Morphological diversity among accessions of Gboma eggplant (*Solanum macrocarpon* L.)

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Abstract

Morphological diversity is a prerequisite for development of new varieties and identification of pollen parents. The pattern of morphological variability pattern was investigated among 13 accessions of S. macrocarpon sourced from Africa and Asia. Our objectives were to quantify diversity among accessions of S. macrocarpon and identify best accessions to be used as pollen parent and conservation. Field experiments took place during 2009 thorough 2010. Two methods of multivariate analysis (Principal Component Analysis and Cluster analysis) were used to analyze the data set. The first principal component axis showed maximum variability and depicts variation associated with receptacle pigmentation (purple). Receptacle pigmentation was important in the assignment of accessions of accessions into cluster, meanwhile pigmentation (purple) contributed to cluster constellation. Dispersion on the plot of PC axes 1 by 2 and grouping on the dendrogram tree correspond to low to moderate variability, phenotypic plasticity and geographic heterogeneity. There was no association between accessions and geographical origin. Pigmentation (purple) and stripped fruit gene could be used by potential users (plant breeders and seed companies) as morphological markers in breeding works. High heritability and heterosis should be expected provided intraspecific hybridization targeting receptacle pigmentation and fruit colour distribution (stripped) traits.

Keywords: Receptacle pigmentation, Cluster analysis, Dispersion, Diversity, Solanum species

Introduction

The common name eggplant includes three closely related cultivated species that belong to subgenus Leptostemonum: *S. melongena* L., brinjal eggplant or aubergine, *S. aethiopicum* L., scarlet eggplant; and *Solanum macrocarpon* L., gboma eggplant (Daunay et al., 2001a). The latter two species result by domestication process that occurred in Africa, starting from two wild ancestors, *Solanum anguivi* and *Solanum dasyphyllum*, (Lester, 1998; Lester and Niakan 1986). *Solanum macrocarpon* (2n=24) (Gboma eggplant) is classified in the section Melongena; series Macrocarpa Bitter, it is cultivated widely throughout tropical Africa, especially in humid regions. The glabrous leaves are important green vegetables, and fruits are consumed fresh. Many cultivars are robust, perennial with deeply lobed leaves; other cultivars from West Africa are smaller and much branched with smaller and often simple leaves and young shoots (Shippers, 2000). Fruits range between 3-12 cm in diameter, spherical or depressed, usually green, whitish or purple or with lighter markings when ready for eating, but at physiological ripeness they turn yellow or orange or brown, and the surface may crack (Duanay *et al.* 2001a, 2001b).

Genetic architecture of a population is generally believed to be the result of breeding system, gene flow within and between population, and prolonged selection by various natural and artificial forces. Assessment of genetic diversity in germplasm collections provides information useful for both germplasm management and breeding. Effective and economic germplasm management requires both morphological and molecular characterization of material. These allow for establishment of core, nonredundant germplasm collections and help to guide future germplasm collection efforts. Multivariate analyses (Principal Component Analysis and cluster dendogram) have been used widely to measure diversity in the germplasm collections, and to assess the relative contribution of characters to total variability in germplasm collection (Baatout 1995). A significant feature of the multivariate model is that it enables researchers to use specific attributes in gene pool for crop improvement. We evaluated the extent of genetic variability among *S. macrocarpon* based on morphological characteristics to provide template for genetic improvement. Our objectives in this study are to quantify the magnitude morphological variability among accessions of *S.*

macrocarpon and identify traits that could sufficiently discriminate germplasm collection for this species and promising accessions as pollen source.

Materials and Methods

Entries evaluated comprised thirteen accessions of *S. macrocarpon* from locations in Africa and Asia, maintained in the gene banks of AVRDC (The World Vegetable Center), Taiwan and French Institute for Agricultural Research, (INRA) Monifavet, France. The genetic stocks are homogenous for most genetic attributes. Field experiments took place at the research field of Horticultural Training Research Institute (HORTI) Arusha (lat 4.8˚S long 3.7˚E; alt. 1290 m) with annual rainfall of 700 to 1000mm; soil type was clay loam with a P^H of between 6.0 and 6.5. Experimental plots were laid out in a randomized complete block design with three replications. Each plot consisted of a double row plot of 7 meters long and 0.75m between rows. Seedlings were raised in multipot seedling trays for four weeks, thereafter transplanted to the sides of the ridges at 0.45 m between plants. Plants were fertilized with NPK (20-10-10) at the rate of 90KgN/ha, 45Kg P_2 O_5/ha and 45Kg K_2O/ha. Urea fertilizer was applied at the rate of 120KgN/ha in three splits i.e. one week after transplanting, at flowering and three weeks thereafter. Ridomil WP (fungicide) was sprayed against damping off in the field at the rate of 20g/15 liters of water 12 days after transplanting. Selecron EC (insecticide) was applied 2 weeks after transplanting at the rate of 20mls/20 liters of water to control insects. The experiment was furrow-irrigated every two days for the first two weeks after transplanting, then once a week thereafter. Weeding was carried out manually and frequently with hoes to maintain weed-free plots.

Every parts of the plant were sampled at all stages of growth, presumably because different parts of the genome affect different suites of traits at different times. Two-state qualitative traits e.g. presence or absence of pubescence/prickles was coded as binary. Ordered multistate qualitative traits were coded as series of discrete states. Morphological scoring was done using according to the AVRDC and Biodiversity descriptors with modifications. Morphological traits were scored at flowering during this time maximum development of vegetative parts ought to have taken place (Karamura and Karamura, 1995). Morphological traits were measured on fifteen plants (five plants per replicate). Traits related to colour (pigmentation gene) was scored using the standard Royal Horticultural Society colour chart. Data was submitted for two methods of multivariate analyses (Principal Component Analysis (referred hereafter as PCA) and Cluster analysis), using PROC-PCA procedure of SAS (1998). Dendogram was constructed, based on distance matrix, using the average linkage between group methods often aptly called UPGMA (Unweighted Pair Group Method of Analysis), with squared euclidean phenotypic distance option Ward's (Sokal and Michener 1958) as grouping criteria, using SPSS version 16. 0.

Results and Discussion

To determine the patterns of variation and to detect the structure in the relationships between morphological traits, PCA was carried out considering all of the 13 variables simultaneously (Table 4).The first to third principal component axes had eigenvalue greater than 2.0 and altogether accounted for 68% of variation (Table 1). The first principal component axis had high eigenvalue (5.08) and accounted for 30% of total variation, it depicts primarily variation associated with receptacle pigmentation, and marked moderate and positive loading charged on leaf and petiole pubescence. Five traits (flower pigmentation, sepal pigmentation, receptacle pigmentation, peduncle pigmentation and fruit cross section) recorded moderate and positive loading though leaf pubescence, petiole pubescence, days to 50% flowering and fruit colour at commercial ripeness showed negative coefficients. The second principal component axis illustrated that both leaf vein and leaf mid rib pigmentation marked negative coefficients, while stem pigmentation and fruit apex shape showed moderate and positive weights. The third principal components axis accounted for additional 15% of variation and showed high contribution of petiole pigmentation, seed colour and fruit colour at commercial ripeness to variability on this axis. They marked moderate and positive weights, though charged on fruit colour distribution. Within *Solanum macrocarpon* receptacle pigmentation (purple) showed high discriminatory ability, this observation confirmed conclusion reached by Lester and Niaken (1998) and

Prohens *et al*. (2005).This trait could be of importance to curators to focus on greater part of *S macrocarpon* collection,. Plant breeders and seed companies may use this trait as morphological marker during selection purposes.

The plot of PC1 by 2 (Fig.1) showed that the first quadrant accommodated four accessions (Acc 55, 53, 54 and 47) sourced from Ghana, Zimbabwe, Mauritania and Burkina Faso. With exception of Acc 55 which contributed moderately to dispersion in the first quadrant, other entries showed very low contribution. Dispersion of accessions in this quadrant is sequel to discriminatory ability of receptacle and peduncle pigmentation and flower size. The second quadrant accommodated four accessions (Acc 50, 45, 51 56), fairly equal distant, dispersion in this quadrant showed discriminatory ability of leaf lobbing and leaf mid rib pigmentation. Two accessions (Acc 46 and 56) from Unknown and Cameroun are located in the third quadrant, dispersion in this quadrant is associated with discriminatory power of petiole pigmentation and fruit colour at physiological maturity. In the fourth quadrant Acc 57 was widely separated from others, traits of discriminatory importance in the fourth quadrant are fruit apex shape, fruit colour at commercial ripeness and stem pigmentation.

Morphological traits were important in the assignment of entries into three distinct clusters at 20% distance (Figure 2). Cluster 1 was divided into two sub cluster ('a' and 'b'), sub cluster 'a' comprised Acc 53 and 54, Acc 50 and 51, they are tightly grouped and linked to Acc 52, they are related for petiole, sepals, peduncle pigmentation (purple), flower colour and fruits colour (purple) at commercial ripeness. On the other hand, Acc 50 and 51 are characterized by strong leaf lobbing, pigmented (purple) leaf vein and petiole. On top of that, fruits are characteristically cream at commercial harvest and fruit colour distribution was uniform. Sub cluster 'b' is linked to 'a' and are similar for purple pigmentation on the petiole, sepals and peduncle. The second cluster comprised 4 accessions, Acc 47 and 48 are similar for lobed leaves and non-pigmented petiole, pigmented sepals (purple), pigmented fruits (purple) at commercial ripeness and circular fruits. The second cluster is characterized by accessions with fairly grooved and cream fruits at commercial ripeness. Acc 56 and 57 in the third cluster are similar for pigmented (purple) petiole and non-pigmented (green) sepals, receptacle and peduncle and circular fruits. Dispersion of the accessions on the plot of PC 1 by 2 (Figure 1) is consistent with groupings on the dendrogram for Acc 45 and 55, Acc 51 and 52, Acc 47 and 48 and Acc 56 and 57. This correspond to low and moderate variability and phenotypic plasticity. Grouping on the dendrogram is consistent with geographic heterogeneity, lack of clustering according to geographic provenance implies that accessions from different locations in Africa and Asia are not significantly different indicating no association between geographic and morphological marker. A similar pattern was also recorded among Uganda, Indonesia and European *Solanum scabrum* (Olet, 2004). These accessions could be maintained to preserve their genetic variability for future genetic studies, and could be used to guide the constitution of the breeding population for future crop improvement. Phenotypic plasticity was evident among accessions; this is similar to overlap among the accessions for morphological traits. In this study, morphological variability and similarity among *S. macrocarpon* accessions beyond geographical limit is similar to previous reports of Polignano *et al*. (2009) in *Solanum melongena* and *Solanum Macrocarpon*. For selection pollen parent for specific trait or multiple traits emphasis should be at the population level rather than geographic location. In this study, twelve accessions were sourced from Africa and one accession (Acc 45) from Malaysia, though the latter grouped with Acc 55 from Ghana. This suggest a possibility that seeds of Acc 45 might have been transported from the West coast of Africa during the transhumance trade in the 19th century or during exchange of germplasm between Africa and Asia. However, a robust molecular technique (AFLP) is required to substantiate this finding. Low variability among *S. macrocarpon* lends weight to findings reported by Bukenya and Hall (1987) and Polignano *et al.* (2009) based on agronomic and fruit quality traits. Low variability in this specie may be attributed to genotypic difference and environmental factors. In another study Bukenya and Caraso (1994) had noted that accessions of *S. macrocarpon* are less

morphologically diverse than *S. aethiopicum* and *S. melongena.*

Reference to the grouping on the dendrogram (Fig. 2) Acc 53 and 54 are related for brown seed colour, cream fruit colour at commercial harvest, pigmented (purple) stem and receptacles and violet petals. On the other hand, strong leaf lobbing, violet petals and grey seed colour are important in grouping Acc 50, 51 and 52. Further Acc 47 and 48 are characterized by purple pigmentation on the stem, leaf mid rib and receptacle, and cream seed colour. Acc 45 and 55 showed relatedness for brown seed colour and absence of pigmentation on the stem and receptacle. Although Acc 56 and 57 had cream fruits at commercial ripeness stage, the fruit epidermis turned brown colour at physiological maturity. Still in cluster 3, Acc 46 and 49 had green fruit epidermis at commercial ripeness; they turned orange in colour at physiological maturity. Fruit pigmentation showed high discriminatory ability in Solanum species (Collonnier et al. 2001, Kashyap et al. 2003, Daunay et al. 2001 and Frary et al. 2007). The color differences in the fruits are basically due to two color pigments' and their effects on appearance and are controlled by more than one gene (Frary et al. 2007). In general purple pigmentation on the receptacle contributed to cluster constellation. Also, since receptacle pigmentation, stripped fruits are located in different clusters, high heritability and heterosis should be expected in subsequent generations. The discriminatory ability of receptacle pigmentation among *S. macrocarpon* showed its importance in delimiting accessions in this species.

Conclusion and Recommendation

Among the accessions of *Solanum macrocarpon*, pigmentation (on the receptacle, flower, sepals, and peduncle) and fruit cross section showed high discriminatory ability. Dispersion (biplot) and grouping (dendrogram) correspond to low and moderate morphological variability and plasticity. These accessions could be conserved to preserve their genetic variability for future genetic studies.

Table 1: Eigen values, variation and vectors for the first six principal components axes for 30 morphological traits among 13 accessions of *Solanum macrocarpon* in Tanzania

Traits	Prin 1	Prin 2	Prin 3	Prin 4	Prin 5	Prin 6
Stem pigmentation	0.17	0.39	0.02	0.02	-0.04	0.41
Leaf pubescence	**-0.34**	0.07	0.08	0.11	0.51	0.03
Leaf lobbing	0.20	-0.14	0.25	-0.36	0.44	0.12
Leaf vein pigmentation	0.12	**-0.46**	0.13	0.07	0.05	0.18
Leaf mid rib pigmentation	0.13	**-0.43**	0.02	-0.04	-0.06	0.42
Petiole pigmentation	-0.11	-0.13	0.54	-0.10	-0.08	-0.24
Petiole pubescence	**-0.34**	0.07	0.08	0.11	0.51	-0.03
Flower pigmentation	**0.31**	0.04	0.14	0.19	0.17	-0.28
Sepal pigmentation	**0.34**	0.20	-0.01	0.12	0.14	0.43
Receptacle pigmentation	**0.36**	0.13	0.11	0.23	0.21	0.22
Peduncle pigmentation	**0.34**	0.03	0.25	0.03	0.27	-0.33
Fruit apex shape	-0.10	**0.34**	0.09	-0.10	-0.06	0.12
Fruit colour at commercial ripeness	-0.18	0.10	0.39	0.43	-0.20	0.09
Fruit colour at physiological maturity	-0.13	**-0.31**	-0.08	0.51	-0.009	0.13
Fruit cross section	**0.30**	0.26	-0.20	-0.11	0.04	-0.08
Fruit colour distribution	0.17	0.02	-0.33	0.48	0.22	-0.27
Seed colour	0.15	0.20	0.44	0.17	-0.12	0.07
Eigen value	5.09	3.77	2.62	1.63	1.29	1.04
Proportion	0.30	0.22	0.15	0.10	0.07	0.06
Cumulative (%)	0.30	0.52	0.68	0.77	0.85	0.91

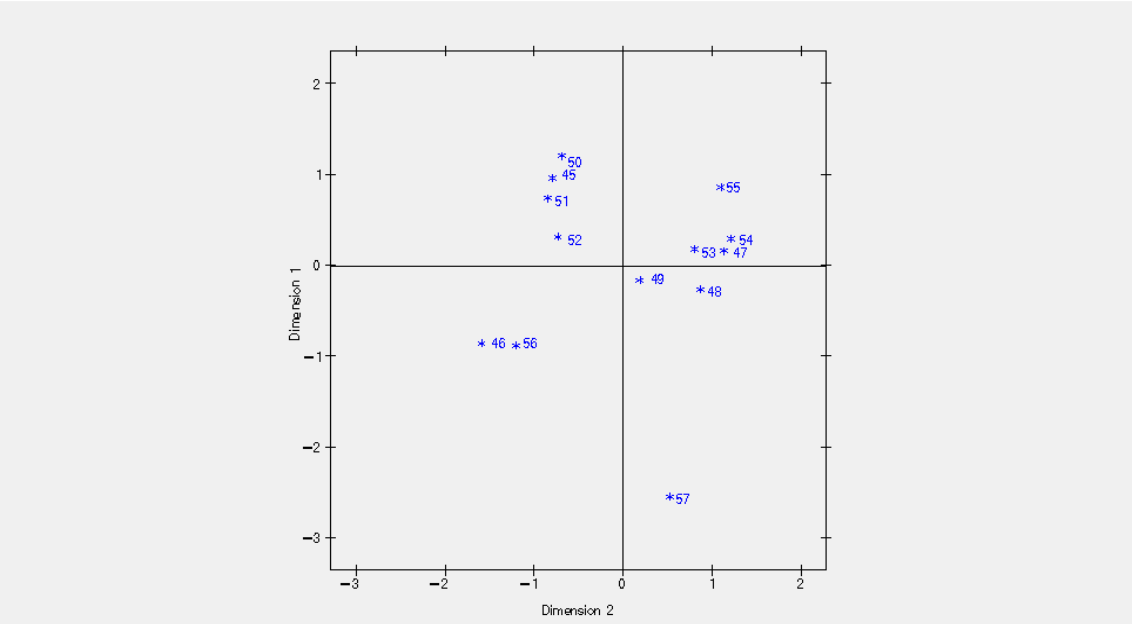

Figure 1: Plot of the first two principal components axes showing spatial distribution of 13 accessions belonging to *Solanum macrocarpon* based on morphological descriptors

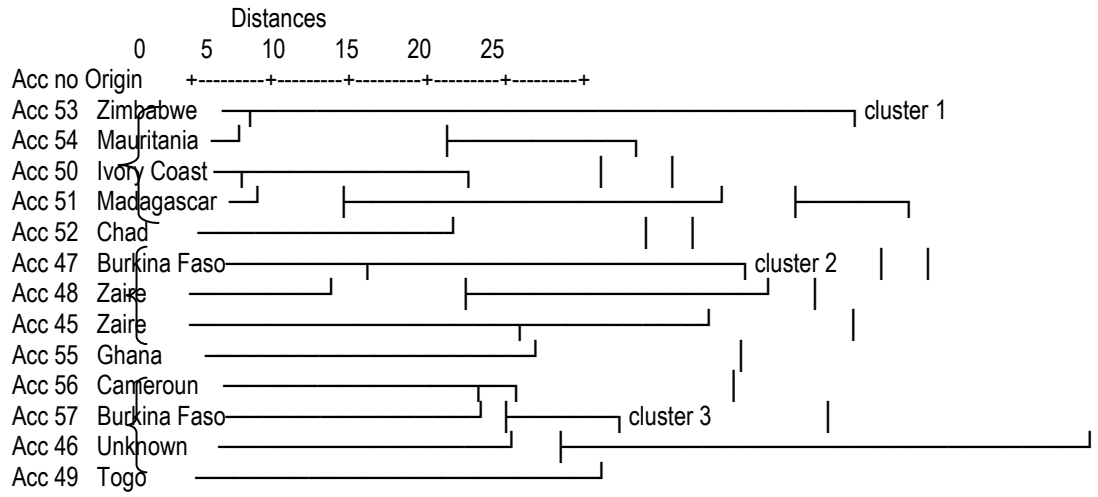

Figure 2: Dendrogram produced for 13 accessions from *Solanum macrocarpon* derived from UPGMA clustering of correlation coefficients for 17 morphological traits by Euclidean distance and Ward's method

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