

MORPHOLOGICAL CHARACTERISATION OF TWO ENDEMIC SPECIES OF *GOMPHOCARPUS* (MOBYDICK) IN KENYA

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Abstract

The genus *Gomphocarpus* comprises 25 – 32 species that occur in Africa and the Arabian Peninsula. In Kenya, two common species; *Gomphocarpus physocarpa* and *Gomphocarpus fruticosus* are commercially cultivated as a cut flower for its inflated green bolls. As a comparison, *Gomphocarpus physocarpa* has larger and more rounded bolls whereas *Gomphocarpus fruticosus* have small bolls with a sharp pointed end. However, these two species can not be differentiated as they easily hybridize. No precise information on morphological or molecular characterization is available locally. The objective of the study was morphological characterization of commercial *Gomphocarpus* species grown in Kenya. *Gomphocarpus* germplasm was collected from Juja farm, Thika, Molo, Narok and Chumvi in Machakos. The experiment was laid out as randomized complete block design with three replications and five treatments per block. The collections were characterized for morphological diversity complimenting with principal component analysis (PCA) and cluster characters using the XLSTAT statistical software. The results showed no morphological variation on qualitative characteristics of leaf, growth habit and stem length. The coefficient of variations (CV) and standard deviations (SD) for all qualitative traits were zero. Boll length was positively correlated to boll weight with a factor of 0.355. Leaf length was inversely correlated to leaf width with a factor of negative 0.064. A PCA based on morphological traits of boll weight and length consistently separates populations of *Gomphocarpus physocarpa* and *Gomphocarpus fruticosus* and reveals a close relationship between them. All the qualitative characteristics of leaf colour, leaf shape, boll shape and flower shape were all clustered at the origin, displaying zero variations. The hierarchical clustering dendrogram revealed a 99.9% similarity among *Gomphocarpus* collections. The study showed that *Gomphocarpus* characteristics did not reveal any significant divergence in morphological qualitative traits observed. This could be an indication of low reproductive isolation in the collections. The two lines also hybridize, creating intermediate forms. Probably, there is need for complimenting similar work with other techniques such as DNA genetic markers to further accurately characterize *Gomphocarpus* germplasm existing in Kenya.

Key words: Characterisation, cut flower, mobydick, morphology

1.0 Introduction

The term *Asclepias* is used to refer to milkweed species grown in American continent and other Western worlds, while *Gomphocarpus* refers to *Asclepias* species found in Africa and Arabian continents (Hodkiss, 2009). The genus *Gomphocarpus* comprises 25 – 32 species that occur in Africa and Arabian Peninsula. Ten other species occur in southern Africa.

Morphologically, *Gomphocarpus* plants demonstrate an erect growth habit with multiple stems of 1 - 3 m in height. The leaves are arranged in pairs and opposite each other along the stem. The leaves are also dull green in colour. Flowers are borne in simple 6 - 10 flowered umbels, each flower being suspended on a pedicel. Flowers are often brightly coloured, with a characteristic five-fold symmetry. The pollen are found in pollinia or 'pollen sacs' rather than being in individual grains or tetrads typically for most plants (Parsons and Cuthbertson, 1992). When the follicles ripen, they split open and the seeds attached to the floss are blown away by wind (Ramanujan and Krishna, 2008).

In Kenya two common species; *Gomphocarpus physocarpa* and *Gomphocarpus fruticosus* are commercially cultivated as a cut flower for its inflated green bolls. As a comparison, *Gomphocarpus physocarpa* has larger and more rounded ornamental seed bolls than *Gomphocarpus fruticosus* whose inflated seed bolls have sharp pointed end and are covered in short, stout hairs. These two species may be easily confused with each other. To estimate levels of morphological variation among natural and commercial populations of *Gomphocarpus* species where the two species coexist with morphologically intermediate plants are almost impossible. No precise information on morphological and molecular characterization is available; therefore, it is difficult to differentiate the *Gomphocarpus* species from different regions in Kenya (Notten, 2010). However, unimproved commercial lines grown in Kenya are a mix of the two cultivars as the two species can hybridize. Since breeding or varietal selection is non-existent, the cultivars grown locally produce bolls which include small green or big purple bolls. Currently, the small green bolls are preferred by buyers and are fetching better prices on the export market (Waiganjo *et al*, 2009). Since no breeding programs or comprehensive morphological characterization are known to exist for *Gomphocarpus* grown as an ornamental plant in Kenya, the use of seeds as planting material leads to character segregation.

This manifests itself to likelihood of non-uniformity of bolls, internode length and plant height. In this project, *Gomphocarpus* germplasm was collected from Juja farm, Thika, Molo, Narok and Chumvi in Machakos and grown in a single uniform environment at Jomo Kenyatta University of Agriculture and Technology. The main objective of the study was to characterize the commercial *Gomphocarpus* grown in Kenya using morphological traits of stem, petiole, leaf, flower, fruit and seed. The

information obtained could form baseline data for selection of high yielding plants with optimal boll formation needed to improve its market value.

2.0 Materials and Methods

The experiment was laid out at Jomo Kenyatta University of Agriculture and Technology farm. The *Gomphocarpus* seeds were planted in trays containing a mixture of sand, cattle manure and forest soil in the ratio 1:2:3, respectively. The trays were covered with wetted newspapers to avoid drying out until the seeds started showing signs of germination. Seven days after germination, the seedlings were transferred into polythene bags containing a similar media mix as before. They were ready for transplanting in the field after six weeks. The seedlings were transplanted on raised beds measuring 1 meter in width with a path of 0.5 meter between adjacent beds. The spacing used for plants was 75cm x 60cm. The design used was randomized complete block design with three replications and five treatments per block. The treatments were the *Gomphocarpus* collections from Juja farm, Thika (Oj Donjo Sabuk), Molo, Narok and Chumvi in Machakos. Other practices for *mobydick* crop culture were followed. In the study, the International Plant Genetic Resources Institute, IPGRI (1997) characterisation descriptors for tea (*Camellia sinensis*) were used as a guideline for data collection.

The plant characters used in *Gomphocarpus* morphological characterization were features of the stem, leaves, flowers, fruits and seeds. Data collection involved qualitative traits on these descriptors such as stem, fruit, flower and seed colour. The colour of flowers and leaves were also recorded as determined by visual appraisal on fully developed open flowers, immature and mature leaves. The plant growth habit was also studied as a morphological feature after tipping of the plants. Branch angle, leaf shape, leaf apex shape, leaf apex habit, leaf base shape and leaf margin were also considered on mature leaves. Plant height, stem diameter, internode length, and leaf length are quantitative traits which are highly influenced by environment. Internode length was the distance between the 5th and 6th leaves from top of a flush growth. The fruit length was recorded at the longest part, using an average of 10 fruits. The fruit diameter was measured from the broadest part with an average of 10 fruits. Leaf length was measured from the base of midrib to tip, for an average of 5 leaves.

3.0 Data Analysis

The five *Gomphocarpus* collections were subjected to analysis of morphological diversity. The variables were also used to perform principal component analysis (PCA) using the XLSTAT 2008 statistical software. The principal components obtained were used to perform cluster analysis with Eigenvalues running from 1.243 to 3.966 using the Neighbor Joining method (Nei, 1973) and Euclidian average distance. The Eigenvalues 1.243 and 3.966 were part of the results in this study. The

figure 3.966 represents the Eigenvalue corresponding with the third principal component. Table 2 covers up to the first three principal components only.

4.0 Results

4.1 Morphological Diversity

The results of *Gomphocarpus* germplasm analysis displayed no variation in the morphological qualitative characteristics (Table 1). These include leaf shape, growth habit, stem type, leaf colour, leaf apex, leaf habit, leaf base shape, leaf margin and leaf arrangement. The coefficient of variations (CV) and standard deviation for all the qualitative traits was zero. Hence, the variance which is a square of standard deviation is also equal to zero (Table 1).

Table 1: Characterization results of *Gomphocarpus* morphological traits using XLSTAT (2008) statistical package

Organ	Qualitative Trait	Resultant Trait	Standard Deviation	Variance	
Stem	Growth habit	Erect shrub	0.000	0.000	
	Stem type	Multi-stem	0.000	0.000	
	Stem colour	Georgian green	0.000	0.000	
	Bark texture	Smooth	0.000	0.000	
	Latex colour	White	0.000	0.000	
Petiole	Colour of immature petiole	Caterpillar (12 E 53)	0.000	0.000	
	Colour of mature petiole	Greenage (12 D 43)	0.000	0.000	
Leaf	Leaf colour	Greenage (12 D 43)	0.000	0.000	
	Leaf shape	Lanceolate	0.000	0.000	
	Upper leaf surface	Smooth	0.000	0.000	
	Leaf apex shape	Acute	0.000	0.000	
	Leaf apex habit	Erect	0.000	0.000	
	Leaf base shape	Cuneat	0.000	0.000	
	Leaf margin	Entire	0.000	0.000	
	Leaf angle	Acute	0.000	0.000	
	Leaf venation	Pinnate/network	0.000	0.000	
	Leaf type	Simple	0.000	0.000	
	Branch angle	Acute	0.000	0.000	
	Leaf attachment to stem	Petiolate	0.000	0.000	
	Leaf arrangement	Opposite	0.000	0.000	
	Flower	Number of sepals	5 sepals	0.000	0.000

	Number of petals	5 sepals	0.000	0.000
	Corolla colour	Soft white (10 B 15)	0.000	0.000
	Corolla forms	Rotate	0.000	0.000
	Position of the ovary	Epigynous	0.000	0.000
	Attachment of anthers	Adnate	0.000	0.000
	Cohesion of stamens	Synandrous	0.000	0.000
	Inflorescence	Umbel	0.000	0.000
	Flower aestivation	Imbricate	0.000	0.000
Fruit	Immature boll colour	Greenage (12 D 43)	0.000	0.000
	Mature boll colour	Caterpillar(12 E 53)	0.000	0.000
	Placentation	Axile	0.000	0.000
	Fruit shape	Ovate	0.000	0.000
	Fruit apex	Mucronate	0.000	0.000
	Colour of silk hairs	white	0.000	0.000
Seeds	Seed colour	Rusty red (04 D 45)	0.000	0.000
	Seed texture	Smooth	0.000	0.000

4.2 Principal Component Analysis

Principal component analysis was used to analyze a number of quantitative traits in *Gomphocarpus* (Figures 1 and 2). The results of the principal component analysis show that the first two principal components accounted for 82.248% of the total variance. The results also display that the first principle component accounted for a fairly large amount of the total variability, which is 56.660%. The second principle component accounted for 25.588% and the third principle component comprised of 17.752%. Each succeeding component accounted for progressively smaller amounts of variance (Table 2).

Table 2: Principal component analysis (PCA) for the first three factors of *Gomphocarpus* germplasm

Principal component	Eigenvalue	Variability (%)	Cumulative (%)
PC1	3.966	56.660	56.660
PC2	1.791	25.588	82.248
PC3	1.243	17.752	100.000

The first two principal components explain a cumulative variability of 82.248%. The further away the variants are from the origin, the stronger their impact on variability (Figure 1). Fruit length was positively correlated to boll weight. The correlation factor between boll length and boll weight was 0.355. Leaf length is inversely correlated to leaf width since these two are directly opposite each other in reference to the origin (Figure 1). The correlation factor between leaf length and leaf width was -0.064.

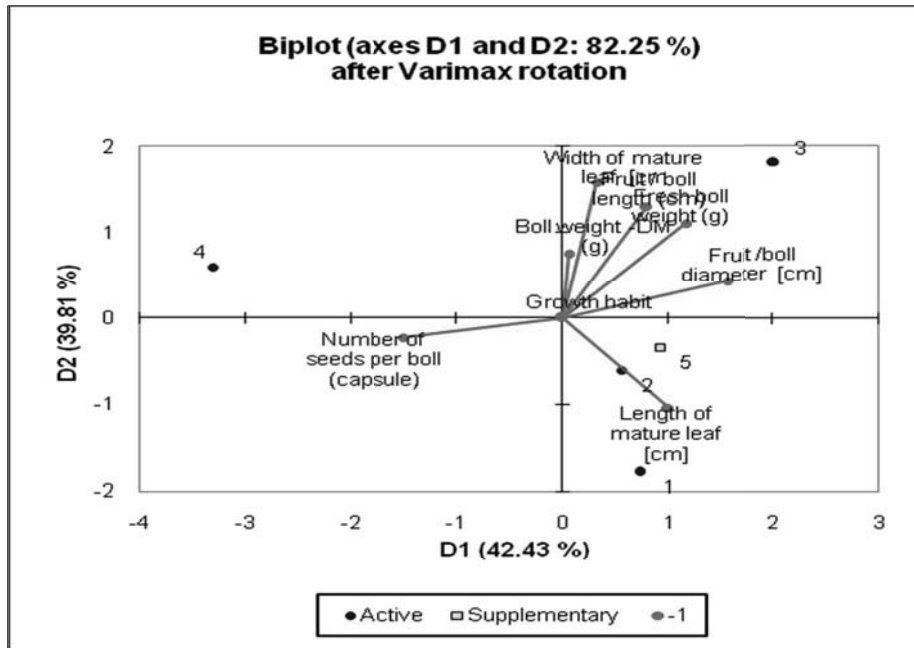


Figure 1: Correlation among traits associated with the first and second Principal Components. The closer the attributes are to each other in the PCA plot, the higher the correlation or the smaller the angle between the attributes, the higher the correlation. In the figure, 1, 2, 3, 4 and 5 stand for Narok, Molo, Thika, Juja and Chumvi *Gomphocarpus* collections.

Figure 1 shows correlations among traits put in three main clusters of characters. In the first cluster, there were traits associated with fruit / boll diameter, fresh boll weight, dried boll weight and mature leaf width. The second cluster comprised of traits associated with length of mature leaf; the third cluster comprised characters related to number of seeds per boll; the third cluster comprised of line 4 exclusively. All the qualitative characters such as leaf colour, leaf shape, boll shape and flower shape clustered at the origin, displaying zero variations within the *Gomphocarpus* germplasm in respect to these characters (Figure 2).

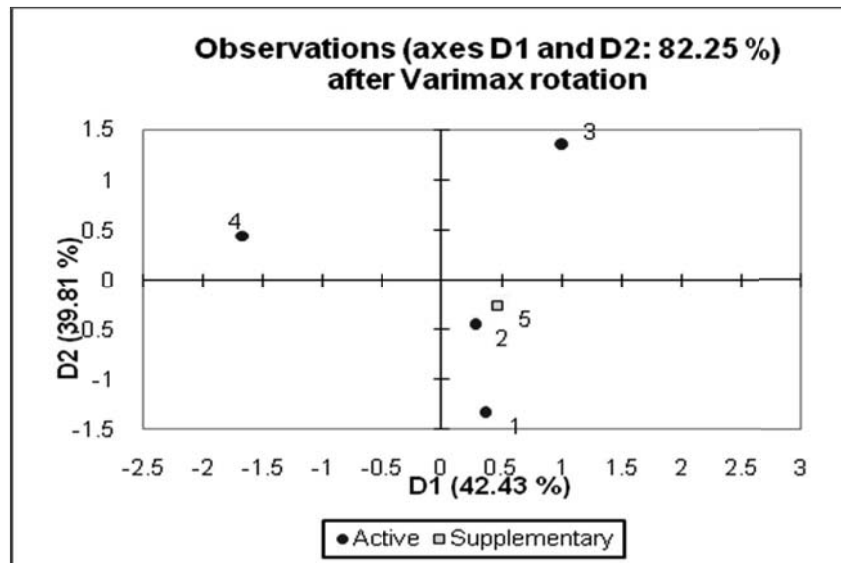


Figure 2: Distribution of variates in principal components PC1 and PC2. In the figure, 1, 2, 3, 4 and 5 stand for Narok, Molo, Thika, Juja and Chumvi *Gomphocarpus* collections.

The PCA plot (Figure 2) indicates that principal components one and two (PC1 and PC2) accounted for 42.43% and 39.81% respectively, giving 82.24% (cumulative) of total variation. The variations arise from quantitative features, such as plant height, fruit diameter, leaf length and width. These features change with environment and are non significant across all *Gomphocarpus* collections.

4.3 Cluster Analysis

The agglomerative hierarchical clustering dendrogram illustrates the relationship among the *Gomphocarpus* collections. The cluster analysis separated the *Gomphocarpus* collections into characters giving Euclidean similarity distance values. The morphological relationship among the *Gomphocarpus* collections ranged from the Euclidean similarity values 0.999977 to 0.999997 with an average of 0.999987 (Figure 3). The dendrogram was divided into three main clusters on the basis of the major morphological characters (Figure 3). At higher similarity levels, the above clusters were further divided into smaller sub-clusters. This occurs at the bottom of the dendrogram. The dendrogram reveals 99.9 % similarity among *Gomphocarpus* collections used in the study.

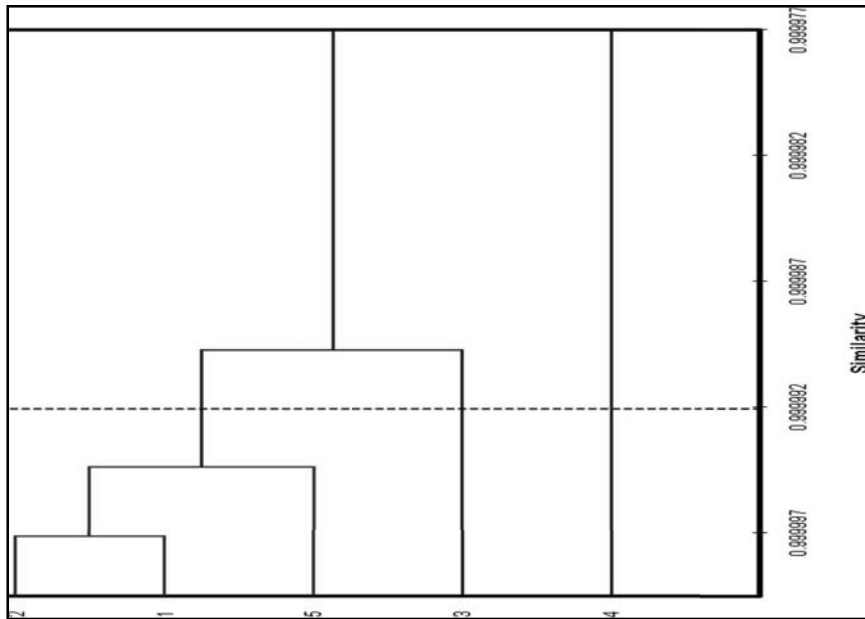


Figure 3: Clustering dendrogram constructed based on morphological characters of *Gomphocarpus* cultivars using the Neighbor Joining method (Nei, 1973) and Euclidian average distance; there are three clusters: the first cluster consists of collections 1, 2 and 5, while cultivars 3 and 4 in second and third clusters respectively. Five collections were used: 1= Narok *Gomphocarpus*, 2=Molo *Gomphocarpus*, 3=Thika *Gomphocarpus*, 4= Juja *Gomphocarpus*, 5= Chumvi (Eastern Kenya) *Gomphocarpus*.

5.0 Discussion

All the morphological qualitative traits including leaf shape, growth habit, position of ovary and number of sepals did not show significant variation in the *Gomphocarpus* collections used in the study (Table 1). *Gomphocarpus* species leaf shape, for instance, were lanceolate for all the collections which confirms similar report by Agrawal *et al.*, (2009).

It is important to decide how many principal components should be retained in order to account for most of the variations. In some circumstances, the last few rather than the first few principal components (PCs) are of interest. The decision could be done by taking the cumulative percentage of total variation. In this study, the first two principal components accounted for a total (cumulative) variation of 82.248 % (Table 2). This falls within a sensible cutoff range of 70% to 90% cumulative percentage of total variation (Jolliffe, 2002).

The study used colour to differentiate the various traits in *Gomphocarpus*. However there was no variation on this trait in all *Gomphocarpus* plants used. Colour traits, shape of central lobe and branching were important salient characters used by farmers to identify varieties (Elias *et al.*, 2001). They reported in their research with

cassava that colour variables played a crucial role in differentiating varieties. The correlation between *Gomphocarpus* mature leaf length and fruit diameter obtained from the results was 0.451 which agrees with Widodo *et al.*, (2009) and Ocampo *et al.*, (2006b) similar correlation figure of 0.583 obtained in *Syzygium* species. Since leaf size correlated positively with fruit size, equilibrium between leaves and fruits should be reached in order to produce adequate size of fruits up to a certain point when leaf expansion is no longer a limiting factor to fruit size (Widodo *et al.*, 2009).

The knowledge of correlations among characters is essential in designing an effective breeding programme. There could be several reasons for using correlations as a means for indirect selection. One of the situations is when the main character is expressed late. In some instances, measurement of the indirect character is relatively much easier than for the direct character. In such cases, identification of highly correlated characters is more commendable.

Gomphocarpus collections from Narok, Molo and Chumvi were closely related in their length of mature leaf. These collections were clustered together in relation to this morphological feature. The *Gomphocarpus* collections from Thika and Juja were quite distinct from the rest. The *Gomphocarpus* collected from Thika which was found in its own cluster seemed to have unique characteristics associated with width of mature boll, dry boll weight, and length of fresh bolls and fruit diameter (Figure 1). All the quantitative traits used in *Gomphocarpus* study were insignificant ($p=0.05$).

Results in this study reveal that *Gomphocarpus* quantitative morphological traits used have limited variation. Thus, it is essential to involve genetic markers to be able to show if any variations exists in *Gomphocarpus* at genetic level.

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References

- Agrawal, A. A., Fishbein, M., Jetter, R., Salminen, J. P., Goldstein, J. B., Freitag, A. E. and Sparks, J. P. (2009). Phylogenetic ecology of leaf surface traits in the milkweeds (*Asclepias spp.*): chemistry, ecophysiology, and insect behavior. *New Phytologist*, **183**, pp. 848–867.
- Elias, M., McKey, D., Panaud, O., Anstett, M.C. and Robert, T. (2001). Traditional management of cassava morphological and genetic diversity by the Makushi Amerindians (Guyana, South America): Perspective for on-farm conservation of crop genetic resources. *Euphytica*, **120**, pp. 143-157.
- Hodkiss, R.J. (2009). *Asclepias* – International Asclepiad Society Journal; *The succulent plant*, **106**, pp. 24-58.
- International Plant Genetic Resources Institute (IPGRI). (1997). Descriptors for Tea (*Camellia sinensis*). Rome, Italy.6.
- Jolliffe, I.T. (2002). Principal Component Analysis, 2nd ed. Springer, 2002.
- Ocampo, J P, D’Eeckenbrugge G C, Bruyère S, D E Bellaire L. L, Ollitrault P. (2006). Organization of morphological and genetic diversity of Caribbean and Venezuelan papaya germplasm. *Fruits*, **61**, pp. 25-37.
- Parsons, W.T. and Cuthbertson, E.G. (1992). Noxious weeds of Australia. (Inkata Press, Melbourne /Sydney), pp. 179-182.
- Nei, M. (1973). Analysis of gene diversity in subdivided populations. Proc. Natl. Acad. Sci. USA, **70**, pp. 3321 – 3323.
- Notten, A. (2010). Kirstenbosch National Botanical Garden. *Gomphocarpus physocarpa* (E.Mey) Schltdl. South Africa.
- Ramanujan, K. (2008). Discoveries: Milkweed evolves to shrug off predation. Northern Woodlands; Centre for Northern Woodlands Education, **15(4)**, pp. 56.
- Waiganjo, M.M., Gikaara, D. N., Kamau, E. and Muthoka, N. M. (2009). Domestication of indigenous ornamentals and the crop production challenges in *mobydick*, *Asclepias sp.* in Kenya. *Acta Horticulture (ISHS)*, **813**, pp.79-86
- Widodo, Y and Sumarno. (2009). Correlations between Degree of Petal Fusion, Leaf Size and Fruit Size: A Case in *Syzygium* (Myrtaceae), *Biodiversities*, **10(3)**, pp.120-123.