. Bamb6 had the heaviest 100 seed weight per plot at 59.3 g, whereas Bamb19 had the lightest at 39.9 g. Bamb8 had the highest seed yield per plant at 71.6 g and the highest number of pods per plant at 141.3, whereas Bamb3 had the least yield per plant and Bamb20 had the lowest number of pods per plant at 44.

### Correlation analysis

Pearson's correlation coefficients showed interrelationships among the traits recorded (Table 5). It was observed that a number of branches per stem had a significant positive correlation with plant height ( $r = 0.50$ ) and seed length, and seed width ( $r = 0.50$ ). Moreover, a highly significant positive correlation was observed between chlorophyll content and

**TABLE 5:** Pearson correlation for all pair-wise comparisons of vegetative and productive growth-related traits.

|       | DTF          | NNS          | NBS           | DM      | PodL  | PodW           | SL            | SW           | HSW   | YPP   | CHL            | TLL            | TLW            | LArea        | PH   | SC   | NPP  |
|-------|--------------|--------------|---------------|---------|-------|----------------|---------------|--------------|-------|-------|----------------|----------------|----------------|--------------|------|------|------|
| DTF   | 1.00         | -            | -             | -       | -     | -              | -             | -            | -     | -     | -              | -              | -              | -            | -    | -    | -    |
| NNS   | -0.09        | 1.00         | -             | -       | -     | -              | -             | -            | -     | -     | -              | -              | -              | -            | -    | -    | -    |
| NBS   | -0.07        | 0.33         | 1.00          | -       | -     | -              | -             | -            | -     | -     | -              | -              | -              | -            | -    | -    | -    |
| DM    | -0.00        | -0.04        | -0.19         | 1.00    | -     | -              | -             | -            | -     | -     | -              | -              | -              | -            | -    | -    | -    |
| PodL  | <b>0.40*</b> | 0.16         | 0.30          | -0.23   | 1.00  | -              | -             | -            | -     | -     | -              | -              | -              | -            | -    | -    | -    |
| PodW  | 0.25         | -0.02        | -0.26         | 0.21    | 0.26  | 1.00           | -             | -            | -     | -     | -              | -              | -              | -            | -    | -    | -    |
| SL    | -0.19        | -0.24        | -0.13         | -0.24   | -0.36 | 0.12           | 1.00          | -            | -     | -     | -              | -              | -              | -            | -    | -    | -    |
| SW    | -0.08        | -0.04        | 0.08          | -0.39   | 0.05  | 0.17           | <b>0.50**</b> | 1.00         | -     | -     | -              | -              | -              | -            | -    | -    | -    |
| HSW   | -0.35        | -0.24        | 0.29          | -0.40   | -0.04 | <b>-0.66**</b> | -0.13         | -0.03        | 1.00  | -     | -              | -              | -              | -            | -    | -    | -    |
| YPP   | -0.38        | -0.34        | 0.22          | -0.40   | -0.13 | -0.03          | 0.37          | 0.07         | 0.35  | 1.00  | -              | -              | -              | -            | -    | -    | -    |
| CHL   | -0.11        | 0.30         | 0.36          | -0.33   | 0.23  | 0.12           | <b>0.45*</b>  | <b>0.41*</b> | -0.01 | 0.34  | 1.00           | -              | -              | -            | -    | -    | -    |
| TLL   | 0.03         | 0.19         | 0.04          | -0.03   | -0.06 | -0.06          | -0.35         | -0.15        | -0.02 | 0.13  | -0.035         | 1.00           | -              | -            | -    | -    | -    |
| TLW   | 0.14         | <b>0.39</b>  | 0.24          | -0.27   | 0.19  | -0.26          | -0.35         | -0.26        | -0.02 | -0.11 | -0.23          | 0.42           | 1.00           | -            | -    | -    | -    |
| LArea | 0.01         | <b>0.44*</b> | 0.27          | -0.25   | 0.05  | -0.29          | -0.29         | -0.16        | -0.03 | 0.02  | -0.08          | <b>0.71***</b> | <b>0.89***</b> | 1.00         | -    | -    | -    |
| PH    | -0.21        | 0.32         | <b>0.50**</b> | -0.13   | -0.02 | -0.39          | -0.01         | 0.02         | 0.13  | -0.15 | 0.19           | 0.10           | <b>0.40*</b>   | <b>0.40*</b> | 1.00 | -    | -    |
| SC    | -0.21        | <b>0.40*</b> | 0.16          | -0.10   | 0.17  | 0.05           | -0.04         | 0.32         | -0.01 | -0.09 | 0.33           | -0.04          | 0.09           | 0.16         | 0.09 | 1.00 | -    |
| NPP   | 0.15         | 0.26         | 0.41*         | -0.51** | 0.34  | 0.01           | 0.23          | <b>0.43*</b> | 0.04  | 0.32  | <b>0.74***</b> | -0.04          | -0.15          | -0.04        | 0.03 | 0.32 | 1.00 |

Note: The bold values indicate the significance values.

DTF, Days to 50% flowering; NNS, Number of nodes per stem; NBP, Number of branches per plant; DM, Days to maturity; PodL\_mm, Pod length; PodWmm, Pod width; SL, Seed length; SW, Seed width; HSW, 100 Seed weight per plot; YPP, seed yield per plant (grams); CHL, Chlorophyll content; TLL, Terminal length; TLW, Terminal leaflet width; LArea, leaf area; PH, Plant height; SC, Stand count per plot; NPP, number of seeds per plot.

\*, highly significant at 0.001; \*\*, highly significant at 0.01; \*\*\* significant at 0.5 probability levels.

the number of pods per plant ( $r = 0.74$ ), terminal leaflet length and leaf area ( $r = 0.71$ ) and terminal leaflet width and leaf area ( $r = 0.89$ ). There was a significant negative correlation between the date of maturity and number of pods per plant ( $r = -0.50$ ), pod width, and 100 seed weight ( $r = -0.66$ ). Furthermore, there was a positive correlation between days to 50% flowering and pod length ( $r = 0.40$ ). The number of nodes per stem had a positive correlation with leaf area ( $r = 0.44$ ), terminal leaflet width ( $r = 0.39$ ) and stand count ( $r = 0.40$ ). The number of branches per stem also showed a significantly positive correlation with the number of pods per plant ( $r = 0.41$ ). Positive correlations were observed for seed length with chlorophyll content ( $r = 0.45$ ), seed width with chlorophyll ( $r = 0.41$ ), and number of pods per plant ( $r = 0.43$ ), 100 seeds weight with yield per plant ( $r = 0.35$ ), terminal leaflet width with plant height ( $r = 0.40$ ), and finally on leaf area and plant height ( $r = 0.40$ ). A set of negative correlation was also observed on days to 50% flowering with 100 seed weight ( $r = -0.3365$ ) and yield per plant ( $r = -0.38$ ), date of maturity with seed width ( $r = -0.39$ ), 100 seed weight ( $r = -0.40$ ), and yield per plant ( $r = -0.40$ ), pod length with seed length ( $r = -0.36$ ), pod width with plant height ( $r = -0.39$ ), seed length with terminal leaflet width ( $r = -0.35$ ), and finally on chlorophyll with terminal leaflet length ( $r = -0.35$ ).

## Principal component analysis

The analysis performed on the 17 traits across the two locations revealed that the first six principal components (PCs) with Eigen values > 1.00 accounted for 78.34% (Table 6) of the variability. Component loadings higher than +0.30 were regarded as significant. The first PC accounted for 20.74% variability and most of the traits that grouped under this component were vegetative traits (Table 6), including the number of nodes per stem, number of branches per stem, plant height, terminal leaflet width, and leaf area. The second

**TABLE 6:** Principal component analysis of 17 quantitative characters in Bambara groundnut landraces.

| Variables       | PC1         | PC2         | PC3          | PC4          | PC5          | PC6          |
|-----------------|-------------|-------------|--------------|--------------|--------------|--------------|
| DTF             | -0.04       | -0.11       | <b>0.38</b>  | <b>-0.44</b> | -0.02        | -0.36        |
| NNS             | <b>0.32</b> | -0.04       | 0.28         | 0.35         | -0.06        | 0.14         |
| NBS             | <b>0.36</b> | 0.13        | -0.02        | -0.04        | -0.29        | 0.07         |
| DM              | -0.25       | -0.27       | 0.11         | <b>0.32</b>  | -0.13        | 0.28         |
| PodL            | 0.16        | 0.05        | <b>0.37</b>  | <b>-0.40</b> | -0.28        | 0.09         |
| PodW            | -0.22       | 0.07        | <b>0.43</b>  | -0.03        | 0.27         | 0.17         |
| SL              | -0.15       | <b>0.36</b> | -0.11        | 0.20         | 0.29         | <b>-0.39</b> |
| SW              | 0.01        | <b>0.37</b> | 0.10         | 0.11         | 0.11         | -0.29        |
| HSW             | 0.14        | 0.09        | <b>-0.47</b> | -0.23        | <b>-0.31</b> | 0.14         |
| YPP             | 0.04        | 0.28        | -0.32        | -0.25        | <b>0.34</b>  | <b>0.32</b>  |
| CHL             | 0.16        | <b>0.43</b> | 0.14         | 0.10         | 0.05         | 0.12         |
| TLL             | 0.24        | -0.20       | -0.02        | -0.09        | 0.49         | 0.27         |
| TLW             | <b>0.39</b> | -0.27       | 0.03         | -0.08        | 0.16         | -0.23        |
| LArea           | <b>0.42</b> | -0.22       | -0.01        | 0.01         | <b>0.35</b>  | -0.07        |
| PH              | <b>0.32</b> | -0.03       | -0.11        | <b>0.30</b>  | -0.18        | <b>-0.37</b> |
| SC              | 0.19        | 0.16        | 0.19         | <b>0.32</b>  | -0.09        | 0.29         |
| NPP             | 0.20        | <b>0.41</b> | 0.18         | -0.17        | -0.02        | 0.08         |
| Eigenvalue      | 3.53        | 3.33        | 2.39         | 1.57         | 1.44         | 1.07         |
| Variability (%) | 20.74       | 19.58       | 14.07        | 9.2          | 8.48         | 6.27         |
| Cumulative (%)  | 20.74       | 40.32       | 54.39        | 63.59        | 62.07        | 78.35        |

Note: The bold values indicated that those traits plays major role in contributing to the variation among the traits in different PCs. Eigenvector values > 0.3 are deemed to be significantly correlated.

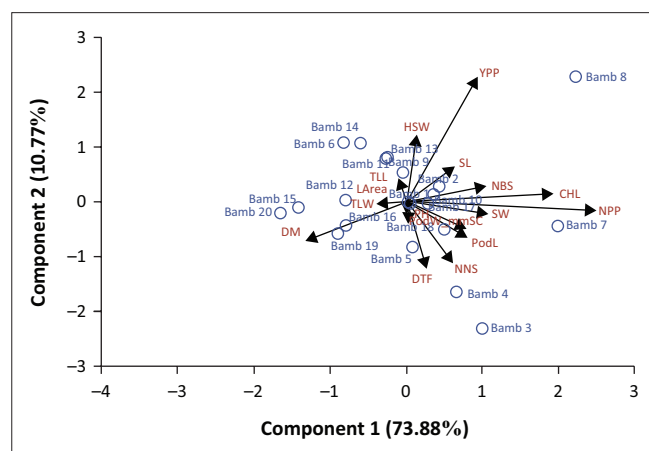
DTF, Days to 50% flowering; NNS, Number of nodes per stem; NBS, Number of branches per plant; DM, Days to maturity; PodL, Pod length; PodW, Pod width; SL, Seed length; SW, Seed width; HSW, 100 Seed weight; YPP, seed yield per plant; CHL, Chlorophyll content; TLL, Terminal length; TLW, Terminal leaf width; LArea, leaflet area; PH, Plant height; SC, Stand count; NPP, number of pods per plant; PC, principal component.

PC accounted for about 19.58% of the total variability and variables found under this component were yield and vegetative traits, namely, seed length, seed width, number of pods per plant, and chlorophyll content. The third PC accounted for 14.07%, which comprised the following yield components: days to 50% flowering, pod width, and pod length. The fourth PC accounted for 9.2% of the total variation, comprising vegetative components such as the number of nodes per stem, date of maturity, plant height, and stand count. The fifth PC accounted for 8.48% of the total

variability comprising both vegetative and yield components, namely, leaf area, seed mass per plant, and terminal leaflet length. The sixth PC accounted for about 6.27% of the total variation, comprising a yield component, namely, the mass of seeds per plant. Finally, the analysis also showed negative loadings above 0.3 which were regarded as variables with negative influence on the PCs, namely, 100 seed weight (third principal); days to 50% flowering, pod length, (fourth principal); 100 seed weight (fifth principal); and days to 50% flowering, seed length, plant height (sixth principal).

### Principal component biplot

The variation observed among the Bambara groundnut landraces used in this study is depicted in the PCA biplot (Figure 1). The landraces were divided into four quadrants, and accounted for 84.65% of the variation for PC1 and PC2, representing 73.88% and 10.77%, respectively. It was observed that landraces Bamb5, Bamb16, Bamb19, and Bamb20 were in the first quadrant, where a strong positive association was observed between terminal leaflet width and date of maturity. Furthermore, Bamb3, Bamb4, Bamb7, Bamb17, and Bamb18 were found in the second quadrant, where a strong positive correlation was observed between plant height and pod width, days to 50% flowering, number of nodes per stem and stand count, and pod length. Plant height and pod width had a strong influence on PC2, whereas a number of pods per plant and chlorophyll had a strong effect on PC1. Bamb1, Bamb2, Bamb10, and Bamb17 were positioned at the centre of the quadrant and were stable accessions for the tested traits across seasons and environments, whereas Bamb3, Bamb4, Bamb7 and Bamb8 were distinct accessions that were located far from the origin. These accessions have peculiar genes and/or alleles that separated them from the test accessions. Hence, Bamb3 and Bamb4 were positively associated with the NNs, Bamb7 and Bamb8 were positively associated with NPP and YPP, respectively. Terminal leaflet length and 100 seed weight



Note: Blue marker indicates genotypes, and red markers indicate traits.

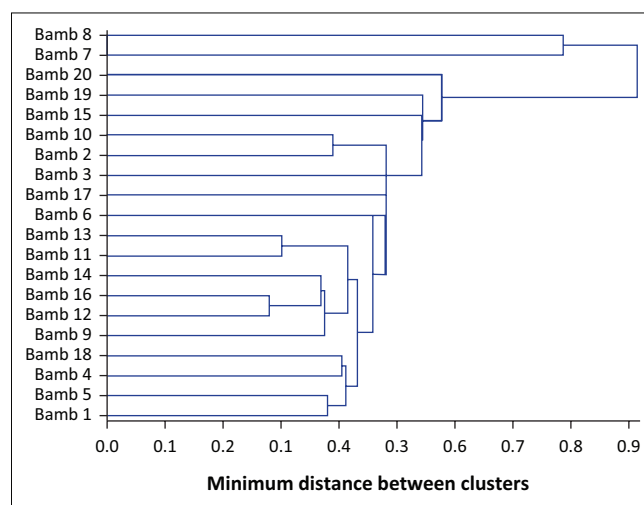
DTF, Days to 50% flowering; NNS, Number of nodes per stem; NBP, Number of branches per plant; DM, Days to maturity; PodL, Pod length; PodW, Pod width; SL, Seed length; SW, Seed width; HSW, 100 Seed weight; YPP, mass of seeds per plant; CHL, Chlorophyll content; TLL, Terminal length; TLW, Terminal leaflet width; LArea, leaflet area; PH, Plant height; SC, Stand count per plot; NPP, number of seeds pods per plot.

**FIGURE 1:** Multidimensional preference analysis of PC1 and PC2 describing the overall variation among Bambara groundnut landraces and agronomic traits.

showed a strong influence on PC2. Finally, the genotypes Bamb6, Bamb9, Bamb11, Bamb12, Bamb13, Bamb14 and Bamb15 were found on the last quadrant where the correlation was observed between terminal leaflet length and leaf area. Bamb3 and Bamb7 had a strong but negative influence on PC2 and the characters that contributed to that influence were mass of seeds per plant, days to 50% flowering and number of nodes per stem. Bamb8 and Bamb20 showed a strong but negative influence on both PC1, and the characters that contributed to that influence were number of pods per plant and date of maturity.

### Cluster analysis

Hierarchical cluster analysis of landraces, based on the similarities of traits, was conducted using the complete linkage method and a dendrogram was constructed (Figure 2). The cluster analysis results corroborate those of the PC biplot analysis (Figure 1). The dendrogram was divided into five major clusters with approximately 0.8 genetic similarities. The dendrogram grouped landraces with similarities as they appear in Table 3, where it was noted that Bamb1, Bamb4, Bamb5, and Bamb18 occurred in the same quadrant in the biplot and are grouped. Their association was due to the following traits: plant height, pod width, pod length, number of nodes per stem, and days to 50% flowering. Another similarity was observed with Bamb9, Bamb11, Bamb12, Bamb13, and Bamb14, which were associated with leaf area, terminal, leaflet length, and leaflet width. Furthermore, few dissimilarities were observed in certain landraces which were noticed to be further away from the other landraces. Bamb3 dissimilarities were due to the number of nodes per stem and days to 50% flowering, and Bamb7 dissimilarities were due to a number of pods per plant and chlorophyll content, while Bamb8's dissimilarities were due to mass of seeds per plant, and finally Bamb20's dissimilarities were due to date of maturity.



DTF, Days to 50% flowering; NNS, Number of nodes per stem; NBS, Number of branches per stem; DM, Days to maturity; PodL, Pod length; PodW, Pod width; SL, Seed length; SW, Seed width; HSW, 100 Seed weight; YPP, mass of seeds per plant; CHL, Chlorophyll content; TLL, Terminal length; TLW, Terminal leaflet width; LArea, leaflet area; PH, Plant height; SC, Stand count; NPP, number of pods per plant.

**FIGURE 2:** Hierarchical cluster analysis showing similarities among Bambara groundnut landraces.

## Genetic parameters

The investigation of variances (genotypic and phenotypic), the genotypic coefficient of variance and phenotypic coefficient of variance (PCV), broad sense heritability, and

**TABLE 7:** Genetic parameters for agronomic traits of Bambara landraces.

| Variables | $\sigma^2$ | $\sigma^2p$ | $\sigma^2e$ | GM     | GCV(%) | PCV   | $h^2$  | GA      |
|-----------|------------|-------------|-------------|--------|--------|-------|--------|---------|
| DTF       | -168.77    | 624.90      | 21.95       | 77.43  | -      | 32.29 | -27.01 | -17.99  |
| NNS       | 22.69      | 70.78       | 17.29       | 19.81  | 24.05  | 42.47 | 32.06  | 28.08   |
| NBS       | 12.97      | 35.70       | 10.04       | 13.36  | 26.95  | 44.72 | 36.32  | 33.51   |
| DM        | 45.05      | 101.23      | 31.49       | 170.87 | 3.93   | 5.89  | 44.51  | 5.41    |
| PodL      | 1.28       | 12.56       | 2.08        | 20.03  | 5.64   | 17.69 | 10.17  | 3.71    |
| PodW      | 1.10       | 6.30        | 1.21        | 12.01  | 8.31   | 20.90 | 15.80  | 6.81    |
| SL        | 1.99       | 15.58       | 0.78        | 12.04  | 11.71  | 33.06 | 12.55  | 8.56    |
| SW        | -2.25      | 13.26       | 0.62        | 10.07  | -      | 36.16 | -16.98 | -12.67  |
| HSW       | 145.90     | 345.12      | 76.22       | 46.52  | 25.97  | 39.93 | 42.28  | 34.83   |
| YPP       | 506.86     | 1134.16     | 400.96      | 39.39  | 57.16  | 85.60 | 44.69  | 78.83   |
| CHL       | 262.28     | 458.41      | 158.23      | 40.7   | 39.79  | 52.61 | 57.22  | 62.09   |
| TLL       | 60.57      | 158.02      | 3.31        | 14.21  | 54.78  | 88.46 | 38.33  | 69.95   |
| TLW       | 0.93       | 3.66        | 1.68        | 6.66   | 14.47  | 28.71 | 25.40  | 15.04   |
| LArea     | -471.01    | 3328.85     | 353.09      | 72.98  | -      | 79.06 | -14.15 | -23.08  |
| PH        | -21.05     | 105.86      | 3.78        | 21.87  | -      | 47.04 | -19.88 | -19.30  |
| SC        | -62.24     | 254.69      | 13.77       | 17.45  | -      | 91.46 | -24.44 | -46.11  |
| NPP       | 3397.93    | 6498.03     | 3651.87     | 86.57  | 67.33  | 93.12 | 52.29  | 5100.45 |

$\sigma^2g$ , genotypic variance;  $\sigma^2p$ , phenotypic variance;  $\sigma^2e$ , Environmental variance; GM, Grand mean; GCV, Genotypic coefficient of variation; PCV, Phenotypic coefficient of variation;  $h^2$ , Broad sense heritability; GA, Genetic advance; Variables-agronomic traits; DTF, Days to 50% flowering; NNS, Number of nodes per stem; NBS, Number of branches per stem; DM, Days to maturity; PodLmm, Pod length; PodW, Pod width; SL, Seed length; SW, Seed width; HSW, 100 Seed weight; YPP, yield per plant; CHL, Chlorophyll content; TLL, Terminal length; TLW, Terminal leaflet width; LArea, leaflet area; PH, Plant height; SC, Stand count; NPP, number of pods per plot.

genetic advance are presented in Table 7. The results show that the genotypic variance reflects on phenotypic variance because relatively high values of both genotypic and phenotypic variance were obtained on 100 seed weight, yield per plant, chlorophyll content, and number of pods per plant. The results indicate that genotypic variance varied from 0.93 for terminal leaflet width to 3397.3 for a number of pods per plant. For phenotypic variance, the least value was also found on the terminal leaf width (3.66) and the top most being on the number of pods per plot (6498.03). The results further showed a greater variation in the PCV, ranging from 3.93% (date of maturity) to 67.33% for number of pods per plant. The highest values of 67.33% and 93.12% were recorded for GCV and PCV in the number of pods per plot. The following traits having values > 20% for GCV and PCV are considered high and those include number of nodes per stem, number of branches per stem, 100 seed weight, yield per plant, chlorophyll, terminal leaflet length, and number of pods per plant. Seed length and terminal leaflet width had medium PCV and GCV values (10% – 20%). Typically, the estimated values of heritability in the broad sense were high ( $h^2b > 30$ ) for almost all traits evaluated (Table 8). The heritability values ranged from 10.17% (pod length) to 57.22% (chlorophyll content). Moderate values ( $30\% < h^2b < 60\%$ ) were obtained for the following traits: number of nodes per stem (32.06%), number of branches per stem (36.32%), date of maturity (44.51%), 100 seed weight (42.28%), yield per plant (44.69%), chlorophyll content (57.22%), and number of pods

**TABLE 8:** Classification of Bambara groundnut landraces based on qualitative morphological traits representing frequency and diversity index.

| Qualitative trait                         | Descriptor        | Number in sample | Frequency (%) | Diversity index $H'$ |
|-------------------------------------------|-------------------|------------------|---------------|----------------------|
| Terminal leaflet shape                    | Round             | 3                | 15            | 1.34                 |
|                                           | Oval              | 6                | 30            | -                    |
|                                           | Lanceolate        | 4                | 20            | -                    |
|                                           | Elliptic          | 7                | 35            | -                    |
| Flower colour                             | Yellow            | 20               | 100           | 0.00                 |
| Dark pigmentation on wings of leaflets    | Absent            | 17               | 85            | 0.42                 |
|                                           | Green             | 03               | 15            | -                    |
| Colour of fully expanded terminal leaflet | Green             | 16               | 80            | 0.50                 |
|                                           | Dark green        | 4                | 20            | -                    |
| Growth habit                              | Bunch type        | 05               | 25            | 0.86                 |
|                                           | Semi bunch type   | 02               | 65            | -                    |
|                                           | Spreading type    | 13               | 10            | -                    |
| Stem hairiness                            | Parse             | 9                | 45            | 0.69                 |
|                                           | Dense             | 11               | 55            | -                    |
| Seed shape                                | Oval              | 20               | 100           | 0.00                 |
| Seed colour                               | Cream             | 5                | 5             | 1.76                 |
|                                           | Brown with purple | 1                | 5             | -                    |
|                                           | Dots              | 3                | 15            | -                    |
|                                           | Black             | 2                | 10            | -                    |
|                                           | Brown             | 3                | 15            | -                    |
|                                           | Red               | 2                | 10            | -                    |
|                                           | Cream & purple    | 1                | 5             | -                    |
|                                           | Light purple      | 3                | 15            | -                    |
|                                           | Yellowish         | -                | -             | -                    |
| Pod colour                                | Purple            | 12               | 60            | 0.94                 |
|                                           | White             | 5                | 5             | -                    |
|                                           | Black             | 3                | 15            | -                    |
| Pod texture                               | Little groove     | 12               | 60            | 0.94                 |
|                                           | Smooth            | 5                | 5             | -                    |
|                                           | Much groove       | 3                | 15            | -                    |

per plant (52.29%). The genetic advance, as a percentage of the mean, ranged from 3.71% for pod length to 100.45% for number of pods per plant. The following traits, listed in their ascending order, had the high values of genetic advance (> 20%): number of nodes per stem (28.08), number of branches per stem (33.51%), hundred seed weight (34.83%), chlorophyll (62.09%), terminal leaflet length (69.95%), yield per plant (78.83%), and number of pods per plant (100.45%).

### Morphological qualitative traits

The leaflet shape of landraces differed significantly during plant growth. It was observed that most of the landraces (Table 8) produced elliptic leaflets (35%) followed by those with oval leaflets (30%), 20% had lanceolate leaflets, and 15% had round leaflets (Figure 3). The results also indicated that the colour of the flower and seed shape was 100%. That is, all the landraces studied produced yellow flowers and oval seed shapes (Figure 4 and Figure 5). A high percentage was also observed on green fully expanded leaves (80%), with the

least being 20% (dark green fully expanded leaflets). With regards to the dark pigmentation on leaves, it was observed that most of the landraces produced leaves with zero pigments (85%) and only a few landraces had dark pigmentation (15%). There was variation in vegetative growth with three different types of growth habit; namely, bunch type, semi-bunch, and spreading type (Figure 5). Among these types, it was observed that semi-bunch type was the most common (65%) followed by the bunch type (25%), and the spreading type was less frequent (10%) (Table 8). It was observed that seed colour (Figure 4) varied significantly, producing eight different colours with different frequencies; namely in their descending order: black, red and yellowish (15%), cream with purple and brown (10%) and cream, light purple, and brown with purple dots (5%). In addition, pod colour and texture were recorded, where purple little grooves recorded the highest frequency of (60%) followed by black much grooved (15%) and the least was observed on yellowish-white pods (5%) (Figure 6). These results are not different from Kambou et al. (2020) who

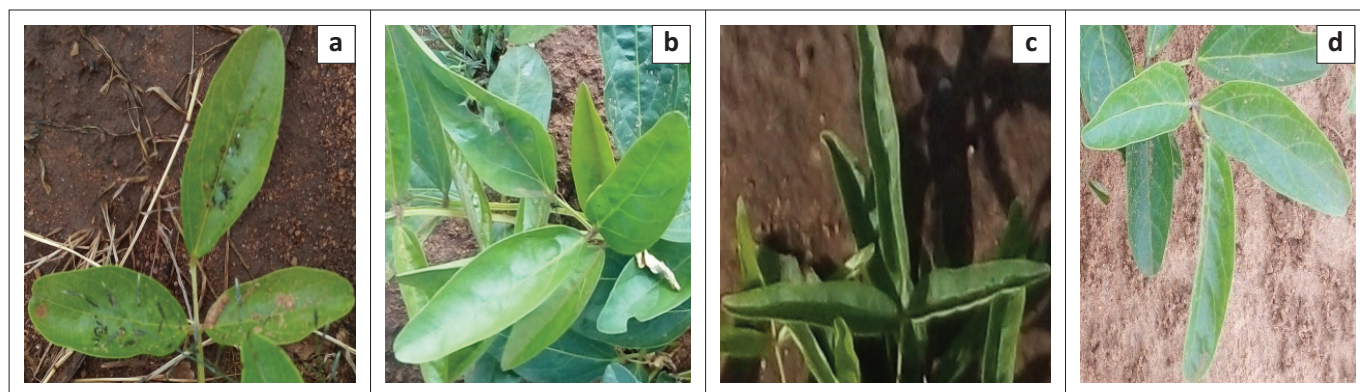
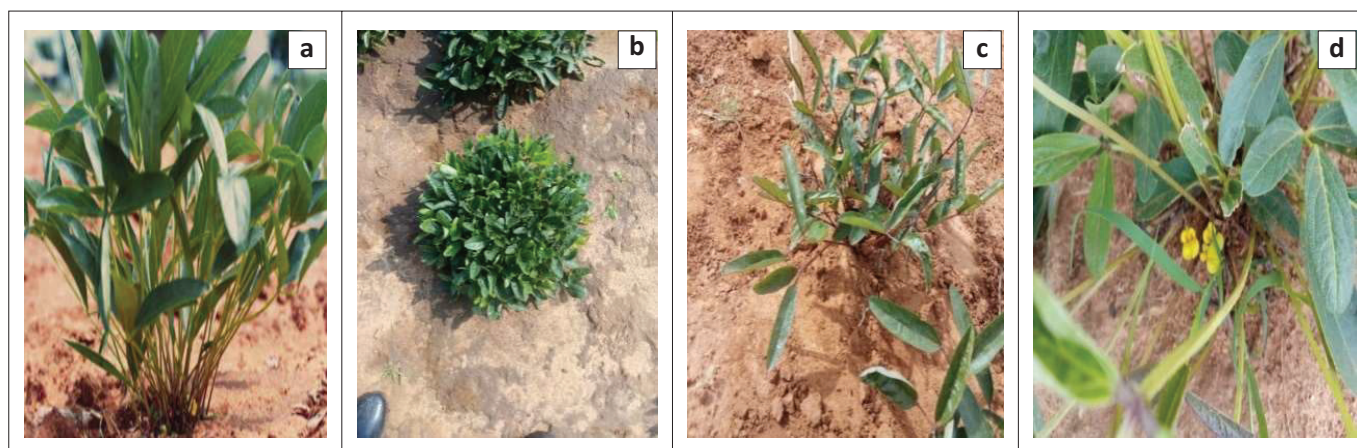


FIGURE 3: Variation in leaflet shapes: (a) round, (b) elliptic, (c) lanceolate, (d) oval.



FIGURE 4: Variation in colours of seeds: (a) Yellowish, (b) Cream and black, (c) light brown, (d) Black (e) red, (f) brown with purple dots, (g) purple cream, (h) cream with purple streaks.



**FIGURE 5:** Vegetative growth showing flower colour and different growth habit (a) yellow flowers, (b) bunch type, (c) spreading type and (d) semi-bunch.



**FIGURE 6:** Pods colour and texture: (a) purple wrinkled, (b) white smooth, (c) black more groove.

also recorded a high frequency of smooth texture on pods followed by smooth textured pods, which were dominant (Figure 6).

### Shannon-Weaver diversity index

The Shannon Weaver index was relatively low (Table 8) for the 10 qualitative traits that were evaluated. The index ranged between 0.01 (leaf colour, seed shape, and flower shape) and 1.76 (seed colour). Compared to the rest of the qualitative traits studied, seed colour was the most polymorphic, followed by terminal leaflet shape (1.34).

## Discussion

### Agro-morphological variability

The mean days to 50% flowering recorded in this study revealed significant variation among the tested Bambara groundnut landraces (Table 3). In the current study, it was observed that days to 50% flowering ranged from 75 to 84 days, which was higher than the range (51–65) that was reported by Abu and Buah (2011); Unigwe et al. (2016) on the characterisation of Bambara groundnut landraces and their evaluation by farmers in the Upper West Region of Ghana; morphological evaluation of selected accessions in South Africa, respectively, which also reported the lowest values

for days to 50% flowering of 41–67 days, respectively. Such variation could be caused by several environmental factors such as temperature, altitude and soil conditions as well as spatial and temporal genotypic factors affecting flowering in Bambara groundnut. According to Unigwe et al. (2016), similar factors may be responsible for the variation in days to flowering. Number of branches per stem ranged from 9.67 to 16.3 (Table 4), which was higher than the values of 10.9–12.3 reported by Effa et al. (2016). This might be due to the presence of variation in the growing environments (Gerrano et al. 2013). One hundred seed weight ranged from 38.7 g to 59.3 g, with an average of 51.8 g. This variation is possibly due to different seed sizes for different landraces and this corroborates the findings by Valombola et al. (2019). Plant height ranged from 19.3 mm to 24.7 mm with an average of 23.2 mm, this was shorter than the range of height 26 mm – 27 mm, and 22.5 mm – 23.8 mm recorded by Effa et al. (2016) on fertilised landraces in south eastern Nigeria which were not significant and were shorter than the accessions selected in a study by Onwubiko, Uguru and Chimdi (2019). The possible explanation for this variation is that no fertiliser was used in the current study (Gerrano et al. 2015). Variations in seed length and seed width may be due to inherent differences in seed size and shape (Gerrano et al. 2017). Landraces showed that pod length and pod width ranged from 18.2 mm to 21.6 mm and 10.1 mm – 12.7 mm,

which is similar to the Bambara accessions from Burkina Faso as reported by Kambou et al. (2020) whose findings were slightly higher than those found in the current study. The yield per plant ranged from 22.8 g to 79 g, which was much less than the findings recorded by Kambou et al. (2020) (100–378 g) and Massawe et al. (2005), which ranged from 84 g to 112.7 g. However, the findings of the current study showed that yield per plant was higher than the results that were recorded by Abedije et al. (2018), which ranged from 13 g to 51 g. These variations could be caused by several factors including environmental features, different seed sizes and the number of pods produced per plant and even the location in which they are planted. Similarly, Gerrano et al. (2013) and Siwale et al. (2022) reported the existence of phenotypic variation among diverse Bambara groundnut accessions in South Africa. Moreover, a study that was conducted in Nigeria also reported the presence of phenotypic variation among tested Bambara groundnut landraces (Esan, Oke & Ogunbode 2023). Zongo et al. (2023) also reported significant differences among the Bambara groundnut genotypes in Niger.

### Correlation analysis

Correlation analysis is important in plant breeding because it measures the degree of genetic and non-genetic association between traits. In this study, the correlation coefficient suggested that the agromorphological traits were positively correlated (Table 5). It was observed that the number of branches per and plant height had a significant correlation ( $r = 0.50$ ), which suggests that selecting plants with a high number of branches may lead to genotypes with longer internodes, and eventually the number of leaves will also be higher, resulting in higher yields. Moreover, a highly significant and positive correlation was observed between chlorophyll content and the number of pods per plant ( $r = 0.74$ ). This simply means that landraces with high chlorophyll content should be considered for selection. This is because the higher the chlorophyll content, the more fruits will be produced. Also, highly significant correlations were observed between terminal leaflet length with leaf area and terminal leaflet width ( $r = 0.71$ ) and leaf area ( $r = 0.89$ ). These results agree with those of Gerrano, Jansen van Rensburg and Adebola (2013) who also recorded a positive correlation between these traits, and attributed their results to the relationship between leaf length and leaf width. The study also showed a negatively significant correlation between the date of maturity and the number of pods per plant ( $r = -0.05$ ). Esan et al. (2023) found significant positive correlations for most of the traits evaluated in Nigerian landraces.

### Principal component analysis

The results of the PCA of the 17 quantitative morphological traits measured are presented in Table 6. The first six PCs contributed 78.35% of the variability among the 20

Bambara groundnut genotypes evaluated. Components with Eigenvalues  $>1$  accounted for 78.34% cumulative variance and component loadings with  $\pm 0.3$  were regarded significant (Kutcher, Ferguson & Cohen 2013) and their traits were measured for the effect each has on the total cumulative variability. The number of nodes per stem, number of branches per stem, terminal leaflet width, leaf area and plant height accounted for 20.74% of the cumulative variance, suggesting that they should be considered in the selection for good agronomic performance (Valombola et al. 2019). Days to 50% flowering, correlated positively with PC3 in the current study, but negatively with PC3 on accessions from North Central Namibia (Valombola et al. 2019). Terminal leaflet width correlated positively with PC1 in the current study, but negatively with genotypes that were evaluated for selection by Mohammed et al. (2019). Generally, the analysis of 17 traits showed that PC1 consisted of several traits that contributed to the greatest variation followed by PC2. Mohammed (2014) recorded similar results where they observed that seed traits like seed length and seed width correlated positively with PC2. Principal biplot analysis showed how strongly each trait is associated with the other. The biplot also showed the association of the traits to the genotypes (Table 3).

### Principal component biplot

The PC further explained agro-morphological similarities and variation among Bambara groundnut landraces relative to 17 measured traits (Figure 1). The quantitative traits led to the grouping of landraces into four quadrants representing 84.65% of the total variation. Bamb5, Bamb16, Bamb19, and Bamb20 were close to each other, implying that they share common traits among them. The shared traits are date of maturity, days to 50% flowering, plant height and terminal leaflet width. Again, landraces that are scattered far apart within the axes would mean that they are distantly related to other landraces within the same quadrant. Similar findings were recorded by Mohammed (2014). PC 1 and PC2 accumulated 73.88% and 10.77% of the total variation, respectively. This suggests that the first two principal components explained the large variations that existed among the accessions based on the extent of the traits contributing towards these variations.

### Cluster analysis

The results indicated that landraces were grouped based on the relationship they have among them (Figure 2). For instance, Bamb1, Bamb4, Bamb5, and Bamb18 were grouped in one cluster and it was noticed that even in the principal biplot they are found in the same quadrant. The same applied to landraces that were scattered far apart from others in the biplot, for example, like Bamb7, Bamb8, and Bamb20. This could be due to some unique traits that

are not shared between those landraces or that those landraces have a common origin in geographical representation and genetic background. These results agree with those recorded by Valombola et al. (2019) where it was found that genotypes grouped in one cluster had a common origin.

### Genetic parameters

According to Owusu et al. (2021), selection for trait improvement does not only depend on available genetic variation but also on the degree of heritability for such variation. The ANOVA between genotypic and phenotypic variance showed that phenotypic values were relatively higher than genotypic values for all traits (Table 7). This is an indication that traits were influenced by the environment. Khan et al. (2020) reported similar findings, where the GCV and PCV were categorised as low as (0% – 10%), intermediate (10% – 20%) and high (> 20%). Both PCV and GCV values in the current study were from medium to high for most of the traits. This indicates that genetic progress could be made on either of the traits with high values through selection (Owusu et al. 2021). The results further indicated that most of the traits had medium to strong heritability and genetic advance, except for traits such as days to 50% flowering, pod length, pod width, seed length, seed width, terminal leaflet width, leaf area, plant height, and stand count. According to Owusu et al. (2021), selection could be easier if heritability is higher than 70%.

### Shannon weaver diversity index

The results showed that, among the three types of growth habit, the semi-bunched morphotype was the most frequent followed by bunch type and the spreading type (Table 8). Similar observations were recorded by Ntundu et al. (2006) on landraces from Tanzania. Eighty percent of the landraces had a green fully expanded leaflet and 35% had elliptic leaflet shape. Bonny et al. (2019) and Khan et al. (2021) observed similar findings. The majority of pod textures had grooves (60%) and few smooth (5%). The Shannon weaver index indicated that seed colour ( $H' = 1.76$ ) and terminal leaflet shape ( $H' = 1.34$ ) were the most diversified traits out of 10 traits measured. These findings are not different from the findings by Zavinon et al. (2019).

### Conclusions

The assessment of morphological variability is the first step in the evaluation of genetic diversity for Bambara groundnut breeding programmes. The strong correlations observed between morphological and agronomic parameters make characters concerned important indices to be used in crop improvement programmes. The Bambara groundnut landraces in this study showed significant variation in phenotypic characters, indicating that the genotypes had high genetic diversity, which can be exploited for direct use

by small-scale and commercial farmers. Furthermore, considering the agronomic performances can be used in the development of new breeding lines. The PCA showed that most of the traits were positively associated, which would help the breeder for simultaneous crossing for the traits of interest. Biplot and dendrogram also agreed with the principal analysis by grouping the landraces based on the agro-morphological traits that they shared. It is therefore important to include genetically unrelated landraces in the breeding programmes in order to widen the genetic pool in the improvement programme. The study also indicated that the GCV, phenotypic coefficient variance, heritability, and genetic advance had medium to strong variability mostly for yield traits. The genetic potential of the landraces as revealed in this study can assist in choosing suitable parental lines, maximising the efficiency of Bambara groundnut-breeding programmes. Further study is needed to understand agronomic practices and nutritional characterisation in Bambara groundnut landraces among smallholder farmer, to realise the full genetic potential and nutritional value of Bambara groundnut cultivars.

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### Competing interests

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### Authors' contributions

S.E.X. was responsible for research trial establishment, data recording, investigation, methodology, formal analysis and original write-up of the article. A.S.G. contributed towards conceptualisation, investigation, methodology, funding acquisition, project fund administration, research trial establishment, data curation, supervision, review and editing. S.M. contributed to the conceptualisation, investigation, methodology, research trial establishment, data curation, formal analysis, supervision, review and editing.

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### Data availability

The authors confirm that the data supporting the findings of this study are available within the article.

## Disclaimer

The views and opinions expressed in this article are those of the authors and do not necessarily reflect the official policy or position of any affiliated agency of the authors.

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