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Full Length Research Paper

Agromorphological characterization of *Sesamum radiatum* (Schum. and Thonn.), a neglected and underutilized species of traditional leafy vegetable of great importance in Benin

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***Sesamum radiatum* (Schum. and Thonn) is a traditional leafy vegetable of national importance in Benin. Although, it is cultivated and consumed in many regions of the country, it is still unfortunately neglected by scientific research. In order to fill in the knowledge gaps necessary for its varietal improvement, 16 accessions collected from different agro-ecological zones were characterized using 16 quantitative morphological traits. A significant variability was found among the accessions. Day to flowering, day to fruiting, total biomass, plant height, noose diameter, number of fruits per plant and number of seeds per fruit were some of the most discriminating parameters. Multivariate analysis (principal component analysis and cluster analysis) revealed four distinct agro-morphological groups of varying performance which will be highly useful in varietal improvement. Correlation analyses between descriptors revealed uniquely desired positive and negative relationships hence, indicating predictable success for eventual breeding program. To further investigate the genetic basis of the phenotypic diversity revealed among *S. radiatum* accessions, the use of molecular markers such as amplified fragment length polymorphisms (AFLPs) is recommended.**

Key words: Benin, *Sesamum radiatum*, diversity, morphological characterization, leafy vegetable.

INTRODUCTION

Throughout the tropical world and particularly, in West Africa, a large number of traditional leafy vegetables (TLVs) have long been known and reported to play important roles in food security for people living in both rural and urban areas (Maundu et al., 1999; Schippers, 2002; Abukutsa, 2007; Francisca and Eyzaguirre, 2007). TLVs are rich in vitamins (especially A, B and C), minerals, fibres, carbohydrates and proteins and some even possess medicinal properties (Almekinders and de Boef, 2000; Schippers, 2002; Grubben and Denton, 2004; Dansi et al., 2008). They represent cheap but quality nutrition for large segments of the populations and

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Figure 1. Vegetative aerial part of a *Sesamum radiatum* plant showing some leave, stem and flower details.

offer an opportunity for improving nutritional status of many families (Chweya and Eyzaguirre, 1999). In many West Africa countries, leafy vegetables are reported to account for 50 to 100% of rural households' income (Mbaye and Moustier, 2000; Diouf et al., 2007) hence, contributing to food security and poverty alleviation.

Recent ethnobotanical investigation of TLVs consumed in Benin revealed 187 species among which *Sesamum radiatum* (Adéoti et al., 2009). *S. radiatum* (Figure 1) is wildly consumed throughout Benin as slimy and nutraceutical vegetable. It is antibiotic and known to treat or prevent hepatic insufficiencies (Shittu et al., 2006). The leaves are also used for treating various sickness including stomach ailments, catarrh, eye pains, bruises and erupted skins (Ogunlesi et al., 2010). Leaves extract was found to have significant myorelaxant effect in

Table 1. List of ecotypes and collecting site (N: North; S: South).

Ecotypes	Status	Collecting sites	Districts
S5	Wild	Atchonsa	Bonou
S6	Wild	Kodé	Adjohoun
S7	Wild	Yoko	Sakété
S8	Wild	Hounti	Lokossa
S12	Wild	Koudokpoe	Zê
S13	Wild	Foli	Za-Kpota
S14	Wild	Sokounhoue	Djakotomè
S15	Wild	Kessounou	Dangbo
S16	Wild	Issaba	Pobè
N1	Cultivated	Gbetebou	Tchaourou
N2	Cultivated	Toume	Sinendé
N3	Cultivated	Saoré	Bembéréké
N4	Cultivated	Goro	Tchaourou
N10	Cultivated	Koutagou	Boukoumbé
N11	Cultivated	Bellefoungou	Djougou
N9	Cultivated	Kotiakou	Tanguieta

guinea-pig thus, supporting the traditional use of the fresh leaves for treating cardiovascular diseases in Cote d'Ivoire (Konan et al., 2006, 2008; Kimiywe et al., 2007). In Western Nigeria, the leaves are used in correcting male sterility by enhancing sperm count and its scientific evidences have been recently provided (Shittu et al., 2007; Ogunlesi et al., 2010). Its cultivation in Benin is the prerogative of women and because of its high market demand appears as one of the vegetables that greatly contribute to household income in rural areas. With regard to the first millennium development goal which calls to combat extreme poverty and hunger, *S. radiatum* is one of the species to consider for promotion in the category of leafy vegetables in the agricultural sector. Despite its nutritional, medicinal and economic importance, the species has been poorly investigated and is currently classified among the so called neglected and under-utilized species (NUS) of Benin (Dansi et al., 2012).

The development of an economically profitable production of any plant species like *S. radiatum* undoubtedly requires the use of improved high yielding varieties. To facilitate breeding efforts, characterisation of available germplasm is a necessary first step since it especially, benefits a plant breeder in choosing proper parental materials (Cilliers and Swanevelder, 2003; Sarutayophat et al., 2007). Also, the knowledge about interrelationships among descriptors (characteristics), from a plant breeder's perspective aid in the selection of superior genotypes from the breeding population and is important in planning and evaluating breeding programmes (Sheela and Gopalan, 2006).

The objectives of this study were to characterize and study the diversity among local ecotypes of *S. radiatum* using agro-morphological descriptors and to study the interrelationship among descriptors used in order to identify outstanding accessions which could be involved in national breeding programs for the benefit of both producers and consumers.

MATERIALS AND METHODS

The study area

Plant accessions were originated from different regions of the Republic of Benin situated in West Africa, between latitudes 6°10' N and 12°25' N and longitudes 0°45' E and 3°55' E (Adam and Boko, 1993). The country covers a total land area of 112,622 km^2 with a population estimated at about 7 millions (Adomou, 2005). It is partitioned into 12 departments inhabited by 29 ethnic groups (Adam and Boko, 1993). The south and the centre are relatively humid agro-ecological zones with two rainy seasons and mean annual rainfall of 1500 mm/year (Adam and Boko, 1993). The North is situated in arid and semi-arid agro-ecological zones characterized by unpredictable and irregular rainfall oscillating between 800 and 950 mm/year with only one rainy season. Mean annual temperatures range from 26 to 28°C and may exceptionally reach 35 to 40°C in the far Northern localities (Adomou, 2005; Akoègninou et al., 2006). The country has about 2,807 plant species (Akoègninou et al., 2006). Vegetation types are semi deciduous forest (South), woodland and savannah woodland (centre East and Northeast), dry semi deciduous forest (centre West and South of Northwest) and tree and shrub savannahs (far north). The experiment was conducted at the station of the International Institute of Tropical Agriculture (IITA) based in Cotonou (South of Benin).

Plant material, experimental design and data collection

Sixteen accessions of local ecotypes collected in the wild and in the cultivated fields were studied (Table 1). Seeds were first sown in polythene bags and after germination; ensuing seedlings were transplanted at the four-leaf stage to 3 × 2 m plots in a completely randomized block design with four replicates, at a spacing of 50 cm between rows and 30 cm within rows as per IPGRI (2001). The field, primarily five years fallow was organic manure-fed. Sixteen (16) qualitative and quantitative parameters of floral and vegetative development, leaf and seed production were studied. These were: days to flowering, total biomass, plant height, node diameter, number of ramification, height of the first branch, leaf length, leaf width, petiole length, days to fruiting, number of fruits per plant, fruit length, fruit width, number of seeds per fruit, thousand seeds weight and seed germination rate. These parameters were measured and data collected as specified in Table 2 from twelve weeks after sowing on five healthy individual plants randomly selected from each of the accessions per repetition following IPGRI (2001).

Data analysis

Collected data was subjected to the one-way analysis of variance using Statistical Analysis System (SAS, 1999) software package and the Student and Newman-Keuls' test was used to separate the treatment means. Using the same software, pairwise distances between accessions were computed and the similarity matrix generated was used to perform a Principal Component Analysis (PCA) and to design a dendrogram using UPGMA cluster analysis (Sneath and Sokal, 1973; Swofford and Olsen, 1990). Computation

Table 2. Quantitative estimation of morphological traits of *Sesamum radiatum.*

Characters	Codes	Technique of measurement
Plant height	PH	Length from soil to the tip of terminal flower head
Leaf length	LL	Distance between the leaf tip and base measured on the third fully opened leaf from the tip
Leaf width	LW	Width of the broadest portion of the third fully opened leaf from tip measured
Petiole length	PL	Measured from main stem to leaf base of the third fully opened leaf from the tip
Noose diameter	ND	An horizontal measure of transition zone between root and stem
Number of ramifications	NR	Count of the total number of ramifications inserted on the stem from soil to the top of plant
Height of first ramification	HFR	Length from soil to the knot of the first ramification
Number of fruits per plant	NFP	Count of the total number of mature fruits obtained from one plant
Thousand seeds weight	TSW	Weight of thousand good seeds measured
Total biomass	TBIO	Weight of the total leaves collected from one plant
Number of seeds per fruit	NSF	Count of the number of seeds obtained per capsule
Fruit length	FL	Measured from the base of fruit to the tip of fruit' horn
Germination rate	SGR	Total number of germinated seeds out of the total number sown
Fruit width	FW	Width of the broadest portion of mature seed
Days to flowering	DTF	Determined by noting the number of days from sowing that 50% of plants had at least one flower
Days to fruiting	DFR	Determined by noting the number of days that 50% of plants had at least one fruit

Table 3. Descriptive statistics of morphological variables measured within accessions of *Sesamum radiatum* collected in Benin.

Characters	Minimum	Maximum	Mean	Standard deviation	Coefficient of variation
Plant height	55	141	106.79	14.97	0.140
Leaf length	1.35	4.35	2.82	0.58	0.206
Leaf width	0.3	0.8	0.54	0.13	0.241
Petiole length	0.1	0.2	0.14	0.04	0.286
Noose diameter	11	23	17.09	2.39	0.140
Number of ramifications	10	36	22.20	4.92	0.222
Height of the first ramification	2	4	3.30	0.66	0.200
Number of fruits per plant	4	567	228.74	129.00	0.564
Thousand seeds weight	2.21	4.1	2.78	0.58	0.209
Total biomass	16.5	450	171.85	112.74	0.656
Number of seeds per fruit	52	108	78.25	12.32	0.157
Fruit length	2.4	3.4	2.86	0.27	0.094
Seed germination rate	0.1	0.6	0.27	0.13	0.481
Fruit width	0.7	0.9	0.76	0.08	0.105
Days to flowering	18	62	38.19	15.95	0.418
Days to fruiting	26	72	46.94	17.03	0.363

of Pearson coefficients of correlation among descriptors was also done using the same software.

RESULTS AND DISCUSSION

Descriptive values (maximum, minimum, mean, standard deviation and coefficient of variation) of the measured quantitative parameters are compiled in Table 3. Data obtained revealed that Coefficients of variation (CV) varied from 9.4% (fruit length) to 65.6% (biomass). The most varying characters as shown by the CV values were: number of fruit (56.4%), germination rate (48.1%), flowering time (41.8%), fruiting time (36.3%), petiole length (28.6%), leaf width (24.1%), number of ramifications (22.2%), and weight of 1,000 seeds (20.9%), leaf length (20.6%), and height of the first ramification (20%). Characters showing weak variation included plant height, noose diameter, number of seeds per fruit, fruit width and fruit length. These very different CV measured for each character is the reflection of some

Table 4. Comparison of means using the test of Student-Newman-Keuls on morphological variables measured in accessions of *Sesamum radiatum* collected in Benin.

Accessions	Height (cm)	Leafy length (cm)	Leafy width (cm)	Total biomass (g)
N1	120.20^{a}	03.52^{ab}	0.68^{a}	225.34^{abc}
N2	103.20^{abc}	02.93^{bcd}	0.63^{a}	338.56^{a}
N3	95.60^{bc}	03.24^{bc}	0.70^{a}	199.24^{abc}
N4	100.80^{abc}	03.12^{bcd}	0.63^{a}	217.00^{abc}
S5	93.80^{bc}	01.94^{f}	0.36^{cd}	26.52^{d}
S6	107.60^{abc}	02.13^{ef}	0.34^{d}	55.48^{cd}
S7	113.40^{ab}	2.52^{de}	0.42^{cd}	127.28^{bcd}
S8	112.20^{ab}	02.98^{bcd}	0.46^{bc}	114.40^{bcd}
N9	94.00^{bc}	03.07^{bcd}	0.56^{ab}	193.36^{abcd}
N10	112.00^{ab}	02.97^{bcd}	0.59^{ab}	309.18^{a}
N11	112.20^{ab}	03.76^{a}	0.68^{a}	267.54^{ab}
S12	122.60^{a}	02.84^{cd}	0.57^{ab}	187.94^{abcd}
S13	122.00^{a}	02.79^{cd}	0.58^{ab}	177.32^{abcd}
S14	84.20^{c}	01.96^{f}	0.48^{bc}	80.56^{cd}
S15	115.60^{ab}	02.76^{cd}	0.42^{cd}	109.68^{bcd}
S16	113.20^{ab}	02.64^{cd}	0.47^{bc}	120.28^{bcd}

Table 5. Principal component analysis of morphological variables measured in various accessions of *Sesamum radiatum* collected in Benin.

Designation	Eigen vectors		
	PC1	PC2	PC3
Eigen value	6.75	2.45	1.71
Proportion	39.73	14.42	10.06
Cumulative	39.73	54.15	64.21

level of variability among accessions.

The analysis of variance revealed significant differences between accessions for the plant height (PH), leaf length (LL), leaf width (LW) and total biomass (TBIO). For these four variables taken individually, the Student-Newman-Keuls' tests classified accessions into five to seven groups (Table 4) hence, indicating significant intraspecific diversity. This means that these descriptors are very important in differentiating between the accessions.

The principal component analysis grouped the 16 variables into various components with the first three explaining 64.21% of the total variation (Table 4). Principal component 1 (PC1) associated with leaf length (LL), leaf width (LW), petiole length (PL), number of fruits (NF), biomass (BIO), number of seed per fruit (NSF), fruit length (FL), fruit width (FW), date to flowering (DTF) and date to fruiting (DFR) accounted for 39.73% of the total variation (Tables 4 and 5). PC2 consisting mainly of noose diameter (NOD), germination rate (GR) and 1000 seeds weight (TSW) accounted for 18% of the total variation while PC3 associated with the number of ramifications and the height of the first ramification explained 10.06% of the total variation (Tables 5 and 6). Based on this data, PC1 appears as one of the most important features in leafy vegetable selection which is vegetative vigour (leaf size, total biomass), earliness (date to flowering) and fruiting (date, number per plant, size). This explains why PC1 alone account for about 40% the total variation. From all the characters, flowering time was found to be the most discriminative parameter differentiating accessions. A relation seems to exist between maturity period and geographical origin. In fact, accessions (N1, N2, N4 and N10) originating from the Northeast were late flowering while accessions (S5, S6, S7) collected from the South showed precocious flowering and those from Northwest and South (N9, S12 and S13) showed an intermediate flowering time. To this regard, the assessment of the influence of the ecology on the reproduction biology of this species is important as such influences have been already reported for many species (Avolio et al., 2011; Bai et al., 2011).

Based on the PC1 and PC2, the sixteen accessions studied were classified into four agromorphological groups (Figure 2) named G1, G2, G3 and G4. The dendrogram (Figure 3) was constructed based on the UPGMA cluster analysis also revealed (with R^2 = 0.53) four groups identical to those obtained with the principal component analysis, hence indicating the reliability of the classification. The four groups obtained have clear different performances (Table 7). Group 1 is early maturing and is characterized by a great number of fruits per plant while Group 4 produces fruits of relatively big size containing several seeds of also big size. Plants of

Table 6. Eigen vectors and values for the three principal component axes.

Characters	Eigen vectors		
	PC1	PC2	PC3
Plant height	0.193	0.773	0.146
Leaf length	0.817	0.176	-0.152
Leaf width	0.915	0.190	-0.206
Petiole length	0.838	0.073	-0.435
Noose diameter	0.293	0.553	0.445
Number of ramifications	0.385	-0.071	0.442
Height of first ramification	-0.116	0.340	0.610
Number of fruits per plant	-0.690	-0.162	-0.058
Thousand seeds weight	0.378	-0.415	-0.061
Total biomass	0.763	0.111	0.343
Number of seeds per fruit	0.643	-0.493	-0.230
Fruit length	0.522	-0.227	0.018
Seed germination rate	0.437	-0.609	0.477
Fruit width	0.605	-0.329	0.128
Days to flowering	0.953	0.109	0.008
Days to fruiting	0.949	0.116	0.002

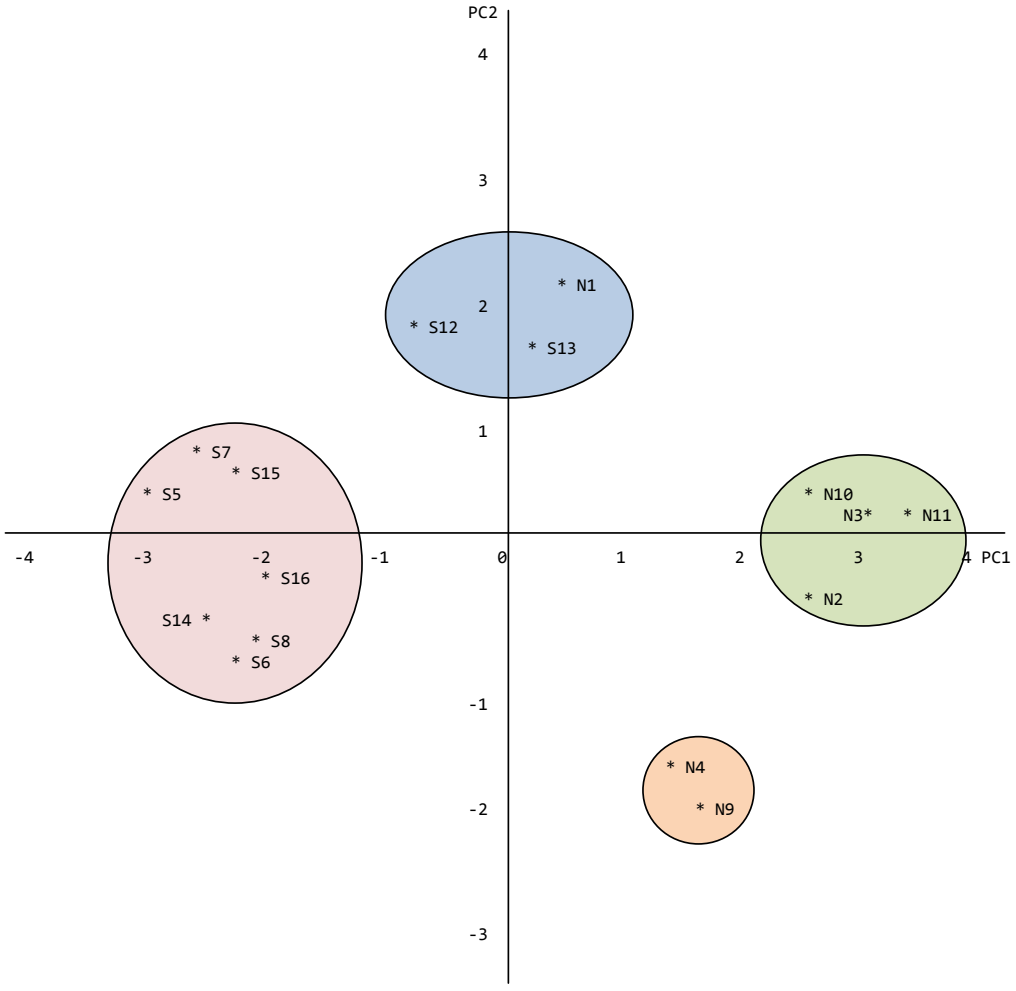

Figure 2. Principal component analysis showing the different grouping of the accessions.

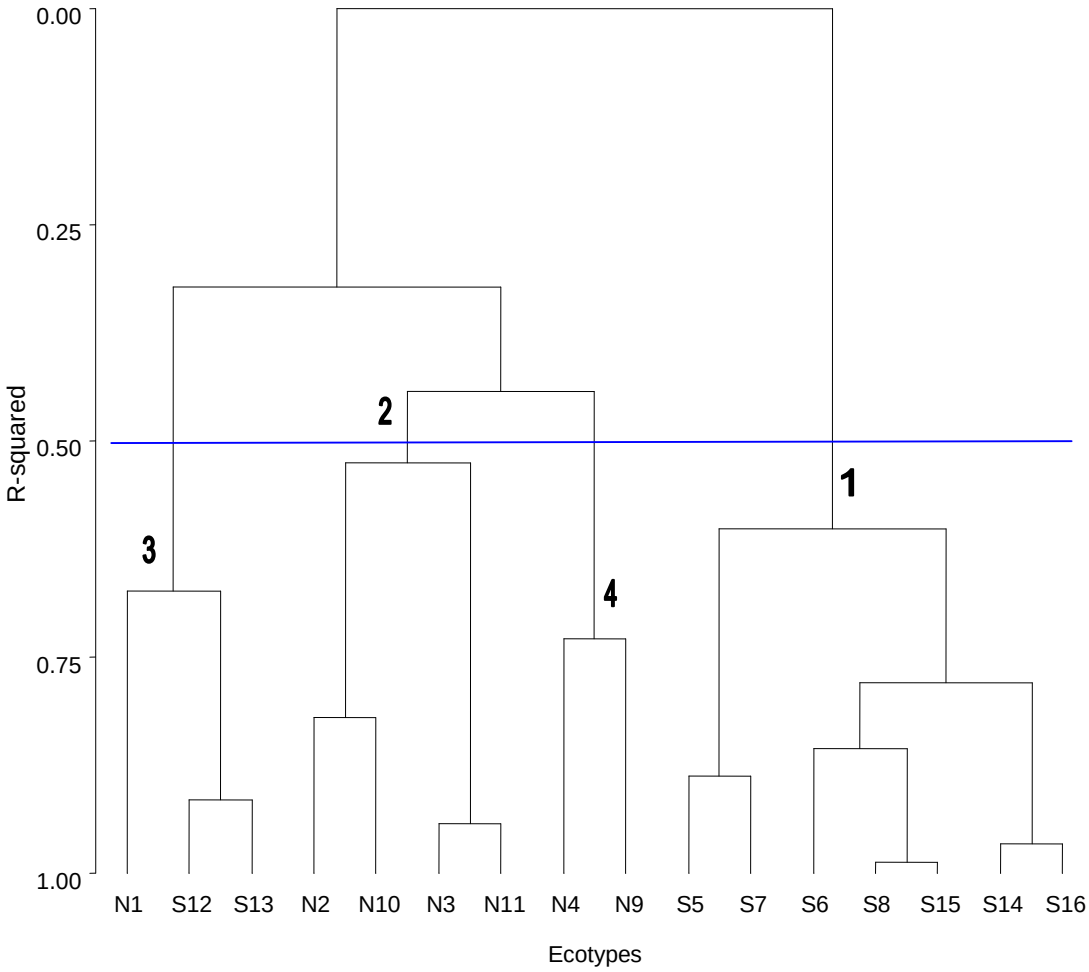

Figure 3. Dendrogram showing the relationship among 16 accessions of *Sesamum radiatum* collected in Benin using the UPGMA methodology.

Group 2 are vigorous (large sized leaves, many ramifications, relatively big noose diameter), produce important biomass and are late maturing in terms of flowering and fruiting. Group 3 is characterised by a high plant height. With leafy vegetable in general, the desired characters for both producers and consumers include high production of total biomass, late flowering and very good fruit with several seeds (Chattopadhyay et al., 1996; Mih et al., 2008). To this regard, data obtained in this study will be highly useful in developing new varieties of S. radiatum by series of crossing between the different group.

Accessions from the groups G2 and G4 exhibiting the best desirable performances are those collected from the home gardens in the North where the species is mostly cultivated and consumed. This may be the results of many years of morphological selection undertaken by farmers with the aim of getting plants with the potential to attract premium prices in local markets. As reported by Vodouhe et al. (2011), getting better performing varieties has been always one key objective of plant domestication for food purposes.

The correlation matrix between variables (Table 8) indicates positive significant correlations between leaf length and leaf width in one hand and between these two parameters and the total biomass in the other hand. Similarly, day to flowering evolves parallel to day to fruiting and the two are also positively correlated to leaf length, leaf width and total biomass. Contrary to this, the number of fruits per plant is negatively correlated to both leaf length and leaf width.

The same significant negative correlation is observed between the number of fruits per plant and the number of seeds per fruit. In terms of plant breeding, these data clearly indicates that with *S. radiatum* selecting varieties with producers and consumers desired characters as indicated above will easily success. Similar results were reported on *Hibiscuss abdariffa* and *Amaranthus* (Diouf et al., 2007), *Vernonia amygdalina* (Mih et al., 2008) and cowpea (Musvosvi, 2009).

To further investigate the genetic basis of the phenotypic diversity revealed among *S. radiatum*

Table 7. Numerical classification of morphological variables measured in accessions of *Sesamum radiatum* collected in Beninand performance of each class.

Characters	Mean value of each class			
	Class 1	Class 2	Class 3	Class 4
Plant height (cm)	105.71	105.75	121.60*	97.40
Leaf length (cm)	2.30	3.23*	3.05	3.10
Leaf breadth (cm)	0.42	0.65*	0.61	0.60
Petiole length (cm)	0.11	0.16*	0.15	0.16
Noose diameter (mm)	16.91	18.20*	17.73	16.00
Number of ramification	21.80	25.50*	19.93	21.40
Height of 1st ramification (cm)	3.26	3.15	3.80	3.00*
Number of fruit per plant	280.57*	137.75	199.00	278.50
Thousand seeds weight	2.48	2.77	2.67	3.99*
Biomass (g)	105.75	256.63*	196.87	205.18
Number of seeds/fruit	71.43	88.00	72.00	92.00*
Capsule length (cm)	2.74	2.85	2.83	3.30*
Rate of germination	0.22	0.38*	0.25	0.35
Fruit breadth (cm)	0.73	0.85*	0.70	0.80
Days to flowering	21.29*	56.75*	46.33	48.00
Days to fruition	28.86*	66.75*	56.00	57.00

*****Performance.

Table 8. Correlation coefficient (Pearson) among descriptors.

Parameters	PH	LL	LB	PL	ND	NR	HFR	NFPP	TSW	B	NSF	FL	SGR	FW	DFL	DFR
PH	1															
LI	0.4552	1														
LB	0.2578	0.8037*	1													
PL	0.1478	0.7069*	0.8839*	1												
ND	0.3909	0.3087	0.2704	0.0413	1											
NR	-0.1653	0.1554	0.1680	0.0817	0.3459	1										
HFR	0.4116	-0.2236	-0.1565	-.2870	0.1606	0-.0083	1									
NFPP	-0.2575	-0.5029*	-0.6339*	-.7017*	-0.2299	-0.2643	-0.916	1								
TSW	-0.1993	0.2867	0.3151	0.2562	-0.0642	-0.0221	-0.0851	0.2108	1							
B	0.2325	0.5237*	0.6681*	0.4141	0.4048	0.4690	0.0928	-0.3658	0.3702	1						
NSF	-0.2736	0.4458	0.4866	0.5605*	-0.1917	0.2159	-0.2361	-0.5001*	0.3434	0.2805	1					
FL	0.2257	0.5214*	0.2857	0.3535	0.0157	0.1361	-0.0361	-0.1976	0.4571	0.2789	0.5167*	1				

Table 8. Contd.

Parameters	PH	LL	LB	PL	ND	NR	HFR	NFPP	TSW	B	NSF	FL	SGR	FW	DFL	DFR
SGR	-0.2218	0.1932	0.2271	0.1921	-0.0330	0.2599	0.0260	-0.2454	0.4013	0.3645	0.3821	0.3702	1			
FW	-0.2343	0.4167	0.4015	0.3685	0.0259	0.4879	-0.1398	-0.4661	0.0976	0.5430*	0.5852	0.2535	0.3245	1		
DFL	0.2058	0.7157*	0.9319*	0.8266*	0.3296	0.3436	-0.0473	-0.6154	0.3555	0.7724*	0.4951*	0.4001	0.3907	0.4654	1	
DFR	0.2046	0.7087*	0.9347*	0.8252*	0.3339	0.3330	-0.0427	-0.6159*	0.3498	0.7674*	0.4990*	0.3876	0.3825	0.4572	0.9995*	1

* = Significant at 0.05 level of probability; PH = plant height; LL = leaf length; LB = leaf width; PL = petiole length; ND = noose diameter; NR = number of ramifications; HFR = height of first ramification; nfpp = number of fruits per plant; TSW = thousand seeds weight; TBIO = total biomass; NSF = number of seeds per fruit; FL = fruit length; SGR = seed germination rate; FW = fruit width; DFL = days to flowering and DFR = days to fruiting.

accessions, the use of molecular markers such as AFLPs (Andru et al., 2011) will be necessary as it was the case for many crops such as fonio (Adoukonou et al., 2007), wheat (Altintas et al., 2008), yam (Sonibare et al., 2010) and rice (Rajkumar et al., 2011).

Conclusion

The agro-morphological characterization of the accessions of *S. radiatum* has contributed to a better assessment of the phenotypes of the collected accessions and an identification of the plants with desired characteristics for breeding. It showed the existence of intra-specific variability. However, it may be necessary to evaluate the characterized accessions over a period of two or more years in many different locations to see if similar observations will be observed. It will be also necessary to enhance the existing germ-plasm through additional collection from different agro-ecological zones and complete this study by molecular analysis for better assessment of genetic diversity with the species.

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