Journal of Natural Sciences Research www.iiste.org
ISSN 2224-3186 (Paper) ISSN 2225-0921 (Online)
Vol.7, No.6, 2017

Morphometric Study of *Senna didymobotrya* (Fresen.) H. S. Irwin and Barneby in Kenya

Pascaline Jeruto*[1] Peter Arama[2] Beatrice Anyango[3] Regina Nyunja[3] Catherine Taracha[4] Sylvia Opiyo[5]

1. University of Eldoret, School of Sciences, P.O BoX 1125-30100 Eldoret
2. Rongo University, School of Agriculture, Natural Resources and Environmental Sciences (SANRES)
3. Jaramogi Oginga Odinga University of Science and Technology, School of Biological and Physical Sciences
4. Kenya Agricultural and Livestock Research Organization, Kabete, Nairobi
5. Maseno University, Chemistry Department

Abstract

Herbal medicine has been used for many years and it remains widespread in developing countries while the use of complementary alternative medicine is increasing rapidly in developed countries. *Senna didymobotrya* is important for its medicinal use among many communities to treat a wide range of ailments. The study examines the patterns of morphological variation and phenetic relationships among 39 populations of *S. didymobotrya* in Kenya using17 quantitative and 17 qualitative characters. A survey was carried out in Siaya, Kisumu, Nandi and Nakuru Counties to collect *S. didymobotrya* plants. Results on morphometric analysis indicated that the standard deviations were highly significant when all quantitative characters were considered. Stem diameter, plant height and stem height had the highest standard deviation of 13.14, 11.74 and 11.15 respectively. Leaf length and Internodal length had the lowest standard deviations of 0.18 each. Principal component analysis indicated that the plant height, stem height, stem diameter, habitat, pod length, pod width and number of seeds accounted for a cumulative percentage of 70% and above of the cumulative variance in a lineal combination of parameters hence can be used to separate the populations. Correlation matrix of the morphological characters indicated a high positive correlations between inflorescence length of basal stalk and inflorescence length (0.83), stem height and plant height (0.62), leaflet width and leaflet length (0.56), leaflet number and leaf number (0.54), pod width and pod length (0.52). The cluster analysis dendrogram placed the plants in four major clusters; Cluster 1 had one plant accession PJ/NK/33 collected from Nakuru County. Cluster II had one plant PJ/KS/13 collected from Kisumu County. In Cluster IV also had one plant accession PJ/SI/2 collected from Siaya County. Cluster III consisted of 36 plants with close relationship (Plants accessions PJ/SI/4 - 10, PJ/KS/11- 18, PJ/NA/19 - 29, PJ/NK/30 - 32 and PJ/NK/34 – 39). The highest similarities and high variations in cluster I, II, and III results that call the relationship between them in question. An identification key has been constructed which, for the first time, can be used to assign herbarium specimens to their respective taxa. Further research should be carried out to collect more samples from other regions within the country and other parts of the world to understand the morphological variations sufficiently to circumscribe the taxonomical doubts on how many species and/or lineages that do exist due to the influence of habitat type on the morphological variation between populations and if other methods like molecular markers (isozymes or microsatellites) are employed ,to reveal patterns of genetic variability. The beneficiaries of this study are the herbalists, pharmacologists, researchers, patients and the general public.

Keywords: *Senna didymobotrya*, Morphometric analysis, Cluster analysis, PCA, Numerical taxonomy.

1. INTRODUCTION

Over the years, plants have been known to synthesize phytochemical compounds, that enable them manufacture their own food and protect them against predators, diseases, pest and insects (Ramakrishna and Ravishankar, 2011). With the upcoming economic crisis, it is approximated that around 70 - 80% of the world's population will rely on herbal medicine to meet their medical needs (Gairola *et al.*, 2010). Biological and pharmacological activities of phytochemical compounds depend on factors such as the ecological factors, age of the plant, species and method of extraction. Thus each plant has different chemical composition, toxicity and bioactivity (Rajakaruna *et al.*, 2002).

The genus *Senna* formerly *Cassia* have been extensively studied for their phyllogenetic diversity, phytoconstituents as well as for their biological activities. Out of 580 species of this genus distributed all over the world, just 46 species have been phytochemically investigated. Phytochemical work on this genus seems to be by no means exhaustive and there still remains a vital scope for study of active molecules (Ganapaty *et al.*, 2002). Emodin, chrysophanol and rhein are widely distributed throughout this genus which suggests that these compounds may be chemotaxonomic markers of the genus *Senna* (Ganapaty *et al.*, 2002).

Current changes in climate have prompted a number of studies to predict future changes in species distributions, ecosystem changes, effect on rare species, and effects on invasive species (Deduke *et al.*, 2012). Population genetic studies are important because they determine not only the degree of diversity but also how

that variation is distributed among regions and within populations. A lot of studies are focused on the diversity among individuals (accessions) and a few are centered on populations (Essilfie and Oteng - Yeboah, 2013). Globally, locations within which *Senna* was naturalized include Australia and parts of America. *S. didymobotrya* was introduced into tropical Asia and America as a fodder, green manure and cover crop, but now it is grown throughout the world as an ornamental (Sunarno, 1997). *S. didymobotrya* is native to tropical Africa (Bett, 2010). It is native in Angola, Ethiopia, Mozambique and Sudan. It was introduced as an ornamental plant into many tropical countries including the Comoros, Madagascar, Mauritius and South Africa (Sunarno, 1997). In South Africa, it has been invasive in grasslands, coastal scrubs, woodlands, road sides, wastelands and on river banks (Nyaberi *et al*., 2013). It was introduced, naturalized and became an invasive species in parts of Kenya, Uganda and Tanzania (Sunarno, 1997). It is also noted that *S. didymobotrya* is an invasive plant which occurs mainly along the roads, riverine/lake shores and fallow lands. These areas are converted to cultivated lands and thus results in destruction of the diversity of the species unknowly. It is hardly attacked by disease or pests (Ngule *et al*., 2013).

Bioactive compounds are also known to vary within one plant. Some plant parts can be more sustainably harvested than others (Harbone, 1984). It is therefore important to assess levels of expression of the phytochemicals in different plant parts. The knowledge gained will ensure possibility of targeted harvesting, conservation and utilization strategies of the species. Assessing the diversity of *S. didymobotrya* populations through morphometric analysis ascertains whether any of the known populations of *S. didymobotrya* are morphologically distinct from one another and whether any such distinction, if significant, reflects geographical range. This information is important for the assessment of the morphormetric taxonomy of the species. The wide medicinal applications of *S. didymobotrya*, it becomes imperative to investigate its phenetic diversity since this affects its phytochemicals and antimicrobial activity as medicinal resource in Kenya.

1.1 Taxonomic information of the genus *Senna* and its botany

The genus *Senna* formerly *Cassia* (*Senna* Mill.) belongs to the kingdom plantae, sub kingdom vividiplantae, infrakindom streptophyta (land plants), superdivision embryophyta, division tracheophyta (vascular plants), subdivision spermatophytina (seed plants/phanerogames), class magnoliopsida, superorder rosanae,order fabales, family Leguminosae (alt. Fabaceae) with three sub - families (Leguminosae, Caesalpinioideae and Cassiinae) (Soladoye *et al*., 2010a). It is a large, widespread, and diverse genus. This genus is native throughout the tropics, with a small number of species in the temperate regions. It comprises of about 580-600 species which are widely distributed throughout the world and is well known for its diverse biological and pharmacological properties (Cláudio *et al*., 2013). Of these, 250 species are found in the tropics (Soladoye *et al*., 2010 a) and fifty (50) species are known to be cultivated. Almost all species were at one time or another placed in *Cassia*, a close relative which until recent decades served as a "wastebin taxon" to hold all Cassiinae.

The genus *Senna* is characterized by a distinctive floral morphology and the presence of extra floral nectaries (EFNs) in numerous species. It has displayed a high diversity of habits, including herbs, shrubs, treelets, tall trees, and lianas, and has successfully colonized a wide range of habitats in different climates and latitudes (Marazzi *et al*., 2006). The genus *Senna* has showed shifting taxonomic boundaries that marked the history of traditional systematics which were best explained by the difficult taxonomic interpretation of morphological variation in *Senna*. For example, the high degree of specialization typical of the buzz - pollinated *Senna* flowers complicates the identification of traits that can be unambiguously used for taxonomic purposes (Marazzi *et al*., 2006).

Plate 1.1.1

Plate 1.1.2

Plate 1.1.3

Plate 1.1.4

Plate 1.1: *Senna didymobotrya* (1.1.1- Inflorescence, 1.1.2- Fruits {immature pods}, 1.1.3 –Roots and 1.1.4- Leaves).

1.1.1 Scientific classification

Kingdom: Plantae
Division: Spermatophyta / Angiospermae
Class: Magniolopsida
Order: Magniolopsidales / Fabales
Family: *Caesalpiniaceae / Fabaceae (*alt. *Leguminosae*)
Sub - family: *Caesalpiniodeae*
Tribe: *Cassieae*
Sub tribe: *Cassiinae*
Genus: *Senna*
Species: *Senna didymobotrya* (Fresen.) H. S. Irwin & Barneby
Synonyms: Cassia didymobotrya Fresen. (1839), *Cassia nairob(i)ensis* L. H. Bailey (1941); *Cassia verdickii* De Wild.; *Chamaesenna didymobotrya* Small (Sunarno, 1997)

1.1.2 Common and local names

Common names include; African senna, African wild sensitive plant, peanut butter cassia (Cherono and Akoo, 2011), peanut butter tree, popcorn cassia, popcorn senna and wild senna. It is also referred to as candelabra tree in English (Rehm, S., 1994) and Séné africain (French). It is called popcorn *Senna* because it smells like fresh cooked buttered popcorn when fingers are run through the leaves (Cherono and Akoo, 2011). In Mozambique, it is referred to as mudlayanhoka, nyocanyokani (Madureira *et al*., 2012) whereas in South African among the Vha - Vendas, it is called Tshiduwana. Different vernacular names are used by different ethnic communities in Kenya to describe the species. For example, the plant is called Murao / Kirao in Meru (Gakuubi and Wanzala, 2012), Senetwet in Nandi and Kipsigis (Jeruto *et al*., 2008), Senetwo in Pokot (Nyaberi *et al*., 2013) Mwino / Mwinu in Kikuyu (Njoroge and Bussmann, 2006 b), Owinu / Ovino / Obino - Luo, Ithaa / Muthaa in Kamba (Wagate *et al*., 2012) Osenetoi in Maasai, Lubino / Luvino - Luhya, Esletoi in Maasai, Mbinu / Mshua in Taita

and Atupa in Swahili (Kokwaro, 2009).

1.1.3 Botanical description

Senna species have yellow, nectarless flowers that offer pollen as a reward to their pollinators, usually large female bees of the genera *Xylocopa* (Marazzi *et al*., 2006). The heterantherous flowers of *Senna* generally have 10 stamens; the three adaxial stamens are typically staminodial, while the remaining seven, or fewer, are fertile. The fertile stamens are poricidal and differentiated into two sets: one set of four middle stamens (between the adaxial staminodes and abaxial stamens) which bees buzz to extract food pollen, and a second set of two or three (often longer) abaxial stamens, whose pollen is deposited on the bee's body during buzzing and transported to the stigma of other flowers (Marazzi *et al*., 2006). Furthermore, many species of *Senna* have assymmetric flowers, with the gynoecium deflected either to the left or to the right within the same inflorescence. This type of floral assymmetry is known as enantiostyly. Although monographic treatments explored the taxonomic utility of extra floral nectaries in *Senna*, little is known about their specific distribution, anatomy, and evolutionary significance in the genus (Marazzi *et al*., 2006).

S. didymobotrya is a shrub native to the tropics with small number of species found in temperate regions. It is native to Eastern and Central Africa which produces golden yellowish flowers with a distinct scent of peanut butter that opens from brown buds (Orwa *et al*., 2009). It is usually a several - stemmed shrub or small tree. It measures 0.5 to 5 (- 9) m tall (Nyaberi *et al*., 2013). Its branches are terete, striate, pubescent to villous and rarely subglabrous (Orwa *et al*., 2009).

The leaves are evergreen that open from brown buds. They are 14 - 50 cm long, pinnate with more than 30 leaflets (Sunarno, 1997). The leaves simply paripinnate, narrowly oblong - elliptical in outline, 10 - 50 cm long; stipules broadly ovate - cordate, 6 - 17 mm x 8 - 10 mm, acuminate, palmately veined, reflexed, tardily caducous; petiole terete, 1 - 8 cm long, rachis up to 40 cm long, both pubescent and glandular; petiolules up to 3 mm long; leaflets in 8 - 18 pairs, chartaceous, elliptical - oblong, 2 - 6.5 cm x 0.5 - 2.5 cm, 2 - 3 times longer than wide, base oblique, apex rounded but mucronate, pubescent to glabrescent, marginal vein distinct (Jeruto, 2009).

The flowers are grouped in 10 - 20 inflorescent. Flowers bright yellow in inflorescences; flower stalks long (Jeruto, 2009). Inflorescence are erect, axillary, 20 - 30 flowered, spike - like raceme, 10 - 50 cm long; peduncle terete, 5 - 8 cm long, pubescent; bracts broadly ovate, 8 - 27 mm x 5 - 15 mm, black green, at first imbricate and enclosing the flower buds. Bracteoles are absent; pedicel slender, 3 - 10 mm long, densely pubescent; sepals 5, subequal, oblong - obovate, 9 - 14 mm long, puberulous, green. There are five petals which are slightly unequal, at first incurved, later on more spreading, ovate to obovate, 17 - 27 mm x 10 - 16 mm, with a slender, about 1 mm long claw, glabrous, bright yellow, delicately veined; stamens 10, filaments shorter than anthers, anthers of 2 lower stamens 9 - 11 mm long, 3 upper stamens staminodial, anthers of 5 median stamens about 5 mm long; ovary and stipe velvety pubescent; style slender, glabrous, recurved, about 1 cm long; stigma punctiform (Kokwaro, 2009).

Orwa *et al*., (2009) described it's fruit as flat, 9 - 16 seeded pod, linear - oblong, 7 - 12 cm x 1.5 - 2.5 cm, glabrescent, short beaked, dehiscent or indehiscent when dry, depressed between the seeds, sutures raised, blackish - brown. Seed flattened, oblongoid, apiculate, 8 - 9 mm x 4 - 5 mm x 2.5 mm, smooth, pale brown; areole elliptical, 3 - 4 mm x 0.7 - 1.5 mm.

1.1.4 Ecology and geographic distribution of *Senna didymobotrya*

The wide variety of species and ecological adaptations makes at least a handful of *Sennas* suitable for any climate warmer than cool - temperate (Sunarno, 1997). *S. didymobotrya* is common in deciduous bush land, along lake shores, streams, rivers and other damp localities, in grassland and woodland, from sea level up to 2500 m altitude (Nyaberi *et al*., 2013). At times it is found in old plantations and in hedges near buildings (Nyaberi *et al*., 2013). In its natural habitat, *S. didymobotrya* is often ruderal in riparian montane wooded grassland or evergreen bush land. It tolerates light frost (Sunarno, 1997; Nyaberi *et al*., 2013) and their biophysical limit in altitude is between 900 - 2400 m. The plant has been found to be exotic in India, Indonesia, Malaysia, and Sri Lanka. *S. didymobotrya* has been naturalised in Australia and parts of America (Sunarno, 1997). The plant is grown indoors or in frost free climate in well drained soils. It is common in undisturbed areas and grows rapidly and is widely distributed in the tropical and subtropical regions (Mazumnder *et al*., 2008).

2. Genetic variation in plants

Researches on genetic diversity have been carried out by various research groups on the diversities of different plants. Different techniques like morphological markers, molecular markers, ethnobotany and use of chemical markers have been used to identify genetic diversity in plants. They have been used singly or incorporation of two or three methods. Morphometric approach is a common strategy of analysing morphological variations below the species level (Gengler - Nowak, 2002; Repka, 2003; Schmalzel *et al*., 2004 and Kropf, 2008). Conventional methods have shown that morphological characteristics are useful to establish phylogenetic

relationships at genus level, but are insufficient to define genetic diversity and relationships among accessions of a species, due to strong influence of the environment on plant traits. Feng *et al*. (2009) reported on the use of molecular markers in determining genetic diversity in species of family Euphorbiaceae and specifically in *Hevea brasiliensis* (Willd. ex A. Juss.) Mull. Arg. Population genetic studies are very important and the study of accessions with a focus on population can be achieved through a combination of morphological, chemical and molecular markers (Essilfie and Oteng - Yeboah, 2013). Ethnobotany has been applied in characterising plants for instance, characterization of Malawian cassava germplasm using ethnobotany, morphological and amplified fragment length polymorphism (AFLP) markers on ninety three accessions collected from farmers field and commercial programs showed a wide diversity according to use, areas and farmers preferences for traits in cassava varieties (Benesi *et al*., 2010). Their ethnobotany and morphological characterisation revealed a wide genetic diversity in the germplasm and AFLP markers for DNA fingerprinting using a subsample of 28 accessions showed a narrow genetic diversity but distinguished all the accessions.

2.1 Morphological markers.

Several studies on phenetic diversity of plants have been done through numerical / morphometric analysis. Numerical taxonomy is also termed as morphometric dealing with grouping by numerical methods of taxonomic units into taxa on the basis of their character state (Rahman and Rahman, 2012; Rahman *et al*., 2013). Numerical taxonomic studies are important for discovering and documenting new character and character states (Rahman, 2013). Cluster analysis (CA) and principal component analysis (PCA) are two techniques commonly used in numerical classification (Sonibare *et al*., 2004). PCA is usually used as an exploratory tool in systematics. It is a method for rotating the axes of a coordinate system such that the first axis (first principal component) passes through the greatest dimension of the swarm of data points, and thus accounts for the greatest amount of variance of any possible axis. The second principal component, orthogonal to the first accounts for the greatest amount of residual variance, and so on. There are as many components as original variables, and these components are linear combinations of the original variables. Most of the variance is usually summarised by the first few components, and PCA thus reduces a larger number of variables to fewer variables, which are often easier to interpret and is thus described as a dimension reducing method. Scores of each specimen on the principal component, usually the first two, can be plotted on bivariate scattergrams, allowing visualization of the relative positions of the populations (Henderson, 2006). PCA has been used in systematics of palm by Boyd (2002).

Cluster analysis (CA) is an exploratory tool for classifying objects with no statistical assumptions about the data. Cluster analysis produces a hierarchical classification of entities (taxa) based on the similarity matrix. It thus provides a logical means of expressing the relationship existing between taxa. Results are usually presented in the form of trees or dendrograms. The two preliminary decisions in CA concern the choice of an association coefficient and of a clustering algorithm. Association matrices for this kind of analysis are produced by either similarity or distance coefficients (Henderson, 2006). Examples of the use of CA in plant systematics are provided by Binns *et al*. (2002) and Gengler - Nowak (2002). Numerical taxonomic studies have been useful in discovery and documentation of new morphological characters, characters states and traits. There have been many attempts to understand phenetic relationships and species relatedness in different groups of plants (Gomez - Campo *et al*., 2001; Henderson and Ferreira, 2002; Pinheiro and Barros, 2007; Bolourian and Pakravan, 2011; Deshmukh, 2011). Work by Maggs - Kolling *et al*. (2000) on *Citrullus lanatus* showed that cluster analysis supported the indigenous classification system in which *Citrullus* types were distinct based on gross morphology, ecology and usage.

Various studies have been carried out in the genus *Senna* Mill., for instance a phenetic study of *Cassia sensu lato* in Thailand using 32 vegetative and reproductive floral morphology showed that in cluster analysis, *Cassia* s. l. was separated into four groups (*Chamaecrista*, *Senna alata*, *Senna* and *Cassia* s / str.) and *Chamaecrista*. *Senna* and *Cassia* s/ str. were found to be distinct taxa separated by filament length, fruit length and ovary stalk length. *Senna* was reported to be a heterogeneous taxon among the the three (Boonkerd *et al*., 2005). Morphometric study of *Senna* in South - western Nigeria using thirteen quantitative characters of the leaves, fruits, seeds and flowers revealed that species had great similarities hence their grouping under the same genus. It also, showed that *S. hirsuta* and *S. sophera* were more closely related. *S. occidentalis*, *S. siamea* and *Senna spectabilis* all shared some resemblance. *Senna occidentalis* was distantly related to *S. sophera* (Soladoye *et al*., 2010a). Another study in Bangladesh using 32 vegetative and floral characters showed that highest similarities was between *S. obtusifolia* and *S. tora* while high variation was observed between *S. alata* and *S. hirsuta* (Rahman *et al*., 2013). The UPGMA tree derived from cluster analysis revealed three major clusters, the first consisting of two species (*S. alata* and *S. auriculata*), the second cluster comprised of four species (*S. hirsuta*, *S. obtusifolia*, *S. tora* and *S. occidentalis*) and the third consisted of five species namely *S. multiglandulosa*, *S. sophera*, *S. siamea*, *S. timoriensis* and *S. surattensis* (Rahman *et al*., 2013).

Infraspecific delimitation of *Acacia senegal* in Uganda using numerical taxonomic principles and

multivariate analysis on 69 characters derived from growth form, branchlets, leaves, flower, pods and seed revealed a wide variation within the variety. It was split into three varieties, namely; *Senegal*, *Leiorhachis* and *Kerensis*. Further, it was found that the most important characters for differentiating the taxa were leaf breadth and length, pinna length and its ratio to pinna breadth, number of leaflet pairs, petiolar gland shape, petiolar and rachis gland size, stem and branch bark texture, stem and branchlet colour, under - bark colour for stem and branches, pod apical shape, growth form, crown shape and prickly state of leaves (Mulumba and Kakudidi, 2009).

Numerical taxonomic studies conducted on five populations of *C. anisata* in the coastal Savana zone of Ghana to determine their patterns of taxonomic variation and identifying diagnostic characters revealed that the populations were classified into two main clusters (Essilfie and Oteng - Yeboah, 2013). The authors reported on morphological characters like peduncle length, ratio of sepal length to width, number of floral branches per inflorescence, anther length, style diameter and length, filament length, plant height, petiole diameter, sepal length and petal length been diagnostic morphological characters.

Phenetic analysis using numerical techniques in evaluating the taxonomic status of the genus *Solanum* from Pakistan using cluster analysis showed that all species of the genus *Solanum* could be divided into two groups at hundred percentage linkage with floral and stem characters playing a significant role in its identification (Yousaf *et al*., 2010). Morphometric analysis of the genus *Ficus* Linn. using foliar parameters subjected to quantitative and principal component analysis produced six groups using leaf length and leaf width, leaf length and lamina length, leaf length and petiole length, lamina length and lamina width (Sonibare *et al*., 2004). A morphometric study of the genus *Caesalpinia* in Kolhapur analyzed with the help of PCA, cluster analysis and coefficient of difference (CD) revealed that *C. cucullata* Roxb. was closely related with *C. sappan* L. while *C. mimosaides* Lam. differed morphologically with other species (Deshmukh *et al*., 2013).

Morphometric analyses also study the effects of environment on the phenetic characters. Results from morphometric and phenetic studies of five geographical populations of *Lutzomyia whitmani* delineated the existence of biogeographical structuring within the species (Dias, 1999). Deshmukh *et al*. (2013) in their study on the genus *Caesalpinia* L. showed that species were different based on the analysed quantitative characters. They concluded that morphometrics permit numerical comparisons between different forms.

A study on evaluating the influence of different environments on the morphology of individuals from *Epidendrum secundum* using morphometry to compare plants under cultivated zones and the ones collected directly from the field showed a large difference between the plants growing in rocky areas and those from the Atlantic rainforest indicating that phenotypes are strongly influenced by habitat (Pinheiro and Barros, 2007). Morphometric and genetic variations in *Pueraria mirifica* cultivars across Thailand using leaves with seed pods and flowers collected from 39 locations revealed a low level of variation between cultivars on morphometric analysis (Suwanvijitr *et al*., 2010). They reported from linear regression analysis that leaves tended to decrease in size from the West to the East whilst pods tended to increase in size from the South to the North and also to the West and East. The genetic analysis conducted by direct sequencing of one nuclear and one chloroplast and by random genome analysis by RAPD - PCR using five primers indicated a low level of variation between isolates though the nuclear sequences displayed a divergence of up to 25.2%. Such phenetic studies have not been done on *S. didymobotrya* in Kenya thus the need of the study.

Objectives

To assess the phenetic diversity of *S. didymobotrya* plants populations collected from different geographical regions in Kenya.

Justification of the research

Senna didymobotrya in East Africa is widely used as a medicinal plant to treat fungal skin infections in Kenya. Its phenetic diversity and its ecological transect is not properly documented. Considering the wide medicinal applications of *S. didymobotrya*, it becomes imperative to investigate its phenetic diversity since it affects its phytochemicals and antimicrobial activity. Given the scarce resources and facilities required to carry out molecular research, phenetic studies have also been used to characterize plants.The information generated will highlight the knowledge gaps which are at the moment addressed. This will generate basic information on the species distribution which will enhance further research on the genetic diversity of the species in Kenya.

MATERIALS AND METHODS

Study areas: Study areas included Nandi, Siaya, Kisumu and Nakuru counties. The sites were purposively selected according to altitude, geographical location and environmental variability

Surveys on phenetic characters of *S. didymobotrya*

Field research was conducted between the 8th July and 9th August, 2011. The field work surveys and data collection were conducted in accordance with the institutional, national and intellectual principles and guidelines of plant use and conservation of biodiversity (WHO, 2003). Surveys were carried out in Siaya, Kisumu, Nandi and Nakuru Counties and stratified random sampling technique was applied. In every County, prior

consultations/ requests for and an endorsement to conduct the study was acquired from the relevant authorities. Field surveys were conducted according to the life forms and flowering period of the plant. The stratum, in each County was represented by the administrative sub-County. In each stratum, sampling was done every five kilometres along the major roads. At each site, the presence of *S. didymobotrya* plants was noted. The phenetic characters of one mature plant with flowers and pods were measured *in situ* (Table 1 part A). The character states were coded and the codes were applied in collecting data (Table 1 part B). If there were no plants at the sampling sites, then the next points were considered. In each County, 30 sampling sites were considered.

Of the three to five or six leaves, the longest leaf length was measured from the petiole base to the tip of the middle leaflet. Average length of three petioles was considered for the analysis. Length and breadth of the leaflet was measured as average of three middle leaflets; the leaflet breadth was taken at the widest point on the lamina. Stem length was measured from the ground level on the tallest stem. Maximum stem thickness / diameter was taken from the same stem used for height. Pod length was taken from the point of stalk attachment to the apex tip. Pod width was taken at mid - length of the pod. Pod surface protrusions (appearance of a pronounced pattern at the seed positions) were scored as present or absent. The seeds per pod were taken as the average from three pods. The leaf number was counted on three branches and their means calculated. Qualitative characters were evaluated by eye observation while texture was determined by feeling. Quantitative measurements were carried out in centimetres, millimetres and elevation was measured in metres.

The measurements were encoded for each plant and considered an operational taxonomic unit (OTU) according to Kropf (2008) and Soladoye *et al*. (2010 a) with modifications (Tables 1 part A and B). Both qualitative characters were coded into states by assigning ordinal values as binary / multistate for carrying out the multivariate analyses. Traits that had only two categories of description were scored normally in the binary matrix. The characters and their binary / multi states used for numerical taxonomic studies are listed in Table 1 part B.

Table1: Characters scored for the study on phenetic diversity of *S. didymobotrya*

Part A	**Phenetic character**	**Code**	**Quantitative measurement**
1	Plant height	PH	cm
2	Leaf number	LN	counts
3	Leaf length	LL	mm
4	Leaf thickness	LT	mm
5	Pod length	PL	mm
6	Pod width	PW	mm
7	Seeds per mature pod	NS	counts
8	Internodal length	IL	cm
9	Leaflet length	LLL	cm
10	Leaflet width at widest point	LLW	mm
11	Stem height	SH	cm
12	Stem diameter at base (thickness)	SD	mm
13	Inflorescence length	FL	cm
14	Inflorescence length of basal stalk (peduncle)	FLP	cm
15	Habitat elevation	E	meters above sea level
16	Inflorescence thickness	FT	mm
17	Leaflet number	LLN	counts
Part B	**Character state**	**Code**	**Qualitative observation**
18	Plant habitat	HT	Fallow land (1); Roadside (2); Homestead/church (3),along a river (4); along lake (5)
19	Plant habit	H	shrub (1); tree (2)
20	Dehiscence of pod	DP	Dehiscent (1), Indehiscent (0)
21	Pod outlook	PO	Flat (1); bulging over seed (2)
22	Pod apical shape	PAS	acuminate (1); acute (2) rostrate (3); rounded (4)
23	Pod veination	PV	pronounced (1); not pronounced (2)
24	Pod texture	PTX	Pubescent (1), Glabrous (0).
25	Pod surface protrusions	PSP	present (1); absent (2)
26	Pod shape	PS	Linear - elliptic to curved (1); Oblong (0).
27	Leaf attachment/arrangement	LA	Alternate (1), Opposite (0)
28	Leaflet laminar shape	LLS	Oblong - obovate to oblong - lanceolate(0); oblanceolate to narrowly elliptic (1)
29	Leaflet attachment/arrangement	LLA	Alternate (1), Opposite (0)
30	No. of stems	S	Several - stems (1); Single (0)
31	Stem bark texture	SBT	Hairy (1), Glabrous (0)
32	Stem - bark colour	SBC	Green - yellow (1); grey- brown (2); bright orange - brown (3); dull grey
33	Inflorescence colour	FC	Yellow – bright yellow (1); dark yellow (2); other colors (3)
34	Inflorescence attachment	FA	Axillary and terminal (1), Terminal (0)

A total of 34 qualitative and quantitative morphological characters were assessed (Table1). The herbarium specimens were also collected simultaneously for phenetically characterized plants and curated using standard procedures (Leenhouts, 1968; Stace, 1993). Plant materials were authenticated by comparison with

herbarium specimens and referring to standard literature (ICRAF, 1992; Agnew, 1994 and Beentje, 1995). Each plant specimen collected was given a herbarium specimen number derived from the collector's name in abbreviation, collection site and the specimen number. The voucher samples kept in Jaramogi Oginga Odinga University of Science and Technology botany laboratory herbarium for future reference.

Morphometric analysis was carried out following procedures of Soladoye *et al.* (2010 a and 2010 b) with modifications. The data on quantitative characters obtained in Table 1 (Part A) were subjected to morphometric analysis using Principal Component Analysis (PCA) and Cluster Analysis methods. The means and standard deviations derived were keyed into a Microsoft excel spread sheet and Statistical Analysis Sysytem (SAS) version 8.02 , copy 1999 - 2001 TS Level 02M0 analysis sheet. The levels of similarities and relationships among the populations of *S. didymobotrya* were computed using Eucildean coefficients derived from product moment correlation coefficient. The similarities were represented in the phenogram by the clustering method and a line drawn to separate the plants at (P) or $\alpha = 0.05$ significance level. Cluster observation was based on the thumb rule and a cumulative of 70% was considered. To identify those morphological characters that were most important in the differentiation of the species, the principal components were analysed with the PROC PRINCOMP (SAS version 8.02, copy 1999 - 2001). The qualitative characters obtained in Table1 (Part B) were subjected to SPSS analysis.

RESULTS

Surveys on phenetic characters of *Senna didymobotrya*

Results presented in Table 2 indicated that a total of 39 plants were sampled during the surveys. Kisumu County had 8 plants while in Siaya and Nakuru Counties, 10 plants were sampled each. Nandi County had the highest number of plants. The specimens collected from Siaya were assigned accessions PJ/SI/1- 10 while specimens collected from Kisumu County were assigned accession numbers PJ/KS/11 –18. Specimens from Nandi County were assigned accession number PJ/NA/19 – 29 and the specimens obtained from Nakuru County were assigned accession numbers PJ/NK/30 – 39.

Table 2: Plant samples collected per County

County	Plants collected	Herbarium specimen number
Siaya	10	PJ/SI/1 – PJ/SI/10
Kisumu	8	PJ/KS/11 – PJ/KS/18
Nandi	11	PJ/NA/19 – PJ/NA/29
Nakuru	10	PJ/NK/30 – PJ/NK/39
Total	39	

Quantitative characters

Seventeen quantitative characters were subjected to morphometric analysis (Table 3).

Table 3: Morphometric analysis (means =+ standard deviations and ANOVA based on the 17 quantitative characters of 39 *S. didymobotrya* plants.

Characters	County				
	Siaya	Kisumu	Nandi	Nakuru	Total
Plant Height (cm)	221.90±24.80	174.13±26.84	207.00±25.41	174.70±15.30	195.79±11.74
Stem Height (cm	135.10±28.77	72.75±11.26	142.72±23.20	97.50±12.34	114.82±11.15
Stem diameter at base (mm)	169.30±28.91	27.13±12.00	50.18±16.16	18.80±2.02	67.95±13.14
Habitat Elevation (m)	1219.30±27.45	1356.88±59.49	1942.63±36.12	2122.10±53.58	1683.03±65.40
Pod Length (mm)	113.10±4.14	95.13±11.19	122.72±5.49	119.30±2.96	113.72±3.35
Pod Width (mm)	16.60±0.87	16.38±2.14	21.00±0.57	16.20±1.71	17.69±0.73
No. of Seeds per mature pod	10.4±1.12	11.88±0.90	12.63±0.31	12.80±0.63	11.95±0.40
Leaf number	15.40±1.15	25.38±6.70	16.45±1.30	33.10±5.24	22.28±2.23
Leaf Length (cm)	20.50±1.79	21.73±3.15	19.04±1.81	23.88±2.35	21.21±1.11
Leaf thickness (mm)	2.56±0.29	2.28±0.35	3.18±0.18	5.70±2.22	3.48±0.60
Internodal length (cm)	1.69±0.20	2.33±0.40	2.17±0.30	3.34±0.37	2.38±0.18
Leaflet length (cm)	4.28±0.34	3.93±0.41	4.48±0.32	4.64±0.38	4.36±0.18
Leaflet width at widest point (mm)	15.60±1.76	15.38±1.52	14.91±0.89	15.50±1.81	15.33±0.73
Leaflet number	16.60±2.00	23.00±3.34	20.91±1.53	25.40±2.09	21.38±1.17
Inflorescence length (cm) long	18.90±2.29	29.60±4.13	22.02±2.41	28.04±4.16	24.32±1.71
Inflorescence length of basal stalk (peduncle) cm	8.80±1.76	17.24±4.87	12.91±2.10	14.95±1.62	13.27±1.35
Inflorescence thickness (mm)	3.99±0.50	7.00±1.60	7.45±2.15	6.80±1.65	6.31±0.82

†† The values are the Means ± SE

The means and standard deviations are presented in Table 3. The standard deviations indicated that the differences were highly significant when all characters were considered. Stem diameter, plant height and stem height had the highest standard deviation of 13.14, 11.74 and 11.15 respectively. Leaflength and Internodal length had the lowest standard deviations of 0.18 each.

Table 4: Correlation matrix of quantitative morphological characters of 39 *S. didymobotrya* plants.

Character	PH	SH	SD	HT	PL	PW	NS	LN	LL	LT	IL	LLL	LLW	LLN	FL	FLP	FT
PH	1																
SH	**0.62**	1															
SD	0.45	0.48	1														
HT	0.34	0.04	0.44	1													
PL	-0.19	0.26	0.13	-0.15	1												
PW	-0.36	0.31	0.06	-0.04	**0.52**	1											
NS	0.18	-0.04	-0.94	0.18	-0.15	0.08	1										
LN	-0.1	-0.16	-0.31	-0.21	0.06	-0.22	-0.11	1									
LL	-0.16	0.06	-0.08	-0.36	0.24	0.06	-0.18	0.21	1								
LT	0.08	0.13	-0.1	0	0.2	0.1	-0.1	0.12	0.22	1							
IL	-0.12	-0.06	-0.28	-0.11	-0.15	-0.21	0.2	0.29	0.21	0.13	1						
LLL	-0.1	-0.07	-0.17	-0.04	0.14	0.09	0.07	0.08	0.49	0.28	0.09	1					
LLW	-0.13	-0.13	-0.11	0.12	0.14	0.09	-0.1	0.28	0.24	-0.2	0.01	**0.56**	1				
LLN	-0.25	-0.3	-0.39	-0.05	0.24	0	0.04	**0.54**	0.42	0.19	0.23	0.28	0.3	1			
FL	-0.23	-0.07	-0.16	0.04	0.01	0.11	0.42	0.04	0.14	0.04	0.38	0.23	0.29	0.15	1		
FLP	-0.28	-0.13	-0.25	-0.04	-0.02	0.06	0.39	-0.01	0.25	-0.02	0.36	0.35	0.25	0.27	**0.83**	1	
FT	-0.12	-0.15	-0.21	-0.18	0.27	0.12	0.12	-0.12	0.09	0.05	-0.12	0.14	0.25	0.04	0.18	0.04	1

Key: Plant height (PH); Stem height (SH); Stem diameter (SD); Habitat (HT); Pod length (PL); Pod width (PW); Number of seeds (NS); Leaf number (LN); Leaf length (LL); Leaf thickness (LT); Internodal length (IL); Leaflet length (LLL); Leaflet width (LLW), leaflet number (LLN); Inflorescence length (FL); Inflorescence length of basal stalk (FLP); Inflorescence thickness (FT).

Table 4 represents the correlation matrix of the morphological characters. The results indicated that there were high positive correlations between inflorescence length of basal stalk and inflorescence length (0.83), stem height and plant height (0.62), leaflet width and leaflet length (0.56), leaflet number and leaf number (0.54), pod width and pod length (0.52).

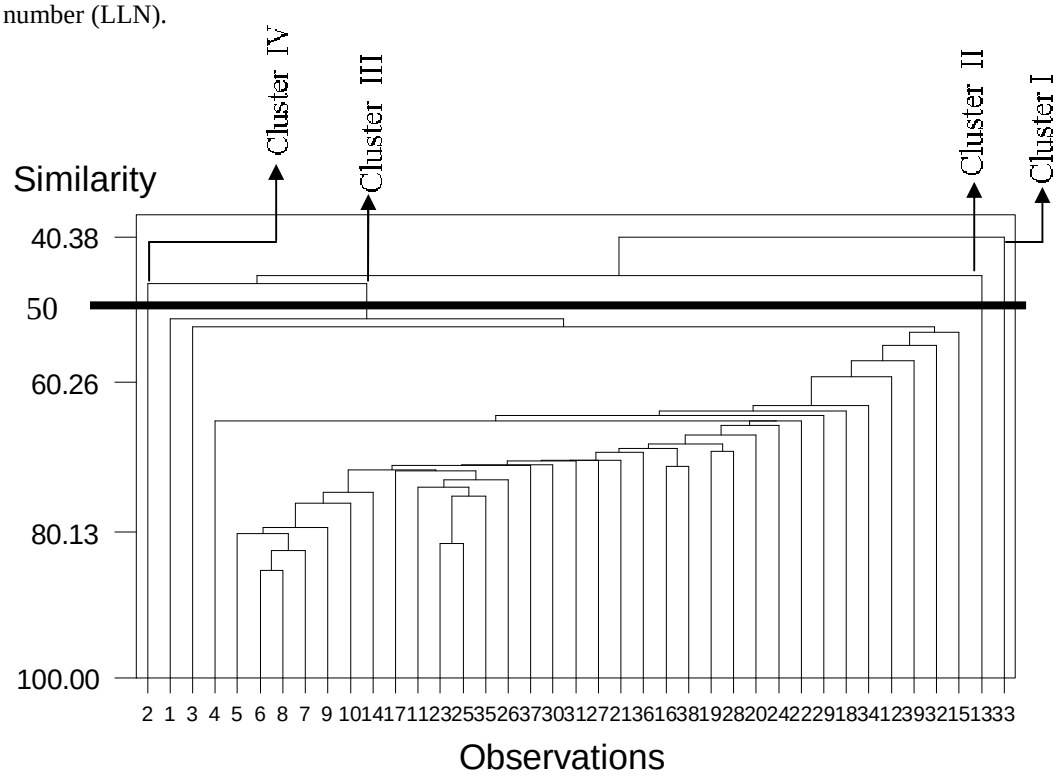
Table 5: Principal Component Analysis loadings, Eigen vector values, and percentage of variance for the 17 Principal Components (PC) obtained from the morphometric characters of 39 plants of *S. didymobotrya*.

Character	PC1	PC2	PC3	PC4	PC5	PC6	PC7	PC8	PC9	PC10	PC11	PC12	PC13	PC14	PC15	PC16	PC17
PH	-0.29	0.09	0.23	**0.37**	0.08	0.14	-0.24	0.4	-0.12	0	-0.08	-0.23	-0.1	0.23	0.5	-0.25	-0.12
SH	-0.23	**0.36**	0.25	0.27	-0.19	-0.18	0.08	0.31	0.02	-0.19	-0.04	-0.08	-0.31	-0.26	-0.48	0.27	0.02
SD	**-0.33**	0.2	0.23	0.15	0.21	-0.17	0.16	-0.13	0.3	0.36	-0.13	0.23	0.34	0.45	-0.18	0.02	0.11
HT	-0.16	-0.1	0.32	0.17	**0.41**	**0.38**	0.09	-0.39	0.17	0.04	0.19	-0.18	-0.01	-0.4	0.12	0.28	-0.11
PL	0.09	**0.52**	-0.01	-0.15	-0.11	0.19	0.32	-0.02	0.23	0.14	0.06	0.4	-0.37	-0.14	0.33	-0.2	-0.04
PW	0.01	**0.41**	0.21	-0.26	-0.17	0.15	0.3	-0.06	-0.36	-0.27	0.23	-0.27	0.4	0.2	0.08	0.12	-0.04
NS	0.07	-0.22	**0.44**	-0.05	-0.16	0.33	-0.07	0.27	-0.33	0.32	0.1	0.38	0.16	-0.2	-0.16	-0.11	0.24
LN	0.24	-0.03	-0.29	**0.36**	0.09	0.22	0.27	0.36	0.07	-0.18	-0.28	0.26	0.35	-0.08	0.14	0.36	-0.06
LL	0.28	0.3	-0.08	0.23	-0.04	**-0.39**	-0.14	0.01	-0.02	0.47	0.15	-0.25	0.32	-0.41	0.16	-0.04	0.04
LT	0.08	0.24	-0.03	0.31	**-0.33**	0.34	-0.4	-0.41	0.13	-0.23	-0.24	0.02	0.13	0	-0.08	-0.16	0.32
IL	0.23	-0.23	0.1	**0.35**	-0.25	-0.07	0.01	0.05	0.33	-0.16	0.67	0.05	-0.02	0.25	0.02	-0.04	0.03
LLL	0.3	0.23	0.13	0.12	0.28	-0.13	-0.41	-0.17	-0.32	-0.09	0.12	0.41	-0.11	0.18	-0.04	0.22	-0.38
LLW	0.26	0.15	0.08	-0.04	**0.63**	-0.06	0.04	0.17	0.04	-0.33	0.06	-0.05	0.01	-0.05	-0.13	-0.36	0.46
LLN	**0.35**	0.07	-0.15	0.21	0.09	**0.37**	0.22	-0.02	-0.16	0.39	-0.06	-0.35	-0.27	0.3	-0.36	-0.07	-0.11
FL	0.31	-0.11	**0.44**	-0.06	-0.1	-0.09	0.11	-0.01	0.26	-0.14	-0.34	-0.08	0.17	-0.1	-0.11	-0.35	-0.52
FLP	**0.35**	-0.12	**0.39**	-0.04	-0.1	-0.22	0.1	-0.1	-0.02	0.05	-0.31	-0.11	-0.26	0.18	0.33	0.39	0.39
FT	0.14	0.15	0.04	-0.42	0.03	0.26	-0.44	0.36	0.48	0.12	0.07	-0.16	0.07	0.11	-0.06	0.3	-0.03
Eigen value	3.63	2.21	2.12	1.64	1.39	1.04	1.02	0.92	0.71	0.63	0.52	0.4	0.32	0.15	0.13	0.1	0.08
Difference	1.42	0.09	0.48	0.25	0.34	0.02	0.1	0.2	0.08	0.11	0.12	0.08	0.17	0.02	0.02	0.03	0
Proportion	21.36	12.99	12.48	9.64	8.15	6.13	6	5.39	4.19	3.69	3.07	2.37	1.88	0.86	0.75	0.61	0.45
Cumulative	21.36	34.36	46.83	56.47	64.62	**70.75**	76.74	82.14	86.33	90.02	93.09	95.46	97.33	98.19	98.94	99.55	100

Key: PC1 = Plant height (PH); PC2 = Stem height (SH); PC3 = Stem diameter (SD); PC4 = Habitat (HT); PC5 = Pod length (PL); PC6 = Pod width (PW); PC7 = Number of seeds (NS); PC8 = Leaf number (LN); PC9 = Leaf length (LL); PC10 = Leaf thickness (LT); PC11 = Internodal length (IL); PC12 = Leaflet length (LLL); PC13 = Leaflet width (LLW), PC14 = leaflet number (LLN); PC15 = Inflorescence length (FL); PC16 = Inflorescence length of basal stalk (FLP); PC17 = Inflorescence thickness (FT).

Information in Table 5 shows values of the 17 quantitative characters that were characterized for each of the 39 plants of *S. didymobotrya* subjected to PCA. A principal component of cumulative percentage of 70% was considered. The first six components expressed 70.75% of the cumulative variance in a lineal combination

of parameters. These corresponded to plant height (PH), stem height (SH), stem diameter (SD), habitat (HT), pod length (PL) and pod width (PW) respectively. The key characters responsible for the variation were expressed by six characters (lineal combinations of parameters). For each factor, parameter/ character with maximum discriminating power and percentage of variance/ proportion they account for are expressed as in Table 5. The first principal component had significant negative loadings for plant height (PH), stem height (SH), habitat (HT) and stem diameter (SD). PC 1 was influenced by leaflet length (LLL), inflorescence length of basal stalk (FLP) and stem diameter (SD). The character responsible for the variation along PC 2 relates to pod length (PL), pod width (PW) and stem height (SH). Number of seeds (NS), inflorescence length (FL) and inflorescence length of basal stalk (FLP) were responsible for the variation on PC 3. PC 4 was influenced by plant height (PH), leaf number (LN) and Internodal length (IL). The leaflet width (LLW), habitat (HT) and leaf thickness (LT) were responsible for the variation along PC 5 and PC 6 was affected by leaf length (LL), habitat (HT) and leaflet number (LLN).



Key: Numbers 1-10 refers to herbarium specimens PJ/SI/1- PJ/SI/10
Numbers 11-18 refers to herbarium specimens PJ/KS/11- PJ/KS/18
Numbers 19-29 refers to herbarium specimens PJ/NA/19- PJ/NASI/29
Numbers 30-39 refers to herbarium specimens PJ/NK/30- PJ/NK/39

Figure 1: Dendrogram showing morphometric relationship of all 39 plants of *S. didymobotrya*.

Figure 1 shows a multivariate summary of plants similarity illustrated by a dendrogram. The cluster analysis dendrogram placed the plants in four major clusters. Cluster 1 had one plant accession PJ/NK/33 collected from Nakuru County. Cluster II had one plant PJ/KS/13 collected from Kisumu County. In Cluster IV also had one plant accession PJ/SI/2 collected from Siaya County. Cluster III consisted of 36 plants (Plants accessions PJ/SI/4 - 10, PJ/KS/11- 18, PJ/NA/19 - 29, PJ/NK/30 - 32 and PJ/NK/34 – 39).

Qualitative characters

Table 6: The similarities in qualitative characteristics.

Character	County			
	Siaya	Kisumu	Nandi	Nakuru
Plant habit	Shrub	Shrub	Shrub	Shrub
Dehiscence of pod	Dehiscent	Dehiscent	Dehiscent	Dehiscent
Pod outlook	Flat	Flat	Flat	Flat
Pod apical shape	Acuminate	Acuminate	Acuminate	Acuminate
Pod veination	Pronounced	Pronounced	Pronounced	Pronounced
Pod texture	Pubescent	Pubescent	Pubescent	Pubescent
Pod surface protrusions	Present	Present	Present	Present
Pod shape	Linear-elliptic to curved	Linear-elliptic to curved	Linear-elliptic to curved	Linear-elliptic to curved
Leaf attachment/arrangement	Alternate	Alternate	Alternate	Alternate
Leaflet laminar shape	Oblong-ovate to oblong-lanceolate	Oblong-ovate to oblong-lanceolate	Oblong-ovate to oblong-lanceolate	Oblong-ovate to oblong-lanceolate
Stem bark texture	Hairy	Hairy	Hairy	Hairy
Stem-bark colour	Green-yellow	Green-yellow	Green-yellow	Green-yellow
Inflorescence colour	Yellow-bright yellow	Yellow-bright yellow	Yellow-bright yellow	Yellow-bright yellow
Inflorescence attachment	Terminal	Terminal	Terminal	Terminal

All the characters presented in Table 6 can not be used to phenetically characterize *S. didymobotrya*.

Table 7: The Differences in qualitative characteristics.

Characters	Plant habitat				Leaflet		No. of stems	
County	Fallow land	Roadside	Homestead/ church	Along lake	Opposite	Alternate	Single stem	Several stems
Siaya	0%	70%	10%	20%	30%	70%		100%
Kisumu	37.5%	50%	12.5%	0%		100%		100%
Nandi	9.1%	63.64%	27.27%	0%		100%	27.27%	72.73%
Nakuru	20%	80%	0%	%	30%	70%		100%

The information presented in Table 7 indicates that at the roadside in Siaya County, 70% of the difference exist on the *S. didymobotrya* populations and the alternate leaflet arrangement gives a 100% difference in Kisumu and Nandi plant samples.

DISCUSSION

Mophometric analysis on quantitative and qualitative characters

In taxonomic studies, diagnostic characters are characters that are constant within a group but vary between groups. Such characters could be used to identify natural plant groups from several others of similar ranking (Davis and Heywood 1963; Kent and Coke, 1992). In numerical analysis, diagnostic characters exhibit high and absolute factor scores and are capable of separating operational taxonomic units (OTUs) under study into distinctive groups. The quantitative diagnostic characters identified in this study could be described as bad taxonomic characters (Davis and Heywood, 1963) because they are easily modified by environmental factors. However, such bad taxonomic characters could still be utilized in any taxonomic study or considerations provided their genetic bases have been ascertained through a series of transplant experiments.

Studies by Mulumba and Kakudidi (2011) showed that there was infraspecific delimitation of *Acacia senegal* with similarity on leaflet length, petiole length and leaflet number having the highest loadings using PCA. Similar correlations have been observed also *Ficus* spp. (Sonibare *et al*., 2004). Their correlation analyses showed highly positive significant correlation on leaf length and leaf width, leaf length and lamina length, leaf length and petiole length, lamina length and lamina width and a negative correlation between leaf width and leaf length / width ration, petiole length and fruit length / petiole length ratio. These results on correlation between characters differ from those of Yousaf *et al*. (2010) who found a significant relationship between floral characters and stem characters which played a significant role in the identification of the genus *Solanum*. Research findings of this study on *S. didymobotrya* showed high positive correlations between plant height and stem height, pod length and pod width, leaf number and leaflet number, leaflet length and leaflet width, inflorescence length and inflorescence length of basal stalk (peduncle), contributed strongly to the delimitation of the species *S. didymobotrya* and therefore significantly contributing to its taxonomy. The positive correlation

between leaflet length and leaflet width was similar to those of Sonibare *et al*. (2004).

In other studies, Essilfie and Oteng - Yeboah (2013), used peduncle length, ratio of sepal length to width, number of floral branches per inflorescence, anther length, style diameter, filament length, style length, plant height, petiole diameter, sepal length and petal length as diagnostic morphological characters for *Clausena anisata*. The use of plant height in this study concurs with their results. Using morphometry on *Epidendrum secundum* populations, Pinheiro and Barros (2007) found that floral characters (inflorescence length, petal length, lateral sepal length, lip length and width, callus of lip length) showed a high correlation. Their work is similar to the results in this study in that inflorescence length and inflorescence length of basal stalk (peduncle) showed a positive correlation as presented in Table 4. Similar studies carried on the genus *Caesalpinia* L. using eight quantitative characters found a significant correlation between leaf length and pod length, pinna length and leaflet length and pedicel length and corolla length (Deshmukh *et al*., 2013). In their work on first comparative phenetic studies of Argentinean species of *Acacia*, Casavi *et al*. (2002) using morphometric, isozymal and random amplified polymorphic DNA (RAPD) approaches, revealed that thorny stipule length had high loadings on all the first three principal components (PC). PCA as a multivariate technique helps in examining relationships among several quantitative variables and as such summarizes data, detects linear relationships, and reduces the number of variables in regression and clustering. Plots of principal components are especially valuable tools in exploratory data analysis (Chiapella, 2000). PC 1 was influenced by inflorescence diameter and seed pod shape, PC 2 was influenced by peduncle length and fruit width while PC 3 was influenced by thorn length, number of pinnae per leaf and fruit length. In this study with *S. didymobotrya*, had 17 PC's whereby PC 1 was influenced by leaflet length, inflorescence length of basal stalk and stem diameter, PC 2 influenced by pod length, pod width and stem height while PC 3 influenced by number of seeds, inflorescence length and inflorescence length of basal stalk.

It was observed that some characters proved constant throughout the specimens hence found not useful in delimiting the species. These characters were plant habit, dehiscence of pod, pod outlook, pod apical shape, pod veination, pod texture, pod surface protusions, pod shape, leaf attachment/arrangement, leaflet laminar shape, stem bark texture, stem bark colour, inflorescence colour and inflorescence attachment. Wherever possible, quantitative characters were used to reduce subjectivity and to avoid artefacts resulting from the conversion of continuous variables into categorical ones. With the help of morphometry used in this study, it was possible to distinguish between the populations of *S. didymobotrya.* Characters in Table 6 should not be used in characterizing whereas characters in Table 7 can be used to qualitatively characterize *S. didymobotrya*.

The cluster analysis (Figure 1) presented the phenetic relationships among the 39 plants of *S. didymobotrya* based on the seventeen quantitative characters. The dendrogram based on cluster analysis showed four clusters though with close relationships. The analysis grouped plants in four clusters. This is important for classification of *S. didymobotrya*. The most important finding is that there is little phenetic variability of *S. didymobotrya* in Kenya. 98% are in group III. Therefore there is need to carry out genetic studies to be able to differentiate the populations within the species.

From the results obtained, the importance of morphometry which is a branch of numerical taxonomy is inherent. In this study, only six out of seventeen quantitative characters have been shown by PCA while the remaining eleven indicated similarities that exist between the plant samples of *S. didymobotrya* studied. These results differed with those observed by Soladoye *et al*. (2010 a) and Rahman *et al*. (2013) and would be explained by the fact that the authors examined the morphological characters of different species of *Senna* whilst in our study only one species of *Senna* was analysed.

The important qualitative characters are plant habitat, leaflet arrangement and the number of stems (refer to Table 7).This correlates to Pinheiro and Barros (2007) report, who evaluated the influence of different environments on the morphology of individuals from *Epidendrum secundum* using morphometric analysis. They compared plants under cultivation and those from the field and found that phenotypes were strongly influenced by the habitat. There were also variations in lengths, shapes and sizes in the inflorescence, leaf, leaflets and pods among plant samples from the different counties. These variations in quantitative characters within species may be due to the following: age of plant, location and place of collection, sunlight intensity and genetic factors which occur as a result of mutation (Jongebloed *et al*., 2004; Soladoye *et al*., 2010 b). Variations in inflorescence, leaf, leaflets, leaf shapes and sizes, plant height, pods, and stem on length and sizes have been shown by the works of previous authors to vary within the same plant (Soladoye *et al*., 2010 b). In their work, they reported that light intensity affects carbohydrates balance which in turn affects the length of the cells along the axis which in turn gives rise to differences in shapes, length and width of the different parts (Campey *et al*., 2000).

The two techniques; (PCA and cluster analysis) used in this work are the most commonly used in numerical taxonomy. Soladoye *et al*. (2010 a) also used these techniques in morphometric study of the genus *Senna* Mill. in South - western Nigeria using 13 quantitative characters of the leaves, fruits, seeds and flowers in the study of eight *Senna* species. Their results revealed that only three characters including leaflet length, leaflet

width and leaflet length/width ratio contributed significantly to the delimitation of the taxa in that *S. sophera* was found to be closely related to *S. hirsuta*. Rahman *et al*. (2013) also using the same techniques found similar results to those of Soladoye *et al*. (2010 a) that *S. sophera* was closely allied to *S. multiglandulosa*. Morphometric techniques have also been used by Leelambika *et al*. (2010) in analyzing variability in twenty six morphometric traits on fifteen accessions belonging to *Mucuna pruriens*, whose results showed that there was a considerable variability for inflorescence, pod and seed characters ($P < 0.0001$) and less variability for leaf characters. Leaf, pod and flower morphometry in *Pueraria mirifica* showed that there was no grouping structure within the species (Suwanvijitr *et al*., 2010). Soladoye *et al*. (2008) also employed these techniques in the phytochemical and morphometric analysis of the genus *Acalypha* in which their results confirmed the similarities and differences that existed between the taxa. The taxonomic delimitation of the genus *Senna* has been a source of contradiction and uncertainties due to wide morphological variation and ambiguous boundaries between taxa therein (Boonkerd *et al*., 2005; Soladoye *et al*., 2010; Rahman *et al*., 2013). Although previous studies (Boonkerd *et al.*, 2005) reduced the delimitation uncertainties, it relied on only vegetative characters and the key developed could not be used to clarify variability within a given variety or species. Results from this research have shown that for most of the quantitative characters there was considerable range of variation.

CONCLUSIONS AND RECOMMENDATIONS

The phenetic characterization/diversity data uncovered in this study can be used in future breeding programs aimed at improving cultivated/domesticated plants of *S. didymobotrya*. From the morphometric analysis, it indicated that *S. didymobotrya* is a multigeographical species. The various plants studied exhibited complex patterns of morphological variations. Therefore, the various morphological types identified in this study could be described and documented for the purposes of communication and future research. The wide morphological variations shown by the plant samples in this study indicates that the species could be used in a wide range of habitats for conservation and restoration programs in disturbed environments.

This study was a first attempt to evaluate the phenetic diversity of *S. didymobotrya* and more questions than answers have been raised. Up to date, it is not possible to tell whether if the morphological variation observed is enough to circumscribe more than one species within *S. didymobotrya* populations, as the variation may be due to phenotypic plasticity related to environmental conditions. Therefore, ecological significance of the differentiation needs further investigations. Since quantitative trait variation has several disadvantages in that obtaining data is time consuming, limited to growing seasons and its expression is rather plastic with environmental effects (Wang *et al*., 2006), there is need to combine both quantitative traits and molecular markers in order to evaluate the relative role of selection drift and gene flow in structuring genetic variation (Szczepaniak *et al*., 2002). The diversity of the species concepts in the biological literature is an asset, not a liability when considering the *S. didymobotrya* and is an integral part of biological theory. The co - varying morphological discontinuities, the phenetic species concept, geographical and ecological isolation and the biological species concept of reproductive isolation should be taken into account. The use of differing concepts has been useful in suggesting multiple lines of evidence for testing taxonomic boundaries.

Principal component analysis determined that the high correlation between plant height and stem height, pod length and pod width, leaf number and leaflet number, leaflet length and leaflet width, inflorescence length and inflorescence length of basal stalk (peduncle) characters, contributes strongly to the delimitation of the *S. didymobotrya* species and therefore, significant contribution to it's taxonomy even though taxonomically, the various diagnostic characters identified could not be considered as suitable taxonomic characters for identification of the various morphological types unless their genetic bases have been ascertained. It also revealed significant variability among the studied plants and produced vital clues on taxonomic groupings within the species. Such a comprehensive study was lacking in Kenya despite numerous reports available on *S. didymobotrya* bioassays, biochemical, and ethnobotany. In conclusion, this research showed the significance of morphometric analysis for detecting variation and taxonomic relationships among the plant samples in Siaya, Kisumu, Nandi and Nakuru counties (ecozones). The taxonomic results also provided a view on the genetic variability of *S. didymobotrya* thus helping in evolving strategies for breeding. Even though the phenetic grouping within *S. didymobotrya* plant samples could not be classified as morphological varieties, they still could be described and documented for general purposes such as communication, management and conservation strategies. Therefore, an application of this method in an elaborate taxonomic review of species *S. didymobotrya* in the future is recommended using characters with the highest loadings in developing a taxonomic identification key.

Morphometric analysis of 39 plant samples of *S. didymobotrya* using 17 quantitative characters provided justification for the existing classification of the species. For the first time such an observation has been reported on quantitative basis. Six characters which, include plant height, stem height, stem diameter, habitat, pod length and pod width contributed significantly to the delimitation of the species of *S. didymobotrya* populations studied. In future, these characters can be evaluated for morphometric analysis. Taxonomic

characters hereby presented cannot be treated as a universal one since the individuals studied were from only four counties in Kenya until similar studies are done across the species range of occurrence. An application of this method in an elaborate taxonomic review of the *S. didymobotrya* species in future is recommended. These results can also be confirmed through molecular techniques and other biological information like reproductive biology and population genetics.

Acknowledgement
The authors would like to acknowledge the late Kamundia John of KALRO- Njoro, Kenya for his assistance in statistical analysis . We thank the technical staff of the Department of Botany, Jaramogi Oginga Odinga University of Science and Technology for their technical support during preparation of the sample materials. Also to collaborators who accorded us moral support and assistance during the study. This work was partially supported by grant from Deutscher Akademischer Austauschdienst (DAAD) through in-country scholarship and the National Commission for Science, Technology and Innovation (NACOSTI) through the Women Scientist Program Call II. We thank all local communities in Nandi, Nakuru and Siaya counties

REFERENCES

Agnew, A. D. Q. (1994). Upland Kenya wild flower: In a flora of the ferns and herbaceous flowering plants of Upland Kenya. Oxford University Press, London.

Beentje, H. (1995). Kenyan trees, shrubs and lianas. National Museums of Kenya, Nairobi.

Benesi, I. R. M., Labuschagne, M. T., Herselman, L. and Mahungu, N. (2010). Ethnobotany, morphology and genotyping of cassava germplasm from Malawi. Journal of Biological Sciences, 10 (7): 616 - 623. ISSN 1727 - 3048.

Bett, S. (2010). Mursik: Indigenous milk preservation technology among the Kalenjins of Kenya. Ng'arua Maarifa Center. Article released on 3rd November 2010.Internet: http:// ngaruamain.blogspot.Com / 2010 / 11 / mursik – indigenous – milk - preservation. Html downloaded on 27th September 2013.

Binns, S. E, Baum, B. R. and Arnason, J. T. (2002). A taxonomic revision of Echinacea (Asteraceae: Heliantheae). Systematic Botany, 27: 610 - 632.

Boonkerd, T., Pechsri, S. and Baum, B. R. (2005). A phenetic study of Cassia sensu lato (Leguminosae - Caesalpinioideae: Cassieae: Cassiinae) in Thailand. Plant Systematics and Evolution, 252: 153 - 165.

Bolourian, S. and Pakravan, M. (2011). A morphometric study of the annual species of Alyssum (Brassicaceae) in Iran based on their macro - and micromorphological characters. Phytologia Balcanica, 17 (3): 283 - 289.

Boyd, A. (2002) Morphological analysis of Sky Island populations of Macromeria viridiflora (Boraginaceae). Systematic Botany, 27: 116 - 126.

Campey, M. L. Waycott, M. and Kendrick, G. A. (2000). Re - evaluating species boundaries among members of the Posidonia ostenfeldii species complex (Posidoniaceae) - morphological and genetic variation. AquaticBotany,66: 41 - 56.

Casavi, P. V., Saidman, B. O., Vilardi, J. C. and Cialdella, A. M. (2002). First comparative phenetic studies of Argentinean species of Acacia (Fabaceae) using morphometric, isozymal and RAPD approaches. American Journal of Botany, 89 (5): 843 - 853.

Cherono, S. and Akoo, A. (2011, July 5). Kenya: Mursik not too sweet for plant. Daily Nation, p. 11

Chiapella J., (2000). The Deschampsia cespitosa complex in Central and Northern Europe: a morphological analysis. Botanical Journal of Linnean Society, 134: 495 - 512.

Cláudio, V. J., Marcos, P. Amanda de, R., Lidilhone, H., Dulce, H. S. S. and Vanderlan da, S. B. (2013). (–) – 7 - Hydroxycassine: a New 2, 6 – Dialkylpiperidin – 3 - ol alkaloid and other constituents isolated from flowers and fruits of Senna spectabilis (Fabaceae). Journal of Brazil Chemical Society, 24 (2): 230 - 235.

Davis, P. H. and Heywood, V. H. (1963). Pricinples of angiosperm taxonomy. Oliver and Boyd, Edinburgh.

Deduke, C., Timsina, B.and Piercey - Normore, M. D. (2012). Effects of environmental change on secondary metabolite production in Lichen - forming fungi. International perspectives on global environmental change. Dr. Stephen Young (Ed.), ISBN: 978 – 953 – 307 – 815 - 1, In Tech, Available from: http: // www. Intechopen. Com / books / international – perspectives – on – global – environmental – change / effect – of – environmental – change – on – secondary – metabolite – production – in – lichen – forming - fungi. Downloaded on 28 / 4 / 2014.

Deshmukh, S. A. (2011). Morphometrics of the genus Cassia L. from Kolhapur district. The Bioscan, 6 (3): 459 - 462.

Deshmukh, S. A., Labhane, N., M., Waghmare, M. B., Chavan, S. R. and Gaikwad, D. K. (2013). Morphometric studies in the genus Caesalpinia L. from Kolhapur District. Research and Reviews: Journal of Botany, 2(1): 25 - 28.

Dias, E. S. D. (1999). Morphometric and phenetic studies of five geographical populations of Lutzomyia whitmani (Diptera: Psychodidae) in Brazil. Journal of Medical Entomology, 36 (6): 846 - 850.

Essilfie, M. K. and Oteng - Yeboah, A. A. (2013). Morphometric studies of Clausena anisata (Willd.) Hook. F. ex. Benth. in Coastal Savanna zone of Ghana. West African Journal of Applied Ecology, 21 (1): 1 - 13.

Feng, S. P., Li, W. G., Huang, H. S., Wang, J. Y. and Wu, Y. T. (2009). Development, characterization and cross – species / genera transferability of EST - SSR markers for rubber tree Hevea brasiliensis. Molecular Breeding, 23: 85 – 97.

Gairola, S., Shariff, N. S., Bhatt, A. and Kala, C. P. (2010). Influence of climate change on production of secondary chemicals in high altitude medicinal plants: Issue needs immediate attention. Journal of Medicinal Plants Research, 4 (18): 1825 - 1829.

Gakuubi, M. M. and Wanzala, W. (2012). A survey of plants and plant products traditionally used in livestock health management in Buuri district, Meru County, Kenya. Journal of Ethnobiology and Ethnomedicine, 8: 39.

Ganapaty, S., Thomas, P. S., Ramana, K. V. Vidyadhar, K. and Chakradhar, V. (2002).A review of phytochemical studies of Cassia species. Journal of Natural Remedies, 2 (2): 102 – 120.

Gengler - Nowak, K. (2002). Phenetic analyses of morphological traits in the Malesherbia humilis complex (Malesherbiaceae). Taxon, 51: 281 - 293.

Gomez - Campo, C., Herranz - Sanz, J. M. and Montero - Riquelme, F. (2001). The genus Coincya rouy (Cruciferae) in South - Central Spain revisited: A morphometric analysis of population structure. Botanical Journal of Linnean Society, 135: 125 - 135.

Harbone, J. B. (1984). Phytochemical methods: A guide to modern techniques of plant analysis, Chapman and Hall Ltd, London.

Henderson, A. and Ferreira, E. (2002). A morphometric study of Synechanthus (Palmae). Systematic Botany, 27 (4): 693 - 702.

Henderson, A. (2006). Traditional morphometrics in plant systematics and its role in palm systematics. Botanical Journal of the Linnean Society, 151: 103 - 111.

ICRAF, (1992). A selection of useful trees and shrubs for Kenya. ICRAF, Nairobi.

Irwin, H. S. and Barneby, R. C. (1982). The American Cassiinae. Mem. New York Botanical Garden. 35: 1 - 918.

Jongebloed, U. Szederkenyi, J. Hartg, K., Schobert, C. and Komor, E. (2004). Sequence of morphological and physiological events during natural ageing and senescence of a castor bean leaf: sieve tube occlusion and carbohydrate back - up precede chlorophyll degradation. PhysiologyPlantarum, 120: 338 - 346.

Jeruto, P., Lukhoba, C., Ouma, G., Mutai, C. and Otieno, D. (2008). An Ethnobotanical study of medicinal plants used by the Nandi people in Kenya. Journal of Ethnopharmacology, 116: 370 - 376.

Jeruto, P. (2009). Ethnobotanical survey, phytochemical analysis, Bioassay and propagation of some endangered medicinal plants from Aldai division, South Nandi district of Kenya. Msc Thesis submitted to Maseno University (Unpublished).

Kokwaro, J. O. (2009). Medicinal plants of East Africa. 3rd Edition. Nairobi, Kenya: University of Nairobi Press; 2009. ISBN 9966 – 846 – 84 - 0.

Kent, M. and Cooke, P. (1992). Vegetation description and analysis: A practical approach. John Wiley, New York.

Kropf, M. (2008). Intraspecific patterns of mountain plants: A morphometric analysis confirms molecular results in the Subterranean Oreophyte Anthyllis montana L. (Fabaceae). Taxon, 57 (2): 511 - 524.

Leelambika, M., Mahesh, S., Jaheer, M. and Sathyanarayana, N. (2010). Comparative evaluation of genetic diversity among Indian Mucuna species using morphometric, biochemical and molecular approaches. World Journal of Agricultural Sciences, 6 (5): 568 - 578.

Leenhouts, P. W. (1968).A guide to practice of herbarium taxonomy. Utreth Netherlands, the International Bureau of Taxonomy and Nomenclature.

Madureira, A. M., Ratmalhete, C., Mulhovo, S., Duarte, A. and Ferreira, M. (2012). Antibacterial activity of some African medicinal plants used traditionally against infectious disesases. Pharmaceutical Biology, 50 (4): 484 - 489. ISSN 1388 - 0209 print / ISSN 1744 - 5116 0nline. DOI:10.3109/13880209.2011.615841.

Maggs - Kolling, G. L., Madesen, S. and Christiansen, J. (2000). A phenetic analysis of morphological variation in Citrullus lanatus in Namibia. Genetic Resources and Crop Evolution, 47 (4): 385 - 393.

Marazzi, B., Endress, K. P., de Queiroz, L. P. and Conti, E. (2006). Phylogenetic relationships within Senna (Leguminosae, Cassiinae) based on three chloroplast DNA regions: patterns in the evolution of floral symmetry and extrafloral nectaries. American Journal of Botany, 93 (20): 288 - 303.

Mazumnder, P. M., Percha, V., Farswan, M. and Upaganlawer, A. (2008). Senna: A wonderful gift of medical sciences. International Journal of Community Pharmacy, 1 (2): 17 - 38.

Mulumba, J. W. and Kakudidi, E. 2009. Numerical taxonomic study of Acacia senegal (Fabaceae) in the cattle corridor of Uganda. South African Journal of Botany, 76: 272 - 278.

Mulumba, J. W. and Kakudidi, E. (2011). Infraspecific delimitation of Acacia senegal (Fabaceae) in Uganda. American Journal of Plant Sciences, 2: 345 - 353.

Ngule, C. M., Swamy, T. A. and Obey, J. K. (2013). Phytochemical and bioactivity evaluation of Senna didymobotrya Fresen Irwin used by the Nandi community in Kenya. International Journal of Bioassays, 2 (7): 1037 - 1043.

Njoroge, G. N. and Bussmann, R. W. (2006). Diversity and utilization of antimalarial ethnophytotherapeutic remedies among the Kikuyus (Central Kenya). Journal of Ethnobiology and Ethnomedicine, 2: 8.

Nyaberi, M. O., Onyango, C. A., Mathoko, F. M. Maina, J. M. Makobe, M. and Mwaura, F. (2013). Bioactive fractions in the stem charcoal of Senna didymobotrya Freasen Irwin and Barney used by pastoral communities in West Pokot to preserve milk. Natural Resource Management: 16: 980 - 985.

Orwa C, Mutua A, Kindt, R., Jamnadass, R. and Simons, A. (2009). Agroforestree Database: a tree reference and selection guide version 4.0 (http: // www. worldagroforestry. org / sites / treedbs / treedatabases. asp) downloaded on 7 / 10/10.

Pinheiro, F. and de Barros, F. (2007). Morphometric analysis of Epidendrum secundum (Orchidaceae) in South-Eastern Brazil. Nordic Journal of Botany, 25: 129 - 136.

Rahman, M. O. (2013). Numerical taxonomy of the genus Senna Mill. from Bangladesh. Journal of Plant Taxonomy, 20 (1): 77 - 83.

Rahman, M. Z. and Rahman, M. O. (2012). A morphometric analysis of Desmodium Desv. (Fabaceae) in Bangladesh. Bangladesh Journal of Botany, 41 (2): 143 - 148.

Rahman, M. O., Zahidur, R. and Begum, A. (2013). Numerical taxonomy of the genus Senna Mill. from Bangladesh. Bangladesh Journal Plant Taxonomy, 20 (1): 77 – 83.

Rajakaruna, N., Harris, S. C. and Towers, G. H. N. (2002). Antimicrobial activity of plants collected from Serpentine outcrops in Sri Lanka. Pharmacological Biology, 40: 235 - 244.

Ramakrishna, A. and Ravishankar, G. A. (2011). Influence of abiotic stress signals on secondary metabolites in plants. Plant Signal Behaviour, 6 (11): 1720 - 1730.

Rehm, S. (1994). Multilingual dictionary of agronomic plants. Kluwer Academic Publishers, Netherlands.

Repka, R. (2003). The Carex muricata aggregate in the Czech Republic: multivariate analysis of quantitative morphological characters. Preslia (Praha), 75: 233 – 248.

Schmalzel, R. J., Nixon, R. T., Best, A. L. and Tress, J. A. Jr. (2004). A morphometric variation in Coryphantha robustispina (Cactaceae). Systematic Botany, 29: 553 – 568.

Soladoye, M. O., Sonibare, M. A. and Rosanwo, T. O. (2008). Phytochemical and morphometric analysis of the genus Acalypha Linn. (Euphorbiaceae). Journal of Applied Science, 8 (17): 3044 – 3049.

Soladoye, M. O., Onakoya, M. A., Chukwuma, E. M. and Sonibare, M. A. (2010 a) Morphometric study of the genus Senna Mill. in South - Western Nigeria. African Journal of Plant Science, 4 (3): 044 - 052. ISSN 1996 - 0824. Http: // www. academicjournals . org / ajps. Academic Journals.

Soladoye, M. O., Sonibare, M. A. and Chukwuma, E. C. (2010 b). Morphometric study of the genus Indigofera Linn. (Leguminosae - Papilionoideae) in South - Western Nigeria. International Journal of Botany, 6(3): 343 - 350.

Sonibare, M. A., Jayeola, A. A. and Egunyomi, A. (2004). A morphometric analysis of the genus Ficus Linn. (Moraceae). African Journal of Biotechnology, 3 (4): 229 - 235.

Stace, C. (1993). Taxonomy and Biosytematics. Second Edition. Oxford University Press, London.

Sunarno, B. (1997). Senna didymobotrya (Fresenianus) Irwin & Barneby. In Faridah Hanum, I. and van derMaesen, L. J. G. (Eds.): Plant Resources of South - East Asia. No. 11. Auxillary Plants. Prosea Foundation, Bogor, Indonesia: 229 – 231.

Suwanvijitr, T., Kaewmuangmoon, J., Cherdshewasart, W. and Chanchao, C. (2010). Morphometric and genetic variation in Pueraria mirifica cultivars across Thailand. Pakistan Journal of Botany, 42 (1): 97 - 109.

Szczepaniak, M., Ciesiak, E. and Bednarek, P. T. (2002). Morphological and AFLP variation of Elymus repens (L.) Gould. (Poaceae). Cell Molecular Biology Letters7: 547 - 558.

Wagate, C. G., Mbaria, M. J., Gakuya, D.W., Nanyingi, M. O., Kareru, P. G., Njuguna, A., Gitau, N., Macharia, J. K. and Njonge, F. (2012). Screening of some Kenyan medicinal plants for antibacterial activity. Phytotherapy Research, 24: 150 - 153. Wiley InterScience.

Wang, J. L., Gao, Y. B., Zhao, N. X., Ren, A. Z., Ruan, W. B., Chen, L., Liu, J. L. and Li, C. L. (2006). Morphological and RAPD analysis of the dominant species Stipa krylovii Roshev. in inner Mongolia steppe. Botanical Studies, 47: 23 - 35.

Yousah, Z., Shinwari, Z. K. and Khan, M. R. (2010). Phenetic analysis of medicinally important species of the genus Solanum from Pakistan. Pakistan Journal of Botany, 42 (3): 1827 - 1833.