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Mass selection of fonio landraces (*Digitaria exilis*) grown in Benin: Pathway, homogeneity assessment and genotypes screening

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Abstract

Fonio production is generally in polyvarietal mixture and no improved varieties are available yet. This work proposed a pathway for homogenization and mass selection from 19 accessions previously collected in Benin, to sort best genotypes. From 2015 to 2017, accessions were submitted to homogenization followed by separated conduct of different phenotypes appeared each year. Each material was evaluated on elementary plot of 3 m^2. Agro-morphological data collected were submitted to descriptive and multivariate analyses. Results showed homogeneity and selection effectiveness varied following characters and accessions. Better homogeneity level was obtained on qualitative characters compared to morphometric traits. Selection led to derivative accessions earlier or more productive than their parents. Thus, compared to parents, shortenings of 10%–30% on plant cycle and progress of 10%–70% in yield were recorded for some derivative accessions. On grains colour, 86% of accessions are homogeneous from 70% to 100%. Three groups characterized by earliness, productivity and grains homogeneity were identified. AS19-1-1, AS1, AS13-1, AS2-1-1, AS15-1-1 and AS18 were sorted genotypes. Multi-local assessments will be conducted for homogeneous and efficient ideotypes responding farmer's needs.

KEYWORDS

Benin | genetic progress | genotypes sorting | Homogenization | ideotypes | original & derivative accessions,

1 | INTRODUCTION

Fonio, one of the neglected and underutilized food species in West Africa, is a cereal of great importance to people due to its medicinal and taste qualities (Cruz et al., 2011). It plays a crucial role in the fight against food insecurity for many people, especially during lean season when early varieties enable to control famine (Dramé & Cruz, 2002; Sekloka et al., 2016).

In Benin, unlike other cereals such as maize, sorghum, pearl millet and rice grown worldwide, fonio appears to be essentially local crop even endemic to the North-West of Benin, in particular Atacora (Dansi et al., 2010; Sekloka et al., 2016). Fonio presents socio-cultural importance, particularly for the Otammari people, in the municipality of Boukombé, the main cropping area, which alone provides 74% of national production (Dramé & Cruz, 2002; Paraïso et al., 2016).

Despite its various uses, fonio has remained long neglected in national research programmes. Very little work is available on varietal breeding of this crop (Abdul & Jideani, 2019; Animasaun et al., 2018). In most parts of West Africa where this cereal is grown, there is no formal system for variety selection, production and

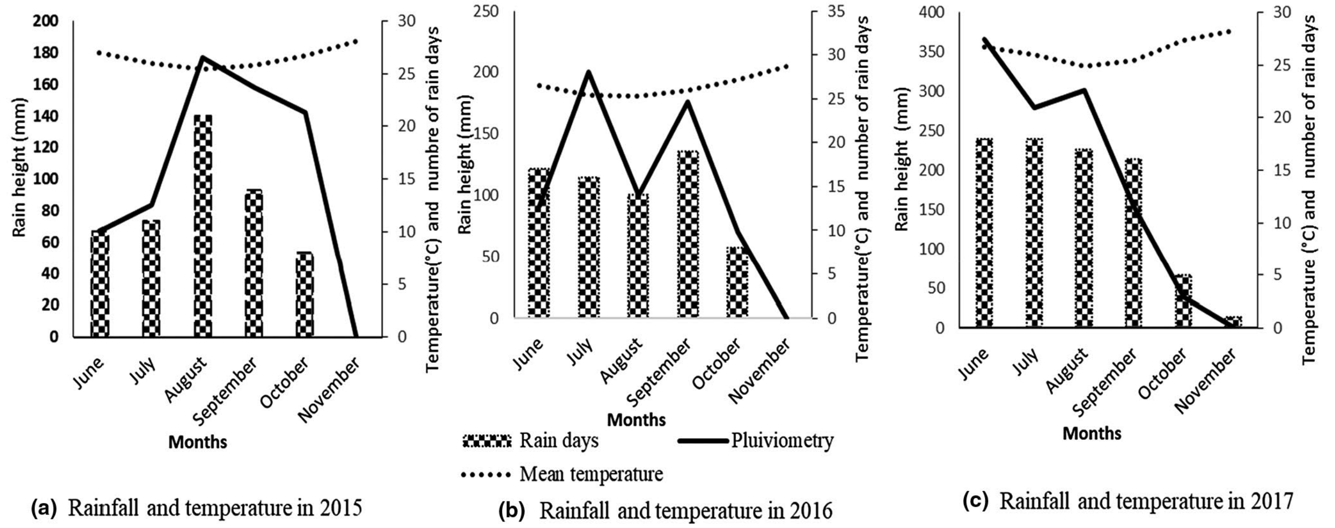

FIGURE 1 Ombrothermic diagram at Parakou during selection process from 2015 to 2017
Source: Data from Meteorology National Agency of Benin

distribution of fonio seeds (Sekloka et al., 2016). The floral organs are extremely tiny and, therefore, difficult to manipulate. Thus, fonio breeding by conventional hybridization encounters technical difficulties. However, breeding attempts have been made in Guinea, but have not yet been conducted to any published scientific results (Ayena et al., 2018; Vodouhè & Achigan Dako, 2006). Consequently, the grown varieties so far are traditional cultivars (landraces) evolving from farmer selection during many centuries of cultivation (Adoukonou-Sagbadja et al., 2007, 2010; Portères, 1976). Access to seeds is essentially traditional, mainly based on the collection from the previous harvest, on exchanges between farmers and sometimes on the seeds purchase at the market (Adoukonou-Sagbadja et al., 2006; Dansi et al., 2010; Sekloka et al., 2015). From the artificial selection of varieties, farming practices and traditional mode of seed management, it results that grown landraces of fonio are often in polyvarietal mixture, and therefore presents intra-varietal heterogeneity (Adoukonou-Sagbadja et al., 2010; Kwon-Ndung & Dachi, 2007; Sekloka et al., 2015).

Faced with these difficulties, scientific research is more called upon to develop homogeneous and performant varieties with regard to enormous potential that this minor cereal abounds. The homogeneity is a main desired criterion in selection given its importance in facilitating cultivation operations, agri-food processing, obtaining uniform products and even in official catalogue registration of new selected varieties (UPOV, 2019). Based on widely qualified mating systems of autogamy or apomixis revealed by molecular studies (Adoukonou-Sagbadja et al., 2010; Barnaud et al., 2017), it could be interesting to resort to homogenization and mass selection on heterogeneous traditional populations of fonio for obtaining homogeneous or improved genotypes. Thus, about 20 fonio accessions were collected in the main cropping area (Boukoumbé), and characterized from agronomic and morphological point of view (Sekloka et al., 2016). These studies revealed that within the same local accession, there were significant morphological and phenological differences, and therefore intra-accession heterogeneity. From 2015 to 2017, successive intra-varietal homogenization operations and mass selection were carried out within these accessions. This work led to the number of accessions which was 19 at the start, to more than 40 accessions.

This work presents the methodological selection approach, assesses the homogeneity of cultivars and selects the best genotypes for variety trials.

2 | MATERIALS AND METHODS

2.1 | Experiment site

The works were conducted during 2015, 2016 and 2017 years at the experiment station of the Faculty of Agronomy of University of Parakou located between 09′20.283 North latitude and 02′902 East longitude. Municipality of Parakou belongs to humid tropical area with Sudanese climate. Generally, it is characterized by alternation of one rainy season (May to October) and one dry season (November to April) (Kora & Guidibi, 2006).

On experiment field, the rains were regular and distributed during selection periods (Figure 1). The rainiest months were June, July, August and September. The cumulative rainfall was 627.8 mm; 638.1 mm and 1141.3 mm for 64, 74 and 75 days of rain, respectively, in the years 2015, 2016 and 2017 (Figure 1). Temperatures vary little with a peak of 28°C-29°C in November of each year.

Of tropical ferruginous type and poor in organic matter, the soil at the experimental site is composed of about 22.40% clay and silt, 1.43% total carbon and 0.167% total nitrogen, or C/N ratio of 8.56 (Azontondé et al., 2009).

2.2 | Plant material

The starting plant material used for the selection consisted of nineteen (19) fonio accessions previously collected in the commune of Boukombé (Benin) (Table 1). These accessions are local varieties theoretically adapted to the growing area.

2.3 | Homogenization and mass selection approach

For homogenization, each accession was evaluated on an elementary plot of 3 m^2. Each plot was individually observed during the different phenological stages of the plants and those which showed morphophenological differences compared to majority of the plants in the plot were isolated and individually harvested (Figure 2). They were then sown the following year on separated plots for observation. Thus, the starting accessions have been purified from one year to another by identification and isolation of the off-types.

Sometimes, the morphological differences at vegetative stage were less noticeable. In these cases, the colour differences at racemes level during maturation, were used to discriminate similar accessions on the vegetative level but intrinsically different (Figure 3).

At the end of each season, and within each plot, the ripe panicles of plants were harvested in homogeneous categories in different bags with labels to ensure the traceability. The harvests have been staggered in order to limit losses by seed shattering. For each homogeneous category, the harvested panicles were dried and ginned. The seeds, thus, obtained were dried again, winnowed, bagged and kept in the refrigerator.

Starting from the examples of two (02) original accessions, the methodology and stages of selection have been summarized as follow (Figure 4). Thus, from 19 original accessions (parents), 7 and 16 new accessions were, respectively, isolated in 2015 and 2016 (Table 2).

2.4 | Selection experiment design and husbandry

During the periods of homogenization, original accessions and those resulting from reselection (derivative accessions) were conducted and separately characterized. Each year, the experimental design consisted of elementary plots of 2 m long by 1.5 m wide (or 3 m^2) arranged in successive bands. Each plot housed an accession drawn at random. Nineteen (19), twenty-six (26) and forty-two (42) accessions were assessed in June of 2015, 2016 and 2017 respectively. Sowing was carried out in continuous rows in each plot of five rows spaced of 0.3 m from each other. For all years, the plots were maintained by manual weeding and hoeing. No fertilizer or pesticide was applied.

2.5 | Data collection

Qualitative and quantitative variables that had shown to be discriminating in previous characterization studies on fonio (Sekloka et al., 2016) were evaluated. Thus, the qualitative variables were

TABLE 1 Identification of collected original accessions

Accessions	Local names	Villages	District	Latitude	Longitude
AS1	Yoro	Mantchari	Dipoli	N 10°15.261′	E000°56.653′
AS2	Iporni	Koutchatahongou	Boukoumbé centre	N 10°09.502′	E001°05.308′
AS3	Iporda	Koumagou-B	Natta	N 10°11.910′	E001°08.028′
AS5	Lamba	Yatié	Tabota	N 10°24.957′	E001°04.119′
AS6	—	Dikoumini	Dipoli	N 10°31.311′	E000°94.941′
AS7	Ipormondé	Otanongou	Dipoli	N 10°33.157′	E000°96.798′
AS8	Ikantoni	Korontière	Korontière	N 10°15.580′	E000°59.513′
AS9	Iporgnirmè	Tassoyota	Korontière	N 10°31.674′	E000°99.290′
AS10	Iporawan	Boukoumbé centre	Boukoumbé centre	N 10°10.399′	E001°06.429′
AS11	Iporapia	Kkoutatiégou	Boukoumbé centre	N 10°10.849′	E001°07.022′
AS12	Iporapia	—	—	—	—
AS13	Iporapia	—	—	—	—
AS14	Ipomonpiété	Otanongou	Dipoli	N 10°32.404′	E000°96.941′
AS15	Tignonpété	Tassayota	Korontière	N 10°32.160′	E000°98.793′
AS16	Iporawan	Koutchatahongou	Boukoumbé centre	N 10°09.200′	E001°05.441′
AS17	Yoro	Dipoli	Dipoli	N 10°16.992′	E000°57.967′
AS18	Tignonpété	Koumagou-B	Natta	N 10°12.161′	E001°08.322′
AS19	Iporni	Dipokor-1	Manta	N 10°18.394′	E001°05.273′
AS20	Iporawan	Yatié	Tabota	N 10°24.612′	E001°03.757′

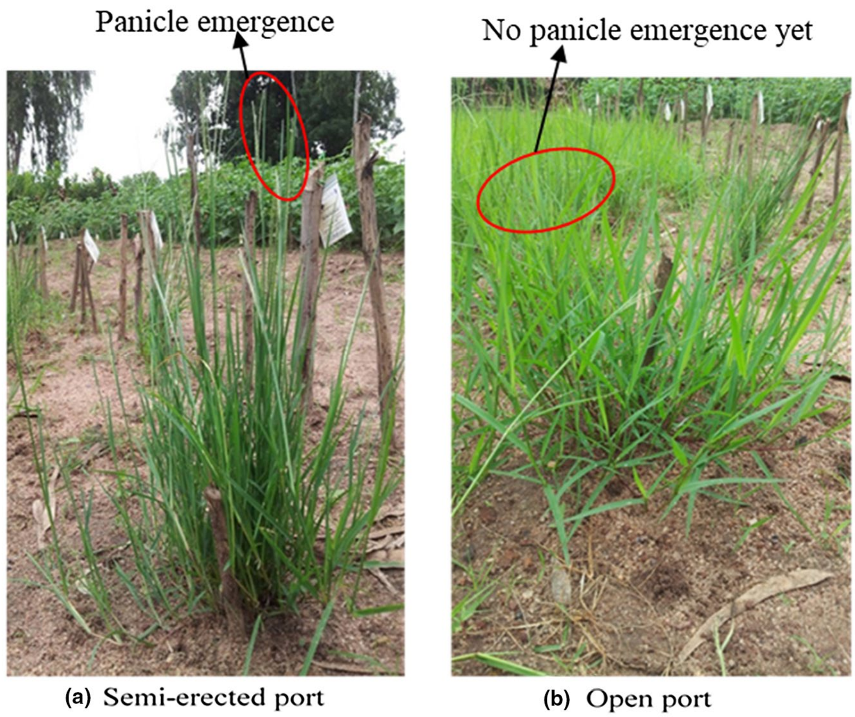

FIGURE 2 Difference of plant ports and heading shifting used in intra-accession homogenization

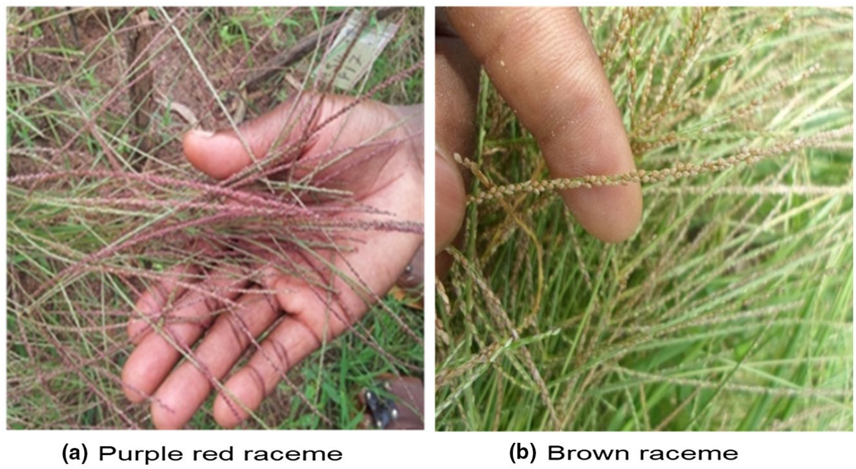

FIGURE 3 Phenotypic screening based on raceme color within accession

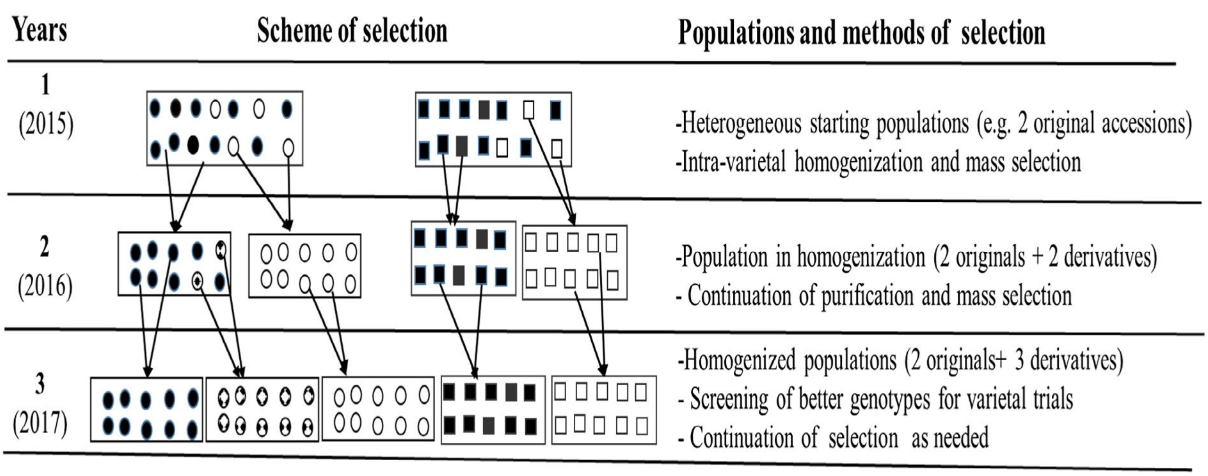

FIGURE 4 Diagram describing the homogenization and mass selection practiced within accessions

assessed visually on the basis of the dominant colour at the scale of each plot (Saidou et al., 2014; Sekloka et al., 2016) (Table 3). Quantitative variables were assessed at plot level for agronomic parameters. For morphometric traits, 10 individual plants were chosen at random from the central rows of each plot and characterized (Table 4).

2.6 | Data analysis

All data were recorded using Excel spreadsheet and analysed with R 3.6.3 software (R Core Team, 2020). For each qualitative trait, visual observations made on plant organs were compared between original accessions and their reselections (derivatives). In order to assess intra and inter-varietal homogeneity on the basis of morphometric variables, the *t* tests of Student were performed to compare each derivative accession to correspondent original accession (parent) basing on individual plants data. The coefficient of variation was also calculated each time.

Similarly, the level of intra-varietal homogeneity within each accessions was assessed on the basis of grains colour:

$DH\,(\%) = (PGD/PGT) \times 100$, with DH: homogeneity rate of grains colour expressed as a percentage, PGD: grains weight of plot presenting colour of accession sown at the start, and PGT: total grains weight of plot.

To estimate the performance progress recorded by derivative accessions compared to their original ones, the gains (G) or losses (P) were calculated on the values of morphometric and agronomic characters as follows:

$G/P\,(\%) = \frac{(y-x)}{x} \times 100$ (Belete et al., 2017; Totok et al., 1998; Zou et al., 2010), with *y*, *x*, respectively, standing for performance of derivative accession and their correspondent original one. The negative and positive values of *G/P* reflect the gains and losses recorded respectively.

A set of relevant variables consisting of cycles sowing–heading and maturation, grain yield, homogeneity rate and racemes colour were submitted to Factor Analysis of Mixed Data followed by Ascending Hierarchical Clustering based on Euclidean distance according to Ward method (Sekloka et al., 2016) in order to form homogeneous groups. Analyses of variance and Chi-square tests (X^2) were performed to describe obtained groups. From these analyses, two best genotypes of each homogeneous group were sorted. The packages 'FactoMineR' (Le et al., 2008) and 'factoextra' (Kassambara & Mundt, 2019) were used for these factor analyses.

TABLE 2 Original accessions and their reselections (derivative accessions)

Originals (parents)	Derivatives_2015	Derivatives_2016
AS1	—	AS1-1
AS2	AS2-1	AS2-2, AS2-1-1
AS3	—	AS3-1
AS5	—	—
AS6	—	AS6-1
AS7	—	AS7-1
AS8	—	—
AS9	—	—
AS10	—	AS10-1
AS11	—	AS11-1
AS12	—	—
AS13	AS13-1	—
AS14	AS14-1	AS14-2
AS15	AS15-1	AS15-1-1
AS16	AS16-1	AS16-1-1
AS17	AS17-1	AS17-2
AS18	—	AS18-1
AS19	AS19-1	AS19-2, AS19-1-1
AS20	—	AS20-1

3 | RESULTS

3.1 | Homogeneity of accessions on the basis of qualitative characteristics

Accessions AS1 and AS7 showed purple coloration at the collar, and their derivatives (AS1-1, AS7-1) light green coloration (Table 5). Others (AS2, AS16, AS17 and AS19) have presented leaf limb of light green colour and their derivatives (AS2-1-1, AS2-2, AS16-1, AS17-1, AS17-2, AS19-1 and AS19-2) dark green colour. Similar discrimination was also observed with the presence and distribution of anthocyanin colouration of leaf limb. These morphological differences were

TABLE 3 Description of qualitative variables

Stages	Qualitative variables	Abbreviations	Categories
Vegetative	Coloration of anthocyanin at leaf limb	ColAnt_LF	Present, absent
	Distribution of anthocyanin colour at leaf limb	DistrAnt-LF	Absent, extremity only, spotted, uniformly
	Intensity of green colour of leaf limb	Coul_LF	Dark green, medium green, light green
	Colour of collar	Coul.collet	light green, purple
Maturation	Colour of racemes	Coul_rac	Brown, purplish red

TABLE 4 Description of quantitative variables

Cycle phases	Variables and abbreviations	Descriptions
Flowering	Cycle sowing–heading (CSE in das)	Number of days after sowing (das) when 50% of plants let discover their panicle on plot
Maturation	Cycle sowing–maturation (CSM in das)	Number of days after sowing when 50% of plants have matured without the grains are dry
	Length of panicle leaf (Long_FP in cm)	Length measured from insertion level of ligula to limb top
	Plant height (HP in cm)	Height measured from soil level to the top of longest panicle
	Raceme length (Long_Rac in cm)	Length from beginning of racemes to the top of longest raceme
Post-harvest	Grain yield (Rdt in kg·ha^{-1})	Ratio of grain weight per plot area after threshing and winnowing

more noticeable with the raceme coloration where the accessions of brown racemes were clearly distinguished from their derivatives to purplish red racemes and vice versa (Table 5).

3.2 | Homogeneity of accessions based on morphometric traits

Of the 23 derivative accessions, five (AS13-1, AS14-1, AS15-1-1, AS19-1 and AS19-1-1) had lower coefficients of variation than those of their original accessions for all three morphometric criteria assessed (Table 6). Also, five other derivative accessions (AS2-1, AS3-1, AS6-1, AS11-1 and AS16-1) had lower coefficients of variation than their original for simultaneously two of the three criteria evaluated. Of these 23 accessions from reselection, significant differences with their original ones (parents) were highlighted in 7 cases for the plants height (30%), 8 for the length of panicle leaf (35%) and 7 for racemes length (30%). Some accessions (AS2-1, AS2-1-1, AS3-1, AS16-1, AS17-1 and AS20-1) were found different from their original simultaneously for the three criteria even if the differences were not always significant (Table 6).

As for the progress on morphological performances, 4, 8 and 4 cases of derivative accessions were, respectively, improved by at least 10% compared to their original ones for the growth in plants height, lengths of panicle leaf and racemes length. The contrary trend was observed, respectively, in 4, 3 and 1 cases of the derivative accessions where the morphometric performances regressed by more than 10% compared to their original ones (Table 6).

3.3 | Agronomic performance of accessions and progress

The evaluation of the precocity parameters showed that the early accessions took approximately 65 days after sowing to heading (AS1, AS1-1, AS7-1, AS6, AS6-1, AS8, AS17, AS17-1, AS17-2 and AS19-1-1) while the late ones took around 80 days. Yields varied from simple (AS1) to quadruple (AS16) (Table 7). For agro-phenological progress, three of the derivative accessions (AS2-1, AS7-1 and AS19-1-1) have shown cycle sowing–heading shortened by more than 10%–15% compared to their original ones. A similar trend was noted for sowing–maturing cycle in five cases of derivative accessions (AS2-1-1, AS2-2, AS3-1, AS10-1 and AS15-1). For yield, progressions and regressions have been noted. Thus, 12 of derivative accessions (52%) had improved grain yield from 10% to more than 70% compared to their original ones, notably for AS1-1, AS3-1, AS7-1, AS11-1, AS14-1, AS14-2, AS15-1-1, AS19-1-1 and AS19-2. However, yield reductions of around 20% to more than 60% were noted on eight derivative accessions (35%) compared to their original ones (Table 7).

3.4 | Homogeneity rate of accessions based on grains colour

The homogenization assessed on the basis of panicles grain colour showed out of all 42 accessions, 16 (≈38%) achieved homogeneity rates greater than 90% (Table 8). Among these, 6 accessions were 100% homogeneous. Twenty (20) accessions (≈48%) were at rates between 70% and 90% and only 6 (≈14%) were at rates from 53.44% to 69.96% (Table 8).

3.5 | Selection of genotypes

All the agronomic variables, homogeneity rates of grains and racemes colour submitted to factor analysis of mixed data showed that the first three factorial axes explained more than 86% of the total variability, and of which the first two axes almost 73% (Table 9).

The cycle characters (CSE and CSM) were strongly and positively correlated with axis 1. The yield (Rdt) is moderately

TABLE 5 Visual comparison of original accessions to their derivatives (reselections)

Accessions		Coul.collet	Coul.LF	ColAnt. LF	Distr-AntLF	Coul_rac
AS1	AS1	Purple	Dark green	Present	Spotted	Purplish red
	AS1-1	Light green	Dark green	Absent	Absent	Brown
AS2	AS2	Purple	Light green	Present	Uniformly	Brown
	AS2-1	Purple	Medium green	Absent	Absent	Brown
	AS2-1-1	Purple	Dark green	Absent	Absent	Purplish red
	AS2-2	Purple	Dark green	Present	Spotted	Purplish red
AS3	AS3	Purple	Light green	Absent	Absent	Brown
	AS3-1	Purple	Light green	Present	Extremity only	Purplish red
AS5	AS5	Purple	Light green	Absent	Absent	Brown
AS6	AS6	Light green	Medium green	Present	Extremity only	Purplish red
	AS6-1	Light green	Dark green	Present	Extremity only	Purplish red
AS7	AS7	Purple	Dark green	Present	Spotted	Purplish red
	AS7-1	Light green	Medium green	Present	Spotted	Brown
AS8	AS8	Light green	Medium green	Absent	Absent	Brown
AS9	AS9	Purple	Light green	Absent	Absent	Brown
AS10	AS10	Purple	Medium green	Absent	Absent	Brown
	AS10-1	Purple	Medium green	Present	Uniformly	Purplish red
AS11	AS11	Purple	Dark green	Absent	Absent	Brown
	AS11-1	Purple	Medium green	Absent	Absent	Rouge
AS12	AS12	Purple	Medium green	Absent	Absent	Brown
AS13	AS13	Purple	Medium green	Present	Spotted	Purplish red
	AS13-1	Purple	Light green	Absent	Absent	Purplish red
AS14	AS14	Purple	Light green	Present	Spotted	Purplish red
	AS14-1	Purple	Medium green	Absent	Absent	Purplish red
	AS14-2	Purple	Medium green	Present	Spotted	Brown
AS15	AS15	Purple	Medium green	Present	Extremity only	Purplish red
	AS15-1	Purple	Dark green	Present	Spotted	Purplish red
	AS15-1-1	Purple	Dark green	Absent	Absent	Brown
AS16	AS16	Purple	Light green	Present	Spotted	Purplish red
	AS16-1	Purple	Dark green	Present	Extremity only	Brown
	AS16-1-1	Purple	Medium green	Absent	Absent	Purplish red
AS17	AS17	Purple	Light green	Absent	Absent	Brown
	AS17-1	Purple	Dark green	Absent	Absent	Purplish red
	AS17-2	Purple	Dark green	Absent	Absent	Purplish red
AS18	AS18	Purple	Light green	Absent	Absent	Brown
	AS18-1	Purple	Medium green	Present	Extremity only	Purplish red
AS19	AS19	Purple	Light green	Present	Extremity only	Purplish red
	AS19-1	Purple	Light green	Absent	Absent	Purplish red
	AS19-1-1	Purple	Dark green	Present	Spotted	Brown
	AS19-2	Purple	Dark green	Absent	Absent	Brown
AS20	AS20	Purple	Medium green	Present	Uniformly	Purplish red
	AS20-1	Purple	Dark green	Absent	Absent	Brown

Abbreviations: ColAnt.LF, Coloration of anthocyanin at leaf limb; Coul.collet, Colour of collar; Coul_LF, Intensity of green colour of leaf limb; Coul_raceme, colour of racemes; DistrAnt-LF, Distribution of anthocyanin colour at leaf limb.

TABLE 6 Homogeneity and progress based on morphometric parameters

Accessions		HP				Long_FP				Long_rac			
		Mean	G/P	*t* value	CV	Mean	G/P	*t* value	CV	Mean	G/P	*t* value	CV
AS1	AS1	79.3			4.7	11.5			9.5	11.8			6.7
	AS1-1	85.4	7.7	**−2.61***[*]	7.0	15.0	30.4	**−5.11***[***]	11.5	12.3	4.2	−1.39	7.3
AS2	AS2	99.0			13.2	8.0			22.5	12.7			10.8
	AS2-1	104.5	5.6	−0.91	12.0	9.8	22.5	**−2.23**[*]	16.6	13.1	3.1	−0.63	9.5
	AS2-1-1	103.2	4.2	−0.74	28.2	10.4	30.0	−2.14	28.2	15.9	25.2	**−6.26**[***]	4.1
	AS2-2	98.8	−0.2	0.03	12.9	7.6	−5.0	0.65	6.3	12.1	−4.7	0.84	14.1
AS3	AS3	92.4			13.9	7.6			18.3	12.5			10.8
	AS3-1	102.8	11.3	−1.47	16.2	11.1	46.1	**−4.86**[***]	15.1	13.8	10.4	**−2.29**[*]	8.1
AS5	AS5	88.1			10.2	10.6			20.1	13.1			9.1
AS6	AS6	94.9			10.8	12.1			20.7	12.1			5.7
	AS6-1	83.4	−12.1	**2.77**[*]	8.5	13.0	7.4	−0.90	12.3	10.9	−9.9	**3.00**[**]	8.6
AS7	AS7	112.7			6.7	8.7			17.5	13.1			11.4
	AS7-1	80.5	−28.6	**10.48**[***]	6.6	11.0	26.4	**−2.63**[*]	19.8	11.1	−15.3	**2.74**[*]	14.4
AS8	AS8	81.4			5.7	11.0			20.5	12.4			9.5
AS9	AS9	96.8			10.7	9.7			11.5	14.3			7.6
AS10	AS10	109.7			11.7	11.6			13.3	13.4			10.4
	AS10-1	95.4	−13.0	**2.36**[*]	13.4	11.1	−4.3	0.69	17.4	13.5	0.7	−0.03	12.2
AS11	AS11	101.4			20.0	9.9			14.3	13.3			18.1
	AS11-1	111.5	10.0	1.16	14.7	10.5	6.1	−0.77	18.7	12.6	−5.3	0.72	11
AS12	AS12	94.6			11.7	10.0			25.0	12.7			8.1
AS13	AS13	97.5			35.5	13.1			17.6	13.1			10.6
	AS13-1	99.9	2.5	−0.20	11.3	9.3	−29.0	**4.26**[**]	13.8	12.6	−3.8	0.88	8.1
AS14	AS14	101.7			8.4	9.6			14.4	12.6			16.4
	AS14-1	101.9	0.2	−0.07	7.2	8.8	−8.3	1.35	13.3	12.4	−1.6	0.30	5.6
	AS14-2	99.9	−1.8	0.28	16.7	11.7	21.9	**−2.28**[*]	20.8	12.7	0.8	−0.16	4
AS15	AS15	106.2			13.7	10.7			16.1	12.3			10.9
	AS15-1	107.2	0.9	−0.14	14.5	9.3	−13.1	1.44	27.1	12.2	−0.8	0.23	13.6
	AS15-1-1	110.0	3.6	−0.67	8.4	10.4	−2.8	0.47	13.2	14.2	15.4	**−3.21**[**]	7.5
AS16	AS16	103.3			9.3	9.3			24.2	12.7			6.8
	AS16-1	122.7	18.8	**−4.65**[***]	6.6	11.5	23.7	**−2.50**[*]	10.6	13.0	2.4	−0.44	12.8
	AS16-1-1	111.7	8.1	−2.03	7.0	9.2	−1.1	0.15	31.2	14.7	15.7	**−3.88**[**]	8.7
AS17	AS17	80.7			8.9	11.6			23.0	12.2			8
	AS17-1	113.3	40.4	**−5.91**[***]	13.2	11.8	1.7	−0.16	24.7	13.3	9.0	−1.24	18.8
	AS17-2	84.8	5.1	−1.02	11.5	11.3	−2.6	0.32	8.2	12.0	−1.6	0.49	7.8

(Continues)

TABLE 6 (Continued)

Accessions		HP				Long_FP				Long_rac			
		Mean	G/P	*t* value	CV	Mean	G/P	*t* value	CV	Mean	G/P	*t* value	CV
AS18	AS18	106.4			9.7	13.2			25.8	13.7			8.4
	AS18-1	106.7	0.3	−0.06	12.0	11.0	−16.7	1.87	12.2	14.4	5.1	−1.12	9.5
AS19	AS19	99.2			16.4	9.9			20.6	13.9			8
	AS19-1	108.9	9.8	−1.49	9.9	9.7	−2.0	0.19	17.1	13.6	−2.2	0.66	7.7
	AS19-1-1	75.6	−23.8	**4.19*****	6.0	13.2	33.3	**−4.55****	5.9	12.6	−9.4	**3.19****	4.6
	AS19-2	103.1	3.9	−0.58	11.3	10.2	3.0	−0.25	22.7	13.7	−1.4	0.33	12.4
AS20	AS20	102.4			9.0	10.4			18.0	12.8			10.3
	AS20-1	106.3	3.8	−0.64	14.5	13.4	28.8	**−2.93***	17.7	13.2	3.1	−0.56	9.1
Mid. parent		97.2			9.8	10.4			14.6	12.9			5.0
Mid. derivative		100.8	3.6	1.52	11.7	10.9	4.2	0.50	15.4	13.0	1.2	0.64	8.7

Abbreviations: G/P, Gain or loss, coefficient of variation (CV) in %; HP, plant height; Long_FP, length of panicle leaf; Long_rac, length of raceme.

The values of *t* test in bold reflect significant differences between derivative accession and their correspondent original one.

***Very highly significant (<0.001).

**Highly significant (<0.01).

*Significant (<0.05).

correlated with it. This axis, therefore, reflects the agronomic performances of accessions. Likewise, homogeneity rate (DH) and racemes colour (coul.rac) are positively and strongly related to axis 2. This axis is, therefore, an axis of homogeneity and of racemic colouration (Figure 5).

The Ascending Hierarchical Clustering of accessions followed by their projection in the first factor plane enabled to structure all of them in three distinct agro-morphological groups (Figure 6). The first axis discriminates enough clearly the accessions of group 1 and those of group 3 located, respectively, on the negative and positive sides of this axis. Group 2 is intermediate, but closer to group 3 for the traits studied. Group 1 is characterized by early accessions with low yields, high homogeneity rates for the majority. Group 3 is constituted of late accessions with overall the highest yields and homogeneity levels. Group 2 is mainly characterized by late and high-yielding accessions with overall lower levels of homogenization.

The quantitative agronomic trait, which discriminate these groups, consisted of the cycles sowing–heading and sowing–maturation, grain yield and homogeneity rate (Table 10).

The Chi-square test (x^2) carried out showed the racemes colour highly discriminated accessions clustering into groups ($p < .001^{***}$) (Table 11). All accessions of Group 3 have brown racemes. Those in Group 2 presented purplish red racemes for the most (about 80%), while Group 1 consisted of combinations of both colours with slight dominance of accessions to brown racemes (Table 11).

Selective sorting in these groups taking into account the cycle traits, yield, racemes colour and intra-varietal homogeneity rate of the grains led to choosing six (06) genotypes of which two (02) sorted in each obtained group (Table 12). The first two varieties (AS19-1-1, AS1) are basically characterized by their earliness and the last two (AS15-1-1, AS18) by their grain yield, but with long cycle (late). The others (AS13-1 and AS2-1-1) were intermediate with particularly purplish red racemes. All these genotypes showed high intra-varietal homogeneity rates (>90%) (Table 12).

4 | DISCUSSION

Characterization and selection of improved varieties are key steps in the management and exploitation of genetic diversity of grown plants (Manzano et al., 2001; Radhouane, 2004; Yobi et al., 2002).

In fonio, a particularly difficult species to select because of the small size of its floral organs and its grains (Vodouhè & Achigan Dako, 2006), the evaluation of visual characters on the landraces analysed in this study showed significant morphological differences between the initial accessions and their reselections (derivatives). This reflects the strong intra-varietal heterogeneity that existed at the level of original accessions (Sekloka et al., 2016) and justifies the process of homogenization undertaken. Adoukonou-Sagbadja et al. (2010) previously observed that cultivated fonio landraces were consisted of polyvarietal mixture due to access

TABLE 7 Accessions performances and progress based on agronomic traits

Accessions		CSE (das)		CSM (das)		Rdt ($kg\cdot ha^{-1}$)	
		Mean	G/P	Mean	G/P	Mean	G/P
AS1	AS1	68	_	82	_	473.3	_
	AS1-1	68	0.0	99	20.7	820.0	73.2
AS2	AS2	89	_	101	_	1003.3	_
	AS2-1	80	−10.1	92	−8.9	766.7	−23.6
	AS2-1-1	88	−1.1	74	−26.7	373.3	−62.8
	AS2-2	87	−2.2	73	−27.7	546.7	−45.5
AS3	AS3	84	_	96	_	910.0	_
	AS3-1	86	2.4	80	−16.7	1213.3	33.3
AS5	AS5	84	_	95	_	706.7	_
AS6	AS6	65	_	73	_	503.3	_
	AS6-1	65	0.0	73	0.0	453.3	−9.9
AS7	AS7	75	_	95	_	903.3	_
	AS7-1	65	−13.3	91	−4.2	1186.7	31.4
AS8	AS8	65	_	98	_	946.7	_
AS9	AS9	81	_	91	_	966.7	_
AS10	AS10	88	_	91	_	1216.7	_
	AS10-1	83	−5.7	73	−19.8	513.3	−57.8
AS11	AS11	78		90		1160.0	
	AS11-1	83	6.4	93	3.3	1476.7	27.3
AS12	AS12	83	_	93	_	666.7	_
AS13	AS13	79	_	95	_	900.0	_
	AS13-1	79	0.0	94	−1.1	996.7	10.7
AS14	AS14	79	_	86	_	826.7	_
	AS14-1	78	−1.3	92	7.0	1080.0	30.6
	AS14-2	81	2.5	94	9.3	1070.0	29.4
AS15	AS15	74	_	87	_	790.0	_
	AS15-1	74	0.0	74	−14.9	543.3	−31.2
	AS15-1-1	78	5.4	91	4.6	1230.0	55.7
AS16	AS16	77	_	90	_	1723.3	_
	AS16-1	83	7.8	104	15.6	640.0	−62.9
	AS16-1-1	84	9.1	91	1.1	833.3	−51.6
AS17	AS17	67	_	90	_	986.7	_
	AS17-1	69	3.0	92	2.2	1090.0	10.5
	AS17-2	65	−3.0	90	0.0	1030.0	4.4
AS18	AS18	80	_	98	_	1033.3	_
	AS18-1	80	0.0	98	0.0	1160.0	12.3
AS19	AS19	79	_	97	_	923.3	_
	AS19-1	81	2.5	99	2.1	676.7	−26.7
	AS19-1-1	67	−15.2	94	−3.1	1190.0	28.9
	AS19-2	79	0.0	98	1.0	1150.0	24.5
AS20	AS20	78		73		950.0	
	AS20-1	81	3.8	90	23.3	906.7	−46
Mid. parent		77.5		89.5		890.9	
Mid. derivative		77.6	0.1	90.0	0.6	939.6	5.5

Abbreviations: CSE, cycle sowing–heading; CSM, cycle sowing–maturation; das, days after sowing; G/P, gain or loss compared to original accessions in %; Rdt, grain yield.

TABLE 8 Level of accessions homogenization based on grains colour

Accessions (originals and derivatives)	Grain colours at the start	Weight brown grains obtained (g)	Weight purplish red grains obtained (g)	Total weight grains obtained (g)	Homogeneity rate (%)
AS1	Purplish red	_	142	142	100.00
AS1-1	Brown	151	_	151	100.00
AS8	Brown	133	_	133	100.00
AS9	Brown	354	_	354	100.00
AS10	Brown	286	_	286	100.00
AS11	Brown	330	_	330	100.00
AS18	Brown	441	1	442	99.77
AS2-1-1	Purplish red	4	342	346	98.84
AS3	Brown	339	7	346	97.98
AS19-2	Brown	358	9	367	97.55
AS19-1-1	Brown	154	4	158	97.47
AS15-1-1	Brown	504	15	519	97.11
AS20-1	Brown	316	11	327	96.64
AS5	Brown	214	15	229	93.45
AS13-1	Purplish red	98	1,384	1,482	93.39
AS3-1	Purplish red	25	318	343	92.71
AS10-1	Purplish red	27	242	269	89.96
AS6-1	Purplish red	12	96	108	88.89
AS11-1	Purplish red	32	235	267	88.01
AS2-2	Purplish red	28	174	202	86.14
AS15-1	Purplish red	35	199	234	85.04
AS16-1-1	Purplish red	45	226	271	83.39
AS7-1	Brown	178	36	214	83.18
AS12	Brown	232	50	282	82.27
AS16-1	Brown	238	69	307	77.52
AS17-2	Purplish red	27	92	119	77.31
AS13	Purplish red	25	84	109	77.06
AS19	Purplish red	55	181	236	76.69
AS2-1	Brown	234	73	307	76.22
AS14-1	Purplish red	35	108	143	75.52
AS14	Purplish red	45	134	179	74.86
AS17-1	Purplish red	66	178	244	72.95
AS18-1	Purplish red	45	117	162	72.22
AS19-1	Purplish red	65	163	228	71.49
AS20	Purplish red	73	178	251	70.92
AS15	Purplish red	40	95	135	70.37
AS14-2	Brown	156	67	223	69.96
AS6	Purplish red	28	58	86	67.44
AS7	Purplish red	41	69	110	62.73
AS17	Brown	173	110	283	61.13
AS16	Purplish red	79	124	203	61.08
AS2	Brown	101	88	189	53.44

TABLE 9 Characteristics of the factor axes

Characteristics	Axis 1	Axis 2	Axis 3
Eigen value	2.25	1.39	0.68
Explained variance (%)	45.04	27.78	13.53
Cumulative variance (%)	45.04	72.83	86.36

mode to seed and the absence of formal seed production and distribution system (Sekloka et al., 2016). Indeed, many authors have emphasized that farmers' seed management practices, particularly exchanges of varieties between farmers, promote gene flow which helps to broaden the genetic diversity between populations of grown plants (Delaunay et al., 2008; Mckeye et al., 2001; Missihoun et al., 2012; Sawadogo et al., 2014). As described in

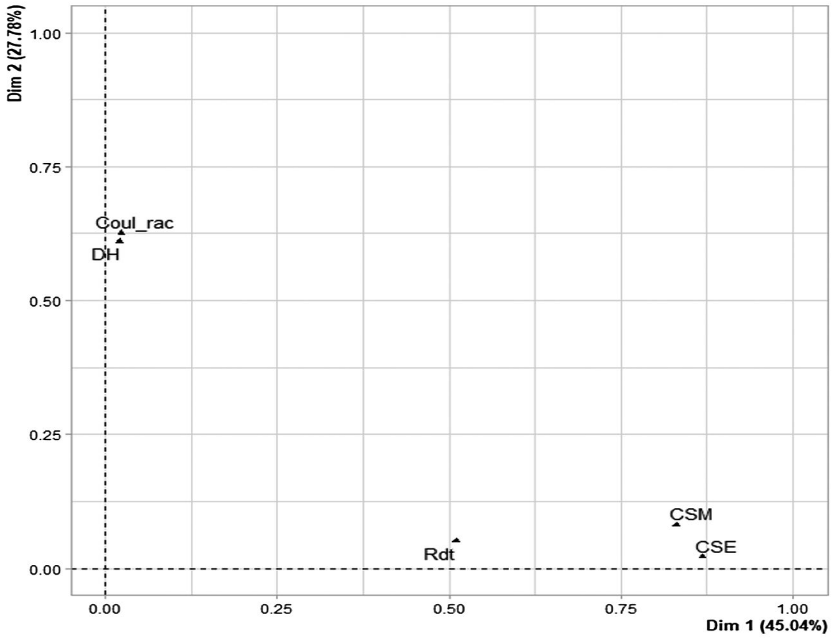

FIGURE 5 Representation showing the relationship of quantitative and qualitative variables with the first two axes

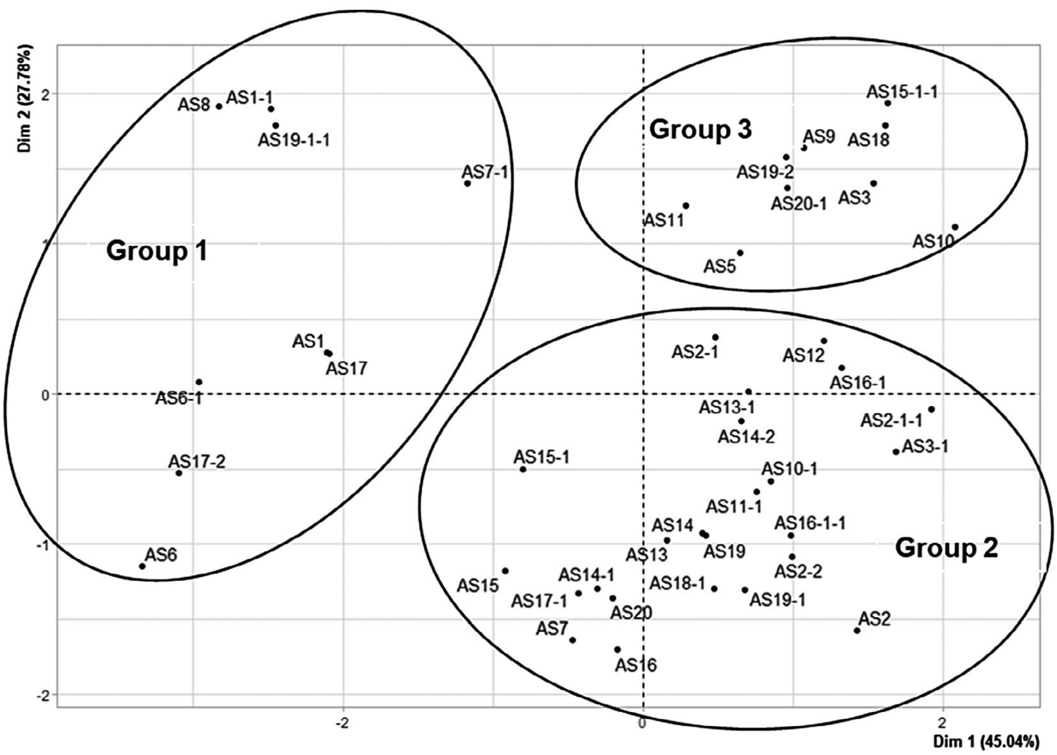

FIGURE 6 Factor map of different accessions clusters projected in the first factor plane

TABLE 10 Agronomic characteristics of the groups obtained

Variables	Group 1	Group 2	Group 3	Mean	SEM	*F* value
CSE (jas)	66.11± 1.4 a	80.38±4.8 b	81.44±3.3 b	77.55	1.1	47.2***
CSM (jas)	75±3.5 a	94±4.4 b	93.22 ±3.3 b	89.76	1.3	78.4***
Rdt (kg·ha^{-1})	618.89±275.5 a	941.39±158.4 b	1152.59±318.8 c	917.5	43.9	12.8***
DH (%)	86.16±14.9 ab	77.62±10.9 a	98.06±2.2 b	83.83	2.1	12.1***

Abbreviations: CSE, cycle sowing–heading; CSM, cycle sowing–maturation; das, days after sowing; DH, homogeneity rate; Rdt, grain yield; SEM, standard error of the mean.

***Very highly significant (<.001).

TABLE 11 Characteristic of racemes coloration of the identified groups

Groups	Brown raceme (% of accessions)	Purplish red raceme (% of accessions)
Group 1	55.6	44.4
Group 2	20.8	79.2
Group 3	100.0	0.0
Result (Chi-square test): X^2 = 17.05; *df* = 2; *p* < .001***		

***Very highly significant (<.001).

Nigeria (Kwon-Ndung & Dachi, 2007), the main access mode to fonio seeds in Benin regions is the collection of seeds from the previous year's harvest and the varieties exchanged between farmers (Adoukonou-Sagbadja et al., 2006; Dansi et al., 2010; Sekloka et al., 2015).

However, if on the major visual characters, the accessions after 3 years of selection, have reached very high levels of homogenization, it was not the same for the morphometric traits where the characters' improvement has been average for some accessions and low for others. This could be explained by the fact that homogenization was much more focused on visual characters than on morphometric traits. But it has also been proven that for most plant species, the quantitative characters are often dependent on several genes influenced by the environment factors (Acquaah, 2007; Falconer & Macky, 1996; Karasu et al., 2009). The response to the selection of such measurable traits also depends on the level of their heritabilities (Acquaah, 2007; Assefa et al., 2001). The morphometric characteristics of such categories could, therefore, have been less homogenized by the mass selection method adopted (Abreu et al., 2010; Solieman & Ragheb, 2014). This result confirms the fact that quantitative characters are more difficult to select than qualitative traits in a breeding programme (Acquaah, 2007).

Likewise, on these morphological traits, the estimate of performances progress showed that, overall, only a few derivative accessions were improved by 10%–15% compared to their correspondent original ones. This once again reflects the fact that selection has moderately or even slightly improved the accessions growth in height and in length. This study, therefore, confirms that mass selection is not very effective on these traits (Abreu et al., 2010; Ngandu-Nyindu, 1981). On agro-phenological traits, some derivative accessions (AS2-1, AS7-1, AS19-1-1, AS2-1-1, AS2-2, AS3-1, AS10-1 and AS15-1) showed that heading or maturation cycles have been shortened by 10%–30% compared to their original ones. Also, more than half of derivative accessions had improved grain yields of 10%–70% compared to their original ones, especially for the derivatives AS1-1, AS3-1, AS7-1, AS11-1, AS14-1, AS14-2, AS15-1-1, AS19-1-1 and AS19-2. Thus, it appears the selection conducted to obtaining some derivative accessions earlier, or more productive than their original ones. These results are similar to those of Belete et al. (2017) who also noted high genetic gains in the yield of eight (08) new varieties compared to one old variety of chickpea in Ethiopia. Unlike morphometric traits, these results on the progress achieved show somewhat the effectiveness of homogenization and mass selection method applied to these agro-phenological traits (Abreu et al., 2010; Belete et al., 2017; Solieman & Ragheb, 2014). These differences in genetic gains between agro-phenological and morphometric traits were mainly related to their degrees or coefficients of heritability (Acquaah, 2007). However, yield is one of quantitative characters often influenced by environmental conditions (Falconer & Macky, 1996; Karasu et al., 2009). Thus, varietal trials in different environments and over several years will allow to validate these performance gains and their stability in the time.

The assessed homogeneity also on the grain colour showed after three selection cycles, approximately 86% of all accessions were at high intra-varietal homogeneity rates of 70%–100%. This also partly highlights the effectiveness of homogenization selection achieved by accessions purification from grains colour on the racemes at maturity. The grains colour has been widely considered as a key criterion for the varietal distinction of fonio (Adoukonou-Sagbadja et al., 2006; Dansi et al., 2010; Sekloka et al., 2015). This shows once again the fact that mass selection is more effective on major visual traits (Abreu et al., 2010; Ngandu-Nyindu, 1981), as they are generally known as more heritable traits (Acquaah, 2007). At last, almost all of sorted genotypes for multi-environment trials exhibited intra-varietal homogeneity rates greater than 97%. These rates comply with standards of 95%, generally accepted and recommended for varieties homogeneity assessment (UPOV, 2019).

TABLE 12 Description of sorted genotypes for multi-local evaluations

Sorted genotypes	CSE (das)	CSM (das)	Rdt (kg·ha^{-1})	Coul_rac	DH (%)
AS19-1-1	67	74	543.34	Brown	97.47
AS1	68	82	473.34	Purplish red	100
AS13-1	79	91	1216.7	Purplish red	93.39
AS2-1-1	88	98	1160	Purplish red	98.84
AS15-1-1	78	90	1723.3	Brown	97.11
AS18	80	93	1476.7	Brown	99.77

Abbreviations: Coul_rac, racemes colour; CSE, cycle sowing–heading; CSM, cycle sowing–maturation; das, days after sowing; DH, homogeneity rate; Rdt, grain yield in station;

5 | CONCLUSION

Varietal homogenization is a process of purification and management of local landraces diversity which quickly leads to improving natural populations. Implemented for three successive years on a collection of fonio landraces, it allowed for the first time in Benin, to obtain homogeneous materials by mass selection. The homogeneity and effectiveness of selection varied following the characters and the accessions. A better level of homogeneity was obtained on the major visual characters compared to morphometric traits. Performance progresses were also variable depending on the characters. The selection better improved the earliness and agronomic traits compared to morphometric characters. It conducted to obtain some derivative accessions earlier or more productive than their original ones. After three selection cycles, and for most accessions, the homogenization level of grain colour ranged from 70% to 100%. With rates higher than 90%, one could estimate satisfactorily the selection and move on to varietal trials in different environments. Thus, the sorting of the genotypes based on the agronomic criteria, racemes colour and homogeneity rates enabled to form three groups within which the genotypes AS19-1-1 and AS1 (group 1), AS13-1 and AS2-1-1 (group 2), AS15-1-1 and AS18 (group 3) were the best retained. Their performances assessment in varietal trials will allow to select homogeneous materials with high performances to respond to production needs in terms of quantity and quality of fonio.

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CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

AUTHORS CONTRIBUTION

CK contributed to study initiation. He participated in all stages including bibliographic research, protocol drafting, field work, data analysis, paper drafting and submission of the article. ES is the initiator of the study. He contributed to protocol drafting, field work, analysis orientations and proofreading of the manuscript. EGA contributed to analysis orientations and proofreading of the manuscript. EK contributed to proofreading of the manuscript.

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