CHAPTER ONE

1.0 INTRODUCTION AND BACKGROUND OF STUDY.

1.0.1 The Genus *Solanum* L.

Solanaceae is an important family of the Angiosperms of the advanced order Solanales in the division Magnoliophyta (Bremer *et al.*, 2003). It includes 91 genera and an estimated number of 2450 species with great variations in habit, morphology and ecology (Mabberley, 2008). The Solanaceae family also informally known as the nightshade or potato family is ranked third in economic importance and is characteristically ethnobotanical, that is, it is extensively used by humans. It is an important source of food, spice and medicine. However, Solanaceae species are often rich in alkaloids whose toxicity to humans and animals range from mildly irritating to causing death in small quantities (Mueller *et al*., 2005; Sekara *et al*., 2007). Tomato, potato, pepper, petunia, datura, tobacco and eggplant are some of the valuable family members of Solanaceae (Doganlar *et al*., 2002b; Knapp *et al*., 2004). The Solanaceae family members adapt well to different agro-ecological environments, hence they disperse across the globe easily (Knapp *et al*., 2004). Considering their worldwide distribution, a remarkably high level of morphological diversity has manifested at the species, the cultivar and the generic levels (Knapp *et al*., 2004).

Solanum L**.** is the largest genus of the family Solanaceae with over 2,000 species. It includes perennial shrubs, vines, herbs, or trees, with or without spines, glabrous or pubescent with unbranched or branched, often glandular hairs. The generic name was first used by Pliny the Elder (23-79AD) for a plant also known as Strychnos, most likely *Solanum nigrum* L (Mueller *et al*., 2005). Its derivation is uncertain, possibly stemming from the Latin word *sol*, meaning "sun", referring to its status as a plant of the sun. Another possibility is that the root was *solare*, meaning "to soothe," or *solamen*, meaning "a comfort," which would refer to the soothing effects of the

plant upon ingestion (Quattrocchi, 2000) or "quieting", alluding to the sedative properties **(**Hyde and Wursten, 2010**).** *Solanum*, being one of the largest genera of flowering plants (Olmstead and Palmer, 1997), contains more than half of all the species in the family including all the species of wild potatoes found in the Western Hemisphere. Most species within *Solanum* are endemic to the Americas; however, close to 20% are Old World species. Many formerly independent genera like *Lycopersicon* Mill. (the tomatoes) or *Cyphomandra* Mart. ex Sendtn. are included in *Solanum* as subgenera or sections today (Bohs, 2001). Thus, the genus nowadays contains roughly 1,500-2,000 species (Bohs, 2001). Several species of the genus are cultivated including three globally important food crops namely tomato (*S. lycopersicum* L.), potato (*S. tuberosum* L.) and eggplant (*S. melongena* L.). Other species are regional significant food crops, such as: Ethiopian eggplant and gilo (*S. aethiopicum* L.), naranjilla or lulo (*S. quitoense* Lam.), Turkey berry (*S. torvum* Sw.), pepino (*S. muricatum* Aiton Hort.), or the "bush tomatoes". About 200 *Solanum* species are known in Africa, with some 32 indigenous in West Tropical Africa out of which 16 are found widely spread throughout Nigeria (Knapp, 2002a). However, the International Solanaceae Genome Project (SOL) listed 24 different *Solanum* species occurring in West Tropical Africa out of which only 13 are found in Nigeria (**Appendix 1**). Most parts of the plants, especially the green parts and unripe fruit, are poisonous to humans (although not necessarily to other animals), but many species in the genus bear some edible parts, such as: fruits, leaves, or tubers. While most medical relevance of *Solanum* is due to the poisoning nature of the plant which is common and may lead to death, several species are locally used in folk medicine, particularly by native people who have long employed them. Giant Devil's fig (*S. chrysotrichum* Schltdl.) has been shown to be an effective treatment for seborrhoeic dermatitis (disease of the sebaceous, or oil glands of the skin) in a scientific study (Donzella *et al*., 2000; Herrera-Arellano *et al*., 2004).

The common name "Eggplant" is given to vegetable *Solanum* which encompasses three closely related cultivated species that belong to *Solanum* subgenus Leptostemonum: *Solanum melongena*, *S. aethiopicum*, and *S. macrocarpon*. (Daunay *et al*., 2001a; Doganlar *et al*. 2002a). The name eggplant comes from the shape (egg-like) and colour (white) of the vegetable's fruit (Lester, 1998). Among these species, *S. melongena* is most commonly referred to as eggplant. There are several other terms used for *S. melongena* and examples include brinjal eggplant, aubergine or guinea squash (Chowdhury 1995, Daunay *et al*., 1999 and Kashyap *et al.*, 2003). However, Brinjal eggplant is the most common name that is used to refer to *S. melongena*. Due to confusion about use of the term "eggplant", Daunay *et al*., (2001b) indicated that eggplant may be used as nomenclature to refer to several *Solanum* species important for human diet and health such as: *Solanum melongena, S. aethiopicum, S. macrocarpon, S. quitoense* Lam. , *S. sessiliflorum* Dunal. and related species. Although most Leptostemonum species are of the New World origin (Daunay *et al*., 1999) all the three eggplant species as well as their wild relatives are endemic to the Old World (Lester, 1998). Phylogenetic classification of species in *Solanum* using chloroplast DNA restriction site variation reveals that within Leptostemonum, the Old World and Australian species form a monophyletic clade (Olmstead and Palmer, 1997). *Solanum aethiopicum* and *S. macrocarpon* have been domesticated in Africa from their wild relatives, *S. anguivi* Lam. and *S. dasyphyllum* Schum. & Thonn. respectively (Lester, 1998). The cultivation of these two species for their fruits and leaves is still primarily limited to Africa (Scippers, 2000; Doganlar *et al*., 2002b). The precise origin of the brinjal eggplant, *S. melongena*, is uncertain; however, it may have been indirectly derived from the wild African species *S. incanum* L. (Daunay *et al*., 2001a) and domesticated in India and Southeast China. Many vegetable *Solanum* species that occur in Nigeria are sources of food and are of medicinal importance (Gbile and Adesina, 1988).

1.0.2 Diversity within vegetable *Solanum*

Members of the genus *Solanum* are varied morphologically, diverse in number and ecogeographically distributed. By far, the greatest number of *Solanum* species occurs in South America, especially on the slopes of the Andes, but secondary centres of diversity and endemism are found in North America, Mexico, Central America, eastern Brazil, the West Indies, Australia, Africa and Madagascar (Levin *et al*., 2005). The domesticated species are often diploids (2n = 24), with many wide-spread escape in the wild (Oyelana and Ugborogho, 2008). The taxonomy of the group has remained challenging due to species' large size, overlapping ecogeographical distribution (Levin *et al*., 2005), morphological plasticity, similar genomes (Okoli, 1988) and existence of swarms of natural hybrids (Obute *et al*., 2006; Oyelana and Ugborogho, 2008). The inconsistencies and misconceptions generated by these factors have made past attempts at taxonomically resolving the complexities associated with the genus difficult.

The extreme diversity of the species belonging to *Solanum* may be attributed to its great antiquity, as well as its extraordinary rate of speciation (Samuels, 1996). The current infrageneric subdivisions within *Solanum* have been challenged by several investigators. *Solanum* is a taxonomical paradox, which exhibits both uniformity and extreme diversity in its morphology at once (Knapp *et al*., 2004). Knapp *et al*., (2004) stated further that this hyperdiversity in one genus is unusual in angiosperms; it makes *Solanum* interesting from an evolutionary standpoint as well as for its usefulness to humans. Bohs (2005) reported that Linnaeus (1753) divided *Solanum* into two groups, Spinosa and Inermia, based on the presence or absence of spines. Bohs (2005) stated further that Dunal (1816) also described two categories, Aculeata and Inermia in his monographs. Sakata and Lester (1997), citing Bitter (1923) who was criticized for splitting the genus to an excessive extent, described more than 60 new *Solanum* species from the Americas. Bohs and Olmstead (1997) stated that previous works [citing Seithe (1962), Danert (1970) and Gilli (1970)] provided elements for D'Arcy's (1991) scheme which is widely used today. Well-defined and

probably monophyletic subgenera and sections exist along with a plethora of poorly circumscribed groups. Significant numbers of *Solanum* species have no conclusive subgeneric or sectional affiliation. Even where well-characterized infrageneric groups exist, their phylogenetic relationships to other groups are unknown (Bohs and Olmstead, 1997). All these contributed to *Solanum* being riddled with taxonomic confusion (Lester, 1997). The difficulty of associating the species names of *Solanum* used by earlier taxonomists with the plants of today is due to brevity and vagueness of the early descriptions, which also lack characters now considered to be diagnostic (Bukenya and Carasco, 1995). Levin *et al*., (2006) in their contributions citing Hepper (1979) stated that another problem is that some of the early names, for example many of the names given by Linnaeus and those before him, are difficult to typify. Several other authors who have also provided schemes for infrageneric groups are Nee (1999), Child and Lester (2001), Hunziker (2001) and Nee *et al*., (2006).

1.2 Statement of Problem

Most of the works done on the *Solanum* species found in Nigeria are mainly based on medicinal and food values (Gbile and Adesina, 1988), genome description (Okoli, 1988), and chromosome morphology (Oyelana and Ugborogho, 2008). For an effective classification through phylogenetic studies, it is essential to have information concerning the extent of genetic diversity within a crop species. This information is particularly useful for characterizing individual accessions and cultivars and as a general guide in the selection of the parents for hybridization. Genetic diversity refers to the variations at the level of individual genes (polymorphism), and provides a mechanism for populations to adapt to their ever-changing environment. The more variations noticed in characters among species of a genus, the better, in order to identify major clades within genus and to gain insight into phylogenetic relationships among these species.

A simple and precise technique for measuring the overall genetic diversity of a crop is not yet available, and no single approach can be considered the best for measuring diversity (Ng and Padulosi, 1997). The classification of crop plants and the determination of their interrelationships require morphological traits together with sophisticated analysis such as the molecular studies. Ogunkanmi (2006) stated that molecular markers are being used in many aspects of plant genetics and breeding and have been applied to basic studies of taxonomy, variability of populations and mating systems. Ogunkanmi (2006) further stated that molecular markers based on differences in DNA sequences between individuals detect more polymorphisms than morphological and protein-based markers and constitute a new generation of genetic markers. In Nigeria, not many works have been done on the nature of genetic diversity and characterization of vegetable *Solanum*, especially using molecular methods. As a result, this study attempts to resolve to a larger extent the taxonomic difficulties associated with vegetable *Solanum* especially among the species found in Nigeria using both morphological and molecular methods, and at the same time provide information needed for characterization and conservation.

1.3 Aim and Objectives

1.3.1 Aim

The aim of this project is to use both morphological and molecular approaches to determine the genetic diversity among eggplants and their related species in Southern Nigeria for the purpose of identification and authentication.

1.3.2 Objectives

1.3.2.1 General Objective

One major focus of this work is on the systematic and evolutionary relationships in vegetable *Solanum*. This will also include establishment of an overall phylogeny for Nigerian species of vegetable *Solanum* using molecular and morphological approaches.

1.3.2.2 Specific Objectives

These include:

1. To explore the variation in germplasm of different species of vegetable *Solanum* in Southern Nigeria, with emphasis on the collections, identification, documentation and preservation of all voucher specimens in secure repositories.
2. To determine the level of genetic diversity among individuals of different and same species (i.e. inter/intra genetic relationships) using both morphological and DNA-based markers.
3. To establish phylogenetic relationships among the taxa studied using both morphological and molecular data obtained**.**
4. To identify agronomic marker traits useful for classification of the taxa using Principal Component Analysis.

1.4 Significance of Study

The significance of this study is to provide essential information for the easy identification of the taxa and the conservation of the species. It is also to gain a better insight into the centre of diversity (endemism) of the plants, locate the probable source in Nigeria with a view to conserving them for maximum genetic diversity and sustainable utilization.

1.5 Research Questions

The classification and phylogeny of eggplant *Solanum* have generated a lot of dispute worldwide; therefore the study of the genetic diversity on this genus in Nigeria is imperative. This current study was therefore, designed to provide answers to the following research questions:

1. What is the extent of variation in germplasm of different species of vegetable *Solanum* in Southern Nigeria?
2. How can the level of genetic diversity among individuals of different and same species be best determined?
3. What kinds of phylogenetic relationships exist among members of the eggplant and related species found in Southern Nigeria?
4. What are the important agronomic traits that most effectively discriminate among eggplant accessions and related species thereby useful for classification of the taxa?

1.6 Operational Definition of Terms and Abbreviations

1.6.1 Definition of Terms

Bootstrapping: This is a statistical method for estimating the sampling distribution of an estimator by sampling with replacement from the original sample, most often with the purpose of deriving robust estimates of standard errors and confidence intervals of a population parameter like a mean, median, proportion, ratio, correlation coefficient or regression coefficient.

Cladistic Analysis: A system of biological taxonomy based on the quantitative analysis of comparative data and used to reconstruct cladograms summarizing the (assumed) phylogenetic relations and evolutionary history of groups of organisms.

Exon: An exon is a nucleic acid sequence that is represented in the mature form of a RNA molecule either after portions of a precursor RNA (introns) has been removed by cis- splicing or when two or more precursor RNA molecules have been ligated by trans- splicing.

Global Positioning System (**GPS):** The **GPS** is a space-based satellite navigation system that provides location and time information in all weather, anywhere on earth or near the earth, where there is an unobstructed line of sight to four or more GPS satellites.

Heuristic Search: It designates a computational method that optimizes a problem by iteratively trying to improve a candidate solution with regard to a given measure of quality.

Intron: An intron is a DNA region within a gene that is not translated into protein.

Jackknifing: It is used to estimate the bias and standard error (variance) of a statistic, when a random sample of observations is used to calculate it, which is, estimating the precision of sample statistics (medians, variances, percentiles) by using subsets of available data.

Maximum Parsimony: Maximum Parsimony is a character-based method that infers a phylogenetic tree by minimizing the total number of evolutionary steps required to explain a given set of data, or in other words by minimizing the total tree length.

Molecular Phylogenetics: This is the use of the structure of molecules to gain information on an organism's evolutionary relationships.

Parsimony: Parsimony implies that simpler hypotheses are preferable to more complicated ones

1.6.2 Abbreviations and Acronyms

BLAST: Basic Local Alignment Search Tool.

BSA: Bovine Serum Albumin

DNA: Deoxyribonucleic acid. It is a nucleic acid that contains the genetic instructions used in the development and functioning of all known living organisms (with the exception of RNA viruses).

ITIS: Integrated Taxonomic Information System

ITS: Internal Transcribed Spacer. It is found in the nucleus.

NCBI: The National Center for Biotechnology Information. It advances science and health by providing access to biomedical and genomic information.

PCR: Polymerase Chain Reaction is the enzymatic synthesis of multiple copies of a specific DNA sequence in a cyclical manner.

PhyML: Phylogenetic estimation using Maximum Likelihood

RNA: Ribonucleic acid.

***s.l.*:** *sensu lato* (in the widest sense)

***s.s.*:** *sensu stricto* (in the strict sense)

SEVAG: 24 chloroform: 1 ethyl alcohol

trnC: Transfer RNA gene for tRNA (Cysteine); found in the chloroplast.

trnF: Transfer RNA gene for tRNA (Phenylalanine); found in the chloroplast.

trnL: Transfer RNA gene for tRNA (Leucine); found in the chloroplast.

UPGMA: Unweighted Pair Group Method with Mathematic Average

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Crop Histories, Origin and Domestication

Bhaduri (1951) and Khan (1979) as cited by Daunay *et al*., (2004) reported that the first records of the use of *S. melongena* are found in Sanskrit documents, and are dated in the beginning of the Christian era. The extraordinarily large number of words for the eggplant in Sanskrit, and the fact that some of them are not only descriptive but also highly complementary suggests that the eggplant was not only common but was also popular in the ancient days with Sanskrit-speaking people. The mention of eggplant is also found in Chinese botanical and agricultural treatises of about the same period such as the 'Atlas of plants in Southern China' written during the Western Jin Dynasty (AD 265- 316), the 'Qimi Yiaoshu' a practical handbook of agriculture written at the time of the Southern and Northern Dynasties (AD 420-581) and in the Ts'i Min Yao Shu, a Chinese work on Agriculture of the fifth century (Bretschneider 1882, quoted by Hedrick, 1919 and cited by Daunay *et al*., 2004). From China, eggplant has reached Japan about the 8th century at the time of the Tung dynasty (Allard, 1996) where it has become a common vegetable. Allard (1996) stated further that when eggplant reached Afghanistan and Persia is still unclear and concluded that further research in historical and archaeological documents would undoubtedly bring further insight to the spread of the species throughout Asia and Middle East. According to the Encyclopaedia Iranica (1996), eggplant has been brought to the Iranian lands at a very early but indeterminable date, and Iranian and Arab sailors have carried it to East Africa in ancient times as shown by the presence of many specific terms for it in Ethiopia. The same source indicates that the Medieval Iranian writers on medicine and botany urged caution in use of the eggplant, but also mentioned its culinary use.

Eggplant has reached the Eastern Mediterranean lands probably after the Arab conquest of Iran. The conquering Turks got to know the plant in Iran. Eggplant was neither known by Greeks nor by Romans. Its spread through the Mediterranean Basin is linked to the Muslim conquests from 8th century onwards. Daunay *et al*., (2004), citing Lobel (1581) and de Candolle (1883), said that the Persian physicians Rhazes (864-925 AD) and Avicenna (also known as Ibn Sina, 980-1037) mentioned eggplant, as well as the Andalusian-Arab physician Averroes (also known as Ibn Rushd or Abu al Walid, 1126-1198). According to Daunay *et al*., (2004), the Moorish Spaniard Ibn Al Awam (12th century) described in his book on agriculture the techniques recommended for growing eggplants, and he also mentioned the presence of the crop in Syria, Egypt and Sicily. According to de Candolle (1883) cited by Daunay *et al*., (2004), the physician Ebn Baitair who lived also in Spain in the 13th century mentioned eggplant, and citing Hedrick (1919), Daunay *et al*., (2001a) reported that the German Albertus Magnus (1193-1280) had written about eggplant. All these historical traces demonstrate the ancient presence of eggplant as medicinal and food crop in the Middle East as well as in Europe. The spread of eggplant to Balkan countries is probably linked to the Ottoman Western conquests (14th to 16th centuries). In the European Medieval medico-botanical treatises (herbals), the eggplant illustrations display medium to large sized fruits, pear shaped to globose, and most often purple coloured (Daunay *et al*., 2007). In the Renaissance herbals of the 16th century, Daunay *et al*., (2007) stated further that these fruit types are still present, together with illustrations deriving from the woodcut by Fuchs (1543), displaying plants bearing egg-shaped fruits.

Many previous authors (e.g. Lester and Hassan, 1991; Bukenya and Carasco, 1999) assumed that ancestral forms of *Solanum* especially *S. melongena* originated from African tropics (where many wild relatives of eggplant are still found) and the Middle East. This assumption was based on morphometrics, crossability, seed coat scanning electron microscopy, leaf isozymes, and seed protein electrophoresis studied on a large set of accessions. It was equally reported that the closest

wild relatives of *S. melongena* which belongs to the *S. incanum* aggregate could be found in these areas (Lester and Hassan, 1991; Daunay *et al*., 2001a). This latter species is a quite complex group and an example of the difficulty to typify it can be found in Lester (1997).

S. incanum aggregate brings together four species (or groups), namely *S. campylacathum* Hochst. (Group A) distributed across the East African tropics, *S. delagoense* Dunal. (Group B) in South-Eastern Africa, *S. incanum sensu stricto* (Group C) in North-Eastern Africa and Middle East, and *S. lichtensteinii* Willd. (Group D) in South-East Africa (Lester and Hassan, 1991). Lester and Hassan (1991) developed the scenario that representatives of *S. incanum* aggregate migrated eastwards, either spontaneously or as a result of human migrations during pre-historic times. Indeed, the roots, leaves and fruits of these species are variously used, for instance, for medicinal purposes, leather tanning and milk curdling (Bukenya and Carasco, 1999; Lester and Hawkes, 2001). According to Lester and Hawkes (2001) the fruits are not eaten. However, Amar (2000) had earlier reported the use of the fruits of a local wild semi-perennial eggplant (possibly *S. incanum* Group C) in the lower Jordan valley at the time of the Mamelouk period (1250-1517) as food.

Deb (1989) reported that *S. incanum* is found as a wild plant in Southern India, but Lester and Hassan (1991), based on a wide experimental and analytical research, ascertained that the Indian putative "*S. incanum*" is a wild form of *S. melongena*. *S. incanum* differentiated progressively in South East Asia into a closely related species, the wild *S. melongena*, which is still found growing spontaneously in a large area from Southern and Eastern India to Southern China, Philippines and Indonesia, and is described by former botanists as *S. cumingii* Benth. Deb (1989) stated further that, this wild form after being subjected to a progressive domestication process gave rise to primitive eggplant cultivars known as *S. ovigerum* Dunal., with small round or oblong fruits, white, green or violet, which evolved progressively into advanced cultivars with large sized fruits. *S. insanum* L., widely spread in India, is probably a form of *S. ovigerum* which has reversed to the

wild state, developing a strong prickliness (character favourable for escaping animals grazing), a low straggling growth habit and a brief life cycle. Lester and Hassan (1991) brought all these taxa under the umbrella of *S. melongena,* structured as Group E (*S. insanum*), Group F (*S. cumingii*), Group G (*S. ovigerum*) and Group H (advanced cultivars). The case of a Thai cultivar type with vigorous plants setting round green ping pong sized berries is interesting because it is considered there as an advanced cultivar type, although the size (and colour) of its berries are much closer to those of the wild *S. incanum* than to those of a classic advanced cultivar, characterized by much larger fruits. This cultivar type can be interpreted as a cultivate survivor of an early domesticated form. The proposed scenario of *S. melongena* origin and domestication seems appropriate based on available data which demonstrate a pseudo-continuum of morphological features, cross compatibility, and genetic distances between Groups A, B, C, D of *S. incanum* and the Groups E, F, G, H of *S. melongena*. However, this scenario is open to refinements anytime; new data will bring new insights on *S. melongena* evolution history.

African eggplants – *S. aethiopicum* and *S. macrocarpon*, are the most popular native, traditional vegetables in West and Central Africa, but the productivity of these crops is still relatively low and the growing area and yields have not been statistically evaluated. The centre of these eggplants' diversity is Western Africa (Lester and Daunay, 2003). African eggplants are grown mainly in gardens and small fields near villages. *S. aethiopicum* is widely grown in South America, and *S. macrocarpon* in Asia and tropical America (Sękara *et al*., 2007). Gbile, (1987) stated that the genus *Solanum* is represented by some 25 species in Nigeria, including five introductions: *S. mammosum* L., *S. tuberosum* L., *S. melongena, S. wrightii* Nees Trans., and *S. seaforthianum* Andr. var. *disjunctum*. *Solanum macrocarpon, S. aethiopicum, S. scabrum, S. melongena, S. gilo, S. indicum, S. anomalum* Thonn. *S. americanum* P. Mill., *S. nigrum,* and others are domesticated, and their leaves or fruits or both are eaten as vegetables and used in traditional medicine. Many other *Solanum* species grow wild and are less known or used (Gbile and Adesina, 1988).

2.2 Distribution and Habitat

Eggplant has a global distribution (Bohs, 2005). By far, the greatest numbers of Solanum melongena species occur in South America, especially on the slopes of the Andes, but secondary centres of diversity and endemism are found in North America and Mexico, Central America, Eastern Brazil, the West Indies, Australia, Africa, and Madagascar (Figure 1). Within this range are several hotspots of high species diversity (Bohs, 2005; Naujeer, 2009).

Within their range, *Solanum* species occupy a huge diversity of habitats, from some of the wettest forests in the world to the driest deserts. *Solanum* species are found throughout a huge altitudinal range, from sea level to over 4500 m (ca. 13,500 ft) in the case of some potatoes (Gbile, 1987).

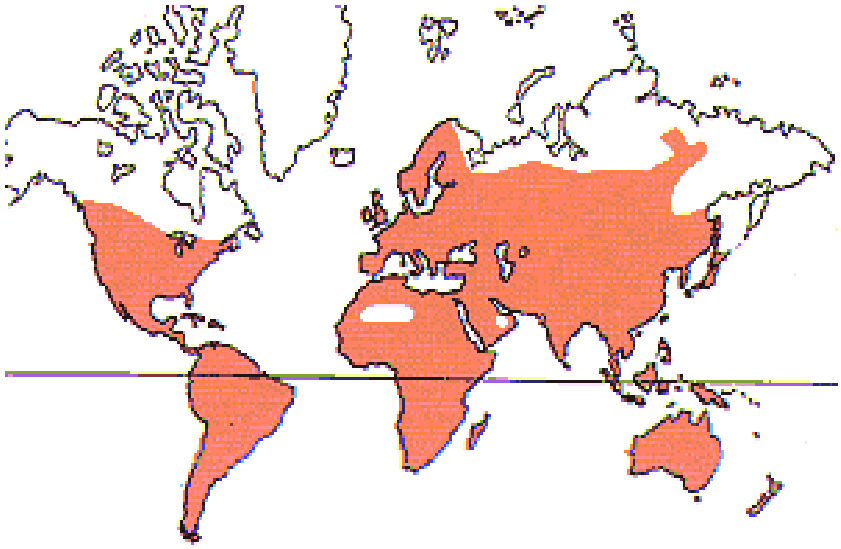


Figure 1: The dispersion of *Solanum* members around the world (Source: Naujeer, 2009)

Legend: Coloured areas indicate area of high *Solanum* species diversity

2.3 Taxonomy of Eggplant

Taxonomy is critical to the study of biodiversity. Taxonomy and systematics have been defined as two completely separate aspects of the science. Taxonomy can be thought of as three interlocking spheres of endeavour — description, identification and phylogeny (Gaston and May, 1992). Each one of these components is enriched by the other two, and without all three the science is incomplete. Phylogeny is the study of the interrelationships of organisms — how species, genera and families are related to one another. It is a known fact that all of life is related in some way; phylogeny is the assembly of the tree of life (Knapp *et al*., 2004). Today, phylogeny is done using the principles of cladistics, first laid out by the German biologist, Willi Hennig, in 1966 (Soltis and Soltis, 2003). Soltis and Soltis (2003) stated further that Hennig's methodology involves using shared, derived characteristics (synapomorphies) to group species into monophyletic clades, sets of taxa all descended from a common ancestor.

Characters can be physical characteristics of the organism, such as fruit size, shape and colour, or flower colour and shape, or can be a sequence of bases in DNA. Today, much phylogeny is done using these latter molecular characters. A phylogenetic tree is a pictorial depiction of nested sets of shared characteristics and is a powerful framework for interpretation of patterns in nature; however, it is said not to be the complete solution (Daly *et al.*, 2001; Soltis and Soltis, 2003; Knapp *et al*., 2004). Each phylogeny is a hypothesis about the relationships of the organism under study, and taxonomists use what Hennig called 'reciprocal illumination' to solve problems and investigate apparent conflicts further (Kitching *et al*., 1998; Knapp *et al*., 2004). More data or a different interpretation of the same data can cause modification of the hypothesis, and thus our ideas of how organisms are related. Knapp *et al*., (2004) further said that the construction of phylogenetic trees for many groups of organisms has revolutionized the field of systematics and has led to new interpretations of how groups are related, as well as making character analysis possible in an evolutionary framework.

Despite the importance of the genus *Solanum*, phylogenetic relationships among the taxa are currently unclear (Daunay and Lester, 1988). Members of the genus are as varied morphologically as they are diverse in number and distribution ecogeographically. *Solanum* taxonomy has been complicated and Systematists have always avoided work on it. This is largely due to species' large size, overlapping ecogeographical distribution (Levin *et al*., 2005), morphological plasticity, similarity of genomes (Okoli, 1988) and existence of swarms of natural hybrids (Obute *et al*., 2006; Oyelana and Ugborogho, 2008). The inconsistencies and misconceptions generated by these factors have made past attempts at taxonomically resolving the complexities associated with the genus difficult (Knapp *et al*., 2004; Levin *et al*., 2005).

The cultivated eggplant (*Solanum melongena*) was originally described by Carl Linnaeus in his *Species Plantarum* in 1753 (Furini and Wunder, 2004; Jarvis, 2007). According to Mace *et al* (1999), Linnaeus described the two important *S. incanum* L. and *S. melongena* L. species which are considered as the cornerstones of the "eggplant complex". However, in later works Linnaeus' concepts of these species changed quite dramatically and that has led to considerable confusion surrounding the exact delimitation of these species. A major cause of Linnaeus' confusion was the high degree of morphological plasticity shown by these species. Frary *et al* (2007) stated that this high level of variation led Dunal, a Victorian taxonomist who was preoccupied with fine details and minute differences, in 1852 to treble the number of *Solanum* species described from Africa. Dammer (1915) as cited by Frary *et al* (2007) increased not only the number (to 200) but also the confusion surrounding these species. According to Prohens *et al*. (2005), Bitter in 1923 subsequently began to unravel this confusion surrounding the African *Solanum* species partly by his use of the concept of species-aggregates, which indicates a close relationship between a group of species but does not force premature nomenclatural decisions for that group. Prohens *et al*. (2005) stated further that the work of Bitter in particular, circumscribed three species-aggregates in his series *incaniformia*, which encompassed the majority of his 81 taxa of the *S. incanum*

aggregate as it is referred to today, namely: *S. campylacanthum* (Hochst.) *sensu ampliore* Bitter, *S. bojeri* (Dunal) *sens. ampl*. Bitter and *S. incanum* L. *sens. ampl*. Bitter. However, Bitter's work was generally ignored, and most of his taxa were indiscriminately lumped together and treated as one species, *S. incanum*.

Lester and Hassan (1991) proposed a modified informal group classification of *S. incanum* and *S. melongena* (Table 1). Within the "eggplant complex" Groups G and H represent the cultivated types of eggplant. Group G is widespread in Southeast Asia and corresponds to primitive cultivars, slightly prickly with small green fruits (3-4 cm in diameter) and white stripes. Group H consists of advanced cultivars with very few tiny prickles bearing large fruits (usually >100g) highly variable in colour (Prohens *et al*., 2005; Naujeer, 2009).

Table 1: Modified informal group classification of *Solanum incanum* and *S. melongena*

Wild taxa of *S. incanum sensu lato*, from Africa		
Group A	*S. campylacanthum* Hochst.	Eastern & Southern Africa
Group B	*S. panduriforme* Dunal	Southern Africa
Group C	*S. incanum* L.	Northern Africa, Arabia
Group D	*S. lichtensteinii* Willd	Southern Africa
Weedy and cultivated taxa of *S. melongena*, from Asia		
Group E	*S. melongena* (*S. insanum*)	India
Group F	*S. melongena* (*S. cumingii* Dunal)	S.E. Asia
Group G	*S. melongena* (*S. ovigerum* Dunal)	S.E. Asia
Group H	*S. melongena* (*S. melongena*)	Worldwide

Source: Lester and Hassan, (1991).

The groupings described by Lester and Hassan (1991) have been widely commented on in literature, e.g. Sakata and Lester (1994) and Samuels (1996). However, other authors, e.g. Karihaloo and Gottlieb (1995), Karihaloo and Rai (1995), and Karihaloo *et al*., (1995), have criticised the distinction of the four groups (morphoforms) of weedy and cultivated forms of *S. melongena* (groups E–H) as being artificial. The work by Samuels (1996) gave further support for the recognition of distinct groups within the species aggregate, and in particular within groups E–H, which led him to recommend recognition at the species level of *S. campylacanthum* Hochst. ex A. Rich. (Group A, with Group B relegated to *S. campylacanthum* subsp. *Panduriforme*), *S. incanum* L. (Group C), *S. lichtensteinii* Willd. (Group D), *S. insanum* L. (Group E), *S. cumingii* Dunal (Group F), and *S. melongena* L. (encompassing Groups G and H). A paper by Sakata and Lester (1997) based on chloroplast DNA diversity gave further support to some of Samuels' claims, in particular the recognition of *S. lichtensteinii* as a distinct species.

S. melongena may have been indirectly derived from its wild *S. incanum* ancestor, domesticated in India and Southeast China. *S. aethiopicum* and *S. macrocarpon* were domesticated in Africa from their wild relatives *S. anguivi* Lam. and *S. dasyphyllum* Schumach. & Thonn respectively (Lester and Hassan, 1991; Kalloo, 1993; Sekara *et al*., 2007). Sakata and Lester (1997) generated a dendrogram from data collected on some *Solanum* samples which showed close genetic affinities between *S. incanum, S. melongena* and *S. macrocarpon* in spite of their highly confusing taxonomic relationships. Lester and Hassan (1991) indicated that although *S. incanum* taxa that occur in India can be distinguished from their wild progenitors in Africa and the Middle East, both *S. melongena* and *S. incanum* have often been confused with their distantly related African eggplant (*S. aethiopicum* and *S. macrocarpon*) and other wild species such as *S. violaceum* Ort.

2.4 Taxonomic Classifications of *Solanum*.

Different authors have contributed to the classification of the genus *Solanum* and this is summarized in the table below:

Table 2: Taxonomic classifications of *Solanum* based on different authors.

TAXONOMIC CLASSIFICATION					
TAXA	**Linnaeus**	**NCBI Taxonomy**	**ITIS**	**PreUnion Cellular Life**	**Species 2000 & ITIS Catalogue of Life 2008**
Kingdom	Plantae	Cellular organisms	Plantae Tracheobionta	Eukaryota Viridaeplantae	Plantae
Division	Magnoliophyta	Eukaryota Viridiplantae	Magnoliophyta Magnoliopsida	Streptophyceae Charales sensu lato	Magnoliophyta
Class	Magnoliopsida	Streptophyta	Asteridae	Embryophytes	Magnoliopsida
Order	Solanales	Streptophytina	Solanales	Tracheophytes	Solanales
Family	Solanaceae	Embryophyta	Solanaceae	Spermatophytes	Solanaceae
Subfamily	Solanoideae	Tracheophyta Euphyllophyta Spermatophyta		Flowering plants Dicots Core eudicots	
Tribe	Solaneae	Magnoliophyta Eudicotyledons		Asterids Euasteridsi	
Genus	*Solanum* L.	Core eudicotyledons	*Solanum* L.	Solanales Dumort. Solanaceae Juss.	*Solanum* L.
Subgenus	Leptostemonum	Asterids Lamiids Solanales		*Solanum* *Solanum* (Leptostemonum)	
Species	*Solanum melongena, S. aethiopicum, S. macrocarpon*	Solanaceae Solanoideae Solaneae	*Solanum melongena*		*Solanum melongena*
Common Name	Eggplants	*Solanum* sp. Martine 805			

NOTE: ITIS = Integrated Taxonomic Information System
NCBI = National Center for Biotechnology Information.

2.5 Phylogeny of genus *Solanum*

Several projects have focused on phylogenetic relationships in the entire genus *Solanum* and in some of its component clades. Many of the subgenera and sections are not yet valid; they are still presently used provisionally as the phylogeny of this genus is not fully resolved yet and many species have not been re-evaluated. Cladistic analyses of DNA sequence data suggest that the present subdivisions and rankings are largely invalid. Far more subgenera would seem to warrant recognition with subgenus *Leptostemonum* being the only one that can at present be clearly subdivided into sections, having 10 clades and 21 sections (Levin *et al*., 2006). Traditional taxonomists have recognized three subfamilies within the Solanaceae (D'Arcy, 1979; Pearce and Lester, 1979):

- The Solanoideae,
- The Nolanoideae, and
- The Cestroideae

Hunziker (1979) excludes the Nolanoideae (i.e., genus *Nolana*) from the Solanaceae and expanded the number of subfamilies to six:

- The Solanoideae,
- The Cestroideae,
- The Juanulloideae,
- The Salpiglossoideae,
- The Schizanthoideae, and
- The Anthocercidoideae.

In both of these schemes, *Solanum* is placed within the subfamily Solanoideae, characterized by flattened seeds with curved embryos (Hunziker, 1979). Within the Solanoideae, *Solanum* belongs to the large and complex tribe Solaneae, which encompasses about 50 genera including *S. melongena* in the scheme of Hunziker (Hunziker, 2001). Previous *Solanum* Systematists

recognized seven subgenera within *Solanum* ranging in size from subgenus *Lyciosolanum* that includes just one species to subgenus *Leptostemonum* that includes hundreds of species. According to widely accepted D'Arcy scheme which is based on the morphological characters and intuitive ideas of relationships of the species of the genus, *Solanum* comprises seven subgenera and approximately 60 to 70 sections (D'Arcy, 1991):

1. *Solanum* subg. *Archaesolanum* Marzell
2. *Solanum* subg. *Bassovia* (Aubl.) Bitter
3. *Solanum* subg. *Leptostemonum* (Dunal) Bitter
4. *Solanum* subg. *Lyciosolanum* Bitter
5. *Solanum* subg. *Minon* Raf. Sections *Brevantherum, Holophylla*
6. *Solanum* subg. *Potatoe* (G. Don) D'Arcy. Sections: *Petota, Neolycopersicon, Dulcamara, Glaucophyllum, Basarthrum, Jasminosolanum, Rhychantherum*
7. *Solanum* subg. *Solanum* Sections Q*uadrangular, Benderianum, Afrosolanum, Lemurisolanum, Macronesiotes, Solanum, Episarcophyllum, Delitescens, Capanulisolanum, Geminata, Pseudocapsicum, Chamaesarachidium*

Based on morphology and biogeography, Whalen (1984) recognized 33 informal species groups within subgenus *Leptostemonum* as well as 36 unplaced *Solanum* species. Whalen (1984) summarized the diagnostic characters, geographical distributions, and component species of each of the 33 species groups and placed them in a hypothetical phylogenetic scheme based on morphological characters, such that his work serves as an invaluable guide to relationships among this large diverse subgenus. Although Whalen's treatment is arguably the most useful, Daunay *et al*., (2004) said further that Whalen (1984) was not the first to recognize taxonomic groups of species within subgenus *Leptostemonum*.

The work of Olmstead and Palmer (1997) on molecular phylogenetic studies of the genus *Solanum* based on chloroplast DNA restriction sites support the monophyly of subgenus

Leptostemonum sensu stricto as earlier observed by Whalen (1984). However, more recent studies, using chloroplast and nuclear DNA sequence data and greater taxon sampling (Bohs and Olmstead, 2001; Bohs, 2005), suggest that the *S. wendlandii* Hook. group and perhaps also the *S. nemorense* Dunal Sol. group both of which have prickles but lack stellate hairs, may not belong to subgenus *Leptostemonum*. Subsequent molecular data from chloroplast ndhF sequences identify about 13 major clades within *Solanum* (Bohs, 2005). An overall phylogeny of *Solanum* using molecular data from three nuclear and chloroplast genes show that few of these subgenera comprise monophyletic groups (i.e. with common ancestors); instead, *Solanum* consists of about 12 to 15 major clades. These have been given informal names and are themselves the subjects of more detailed phylogenetic studies (Weese and Bohs, 2007).

Molecular phylogenetic analyses have established that the formerly segregate genera *Lycopersicum, Cyphomandra, Normania,* and *Triguera* are nested within *Solanum,* and all species of these four genera have been transferred to *Solanum*. Conversely, molecular data confirm that *Lycianthes,* sometimes considered to belong within *Solanum,* should be maintained as a separate genus. Molecular data and phylogenetic analyses challenge the traditional view of Solanaceae subfamilies (Olmstead and Palmer, 1992; Olmstead and Sweere, 1994; Fay *et al*., 1998 and Olmstead *et al*., 1999), but the precise number of monophyletic groups in the family and their names and circumscription are still under investigation. A major finding is that a member of the tribe Anthocercidae, the genus *Nicotiana,* and the subfamily Solanoideae (including *Solanum*) form a strongly supported monophyletic group characterized by a chromosome number based on x = 12 (Olmstead and Palmer, 1992; Olmstead and Sweere, 1994).

2.6 Characterization of Eggplant

The ability to characterize morphological diversity is indispensable for effective management, sustainable use of eggplant genetic resources and most importantly in phylogenetic studies. Primary characterization, as used in this study, involves measuring simple plant characters (e.g. leaf area, fruit shape, size and colour, plant prickliness and hairiness) that can be easily recorded through visual observations at different plant growth stages. Secondary characterization deals with more complicated morphological traits of agronomic importance such as pest and disease resistance, fruit set, yield potential and biochemical (glycoalkaloid or antioxidant) properties (Ayad *et al*., 1995; Naujeer, 2009).

Several workers have contributed to the characterization of the largest genus *Solanum* of *Solanaceae* family (Seithe and Anderson, 1982; Whalen, 1984; Furini and Wunder, 2004). The historical difficulties and taxonomic confusions as regard the classification of the genus *Solanum* are due to the fact that more than 3000 binomial names have been used to describe 1000 to 1400 species (Furini and Wunder, 2004). This taxonomic confusion surrounding the much crowded and complex genus *Solanum* has still not been completely resolved (Naujeer, 2009). Eggplant collections have been evaluated mostly for morphological and agronomic characters (Karihaloo and Gottlieb, 1995) revealing wide diversity in plant morphology (plant growth habit, vigour, hairiness, prickliness, fruit shape, size and colour, and yield potentials), physiology (flowering behaviour, water use) and biochemical properties (fruit bitterness, glycoalkaloid content) (Collonier *et al*., 2001; Naujeer, 2009). Fruit colour, size and shape are the most distinctive characters that vary between the cultivated *Solanum* species and their wild types (Kumar *et al*., 2008). Chowdhury (1976) had earlier classified eggplant into three main groups based on their fruit shapes: round, oval or egg-shaped (*S. melongena* var. *esculentum* Martin & Rhodes), long

slender shaped (*S. melongena* var. *serpentinum* Martin & Rhodes) and dwarf types (*S. melongena* var. *depressum* Martin & Rhodes) as cited by Martin and Rhodes (1979).

Karihaloo and Gottlieb (1995) referred to a number of wild and weedy taxa occurring in India and bearing close morphological similarities to *S. melongena* which further complicates the eggplant systematics. Lester and Hassan (1990) also reported the taxonomic confusion between *S. melongena* and its weedy form, *S. incanum.* The high level of morphological plasticity manifested at the generic, species and cultivar levels within the eggplant complex (Furini and Wunder, 2004), and the crossability affinities between *S. melongena* and other distantly related *Solanum* species producing fertile F1 hybrids (Daunay *et al*., 1991) makes classification much more complicated. India or Indochina is reported to be the centre of eggplant diversity (Lester and Hassan, 1991) but the affinities of *S. melongena* to related species are uncertain (Furini and Wunder, 2004).

2.7 Molecular tools in Eggplant *Solanum* Characterization

The taxonomic uncertainties still persist in this genus largely because previous studies to address the taxonomic problem of vegetable *Solanum* have focused mainly on morphology (Deb 1989; Karihaloo and Rai 1995), crossability and F1 fertility (Baksh, 1979; Hassan and Lester 1990a; Lester and Hassan, 1991; Furini and Wunder, 2004) and anatomy (Hassan and Lester 1990b). Establishing genetic affinities on such parameters are insufficient, as *Solanum* makes successful crosses with putative progenitors as well as distantly related species. Later taxonomic studies were based on (1) diverse combinations of accessions belonging to groups A, B, C, D, E, F, G or H (Lester and Hassan, 1991), together with other *Solanum* species, and (2) the use of allozymes (Isshiki *et al*., 1994; Karihaloo and Gottlieb, 1995), enzymes (Kaur *et al*., 2004), seed proteins (Karihaloo *et al*., 2002), and chloroplast DNA diversity (Sakata *et al*., 1991; Sakata, 1992; Sakata and Lester, 1994; 1997).

However, very little work has been done, so far, to address the problem of the status of the cultivated eggplant, and its relationship with near relatives, by utilizing nuclear DNA diversity. Gepts (1993) observed that DNA-based markers provide powerful tools for discerning variations within crop germplasm and for studying evolutionary relationships. The advent of molecular biology has revolutionized the field of plant systematics and has led to new insights into phylogenetic relationships at all taxonomic levels (Bohs, 2005). The use of molecular techniques in genetic diversity studies is supported by the finding that evolutionary forces such as natural selection and genetic drift produce divergent phylogenetic branching which can be recognized because the molecular sequences, on which they are based, share a common ancestor (Singh *et al*., 2006). Among these was the work of Karihaloo *et al*., (1995) which focused directly on nuclear genomic diversity by undertaking RAPD analysis. Karihaloo and Gottlieb (1995) also reported that greater DNA polymorphism exists in weedy *S. insanum* than in advanced cultivars of eggplant. Molecular genetics techniques were also used in the studies of Olmstead and Palmer (1997) and Olmstead *et al*., (1999) to investigate relationships across the Solanaceae. Other studies such as that of Tohme *et al*., (1996), Bohs and Olmstead (1999; 2001), Jacoby *et al*. (2003), Stedje and Bukenya-Ziraba (2003), Furini and Wunder (2004), Levin *et al.* (2005; 2006) and Polignano *et al*., (2009) provide information at the subgeneric and sectional levels by using molecular techniques.

The study of Bohs (2005) identified about 13 major clades within *Solanum* using molecular data from chloroplast ndhF sequences and a broad spectrum of samples from *Solanum* subgroups. Sampling of Bohs (2005) included all the seven subgenera listed in D'Arcy's (1972) conspectus, and 40 of the 62 sections in D'Arcy (1991). The study recognized several new clades such as the Dulcamaroid and Morelloid clades which include species from different taxonomical groups. Even though the clades identified by Bohs (2005) are well supported, they need to be corroborated by data from other genes and both morphological and biochemical characters

should be examined together. Moreover, new formal taxonomic designations for infrageneric categories in *Solanum* are strongly discouraged without more extensive data and sampling (Bohs 2005). Most papers provide information about phylogenetic relationships using single-locus methods (Bohs and Olmstead 1997; Bohs 2005). Other studies use DNA sequence data from nuclear regions such as ITS and granule-bound starch synthase gene (GBSSI or waxy) or chloroplast regions (trnT-trnF and trnS-trnG) or combinations of these data (Levin *et al*., 2005; 2006). Such single-locus-based species trees contain some errors (Takahata and Nei, 1985; Neigel and Avise, 1986). A substantial number of errors can be avoided by using multilocus methods such as the Random Amplified Polymorphic DNA (RAPD) or Restriction Fragment Length Polymorphism (RFLP). These methods provide species trees based on the phylogenetically relevant information contained in many loci or ideally in the whole genome (Hampl *et al*., 2001). The RAPD technique (Williams *et al*., 1990; Welsh and McClelland, 1990) is a frequently used tool in population and evolutionary genetics, since no prior knowledge of the genome structure or sequence data is required. Nevertheless, there are several technical and theoretical concerns regarding the applicability of the RAPD method (Van de Zande and Bijlsma, 1995). Reproducibility can be a problem, but this can be reduced when only the reproduceable and unambiguous bands are used (Pellissier *et al*., 1992).

Several studies have used multi-locus techniques to investigate phylogenetic relationships in *Solanum*, including the Amplified Fragment Length Polymorphism (AFLP) analysis of *S. melongena* L. and wild relatives (Mace *et al*., 1999), and *S. retroflexum* Dun. and related species (Jacoby *et al*., 2003). Furini and Wunder (2004) as well as Fory *et al*., (2010) have used AFLP to analyze infrageneric relationships. RAPD data were used in several studies such as Karihaloo *et al*., (1995), Spooner *et al*., (1996; 1997), Miller and Spooner (1999), Van den Berg *et al*., (2002), Stedje and Bukenya-Ziraba (2003), and Singh *et al*., (2006) to clarify phylogenetic relationships. Other studies have also used mitochondrial DNA (mtDNA) markers (Isshiki *et al*., 2003), as well

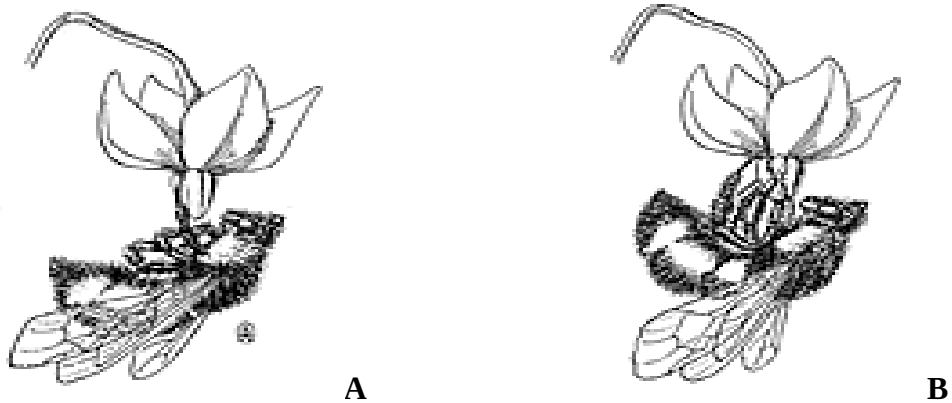
as Sequence Tagged Micro Satellite (STMS) markers (Behera *et al*., 2006), etc. In most cases multi-locus methods were applied to explore relationships at sectional levels. It is rarely used for the analysis of infrageneric groups. All these studies converge for confirming the close relationships between the A to H groups, and demonstrate the great diversity in the *S. incanum* group and the narrow variability within the *S. melongena* group. These observations further confirm a genetic bottleneck event during eggplant evolutionary history and domestication from a narrow gene pool as reported by Lester and Hassan (1991).

2.8 Botanical Description

Eggplant and its related species belong to subgenus *Leptostemonum*, the largest subgenus of genus *Solanum*. The plant is woody and develops several branches according to a roughly dichotomic ramification pattern. Anthocyanin, prickles and hairiness on vegetative parts vary quantitatively. Inflorescences are 1 to 5 andromonoecious cymes, although most modern cultivars display solitary hermaphrodite flowers. The basic flower type is pentamerous (5 sepals, 5 petals, 5 stamens) but hexa-, hepta-, and octa-merous flowers are commonly found in globose and round fruited types. Eggplant is generally considered to be an autogamous species; however, in open fields and warm conditions, flowers are visited by insects and the rate of allogamy can reach 70% or more. The fruits are berries of highly variable shape (round, intermediate, long, snake-like) and size (tens of grams to more than a kilo). The absence or presence as well as the distribution pattern of two kinds of pigments, chlorophylls and anthocyanins, control a wide diversity of fruit colours (Daunay *et al*., 2004). Eggplant is a diploid (2n=24) species, with a basic chromosome number of 12 and a genome size of approximately 956 Mbp (Bennett and Leitch, 2004).

Eggplant exhibits great morphological and reproductive diversity and many species of the genus have been used as model organisms for the examination of many biological questions. Some examples of these include investigations of the developmental evolution of leaf shape (Sinha, 1997; Bharathan *et al*., 2002), fruit morphology and chemistry (Knapp, 1986; Brown, 1987; Cipollini *et al*., 2002), a wide variety of floral syndromes including zygomorphy (bilaterally symmetrical) and heterandry (Knapp, 1989, 1991, 2002a), derived reproductive systems such as andromonoecy and dioecy (sexes borne on different plant) (Symon, 1979; Whalen and Costich, 1986; Anderson and Symon, 1989; Knapp *et al*., 1998; Lester *et al*., 1999), and the evolution of plant form, self-incompatibility, and polyploidy (e.g., Bell and Dines, 1995; Richman and Kohn, 2000; and Stone, 2002).

The poricidally dehiscent anthers of nearly all *Solanum* species make this genus an exemplar of the buzz pollination syndrome found in about 200 genera of flowering plants (**Figure 2**) (Buchmann, 1983). In these flowers, nectar is absent and pollen is the sole floral reward. However, at least one group, *Solanum* section Pachyphylla, exhibits the male euglossine syndrome in which floral osmophores secrete scents that are gathered by male "orchid" bees (Hammer *et al*., 1992; Sazima *et al*., 1993).



A **B**

Figure 2: Pollination in *Solanum*

(A) Bombus worker bee buzzing a *Solanum pastillum* S. Knapp. flower

(B) Bombus worker cleaning and packing pollen in the scopa after buzzing a *Solanum pastillum* flower (adapted from Knapp, 2002b).

Dioecy has evolved independently several times in the large, mostly tropical genus *Solanum.* In all cases of dioecy in *Solanum*, functionally male flowers have normal anthers, normal pollen and reduced stigmas; while functionally female flowers have stigmas and anthers that appear normal but contain non-functional, usually inaperturate pollen. The inaperturate pollen has living cytoplasm, but apparently never germinates and it has been hypothesised that the pollen in these functionally female flowers is retained as a pollinator reward (Knapp *et al*., 1998).

2.9 Economic Importance

Eggplant is a very popular native vegetable in Asia and the Mediterranean basin. In 2003, eggplant world production was 29 million tons (t) from 1.6 million hectares (ha) (FAO 2003). Asia is the major eggplant producer, with China (16 million t from 851,000 ha) and India (8 million t from 510,000 ha) as leaders, followed far behind, by Turkey (970,000 t from 37,000 ha), Egypt (703,000 t from 31,000 ha), Japan (420,000 t from 12,000 ha), and Italy (377,000 t from 13,000 ha). The average yield (18 t/ha) is extremely variable, depending on climate, cultural system, crop duration and grower technology. The Netherlands is the absolute champion with yields of 390 t per hectare.

The nutritional energy value of eggplant is limited (Gebhardt and Thomas, 2002), but the presence of good fibre and various vitamins (e.g. vitamin c) and minerals (e.g. potassium) in the fruit is beneficial to human health. Furthermore, the fruit contains phenolic compounds such as anthocyanins and phenolic acids which have antioxidant properties (Cao *et al*., 1996; Stommel and Whitaker, 2003) as well as alkaloids (Aubert *et al*., 1989), which have several beneficial biological and pharmaceutical properties. Frary *et al*., (2007) in their contributions stated that the beneficial effects of eggplant alkaloids are further supported by the widespread use of eggplant

in traditional medicine. Alkaloids can, however, have negative effects as excessive levels can result in extremely bitter, unpalatable fruit. Fortunately, the fruits of cultivated species normally do not contain such high levels of alkaloids unless the plants have been subjected to extreme stress (Drummond, 1986; Drummond and Brown, 1987).

In popular medicine, eggplant is indicated for the treatment of several diseases including diabetes, arthritis, asthma, bronchitis, dysuria and dysentery. In addition, evidence has it that eggplant extracts have a significant effect in reducing blood and liver cholesterol rates in humans and adult rats (Silva *et al*., 1999). In particular, the fruit helps to lower blood cholesterol levels and is suitable as part of a diet to help regulate high blood pressure. Eggplant fruit is said to be very nutritious and is a good source of vitamin C and potassium. It is usually cut into strips or cubes and fried, baked, stewed or added to soups, curries, etc. The fruit is also used as an antidote to poisonous mushrooms. It can be pounded with vinegar to create a poultice for cracked nipples, abscesses and haemorrhoids. A decoction made from the leaves can be applied to discharging sores and internal haemorrhages. A soothing poultice for the treatment of burns, abscesses, cold sores and similar conditions can also be made from the leaves. The ashes of the peduncle are used in the treatment of intestinal haemorrhages, piles and toothache (Donzella *et al*., 2000; Herrera-Arellano, 2004).

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Sample Collections.

Sample collections were made during various field trips to places known as primary centres of eggplant diversity and endemism within the Southern part of Nigeria (Figure 3). These were done during the rainy season (between June and October), a period when eggplant *Solanum* will be flowering and fruiting. The materials collected from each plant include young fresh leaves, mature leaves (for herbarium), fruits and seeds. The fresh leaves of each species collected were kept in small and sealable polythene nylon, and silica gel was added to each to dry the leaves gradually and preserve the DNA content. Each sample was labelled accordingly as follow:

(a) Name: Generic, specific, common and local names (temporary for the purpose of identification because they are still subject to verification)

(b) State of collection

(c) Location: (i) Latitude

(ii) Longitude

(iii) Elevation (in metres above sea level)

(d) Name of the town / village of collection

(e) Date of collection – including the season (rainy or dry)

(f) Time of the day of collection

Evaluations of vegetative parts (leaves and stem) were also carried out on each type in their natural environment and their photographs taken prior to sample collection. Collections were made in at least two different locations for effective sampling in areas known to be the centre of eggplant diversity within the Southern part of Nigeria.

3.1.1 Methods of evaluation

The following instruments were used in evaluation:

1. Global Positioning System (GPS) — used to determine the location including the latitude, longitude and elevation of the place of each collection.
2. Portable weighing balance — used to measure fresh fruit weight.
3. Tape rule —used to measure the height, length, breadth and width.
4. Portable digital clock --- to record accurately the date and time of the day.
5. Calculator —used to get the average value for each quantitative character measured.

3.2 Morphological Study

Morphological crop descriptors allow a quick and easy discrimination between phenotypes. They are generally highly heritable traits that can be easily recorded through visual observations and are equally expressed in all environments. Naujeer (2009) stated further that morphological descriptors for eggplants have been developed by IPGRI/ FAO (now known as Bioversity International), EGGNET and UPOV which provide internationally accepted definitions for these descriptors and include a complete description of important quantitative and qualitative traits illustrated by figures and measured either in metric or arbitrary scale.

A comprehensive morphological study was carried out on at least ten different plants of each species in their natural environment. This was done so that the average value for each character measured could be obtained. The study involved the measurement and recording of both quantitative and qualitative characters on these plants at flowering or fruiting stage when further

growth was temporarily suspended. *Solanum* descriptors according to IPGRI (International Plant Genetic Resources Institute, Rome, Italy) standard were used for this study (**Appendix 2**).

3.3 Identification of the Plant Samples Collected

This involved the determination of the plant as being identical with or similar to another already known plant. First, the collected plant specimen was compared with photographs, drawings and illustrations from existing sample collections. Then proceeded to the use of details from the database of the sample (using flora of the particular region from which the plant was collected) in order to determine the family and genera to which the plant belongs. This identification was achieved through the use of keys in the flora. Finally, voucher specimen i.e. herbarium specimens were prepared following the method of Ogundipe and Daramola (1998) and Ogundipe *et al*., 2009 and sent to Forestry Herbarium Ibadan (FHI) where they were authenticated by taxonomists, and then deposited at both the University of Lagos Herbarium (LUH) and Forestry Herbarium Ibadan (FHI) for reference purposes.

3.4 Molecular Studies

These followed the method of Doyle and Doyle (1990), Harrington *et al*., (2005), Ronquist *et al*., (2009), with some modifications and involved the following processes:

3.4.1 Total Genomic DNA Extraction

Total genomic DNA extraction was carried out on young fresh leaves of each sample dried with silica gel and kept in sample bag, using the modified Cetyltrimethylammonium bromide (CTAB) extraction protocol (Doyle and Doyle, 1990) followed by additional purification. The protocol involved the following:

10ml of Isolation Buffer (10X CTAB made up of 100mM Tris HCl pH 8.0, 1.4M NaCl, 20mM EDTA and 10% CTAB) containing 80µl of betamercaptoethanol in 50ml Blue Cap tubes was preheated in a 65°C water bath. Mortar and pestles were equally preheated at this temperature. Then 0.3—0.5g of silica-dried leaf tissue was ground in a mortar and pestle (preheated to 65°C) using a portion (500µl) of the isolation buffer. For tough tissues a pinch of silica gel was added to aid grinding. Then, another 500µl of the buffer was added and swirled to suspend the slurry. The slurry was then poured into 1.5ml Eppendorf tube and incubated at 65°C for 20 minutes with optional occasional gentle swirling. Thereafter, an equal volume (1ml) of SEVAG mixing was added, mixed gently but thoroughly. The cap of the tube was opened to release gas, and then re-tightened. The tube was rocked using an orbital shaker at 350 rpm for 60 minutes. The tube was centrifuged at 4000rpm at 25°C for 20 minutes. Ideally the aqueous (top) phase, containing the DNA, was clear and colourless. The aqueous (top) phase containing DNA was then removed with a plastic transfer pipette and transferred to another clean and labelled 1.5ml Eppendorf tube. SEVAG and plant debris was disposed of in the designated SEVAG waste container. The volume of the aqueous phase (typically c. 0.5ml) was estimated and then 2/3 volume of -20°C iso-propanol was added and mixed gently. It was put in -20°C freezer for a few hours for fresh material and at least one week for silica dried material samples to precipitate DNA. The tube was again centrifuged at 3000rpm for 5 minutes to collect precipitate. The liquid was poured off after spin and 0.5ml of 70% ethanol was added. The pellet was dislodged to facilitate "washing" and washed for 30 minutes. This was repeated twice. Thereafter, DNA was spurned down at 3000rpm for 5 minutes; the liquid was poured off gently and carefully drained upside down to allow alcohol to evaporate. At times the tube was left on its side to dry in order not to lose the DNA pellet. Finally, DNA was resuspended and eluted in 100µl low TE salt and then transferred into another clean and labelled 1.5ml Eppendorf tube and stored at -20°C (or -80°C for long periods).

3.4.2 Purification of extracted DNA

Purification was done in a Silica-column inserted into vacuum manifold connected to a vacuum pump using QIAquick purification kit. The following procedures were followed:

Labelled silica-columns were inserted into a vacuum manifold. Then 750µl of labelled Buffer PB was added to 150µl of total DNA, then samples were loaded into the silica-columns and vacuum was applied until the entire sample has passed through the column. The sample was washed by adding 750µl of PE Buffer (to which ethanol has been previously added) to each column and vacuum was again applied. After about 10 seconds, vacuum was switched off and columns transferred to the provided 200µl collection tubes. It was then centrifuged at 13000rpm for 1 minute to remove residual ethanol (Buffer PE). The lids of sterilised (and labelled) 1.5ml Eppendorf tubes were cut and the column placed into the tubes. 50µl of labelled Buffer EB (elution buffer) was added directly onto the membrane of the columns and left for 1 minute at room temperature to dissolve the DNA in the elution buffer. This was centrifuged for 1 minute at 13000rpm; caps were replaced onto tubes and DNA quantity was verified on an agarose gel.

3.4.3 Verification of DNA Quality by Electrophoresis

This was done on a 1% agarose gel to check the quality and purity of the extracted DNA samples. Sides of gel tray were taped to hold the gel while setting and well-forming combs were placed into the tray. A 1% agarose gel was prepared by mixing 1.5g agarose with 150ml of 1X TBE Buffer. It was melted in microwave until all agarose was dissolved (1-2 minutes). It was allowed to cool down under the air-conditioner to about 36^{o}C. Once cooled, 3µl of Ethidium Bromide was added (in the fumehood) and swirled to mix properly. The gel was poured into the tray and left to stand for at least 30 minutes before loading samples. Combs were carefully and gently removed, and the gel placed into an electrophoresis tank. 3µl of loading dye (made of

0.25% bromophenol blue, 0.25% xylene cyanol FF and 30% glycerol in water) for each DNA sample was spotted on a strip of parafilm. Using a 1- 20 µl pipette range, 5µl of DNA was mixed with the loading buffer and the mixture was loaded into the well on the gel. This was run for about 30 minutes at 110Milli Amps. Thereafter, the gel was viewed (with the use of eye protection) under the ultraviolet transilluminator attached to a computer system and photographs of the gel were taken with the aid of gel documentation units (Uvitec) and saved.

3.4.4 Spectrophotometric Analysis

DNA quantification was performed by ultraviolet (UV) spectrophotometric measurement of the absorption at 260 nm on a sample of the DNA in solution. The fraction of light passing through the solution was measured. To quantify DNA in all the samples, readings were taken at wavelengths of 260 nm and 280 nm. The reading at 260 nm allows calculation of the concentration of nucleic acid in the sample. The reading at 280 nm gives the amount of protein in the sample. Pure preparations of DNA have OD260/OD280 values of 1.8 to 2.0. If there is contamination with protein or phenol, this ratio will be significantly less than these values, and accurate quantification of the amount of nucleic acid will not be possible.

Spectrophotometric analysis of DNA solutions requires an instrument with a deuterium source or other means of determining absorbances in the UV range. This was achieved with the Eppendorf BioPhotometer plus (UV/Vis Photometer) to determine the quantity (concentration) and quality (purity) of the isolated DNA from samples. The spectrophotometer was first standardized (blanked) with 100 µl of distilled water before proceeding with the DNA samples. For each sample, 5 µl of DNA was added to 95 µl of distilled water in a cuvette then placed in the cuvette compartment of the spectrophotometer. Both measurement and calculation of results were

performed at the press of the sample button on the equipment. The large LCD screen displayed the sample concentrations, absorption values, OD260/OD280 and OD260/OD230 and sample dilutions at a glance. These were recorded for each sample (**Appendix 8**).

3.4.5 Polymerase Chain Reactions (PCR) and Amplification

Polymerase Chain Reactions (PCR) is referred to as the enzymatic synthesis of multiple copies of a specific DNA sequence in a cyclical manner. It involves initiation, denaturation, annealing, elongation and final extension of the DNA fragment over varying temperatures of 94^{o} C, 94^{o} C, 65^{o} C, 72^{o} C and 72^{o} C respectively. The reaction mixture is then held at 4^{o} C. The study here involved the amplification of the internal transcribed spacer (ITS) region of nuclear ribosomal DNA, and the non-coding chloroplast DNA (cpDNA) regions trnlE-trnlF spacer plus trnl intron from the extracted total genomic DNA. PCR experiment sheet was filled, and the appropriate number of 0.2ml PCR tubes was labelled. A positive (a known sample which has worked previously for the region being amplified) and negative (PCR mix but no DNA added, to check for contamination) controls were prepared. A master mix including every cocktail except the template DNA (sample) was made up in a 1.5ml eppendorf tube, allowing for one extra sample (i.e. for six samples enough master mix for seven was made).

3.4.5.1 Amplification of the internal transcribed spacer (ITS) region.

Amplification of the internal transcribed spacer (ITS) region of nuclear ribosomal DNA, composed of ITS1, the 5.8S gene, and ITS2, was done using two primers ITSleu1 (5'-GTC CAC TGA ACC TTA TCA TTT AG-3') and ITS4 (5'- TCC TCC GCT TAT TGA TAT GC-3') as described by White *et al*.(1990) and Baldwin *et al*. (1995). The fragment size amplified for ITS was between 1236 - 1280bp. Reaction mixture of 25µl included 22.5µl of PCR Master Mix (Red-top premix), 0.5µl Bovine Serum Albumin (BSA), 0.5µl of primer 1 (ITSleu1), 0.5µl of

primer 2 (ITS4), and 1µl of DNA sample. The reactions were performed in an Applied Biosystems thermal cycler (Gene Amp PCR System 2400). The DNA amplifications were carried out using the ‘‘Hot Start’’ PCR method: 94^0C for 2 min, 72^0C for 3 min (initial strand separation); 30 cycles at 94^0C for 1min (Denaturation), 50^0C for 1 min (annealing), 72^0C for 1 min and 30s (primer extension); with a final extension at 72^0C for 7 min. PCR products were cleaned using the QIAquick PCR purification kit (Qiagen, Valencia, California, USA).

3.4.5.2 Amplification of trnL-trnF spacer plus trnL intron

The non-coding cpDNA regions trnL-trnF spacer plus trnL intron are adjacent and were amplified separately. TrnL includes region between trnlC- trnlD, and trnF includes region between trnlE–trnlF. Primers used for amplification were UniC (5'-CGAAATCGGTAGACGCTACG) and UniF (5'-ATTTGAACTGGTGACACGAG) of Taberlet *et al*., (1991). The fragment size amplified was between 750 – 800bp. Reactions of 25µl were obtained with 22.5 µl of PCR Master Mix (Green-top premix), 0.5µl Bovine Serum Albumin (BSA), 0.5µl of primer 1 (UniC), 0.5µl of primer 2 (UniF), and 1µl of mM DNA sample, to complete a volume of 25 µl. The thermal cycler program included initial denaturing at 94^0C for 3 min; 28 cycles at 94^0C for 1 min (Denaturation), 48^0C for 1 min (annealing), 72^0C for 1 min (primer extension) ending with a final extension at 72^0C for 7 min and final hold at 4^0C.

3.4.5.3 Purification of amplified DNA and Gel Electrophoresis

Purification of amplified DNA was done in a Silica-column inserted into vacuum manifold connected to a vacuum pump using QIAquick purification kit the same way it was done for extracted DNA. The integrity and purity of the amplified DNA samples were also checked on 2% agarose gel (i.e. 3.0g of agarose dissolved in 150ml of 1X Tris-Borate-EDTA (TBE) buffer). The procedure is the same as described for purified DNA extract. The concentration of the PCR

product (i.e. the amplicon) was also determined by adding 2µl of the amplicon to 55 µl of sterile water in a cuvette and verified on the spectrophotometer.

3.4.6 Cycle Sequencing

Cycle sequencing is a modification of the traditional Sanger sequencing method. The principles are the same as in Sanger sequencing; Dideoxynucleotides are used in a polymerization reaction to create a nested set of DNA fragments with dideoxynucleotides at the 3' terminus of each fragment. Cycle sequencing is a method used to increase the sensitivity of the DNA sequencing process and permits the use of very small amounts of DNA starting material. This is accomplished by using a temperature cycling process similar to that employed in the polymerase chain reaction (Sanger *et al*., 1977). Amplification of selected regions was achieved in a 10 µl reaction mixtures containing 0.5 µl pink juice (Big Dye Terminator, Applied Biosystems Inc.), 3.0 µl 5X sequencing buffer (Bioloine), 0.75 µl primer (1:10 dilution; forward or reverse for each primer pair) and varying quantities of purified PCR products depending on the strength of the reaction, made up to 10 µl with sterile water. The amplification of ITS region was improved by the addition of 4% Dimethyl Sulfoxide (DMSO) in the total volume of the sequencing mix. Cycle sequencing was done in a Gene Amp® PCR System 9700 Thermocycler (Applied Biosystems Inc.) using the following programme: Initial denaturation at 95^{o}C for 30s followed by one cycle of denaturation at 95^{o}C for 60s, annealing at 55^{o}C for 30s and extension at 72^{o}C for 60s; ran for 30 cycles and thereafter held the reaction mixture for 7mins at 72^{o}C to allow complete extension of the PCR products with a final hold at 4^{o}C.

3.4.6.1 DNA Sequencing, Editing and Alignment

DNA sequencing was done to determine all or part of the nucleotide sequence of a specific deoxyribonucleic acid (DNA) molecule. Knowledge of DNA sequences has become indispensable for basic biological research, other research branches utilizing DNA sequencing, and in numerous applied fields such as diagnostic, biotechnology, forensic biology and biological systematics. The advent of DNA sequencing has significantly accelerated biological research and discovery.

3.4.6.2 Sequencing of ITS region

Sequencing of the purified cycle sequencing products (following a modification of ABI protocol, 2009) was done in both directions on an automated sequencer (ABI PRISM® 3730 DNA Analyzer) at the Royal Botanical Garden, Kew DNA Sequencing Core Facility following the manufacturer's instructions. Primers used included ITS4 and ITS5HP (5'-GGA AGG AGA AGT CGT AAC AAG G-3').

3.4.6.3 Sequencing of trnL-trnF spacer plus trnl intron

These two regions were cleaned after cycle sequencing using the QIAquick PCR purification kit and sequenced on an ABI PRISM® 3730 automated sequencer as before using the same primers (UniC and UniF) as for PCR.

3.4.6.4 Sequence Editing and alignment

Sequences were analyzed and edited, and a consensus sequence for each species (contigs) was constructed using the program Sequencher version 4.5 (Gene Codes Corporation, Ann Arbor, Michigan, USA). The program was used to assemble complementary strands and verify software base-calling on a Macintosh computer. Species sequences were aligned manually by eye in Se-Al

(Rambaut, 1996) and MacClade 4.0 (Maddison and Maddison, 2000). Indel alignments took into account the mechanisms and patterns of evolution in non-coding sequences outlined in Kelchner, (2000). At the end, data generated for ITS and trnlE-trnlF plus intron trnl matrices included sequences from 20 selected samples, which comprised selections from eggplants *Solanum* and representatives of its related species and later used in the analyses.

3.4.6.5 Basic Local Alignment Search Tool (BLAST)

The problem of synonymy and taxa mis-identification in the eggplant *Solanum* accessions studied was resolved by subjecting the nucleotide sequence obtained from the two plastid (non-coding trnL intron and intergenic spacer trnL-trnF) regions to BLAST. This was done in order to get the maximum identity/similarity in percentage (%) between the sequenced regions of samples studied and the sequenced data of the same regions already in the NCBI Database.

3.5 Data Analysis

3.5.1 Morphological Data

All calculations and analyses were made using the appropriate options of the Statistical Package for Social Sciences (SPSS) statistical software version 15.0 (Apache Software Foundation, Chicago, IL) and Numerical Taxonomic System Software (NTSYS) pc version 2.02j by Applied Biostatistics Inc. (Rohlf, 1996). The Statistical Package for Social Sciences (SPSS) software for Windows Evaluation Version 15.0 was used to evaluate the values obtained for a particular character on ten different plants of each species, especially quantitative characters. This yielded mean, standard error of the mean (SEM), standard variation and variance for each character. The average (mean) values obtained for quantitative and qualitative morphological characters were analyzed separately to compute pairwise distance (similarity) matrices using Sequential

Hierarchical and Nested (SAHN) clustering option of the NTSYS-pc 2.02j software package (Rohlf, 1996); then they were combined and analyzed together. The software generated dendrograms, which grouped the test lines using Unweighted Pair Group Method with Mathematic Average (UPGMA) on the basis of genetic similarity and Jaccard's coefficient. Principal Component Analysis (PCA) option of SPSS was also used on the morphological data to determine the relationship between plant traits and accessions. To achieve this, accessions were compared through ordination analysis and projected on two-dimensional matrix. The bivariate matrix plot was also projected on a three dimensional scale to display the similarity or dissimilarity distance between different accessions in space.

3.5.2 Molecular Data

3.5.2.1 Phylogenetic Estimation using Maximum Likelihood (PhyML)

The data sets obtained from the sequenced nuclear (ITS) and chloroplast (trnL-trnF spacer plus trnL intron) gene regions of the selected samples were analyzed by Cladistic method using PhyML (Phylogenetic estimation using Maximum Likelihood). The core of the heuristic is based on a well-known tree-swapping operation, namely 'nearest neighbour interchange', which defines three possible topological configurations around each internal branch (Swofford *et al*., 1996). PhyML was used to analyze each data set separately prior to combining. For each of these configurations, the length of the internal branch that maximizes the likelihood was estimated using numerical optimization. The difference of likelihood obtained under the best alternative topological configuration and the current one defines a score. A score with positive value indicates that the best alternative topological configuration yields an improvement of likelihood. A score with negative value indicates that the current topological configuration cannot be improved at this stage and only the length of the internal branch is adjusted. Each internal branch was examined in this manner and ranked according to its score. The optimal length of external

branches was also computed. These calculations were performed independently for every branch and they defined a set of (topological or numerical) modifications, each of which corresponds to an improvement of the current tree regarding the likelihood function.

The standard approach would only apply one of these modifications, typically that corresponds to the internal branch with best score. Here, a large proportion of all modifications computed previously were performed instead. This proportion was adjusted so as to increase the likelihood at each step, ensuring convergence of the algorithm. This way, the current tree was improved at each step, both in terms of topology and branch length, and only a few steps (usually a few dozen or less) were necessary to reach an optimum of the likelihood function.

CHAPTER FOUR

4.0 RESULTS

4.1 Morphological Characterization.

4.1.1 Samples Collected.

A total of 49 samples of different species of vegetable *Solanum* (eggplant) were collected and used for this study. The coordinates (GPS) reading of exact collection point of each sample in the zones visited and the codes used (**Appendix 3**) were used to generate the map as shown in Figure 3. It should be noted that some points of collection were not shown in this map because their coordinates were so close to each other. Therefore collection sites close to one another were represented with one coordinate for clarity.

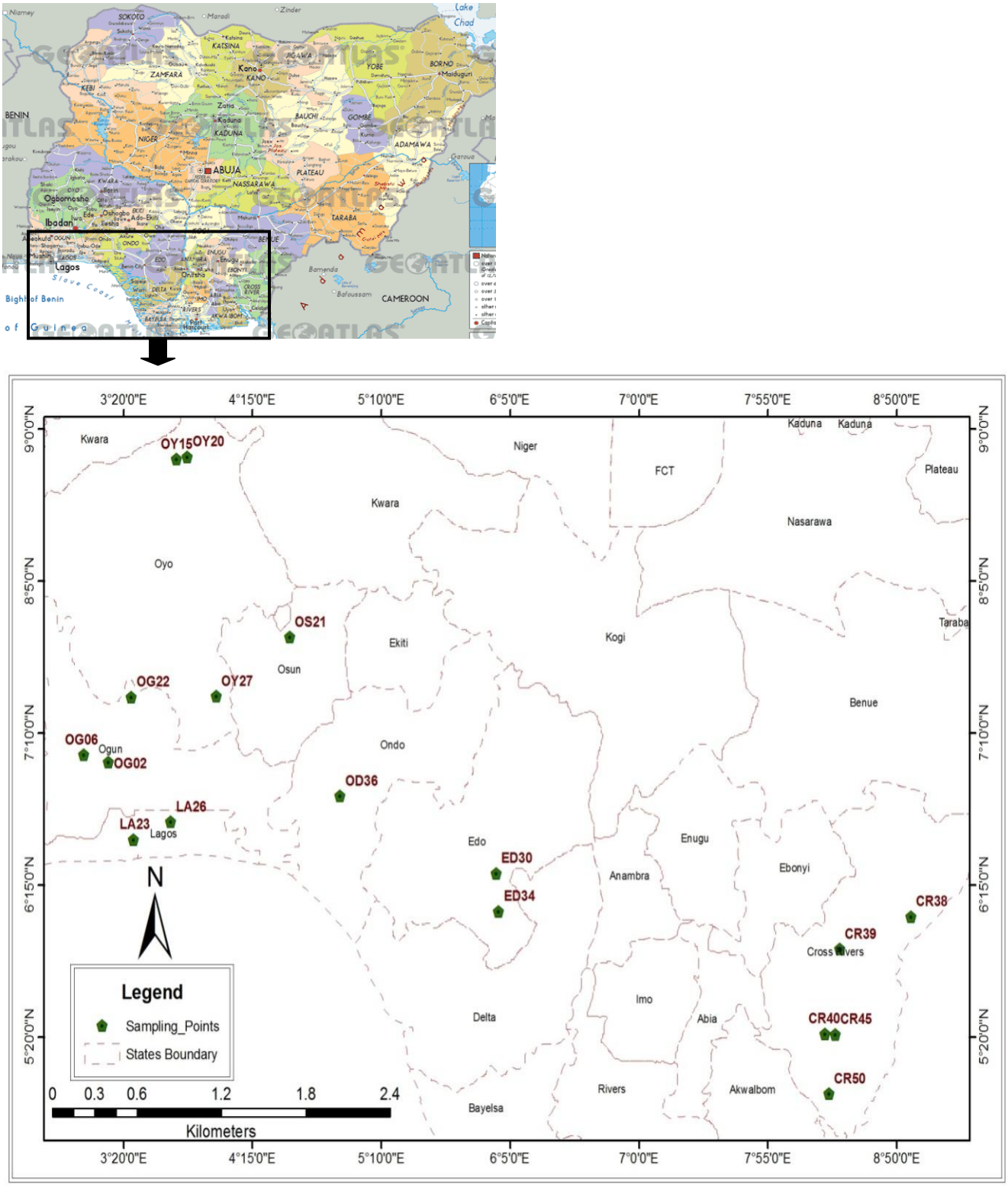


Figure 3: Map of Southern Nigeria showing some Points of Sample Collection.

4.1.2 Authenticated Voucher Specimens

The authenticated voucher specimens are as shown in Table 3. The authentication showed that the 49 *Solanum* samples collected represent 12 different species and were made up of 3 samples of *Solanum dasyphyllum*, 2 of *S. nigrum*, 6 of *S. macrocarpon*, 5 of *S. torvum*, 1 of *S. erianthum*, 13 of *S. melongena*, 10 of *S. gilo*, 1 of *S. incanum*, 2 of *S. scabrum*, 2 of *S. aethiopicum*, 3 of *S. indicum* subsp. *distichum* var. *distichum* and 1 sample of *S. macranthum*.

Table 3: List of samples of *Solanum* species collected and their Identification

Sample I.D No.	Identification	Sample I.D No.	Identification	Sample I.D No.	Identification
OG02	*Solanum dasyphyllum*	OY14	*S. gilo* Raddi (White fruit)	LA26	*S. macrocarpon* (Green fruit)
OG03	*S. nigrum*	OY15	*S. incanum* (Green small fruit)	OY27	*S. melongena* (Purple fruit)
OG04	*S. dasyphyllum*	OY16	*S. scabrum*	OY28	*S. macrocarpon* (Brown fruit)
OG05	*S. nigrum*	OY17	*S. aethiopicum*	OY29	*S. torvum*
OG06	*S. macrocarpon* (White fruit)	OY18	*S. scabrum*	ED30	*S. melongena* (Purple fruit)
OG07	*S. macrocarpon* (Green fruit)	OY19	*S. melongena* (White fruit)	ED31	*S. melongena* (Green fruit)
OG08	*S. torvum*	OY20	*S. aethiopicum*	ED32	*S. macrocarpon* (Green fruit)
OG09	*S. erianthum*	OS21	*S. torvum*	ED33	*S. gilo* Raddi (Green fruit)
OG10	*S. melongena* (Green fruit)	OG22	*S. melongena* (Green fruit)	ED34	*S. dasyphyllum*
OY11	*S. gilo* Raddi (White fruit)	LA23	*S. gilo* Raddi (Green egg-shaped fruit)	OD35	*S. macrocarpon* (Green fruit)
OY12	*S. gilo* Raddi (White fruit)	LA24	*S. gilo* Raddi (Green round fruit)	OD36	*S. melongena* (Green fruit)
OY13	*S. gilo* Raddi (White fruit)	LA25	*S. gilo* Raddi (Green round fruit with greenish purple stem)	CR37	*S. torvum*

Table 3: List of samples of *Solanum* species collected and their Identification (Contd.)

Sample I.D No.	Identification	Sample I.D No.	Identification
CR38	*S. indicum* subsp.*distichum* var. *distichum*	CR45	*S. gilo* (Green,white striped fruits, & no thorns)
CR39	*S. torvum*	CR46	*S. indicum* subsp. *distichum* var. *distichum*
CR40	*S. melongena* (long&white fruits;thorny)	CR47	*S. melongena* (Purplish-white fruits with depression at the top & no thorns)
CR41	*S. melongena* (long & purple fruits,no thorns,white flowers)	CR48	*S. melongena* (Long purple fruits with white patches at the head & no thorns)
CR42	*S. melongena* (long&purple fruits,thorny,white flowers)	CR49	*S. gilo* (Pure white fruits & no thorns)
CR43	*S.indicum* subsp.*distichum* var. *distichum*	CR50	*S. macranthum*
CR44	*S.melongena* (Round,purple white striped fruits, no thorns)		

4.2 Description of Morphological Characters of Collected Samples

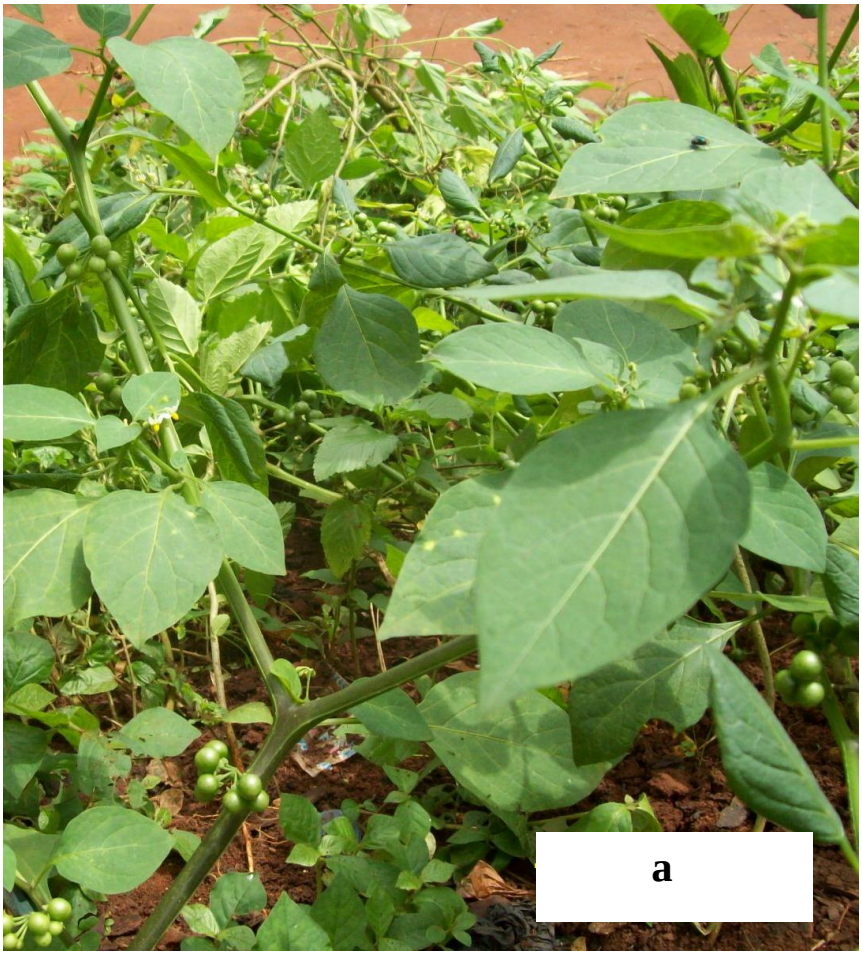
Solanum species can assume a bewildering array of growth forms. Sample exploration showed that members of eggplant family generally occur as herbs, shrubs, trees (**Plate 1**) or vines, with or without spines, glabrous or pubescent with unbranched or branched, often glandular hairs. Leaves are alternate or paired and frequently unequal in size, simple to pinnately lobed, petiolate or sessile, without stipules. Inflorescences are cymose, could be branched or unbranched. Flowers are usually perfect, (4-) 5-merous, actinomorphic or zygomorphic; calyx campanulate, sometimes accrescent in fruit; corolla rotate, campanulate, stellate, or urceolate, white, green, yellow, pink, or purple (**Plates 4, 7** and **12d**); Fruits are berry, usually fleshy but occasionally dry, usually green, white or purple in colour turning orange or red as they become ripe (**Plates 3c, 5, 9, 10, 11b** & **c, 14c, 17c**). The seeds are many and often flattened, embedded in abundant endosperm.

Plate 1: **Photographs of some vegetable *Solanum* species (X ¼).**

Legend: (a) *S. nigrum*, a herb; (b) *S. torvum*, a shrub; (c) *S. gilo*, an annual showing white fruits; (d) *S. macranthum*, a tree; (e) *S. melongena*, a shrub; (f) *S. macrocarpon*, a shrub showing white fruits and purple flowers.

4.2.1 *Solanum nigrum*

Plant body is generally glabrous and is commonly called “Black nightshade”. It is a fairly common herb or short-lived perennial shrub, found in many wooded areas, as well as disturbed habitats. It has a height of 30–120 cm, leaves 4-7.5 cm long and 2–5 cm wide; ovate to heart-shaped, with wavy or entire edges; both surfaces hairless; petiole 1–3 cm long with a winged upper portion. Petals greenish to whitish, recurved when aged and surround prominent bright yellow anthers. The berry is mostly 6–8 mm diameter, greenish in colour and turns dull black or purple-black when ripe (**Plate 2**).



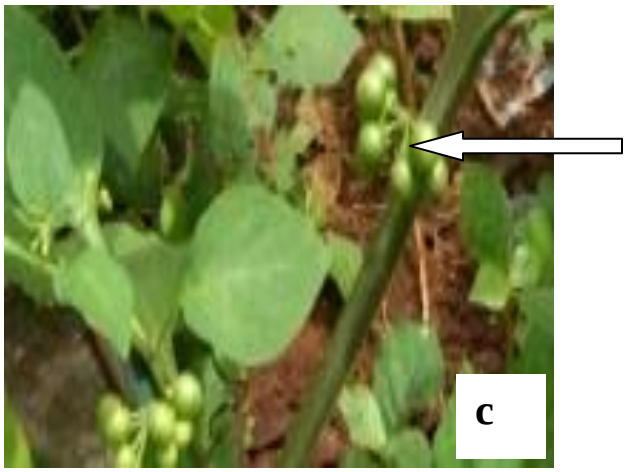


Plate 2: *Solanum nigrum* (X ¼)

Legend: (a) whole plant (b) flowers and (c) arrow showing a cluster of fruits

4.2.2 *Solanum scabrum*

S. scabrum belongs to the *Solanum nigrum* complex. Most species in this group are difficult to identify but *S. scabrum*, commonly called “Garden Huckleberry” can be recognized with relative ease by its large sized leaves and strong stem with distinct dented wings. The flowers are either white or light purple in colour and have brown anthers. Its berries are broadly ovoid, dark purple and 12-17mm broad (**Plate 3**). In contrast to most other members of *S. nigrum* complex, these berries remain on the plant at maturity.

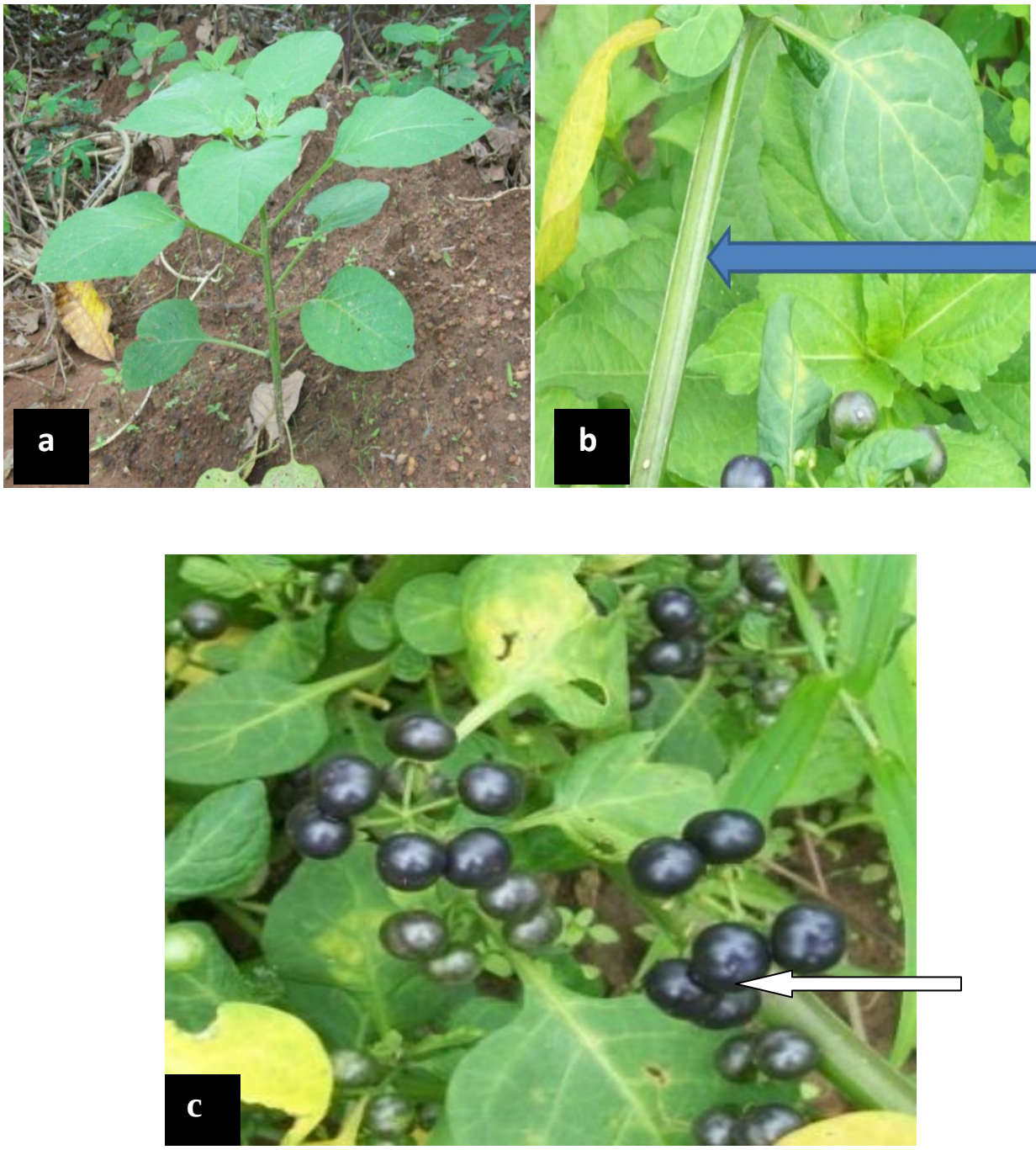


Plate 3: *S. scabrum* (X ¼)

Legend: (a) whole plant (b) arrow showing stem with distinct dented wings (c) arrow showing a cluster of dark purple fruits

4.2.3 *S. macrocarpon*

A glabrous, erect or climbing, unarmed, herbaceous plant reaching up to 1.5 m tall with green stem, woody at the base. Leaves are sessile, oblong-lanceolate, 10-30 cm long x 4-15 cm wide. White-pink or light purple flowers present. Flower shape could be stellate or semi-stellate (**Plate 4**). Fruits have greenish or pale creamy - white skin, depressed globose or round in shape, 5-6 cm long x 7-8 cm wide, with bright green calyx, turning orange-yellow when ripe (**Plate 5**). Common names include African eggplant, Native eggplant, Local garden egg, and Gboma eggplant.

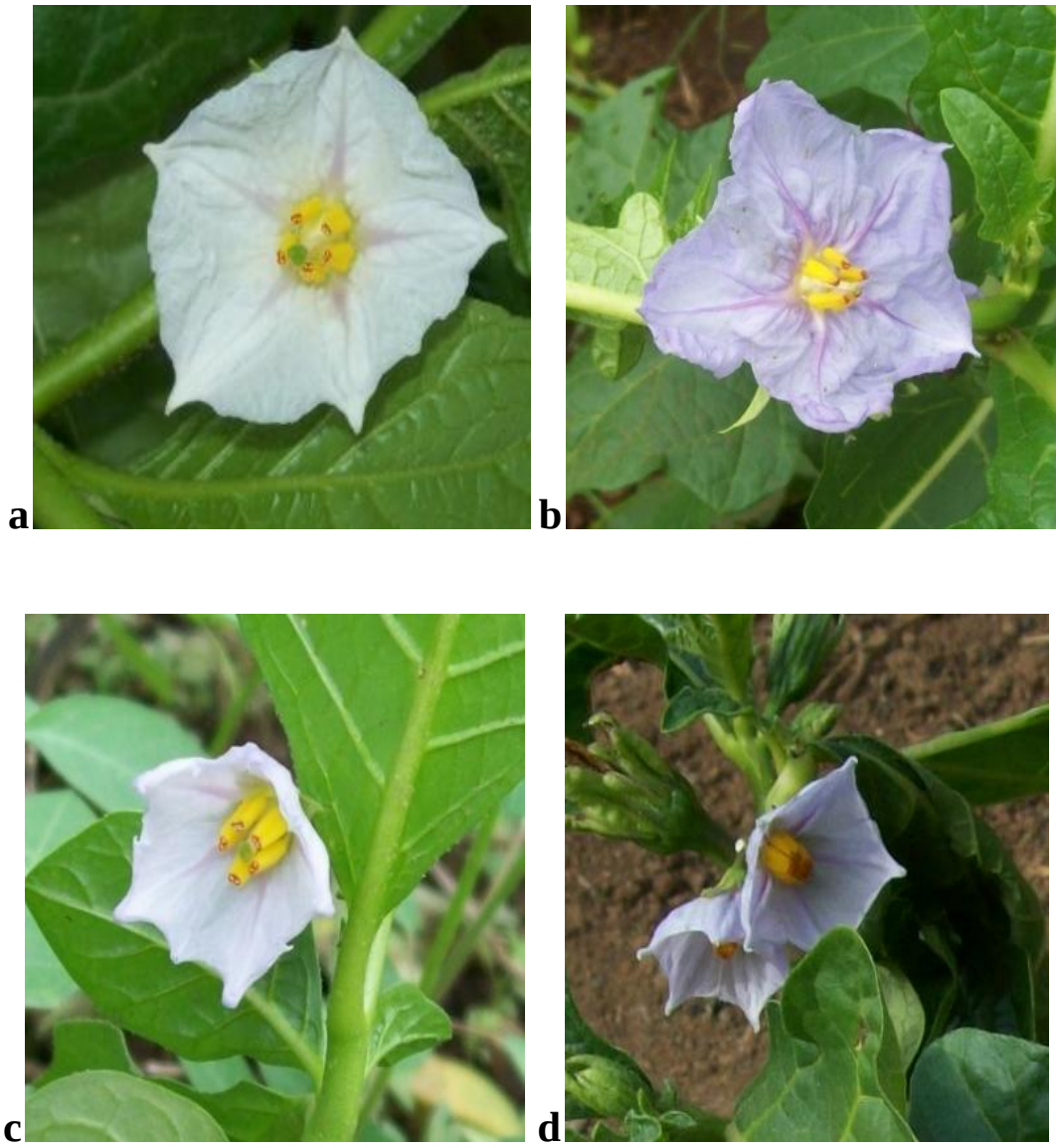


Plate 4: Flower morphology in *S. macrocarpon* (X ¼)

Legend: (a) semi-stellate white; (b) semi-stellate purplish-white; (c) rotate white; (d) rotate purplish-white.

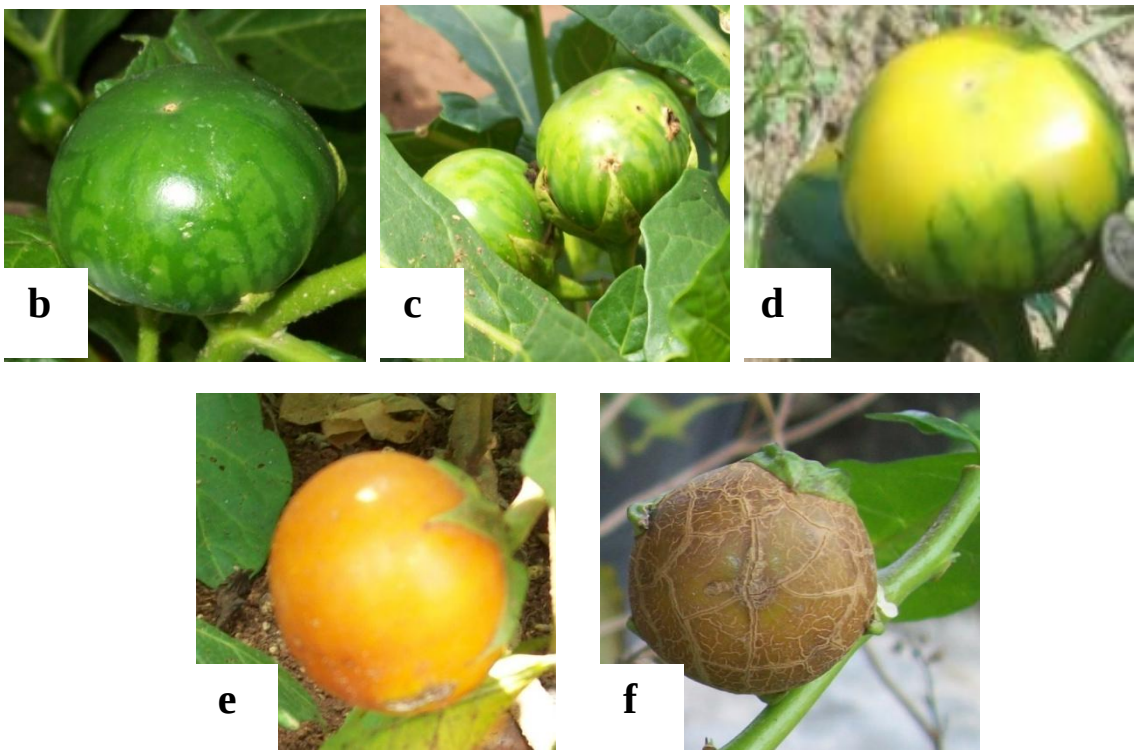


Plate 5: ***S. macrocarpon*** **(X ¼)**

Legend: (a) whole plant with white fruits (b) a green fruit (c) mature and greenish fruit (d) a ripening fruit (e) a ripened fruit turning orange-yellow (f) a cracked and brownish fruit.

4.2.4 *S. melongena*

A lot of morphological diversity and variability was observed during the collection of these samples. It is a bushy plant and grows to a height of 60 to 120 centimetres. *S. melongena* is a perennial but grown commercially as an annual crop. The plant is erect, compact, and well branched. The leaves are large, simple, lobed and alternate (**Plate 6**). It is commonly called Aubergine in (France and England) or Brinjal eggplant (in the United States).

(X 1/5)

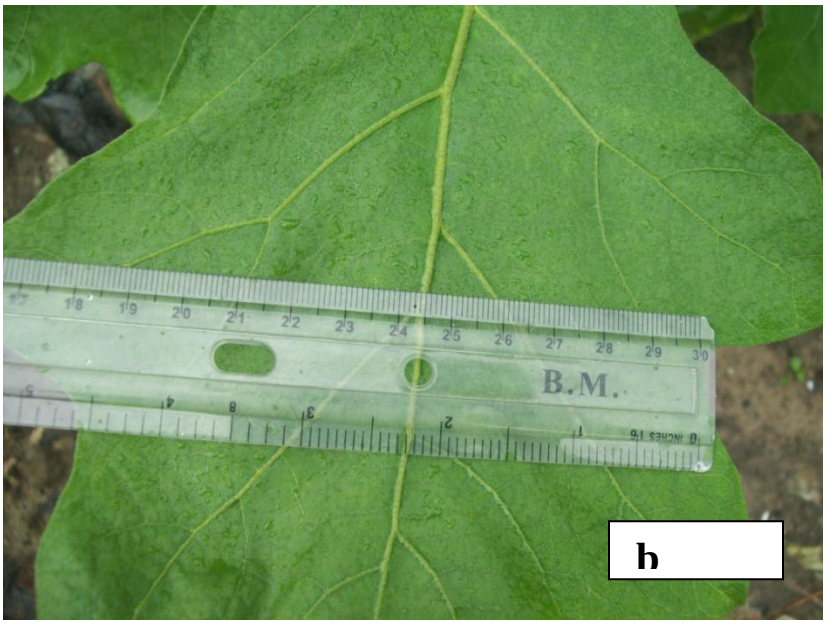


Plate 6: (a) Clusters of *S. melongena* plant (b) a leaf being evaluated

The morphological diversity noticed in *S. melongena* ranged from differences in flower colour, shape and size to fruit colour, shape, and size. The presence or absence of prickles/thorns is another distinguishing feature. The flowers are large, violet- or white-coloured, and solitary, or in clusters of two or more (**Plate 7**). The stems, leaves, and calyx of some cultivars are spined (**Plate 8**). The fruit is a pendant, fleshy berry.

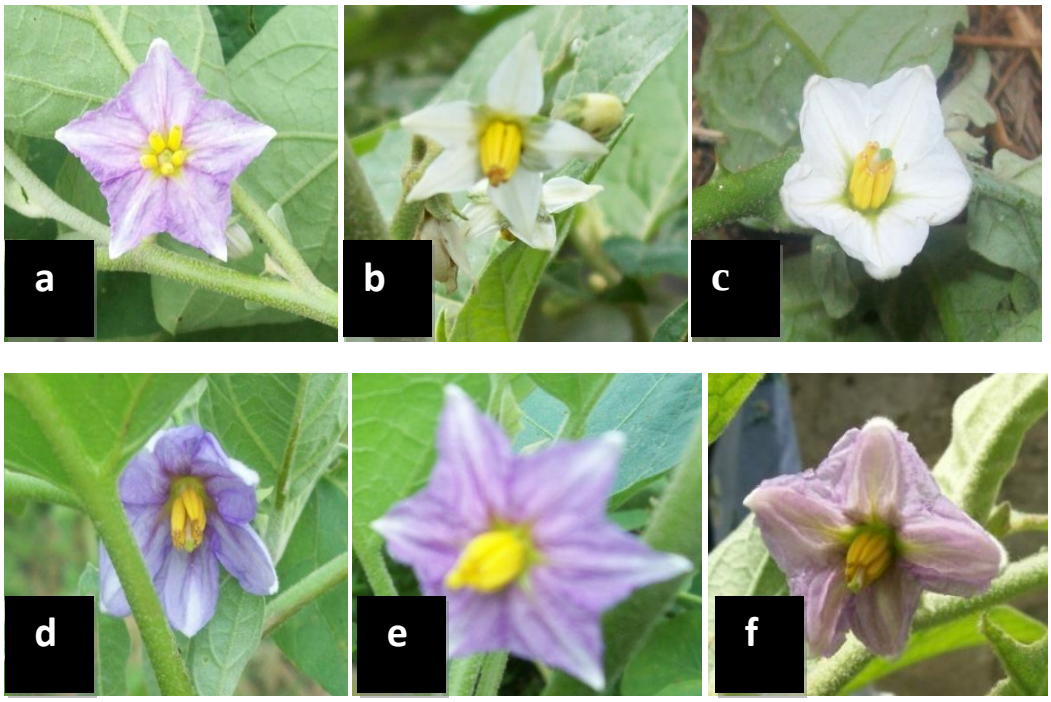


Plate 7: *S. melongena* showing different flower colours and shapes (X ¼)

Legend: (a) light purple and semi-stellate shape (b) white and stellate (c) white and semi-stellate (d) light purple and rotate (e) purple and stellate (f) purple and semi-stellate.

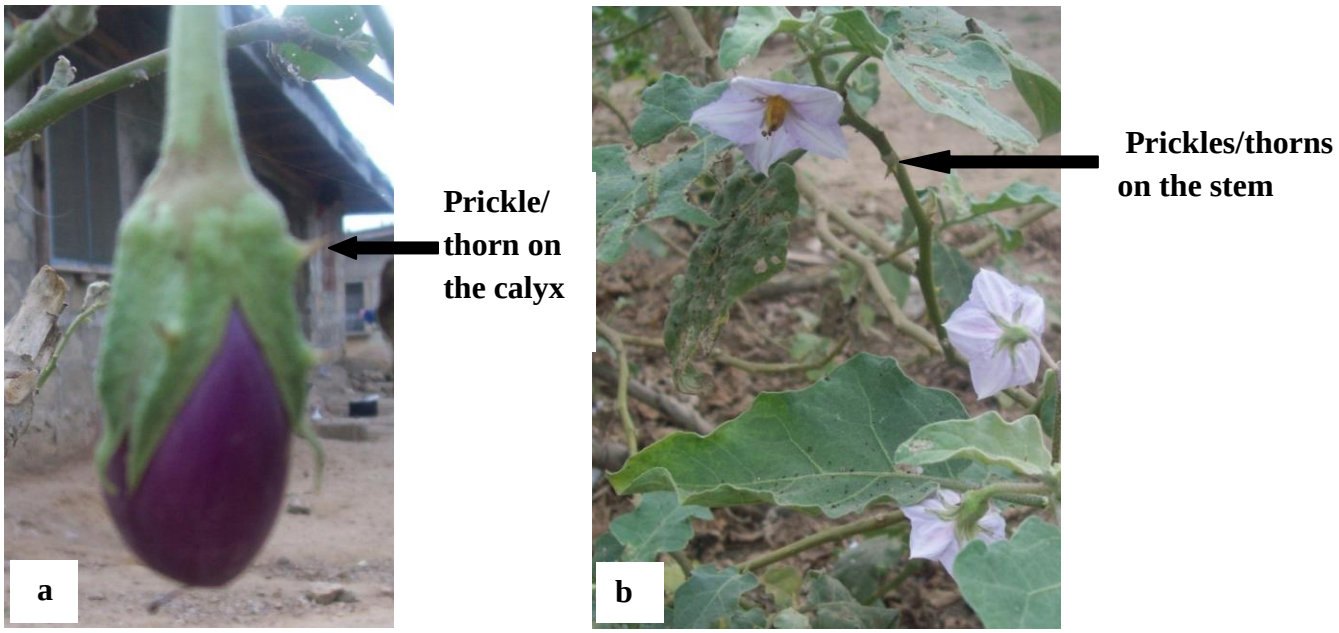


Plate 8: *S. melongena* showing prickles or spines(X ¼)

Legend: (a) fruit and (b) stem with prickles or spines

The colour of fruit varies from (shiny) purple, white, green, yellowish, or striped (**Plate 9**); the shape of fruit varies from ovoid, oblong, obovoid, or long cylindrical (**Plate 10**). Common names include *brinjal* in India, and *aubergine* in Europe. The name eggplant derives from the shape of the fruit of some varieties, which are white and shaped very similarly to chicken eggs.

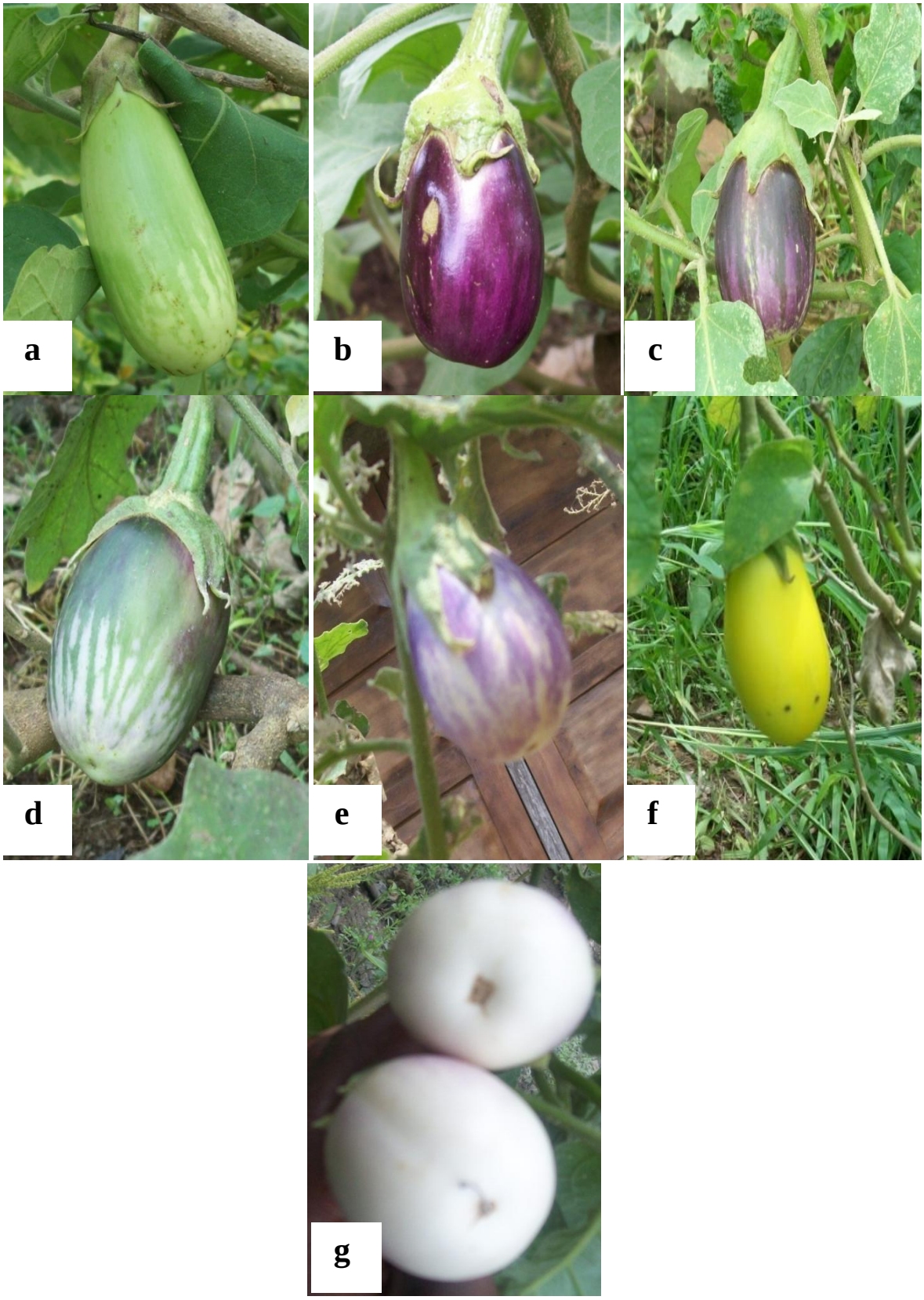


Plate 9: Different colours of *S. melongena* fruits (X 1/5)

Legend: (a) light green (b) shining purple (c) dark purple (d) greenish purple with white stripes (e) whitish purple (f) whitish turning yellow when ripe (g) pure white.

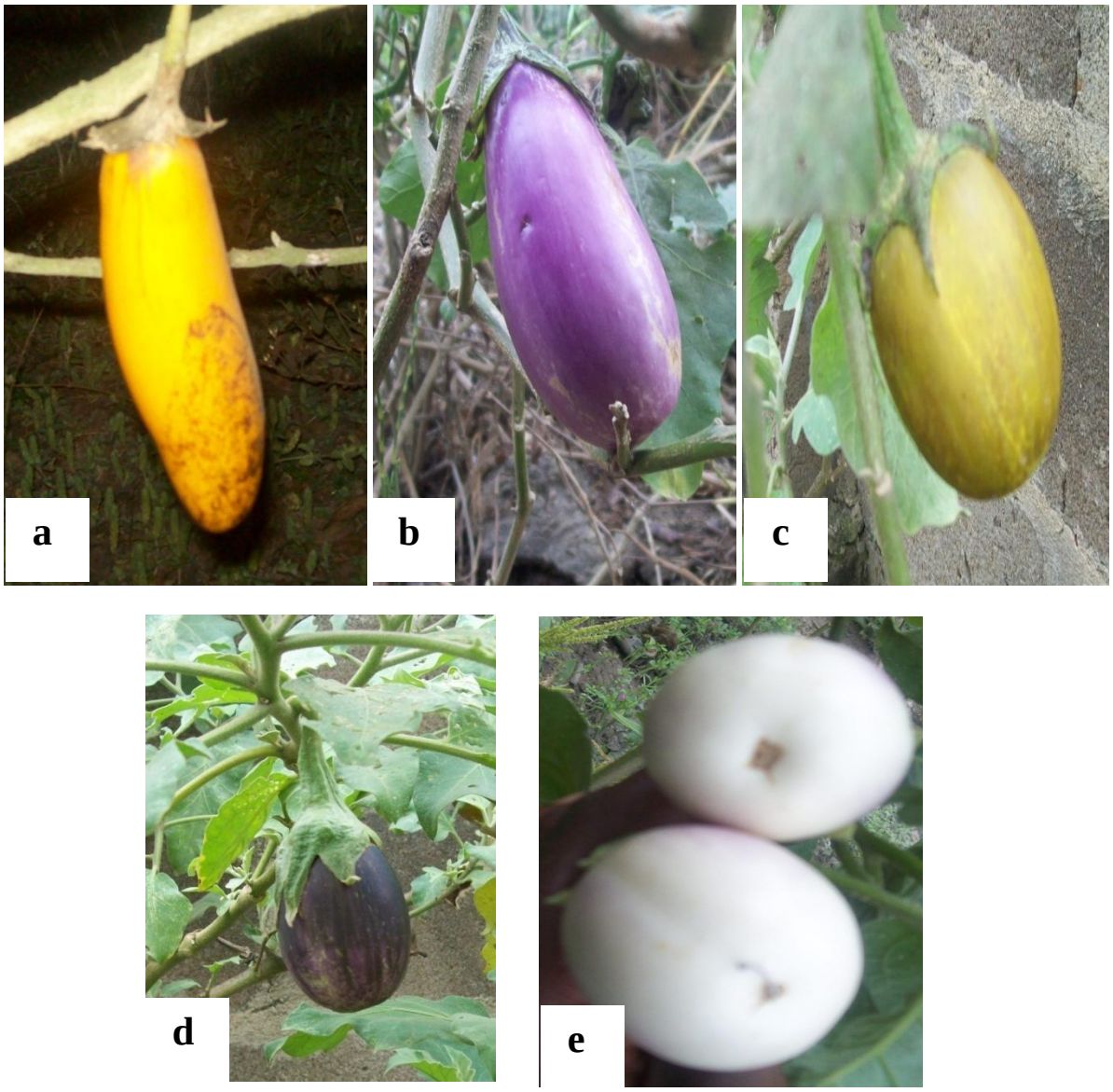


Plate 10: *S. melongena* showing different predominant fruit shapes (X 1/5)

Legend: (a) slightly curved and elongate (b) obovate (c) ellipsoid (d) ovate (e) egg-shaped with depression on top.

4.2.5 *S. gilo* Raddi

S. gilo resembles brinjal eggplant (*S. melongena*) in growth habit. Two basic types were encountered during collections. One type had round green fruits while the second type had long and/or round, white with green-striped fruits (**Plate 11**). Fruits of both turn orange-red when ripe. Another variation noticed in the morphology of *S. gilo* plants was the stem colour which could be purple or green. The flower is white and stellate in shape (**Plate 12**). Common names include Jiló and in some countries in West Africa (e.g. Nigeria) it is known as "garden eggs".

a 

b 

c 

Plate 11: *S. gilo* (X ¼)

Legend: Different fruit colours and shapes (a) whole plant with white fruits (b) round green fruits; (c) long and/or round, white with green-striped fruits.

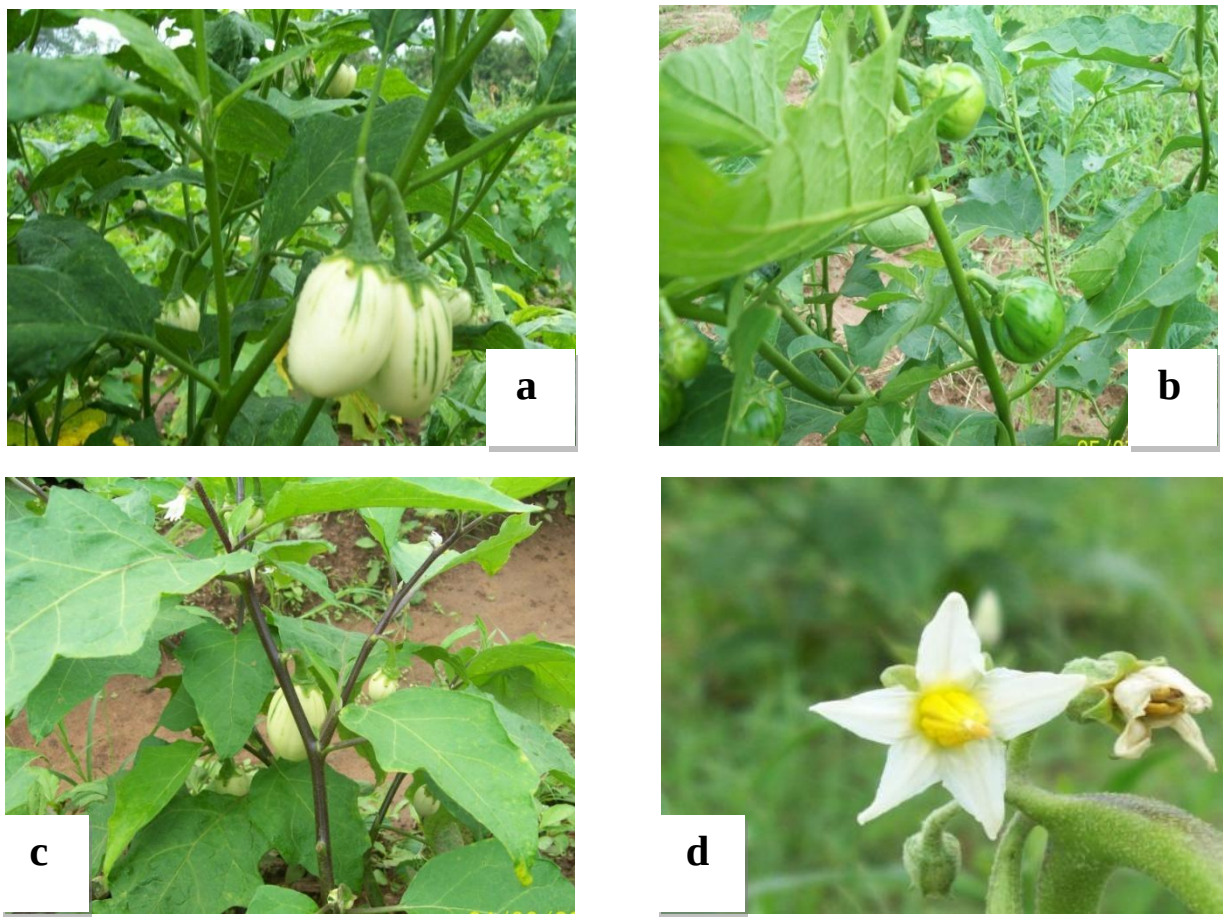


Plate 12: *S. gilo* plants showing different morphological features (X ¼)

Legend: (a) green stem with white fruits (b) green stem with green fruits and (c) purple stem with white fruits (d) white stellate flower.

4.2.6 *S. aethiopicum*

Ethiopian Eggplant, nakati (Osun in South West Nigeria) or *Solanum aethiopicum* is a fruiting plant that grows all year long. It is also known as Mock Tomato, Garden Eggs and Ethiopian Nightshade. These names are a result of its varied morphology. The ripe fruit often looks like a cross between an eggplant and a tomato, which is also a species of *Solanum*. The flower is white and stellate-shaped. The fruits are about two inches in diameter and usually green. When the berries mature and ripe, they turn bright orange-red (**Plate 13**) because of high carotene content.

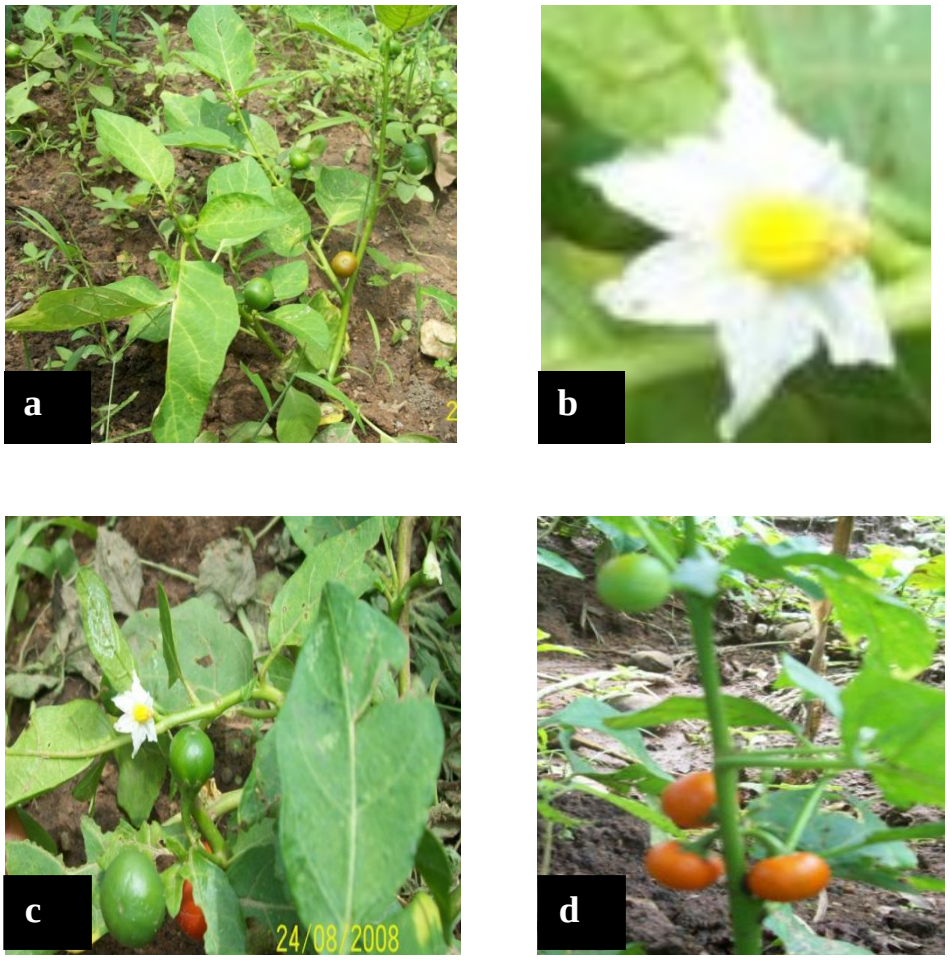


Plate 13: *S. aethiopicum* (X ¼)

Legend: (a) whole plant (b) white stellate flower (c) mature green fruit (d) ripe, bright orange-red fruit.

4.2.7 *S. dasyphyllum*

A semi-woody under shrub which can grow up to 1½ m high, with stem, leaves and corolla hirsute or armed with prickles (**Plate 14**), naturalized and not known in the wild. It is very closely related to *S. macrocarpon* which is glabrous. It is commonly called Wild African eggplant.

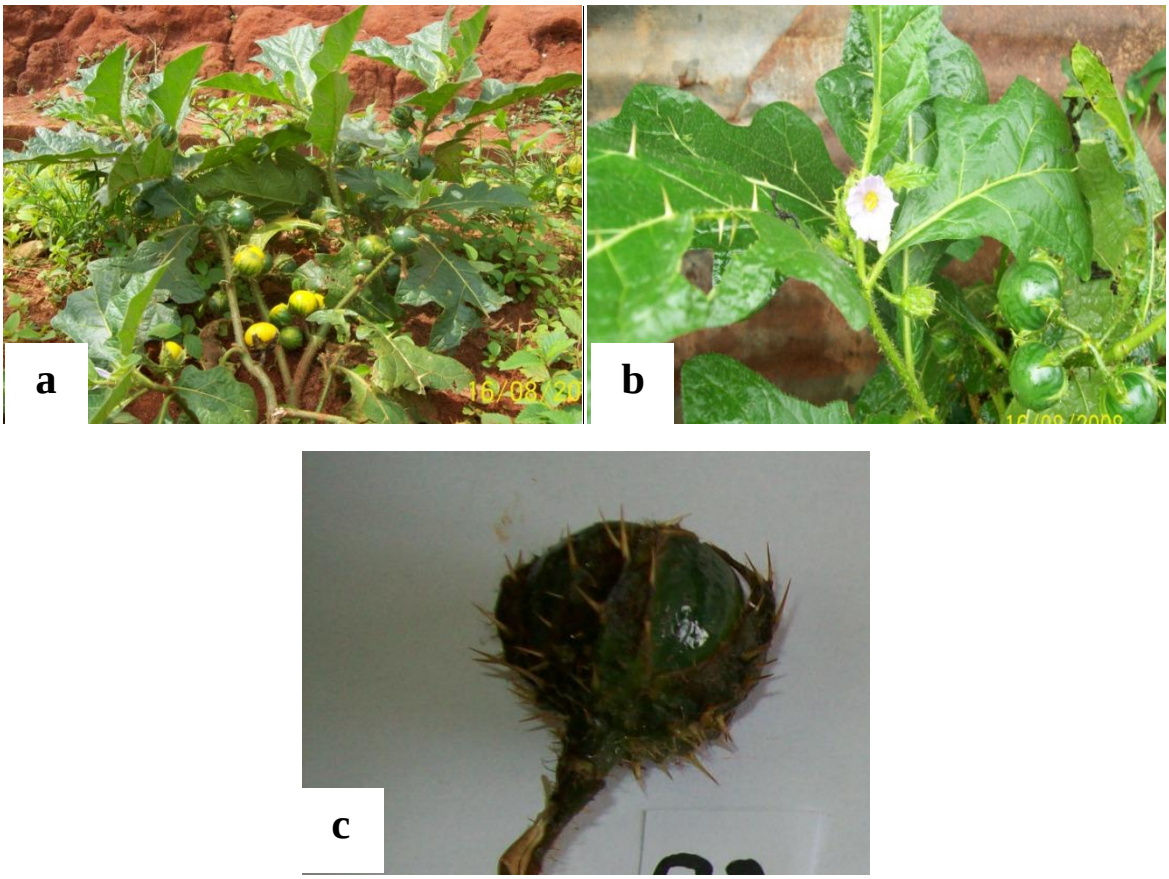


Plate 14: *S. dasyphyllum* (X ¼)

Legend: (a) whole plant with ripe fruits (b) leaves with prickles, flower and mature green fruits (c) fruit with calyx covered with prickles.

4.2.8 *S. torvum* Sw.

Solanum torvum (Turkey Berry) is a bushy, erect and spiny perennial plant. It is also known as Devil's Fig, Prickly Nightshade, Shoo-shoo Bush, Wild Eggplant, Pea Eggplant and many other names. The plant is usually 2 or 3 m in height and 2 cm in basal diameter, but may reach 5m in height and 8 cm in basal diameter. The shrub usually has a single stem at ground level, but it may branch on the lower stem. The stem bark is grey and nearly smooth with raised lenticels. The inner bark has a green layer over an ivory colour. Foliage is confined to the growing twigs. The twigs are grey-green and covered with star-shaped hairs. The spines are short and slightly curved and vary from thick throughout the plant, including the leaf midrib, to entirely absent. The leaves are opposite or one per node, broadly ovate with the border entire or deeply lobed. The petioles are 1 to 6 cm long and the blades are 7 to 23 by 5 to 18 cm covered with short hairs. The flowers are white, tubular with 5 pointed lobes, and grouped in corymbiform cymes. They are shed soon after opening. The fruits are berries that grow in clusters of tiny green spheres of about 1 cm in diameter) that look like green peas (**Plate 15**). They become yellow when fully ripe. They are thin-fleshed and contain numerous flat, round, brown seeds.

Plate 15: ***S. torvum*** **plant showing leaves, flowers and fruits. Inset is a fruit with long stalk (X ¼).**

4.2.9 *S. erianthum*

Common names include Potato Tree, Mullein Nightshade, Velvet Nightshade, and Salvadora. *S. erianthum* is a fast growing evergreen shrub or small tree, reaching a height of 2–8 m. The grey or brown bark is smooth-lenticellate and the trunk is 2–5 cm thick. The crown is flat-topped and spreading. Although the wood is soft and brittle, the limbs are strong enough to support birds. The simple leaves are alternate, ovate or elliptic, and 12–37 cm long. Flowers are in lateral cymes and are 1.1–1.8 cm in diameter. The five-lobed corolla is white and the five stamens have yellow anthers. The fruit is a yellow berry 1–1.2 cm in diameter with many seeds. The specific name, *erianthum*, is derived from the Greek words '*erion*', meaning "wooly", and '*anthos*', meaning "flower," referring to the dense trichomes (hairs) on the flowers. Other parts of the plant are also covered with trichomes, including the berries, leaves, stem tips, and petioles. Trichomes on the leaves, stems, and petioles release an odour similar to tar when rubbed (**Plate 16**).

Plate 16: *S. erianthum* showing simple leaves, white flowers and fruits (X ¼).

4.2.10 *S. macranthum* A.Rich.

Solanum macranthum, which goes by the whimsical common name of Potato Tree or even Giant Potato Tree, is a bushy tree with purple-white flowers that can grow to a height of 15 feet. The fruits are large and round in shape. The leaves are highly lobed, with midrib and stalk covered with long and very sharp thorns/ prickles (**Plate 17**).

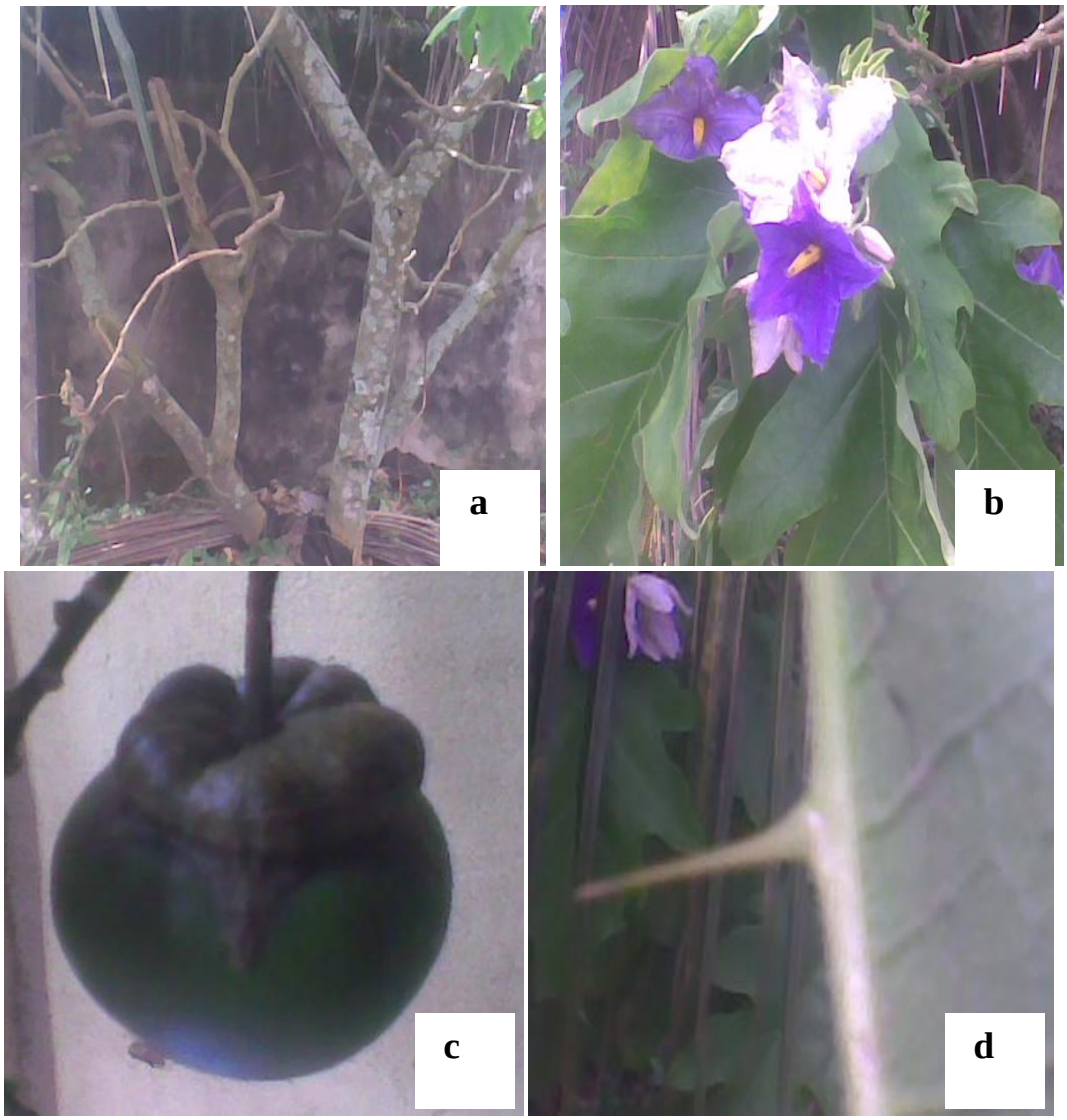


Plate 17: *S. macranthum* plant (X 1/2).

Legend: (a) stem (b) purple-white flowers (c) big round fruit (d) sharp thorn/ prickle on the midrib

4.2.11 *S. incanum*

S. incanum, commonly called bitter apple, is a shrub that grows to a height of about 1m. The stems are purplish in colour and the leaves are simple and lobed. The flower is stellate and whitish in colour. The fruits are small, round and light-green in colour. They are arranged opposite to one another (**Plate 18**).

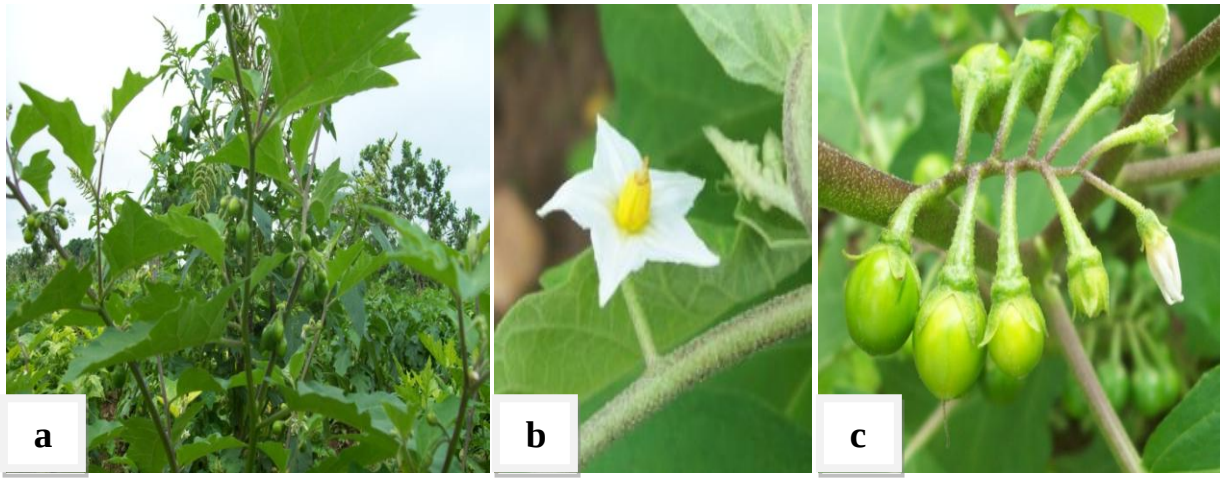


Plate 18: *S. incanum* (X ¼)

Legend: (a) plant with fruits showing purple-coloured stem (b) white stellate flower and (c) fruits arranged opposite to each other.

4.2.12 *S. indicum* subsp.*distichum* var. *distichum*

African nightshade/Bush tomato is an erect undershrub 0.30 to 1.5 m in height. The stems are much branched and the leaves are ovate, 3.5 to 15 cm long, and 2.5 to 8 cm wide, lobed or pinnatifid in the margins, blunt or pointed at the tip, pointed at the base, and stellately wooly beneath. The leaves in the branchlets are much smaller. The fruits are very small, greenish and round. They become orange-coloured when ripe (**Plate 19**).

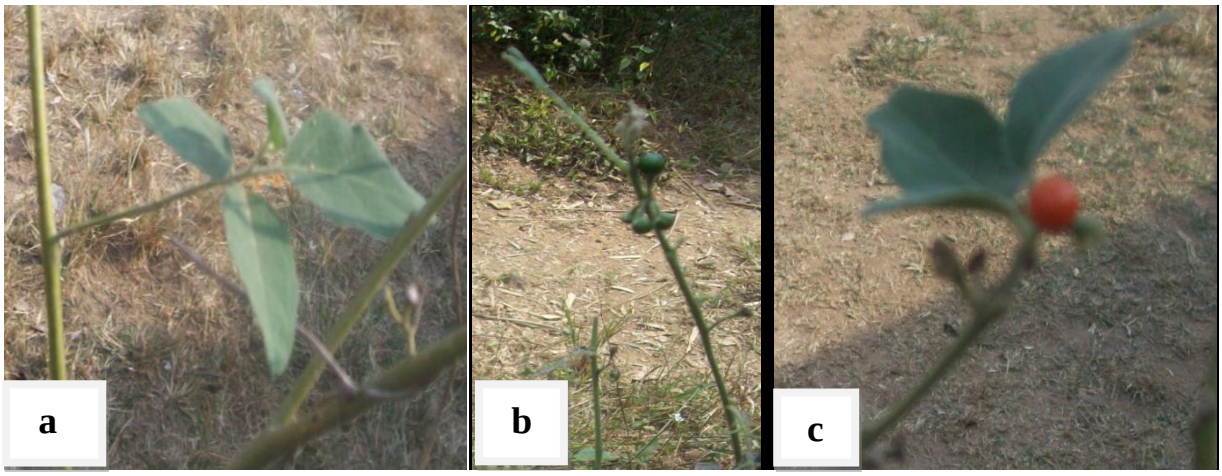


Plate 19: *S. indicum* (X ¼).

Legend: (a) plant with ovate leaf shape (b) small, green, round fruits (c) ripe, orange-coloured fruit

4.3 Taxonomic Key

An indented dichotomous key is presented below for the identification of the species:

1a. Plant size small or intermediate, stem colour green ---------- 2

2a. Corolla type semi-stellate, fruit fresh colour white ---------- 3

3a. Leaf upper surface colouration green, leaf blade lobe strong ---------- 4

4a. Leaf sessile, stem densely spinosus ---------- *S. dasyphyllum*

4b. Leaf petiolate, stem smooth ---------- *S. macrocarpon*

3b. Leaf upper surface colouration light-green, leaf blade lobe weak ---------- *S. indicum subsp.distichum var. distichum*

2b. Corolla type stellate, fruit fresh colour light-green ---------- 5

5a. Foliage density sparse, leaf surface with spine ---------- *S. torvum*

5b. Foliage density dense, leaf surface without spine ---------- 6

6a. Leaf shape lanceolate, leaf lower surface indumentum velutinous ----- *S. erianthum*

6b. Leaf shape ovate, leaf lower surface indumentum velutinous puberulent ---------- 7

7a. Growth habit intermediate, corolla colour white ---------- 8

8a. Leaf orientation horizontal, stem pubescence density glabrous ---------- 9

9a. Leaf blade widest part base, foliage density dense ---------- *S. nigrum*

9b. Leaf blade widest part middle, foliage density intermediate ----- *S. scabrum*

8b. Leaf orientation semi erect, stem pubescence density sparse or intermediate-10

10a. Predominant fruit shape obovate, fruit size intermediate ---- *S. melongena*

10b. Predominant fruit shape flattened, fruit size large ---------- *S. aethiopicum*

7b. Growth habit upright, corolla colour purple-white ---------- 11

11a. Fruit predominant colour milk white, fruit widest part between 1/4 and 1/2 - *S. gilo*

11b. Fruit predominant colour light green, fruit widest part more than 1/2 -------- *S. incanum*

1b. Plant size large, stem colour green purple ---------- *S. macranthum*

4.4 Analyses of Morphological Data.

A total of 37 morphological traits, made up of 26 qualitative (measured by visual observation of the plants according to IPGRI standard) and 11 quantitative characters were measured and compared among the 49 samples studied (**Table 4**). All these descriptor states characterized showed high level of morphological diversity among the samples studied. Morphological characters recorded were grouped accordingly under: (i) Plant's stem/branches (ii) Leaves and (iii) Inflorescence/flower and fruits. Wide variability for all the quantitative descriptors studied was revealed by the range of variation for the different quantitative descriptors. This led to the observation of a large coefficient of variation (> 30%) for all the quantitative characters studied (**Table 5**). Mean values for all the quantitative descriptors together with their standard deviations are presented in **(Table 6)**.

Table 4: List of both Qualitative and Quantitative Characters evaluated in this study

QUALITATIVE CHARACTER		QUANTITATIVE CHARACTER	
Trait	Abbreviation	Trait	Abbreviation
Plant growth habit	PGH	Plant height at Flowering (cm)	PHF
Plant Size	PS	Stem length at 1st inflorescence (cm)	SLFI
Stem pubescence density	SPD	Internode length (cm)	IL
Stem colour	SC	Leave blade length (cm)	LBL
Type of leaf	TOL	Leave blade width (cm)	LBW
leaf attitude (i.e. orientation)	LA	Petiole length	PL
Leaf shape	LS	Mean fresh fruit weight (g)	MFFW
Foliage density	FD	Fruit length (cm)	FL
Position of widest part of leaf blade	PWPLB	Fruit width(cm)	FW
Leaf colour on the adaxial surface	LCAS	LBL to LBW Ratio	LBL/LBW
Leaf Surface Attitude	LSA	FL to FW Ratio	FL/FW
Leaf Blade Lobing	LBLO		
Leaf Apex Shape	LAS		
Leaf Hairiness type on Abaxial Side	LHTAS		
Anthocyanin Colouration of Leaf Veins	ACLV		
Petiole colour	PC		
Corolla type	CT		
Corolla colour	CC		
Inflorescence type	IT		
Predominant fruit shape	PFS		
Fruits predominant colour at commercial ripeness	FPCCR		
Fruits flesh colour	FFC		
Position of widest part of the fruit	PWF		
Fruit secondary colour at commercial ripeness	FSCCR		
Fruit size uniformity	FSU		
Fruit curvature	FC		

Table 5: Range of Variation in Quantitative Descriptors and Coefficient of Variations given by the Ratio of the Standard Deviation to the Mean

	PHF	SLFI	IL	PL	LBL	LBW	FL	FW	MFFW	LBL/ LBW	FL/FW
N Valid	49	49	49	49	49	49	49	49	49	49	49
Missing	0	0	0	0	0	0	0	0	0	0	0
Mean	124.99	79.17	8.15	3.81	17.63	12.03	3.96	3.20	38.74	1.51	1.23
Std. Error of Mean	7.56	8.20	0.62	0.34	1.00	0.74	0.39	0.24	4.58	0.03	0.07
Std. Deviation	52.95	57.40	4.33	2.35	7.02	5.15	2.70	1.70	32.06	0.24	0.48
Variance	2804.19	3295.04	18.77	5.50	49.34	26.57	7.27	2.880	1027.54	0.06	0.23
Range	310.40	328.00	18.00	7.60	26.60	21.10	11.30	6.80	92.50	.90	2.40
Minimum	43.10	3.20	3.10	0.00	4.90	2.80	0.50	0.50	0.50	1.00	0.50
Maximum	353.50	331.20	21.10	7.60	31.50	23.90	11.80	7.30	93.00	1.90	2.90
Sum	6124.50	3879.40	399.10	186.80	863.70	589.70	194.00	156.80	1898.20	74.20	60.50
Coefficient of Variation (%)	42.37	72.50	53.19	61.51	39.85	42.83	68.11	53.03	82.75	0.16	0.38

PHF= Plant height at Flowering; SLFI= Stem length at 1st Inflorescence; IL= Internode length; PL= Petiole length; LBL=Leaf blade length; LBW= Leaf width length; FL=Fruit length; FW= Fruit width; MFFW= Mean Fresh Fruit weight.

Table 6: Mean values and Standard Deviation of Quantitative Descriptors

PLANT'S ID	PHF (cm)	SLFI (cm)	IL (cm)	PL (cm)	LBL (cm)	LBW (cm)	FL (cm)	FW (cm)	MFFW (g)	LBL/LBW	FL/FW
OG02	80.5	41.9	12.3	0.0	19.6	12.0	3.7	4.4	22.4	1.6	0.9
OG03	59.0	32.6	11.5	3.3	11.3	7.0	1.1	1.2	0.6	1.6	0.9
OG04	81.8	52.0	6.1	0	31.5	17.7	3.0	3.5	19.5	1.8	0.9
OG05	60.5	36.5	7.7	3.3	7.6	6.3	1.2	1.2	0.7	1.2	1.0
OG06	80.5	41.2	6.6	0.0	30.6	19.5	3.6	7.3	90.4	1.6	0.5
OG07	70.4	42.0	6.1	0.0	12.3	6.8	3.1	4.5	33.8	1.8	0.7
OG08	160.0	145.6	5.9	7.2	22.2	15.5	1.1	1.2	9.6	1.4	1.0
OG09	353.5	331.2	15.5	5.5	21.9	12.5	1.2	1.0	8.4	1.6	1.2
OG10	138.8	98.6	8.5	5.5	19.4	10.6	6.3	4.8	71.4	1.8	1.3
OY11	150.0	140.8	21.1	7.6	22.9	16.2	5.9	4.2	31.7	1.4	1.4
OY12	124.8	88.2	20.4	6.1	25.9	19.0	4.3	4.4	36.9	1.4	1.0
OY13	124.9	99.0	16.0	5.6	29.7	21.9	4.3	4.9	42.4	1.4	0.9
OY14	109.4	69.3	15.2	5.4	20.3	12.3	5.5	4.0	21.5	1.7	1.4
OY15	142.5	115.1	15.4	4.4	20.9	20.5	1.6	1.6	1.7	1.0	1.0
OY16	113.4	98.6	8.2	5.3	6.4	5.6	1.2	1.3	0.7	1.0	0.8
OY17	71.6	42.5	4.9	6.2	11.2	7.4	1.5	2.3	2.6	1.5	0.7
OY18	137.7	112.6	15.5	4.4	10.3	7.1	1.1	1.2	0.5	1.5	0.9
OY19	130.7	87.3	15.4	6.5	15.1	10.7	9.0	5.2	91.4	1.4	1.7
OY20	65.4	32.1	6.9	6.0	11.5	7.9	1.5	1.7	1.9	1.5	0.9
OS21	194.6	161.7	7.2	6.0	21.9	18.2	1.2	1.2	10.0	1.2	1.1
OG22	161.3	122.0	4.6	1.4	10.8	7.9	8.2	3.8	34.2	1.4	2.2

Table 6 (Contd.): Mean values and Standard Deviation of Quantitative Descriptors

PLANT'S ID	PHF (cm)	SLFI (cm)	IL (cm)	PL (cm)	LBL (cm)	LBW (cm)	FL (cm)	FW (cm)	MFFW (g)	LBL/LBW	FL/FW
LA23	96.1	19.9	7.6	4.3	17.7	10.5	3.3	3.5	30.6	1.7	0.9
LA24	95.1	9.2	7.8	4.2	18.7	11.0	3.3	3.2	30.5	1.7	1.1
LA25	124.5	3.2	6.4	2.6	16.2	10.9	2.9	3.1	31.1	1.6	0.9
LA26	62.8	7.1	7.1	1.0	18.9	10.4	3.4	3.7	93.0	1.8	0.9
OY27	146.0	105.5	8.4	4.2	15.0	9.2	7.2	5.0	80.5	1.7	1.5
OY28	121.7	15.2	4.6	1.1	28.9	15.5	2.3	2.4	33.3	1.9	1.0
OY29	165.5	138.0	4.7	7.5	25.6	23.9	1.3	1.2	9.4	1.1	1.0
ED30	155.0	106.0	4.4	6.2	21.6	16.4	6.3	3.9	70.5	1.3	1.7
ED31	167.0	161.0	4.6	5.0	17.2	11.8	4.7	2.8	70.7	1.5	1.7
ED32	74.1	22.1	4.8	0.0	28.8	15.6	4.2	5.6	41.8	1.9	0.8
ED33	61.5	30.9	6.1	2.2	10.9	6.7	7.3	7.2	30.6	1.7	1.0
ED34	100.0	30.0	5.0	0.0	29.2	17.0	3.5	3.2	21.5	1.7	1.1
OD35	43.1	30.1	3.1	0.0	20.8	11.7	4.2	5.5	42.7	1.8	0.8
OD36	116.0	79.5	5.6	4.8	18.6	13.9	8.7	5.0	73.5	1.3	1.8
CR37	136.4	123.0	8.2	6.2	18.5	15.6	1.2	1.2	9.3	1.2	1.0
CR38	176.5	85.0	7.9	1.1	4.9	2.8	0.5	0.5	6.5	1.8	1.1
CR39	194.0	151.0	7.4	6.3	22.4	19.5	1.3	1.2	8.9	1.2	1.1
CR40	135.0	80.4	6.5	3.4	13.1	9.1	11.8	4.2	90.8	1.5	2.9
CR41	97.2	61.7	5.8	3.8	12.7	9.1	5.1	3.3	78.1	1.4	1.6
CR42	111.5	60.2	7.1	2.7	11.7	7.5	7.8	3.3	86.4	1.6	2.4
CR43	134.3	55.5	8.1	3.3	9.3	5.1	0.8	0.8	6.6	1.9	1.0
CR44	156.8	62.9	6.0	4.3	12.3	10.2	7.7	4.4	86.8	1.2	1.8

Table 6 (Contd.): Mean values and Standard Deviation of Quantitative Descriptors

PLANT'S ID	PHF (cm)	SLFI (cm)	IL (cm)	PL (cm)	LBL (cm)	LBW (cm)	FL (cm)	FW (cm)	MFFW (g)	LBL/LBW	FL/FW
CR45	**121.5**	**60.1**	**7.1**	**2.0**	**8.7**	**5.6**	**3.1**	**2.3**	**40.5**	**1.6**	**1.4**
CR46	**109.9**	**72.7**	**5.4**	**0.9**	**5.6**	**3.2**	**1.0**	**0.8**	**6.8**	**1.8**	**1.2**
CR47	**148.5**	**42.2**	**5.0**	**5.3**	**19.5**	**14.6**	**5.6**	**4.4**	**90.5**	**1.4**	**1.3**
CR48	**97.2**	**62.5**	**4.1**	**4.3**	**13.4**	**9.6**	**8.0**	**3.9**	**92.7**	**1.4**	**2.1**
CR49	**123.0**	**63.0**	**5.4**	**3.3**	**15.2**	**11.0**	**4.1**	**3.0**	**38.2**	**1.4**	**1.4**
CR50	**243.0**	**150.7**	**3.9**	**7.1**	**25.0**	**19.7**	**3.8**	**3.3**	**64.1**	**1.3**	**1.6**
MEAN	**125.0**	**79.2**	**8.1**	**3.8**	**17.6**	**12.0**	**4.0**	**3.2**	**38.7**	**1.5**	**1.2**
SD	**52.95**	**57.40**	**4.33**	**2.35**	**7.02**	**5.15**	**2.70**	**1.70**	**32.06**	**0.24**	**0.48**
P(<0.05)	*	*	*	*	*	*	*	*	*	*	*

PHF= Plant height at Flowering; SLFI= Stem length at 1st Inflorescence; IL= Internode length; PL= Petiole length; LBL=Leaf blade length; LBW= Leaf width length; FL=Fruit length; FW= Fruit width; MFFW= Mean Fresh Fruit weight.

*Significant at $P < 0.05$

4.4.1 Plant's Stem/Branches Characteristics

The Plant's stem/branches characters varied considerably among the *Solanum* samples studied. Analysis of variances with value 2804.19 showed significant difference ($P < 0.05$) in plant height at flowering/fruiting and stem length at first inflorescence between different accessions **(Table 5).** Plant growth habit ranged from upright to intermediate types. Stem colour increased from green to green with purple spots and greenish-purple. **Table 7** below shows the detailed stem/branches characteristics for both qualitative and quantitative traits.

Table 7: Plant's Stem/Branches Characteristics observed for the *Solanum* samples Characterized.

PLANT'S ID	PHF (cm)	SLFI (cm)	PGH	SC
OG02	80.5	41.9	Intermediate	Green
OG03	59.0	32.6	Intermediate	Green
OG04	81.8	52.0	Upright	Green
OG05	60.5	36.5	Intermediate	Green
OG06	80.5	41.2	Upright	Green
OG07	70.4	42.0	Intermediate	Green
OG08	160.0	145.6	Upright	Green
OG09	353.5	331.2	Upright	Green
OG10	138.8	98.6	Upright	Green
OY11	150.0	140.8	Upright	Green
OY12	124.8	88.2	Upright	Green
OY13	124.9	99.0	Upright	Green
OY14	109.4	69.3	Intermediate	Green purple
OY15	142.5	115.1	Upright	Green purple
OY16	113.4	98.6	Intermediate	Green
OY17	71.6	42.5	Intermediate	Green
OY18	137.7	112.6	Upright	Green
OY19	130.7	87.3	Intermediate	Green
OY20	65.4	32.1	Intermediate	Green
OS21	194.6	161.7	Upright	Green
OG22	161.3	122.0	Upright	Green
LA23	96.1	19.9	Upright	Green
LA24	95.1	9.2	Upright	Green
LA25	124.5	3.2	Upright	Green purple
LA26	62.8	7.1	Upright	Green
OY27	146.0	105.5	Intermediate	Green
OY28	121.7	15.2	Intermediate	Green
OY29	165.5	138.0	Upright	Green with purple spots
ED30	155.0	106.0	Upright	Green purple
ED31	167.0	161.0	Upright	Green
ED32	74.1	22.1	Upright	Green
ED33	61.5	30.9	Upright	Green
ED34	100.0	30.0	Upright	Green
OD35	43.1	30.1	Upright	Green
OD36	116.0	79.5	Upright	Green
CR37	136.4	123.0	Upright	Green with purple spots
CR38	176.5	85.0	Upright	Green
CR39	194.0	151.0	Upright	Green
CR40	135.0	80.4	Upright	Green
CR41	97.2	61.7	Intermediate	Green
CR42	111.5	60.2	Upright	Green
CR43	134.3	55.5	Upright	Green
CR44	156.8	62.9	Upright	Green
CR45	121.5	60.1	Upright	Green
CR46	109.9	72.7	Upright	Green
CR47	148.5	42.2	Upright	Green
CR48	97.2	62.5	Intermediate	Green with purple spots
CR49	123.0	63.0	Upright	Green
CR50	243.0	150.7	Upright	Green

PGH = Plant Growth Habit; SC = Stem Colour.

4.4.2 Leaf Characteristics

All the accessions of *S. melongena* and most of the other related species studied had broad simple leaves. All the samples also have prickle-less leaves except for one accession of *S. melongena* (CR40) and all accessions of *S. dasyphyllum*, *S. torvum* and *S. macranthum*. Many samples have acute leaf blade tip angle, while others have leaf blade tip angle ranging from very acute to intermediate and obtuse (**Table 8**). Leaf blade length (LBL) varied from 4.90cm in CR38 (*S. indicum* subsp.*distichum* var. *distichum*) to 31.50cm in OG04 (*S. dasyphyllum*) while leaf blade width (LBW) ranged from 2.80cm in CR38 (*S. indicum* subsp.*distichum* var. *distichum*) to a maximum of 23.90cm in OY29 (*S. torvum*). Leaf blade length was the least diverse among all the traits phenotyped with a low coefficient of variation of 39.85% followed by plant height at flowering and leaf blade width (42.83%) (**Table 5**). The smallest leaf size was noted in *S. nigrum* and *S. scabrum* while the largest was noted in *S. macranthum*. The sharpest and longest prickles were also found in *S. macranthum*. Leaves of all accessions of *S. macrocarpon* and *S. dasyphyllum* were sessile.

Table 8: Leaf characteristics recorded for all the samples.

PLANT'S ID	LBL (cm)	LBW (cm)	LBLO	LAS	LHTAS	ACLV
OG02	19.6	12.0	Strong	Acute	Pilose	Green
OG03	11.3	7.0	Very weak	Acute	Glabrous	Green
OG04	31.5	17.7	Strong	Acute	Velutinous	Green
OG05	7.6	6.3	Very weak	Acute	Glabrous	Green
OG06	30.6	19.5	Strong	Acute	Glabrous	Green
OG07	12.3	6.8	Weak	Acute	Glabrous	Green
OG08	22.2	15.5	Intermediate	Acute	Puberulent	Green
OG09	21.9	12.5	Very weak	Acute	Puberulent	Green
OG10	19.4	10.6	Intermediate	Acute	Puberulent	Green
OY11	22.9	16.2	Strong	Acute	Puberulent	Green
OY12	25.9	19.0	Strong	Acute	glabrous	Green
OY13	29.7	21.9	Intermediate	Acute	Puberulent	Green
OY14	20.3	12.3	Intermediate	Acute	Puberulent	Main vein purple rest green
OY15	20.9	20.5	Strong	Acute	Puberulent	Green
OY16	6.4	5.6	Very weak	Acute	Glabrous	Green
OY17	11.2	7.4	Weak	Very acute	Glabrous	Green
OY18	10.3	7.1	Very weak	Very acute	Glabrous	Green
OY19	15.1	10.7	Intermediate	Acute	Puberulent	Green
OY20	11.5	7.9	Weak	Acute	Glabrous	Green
OS21	21.9	18.2	Intermediate	Acute	Puberulent	Green
OG22	10.8	7.9	Intermediate	Intermediate	Puberulent	Green
LA23	17.7	10.5	Intermediate	Acute	Puberulent	Green
LA24	18.7	11.0	Intermediate	Acute	Puberulent	Green
LA25	16.2	10.9	Strong	Acute	Puberulent	Green
LA26	18.9	10.4	Intermediate	Acute	Glabrous	Green
OY27	15.0	9.2	Intermediate	Intermediate	Puberulent	Green
OY28	28.9	15.5	Weak	Obtuse	Glabrous	Green
OY29	25.6	23.9	Strong	Acute	Puberulent	Green
ED30	21.6	16.4	Intermediate	Obtuse	Puberulent	Main vein purple rest green
ED31	17.2	11.8	Intermediate	Intermediate	Puberulent	Green
ED32	28.8	15.6	Intermediate	Intermediate	Glabrous	Green
ED33	10.9	6.7	Intermediate	Acute	Puberulent	Green
ED34	29.2	17.0	Strong	Acute	Velutinous	Green
OD35	20.8	11.7	Intermediate	Intermediate	Glabrous	Green
OD36	18.6	13.9	Intermediate	Intermediate	Puberulent	Green
CR37	18.5	15.6	Strong	Acute	Puberulent	Green
CR38	4.9	2.8	Weak	Acute	Puberulent	Green

Table 8 (Contd.): Leaf characteristics recorded for all the samples

PLANT'S ID	LBL (cm)	LBW (cm)	LBLO	LAS	LHTAS	ACLV
CR39	22.4	19.5	Strong	Intermediate	Puberulent	Green
CR40	13.1	9.1	Strong	Intermediate	Puberulent	Green
CR41	12.7	9.1	Intermediate	Intermediate	Puberulent	Green
CR42	11.7	7.5	Intermediate	Intermediate	Puberulent	Main vein purple rest green
CR43	9.3	5.1	Weak	Acute	Puberulent	Green
CR44	12.3	10.2	Strong	Intermediate	Puberulent	Main vein purple rest green
CR45	8.7	5.6	Weak	Intermediate	Puberulent	Green
CR46	5.6	3.2	Intermediate	Acute	Puberulent	Green
CR47	19.5	14.6	Intermediate	Intermediate	Puberulent	Green
CR48	13.4	9.6	Strong	Intermediate	Puberulent	Main vein purple rest green
CR49	15.2	11.0	Intermediate	Obtuse	Puberulent	Green
CR50	25.0	19.7	Very strong	Acute	Velutinous	Green

LBLO = Leaf Blade Lobing; LAS = Leaf Apex Shape; LHTAS = Leaf Hairiness type on Abaxial Side; ACLV = Anthocyanin Colouration of Leaf Veins.

4.4.3 Flower and Fruit Characteristics

Flower (corolla) colour intensity observed in all the accessions increased from pure white to light purple and dark purple. Deep bluish purple corolla colour was noted in *S. macranthum* and bluish purple flower colour in *S. macrocarpon*. Corolla colour in *S. melongena* varied from white to light purple and corolla shape ranged from semi-stellate to stellate and rotate. Fruit shape, size and colour were the most variable traits recorded among the samples (**Table 9**). A large coefficient of variation above 50% applied to fruit length, width and weight suggesting that fruit traits were the most diverse quantitative morphological characters observed. The five characteristic fruit shapes recorded were flattened, round, ellipsoid, ovate or egg-shaped and obovate or oblong types. Fruits that are round in shape occurred most frequently (40.8%) among the samples characterized. The maximum fruit length and width recorded were 11.80cm in CR40 and 7.30cm in OG06 respectively while their minimum values were both the same (0.50cm) both in CR38. Fruits predominant colour at commercial ripeness (FPCCR) ranged from milk-white to light green and dark green, and purple to purple-black. All samples of *S. macrocarpon* when compared with *S. melongena* produced relatively smaller round to spherical either dark green fruits with pale green stripes or milk white in an erect position. Similarly, the fruits of *S. torvum* and *S. nigrum* accessions were dark green with uniform colour distribution. Mean Fresh fruit weight of all the accessions studied ranged from 0.5g in OY18 to 93.0g in LA26.

Table 9: Wide diversity in fruit Characters observed in eggplant *Solanum* accessions

PLANT'S ID	PFS	FPCCR	FSCCR	FFC	FSU	PWF	FL (cm)	FW (cm)	MFFW (g)
OG02	Flattened	Light green	Orange yellow	White	Intermediate	Btw 1/4 and 1/2	3.7	4.4	22.4
OG03	Rounded	Light green	Dark green	Light green	High	Btw 1/4 and 1/2	1.1	1.2	0.6
OG04	Flattened	Dark green	Orange yellow	White	Intermediate	Btw 1/4 and 1/2	3.0	3.5	19.5
OG05	Rounded	Light green	Dark green	Light green	High	Btw 1/4 and 1/2	1.2	1.2	0.7
OG06	Flattened	Milk white	Orange yellow	White	Intermediate	Btw 1/4 and 1/2	3.6	7.3	90.4
OG07	Flattened	Dark green	Orange yellow	White	Intermediate	Btw 1/4 and 1/2	3.1	4.5	33.8
OG08	Rounded	Light green	Orange yellow	Light green	High	Btw 1/4 and 1/2	1.1	1.2	9.6
OG09	Rounded	Light green	Orange yellow	Light green	High	Btw 1/4 and 1/2	1.2	1.0	8.4
OG10	Obovate	Light green	Orange yellow	White	Intermediate	More than 1/2	6.3	4.8	71.4
OY11	Ovate	Milk white	Orange yellow	White	High	Btw 1/4 and 1/2	5.9	4.2	31.7
OY12	Rounded	Milk white	Orange yellow	White	Intermediate	Btw 1/4 and 1/2	4.3	4.4	36.9
OY13	Rounded	Milk white	Orange yellow	White	High	Btw 1/4 and 1/2	4.3	4.9	42.4
OY14	Obovate	Milk white	Orange yellow	White	Intermediate	Btw 1/4 and 1/2	5.5	4.0	21.5
OY15	Obovate	Light green	Orange yellow	Light green	High	More than 1/2	1.6	1.6	1.7
OY16	Flattened	Dark green	Purple	Light green	High	Btw 1/4 and 1/2	1.2	1.3	0.7

Table 9 (Contd.): Wide diversity in fruit Characters observed in eggplant *Solanum* accessions

PLANT'S ID	PFS	FPCCR	FSCCR	FFC	FSU	PWF	FL (cm)	FW (cm)	MFFW (g)
OY17	Flattened	Light green	Orange yellow	White	High	More than 1/2	1.5	2.3	2.6
OY18	Flattened	Dark green	Purple	Light green	High	Btw 1/4 and 1/2	1.1	1.2	0.5
OY19	Obovate	Milk white	Orange yellow	White	Intermediate	More than 1/2	9.0	5.2	91.4
OY20	Flattened	Light green	Orange yellow	White	High	Btw 1/4 and 1/2	1.5	1.7	1.9
OS21	Rounded	Light green	Orange yellow	White	High	Btw 1/4 and 1/2	1.2	1.2	10.0
OG22	Ellipsoid	Light green	Orange yellow	White	Intermediate	More than 1/2	8.2	3.8	34.2
LA23	Rounded	Light green	Orange yellow	White	High	Btw 1/4 and 1/2	3.3	3.5	30.6
LA24	Obovate	Light green	Orange yellow	White	Intermediate	Btw 1/4 and 1/2	3.3	3.2	30.5
LA25	Rounded	Light green	Orange yellow	White	Intermediate	Btw 1/4 and 1/2	2.9	3.1	31.1
LA26	Flattened	Light green	Orange yellow	White	Intermediate	Btw 1/4 and 1/2	3.4	3.7	93.0
OY27	Ellipsoid	Purple black	Orange yellow	White	High	Btw 1/4 and 1/2	7.2	5.0	80.5
OY28	Rounded	Dark green	Orange yellow	White	Intermediate	Btw 1/4 and 1/2	2.3	2.4	33.3
OY29	Rounded	Light green	Orange yellow	White	High	Btw 1/4 and 1/2	1.3	1.2	9.4
ED30	Obovate	Purple	Orange yellow	White	Intermediate	More than 1/2	6.3	3.9	70.5
ED31	Obovate	Light green	Orange yellow	White	Intermediate	More than 1/2	4.7	2.8	70.7
ED32	Rounded	Dark green	Orange yellow	White	Intermediate	Btw 1/4 and 1/2	4.2	5.6	41.8
ED33	Ellipsoid	Light green	Orange yellow	White	Intermediate	Btw 1/4 and 1/2	7.3	7.2	30.6
ED34	Rounded	Light green	Orange yellow	White	Intermediate	Btw 1/4 and 1/2	3.5	3.2	21.5

Table 9 (Contd.): Wide diversity in fruit Characters observed in eggplant *Solanum* accessions

PLANT'S ID	PFS	FPCCR	FSCCR	FFC	FSU	PWF	FL (cm)	FW (cm)	MFFW (g)
OD35	Rounded	Dark green	Orange yellow	White	Intermediate	Btw 1/4 and 1/2	4.2	5.5	42.7
OD36	Obovate	Light green	Orange yellow	White	Intermediate	More than 1/2	8.7	5.0	73.5
CR37	Rounded	Light green	Orange yellow	White	High	Btw 1/4 and 1/2	1.2	1.2	9.3
CR38	Rounded	Light green	Orange yellow	White	High	Btw 1/4 and 1/2	0.5	0.5	6.5
CR39	Rounded	Light green	Orange yellow	White	High	Btw 1/4 and 1/2	1.3	1.2	8.9
CR40	Elongate	Milk white	Orange yellow	White	High	Btw 1/4 and 1/2	11.8	4.2	90.8
CR41	Ellipsoid	Purple	Orange yellow	White	Intermediate	More than ½	5.1	3.3	78.1
CR42	Obovate	Purple	Orange yellow	Pale yellow	Intermediate	More than 1/2	7.8	3.3	86.4
CR43	Rounded	Dark green	Orange yellow	white	Intermediate	Btw 1/4 and 1/2	0.8	0.8	6.6
CR44	Obovate	Purple	Orange yellow	Pale yellow	Intermediate	More than 1/2	7.7	4.4	86.8
CR45	Ellipsoid	Purple	Orange yellow	Pale yellow	Low	Btw 1/4 and 1/2	3.1	2.3	40.5
CR46	Rounded	Dark green	Orange yellow	White	Intermediate	Btw 1/4 and 1/2	1.0	0.8	6.8
CR47	Obovate	Purple	Orange yellow	Pale yellow	High	More than 1/2	5.6	4.4	90.5
CR48	Obovate	Purple	Orange yellow	Pale yellow	Intermediate	More than 1/2	8.0	3.9	92.7
CR49	Ellipsoid	Milk white	Orange yellow	Pale yellow	Intermediate	Btw 1/4 and 1/2	4.1	3.0	38.2
CR50	Rounded	Light green	Orange yellow	white	Intermediate	Btw 1/4 and 1/2	3.8	3.3	64.1

PFS = Predominant fruit shape; FPCCR = Fruit predominant colour at commercial ripeness; FSCCR = Fruit secondary colour at commercial ripeness; FFC = Fruits flesh colour; FSU = Fruit size uniformity; PWF = Position of widest part of the fruit.

4.5 Pair-wise Analysis of Morphological Data

The data obtained for each of qualitative and quantitative characters were analysed separately and then combined. The average value for each quantitative character measured on each sample was used in calculating total variance explained by Principal Axis (**Appendix 4**). These data were subjected to pair-wise analyses (**Appendix 5**) and dendrograms generated showing relationship among the species based on morphological data are represented in **Figures 4a, b** and **c**. However, detailed result of morphological characterization is shown in **Appendices 6A, B, C** and **7**. **Figure 4a** shows 13 major clusters and two ungrouped samples (OY27 and CR45) at a coefficient of 0.72 (72%) when qualitative characters only were analysed.

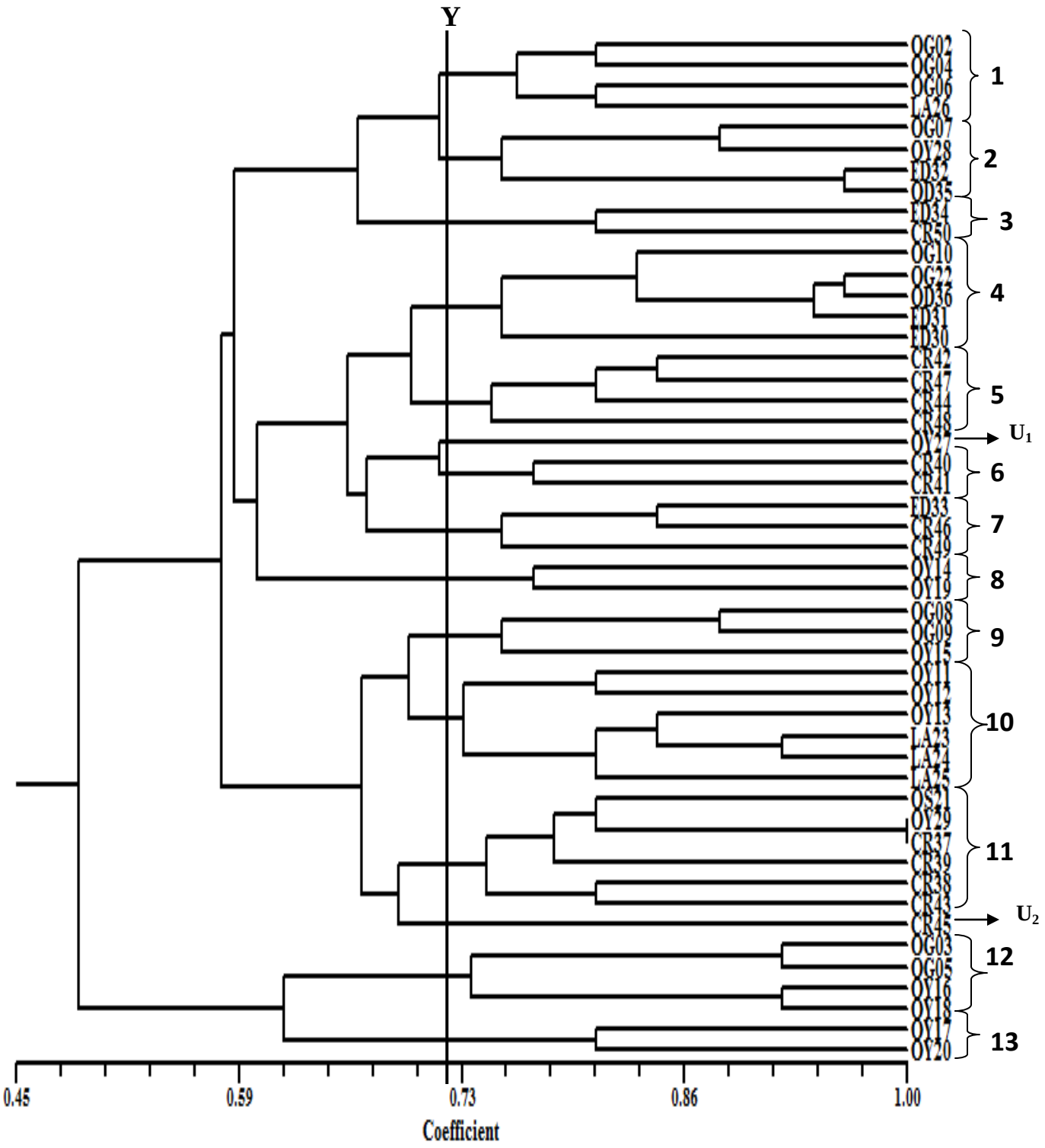


Figure 4a: A UPGMA dendrogram of Pair-wise (Similarity) analysis showing relationships among the samples using Data for Qualitative characters only.

Legend: Y represents truncated line at a Co-efficient of Similarity of about 0.72 (or 72%); thirteen (13) Clusters and 2 Ungrouped (U_1 and U_2) samples were equally distinguishable at that Similarity Coefficient.

The dendrogram generated from the combined data revealed that at truncated line of about 43% (i.e. a co-efficient of 0.43), six major clusters and one ungrouped (i.e. sample CR50) were formed (**Figure 4b**).

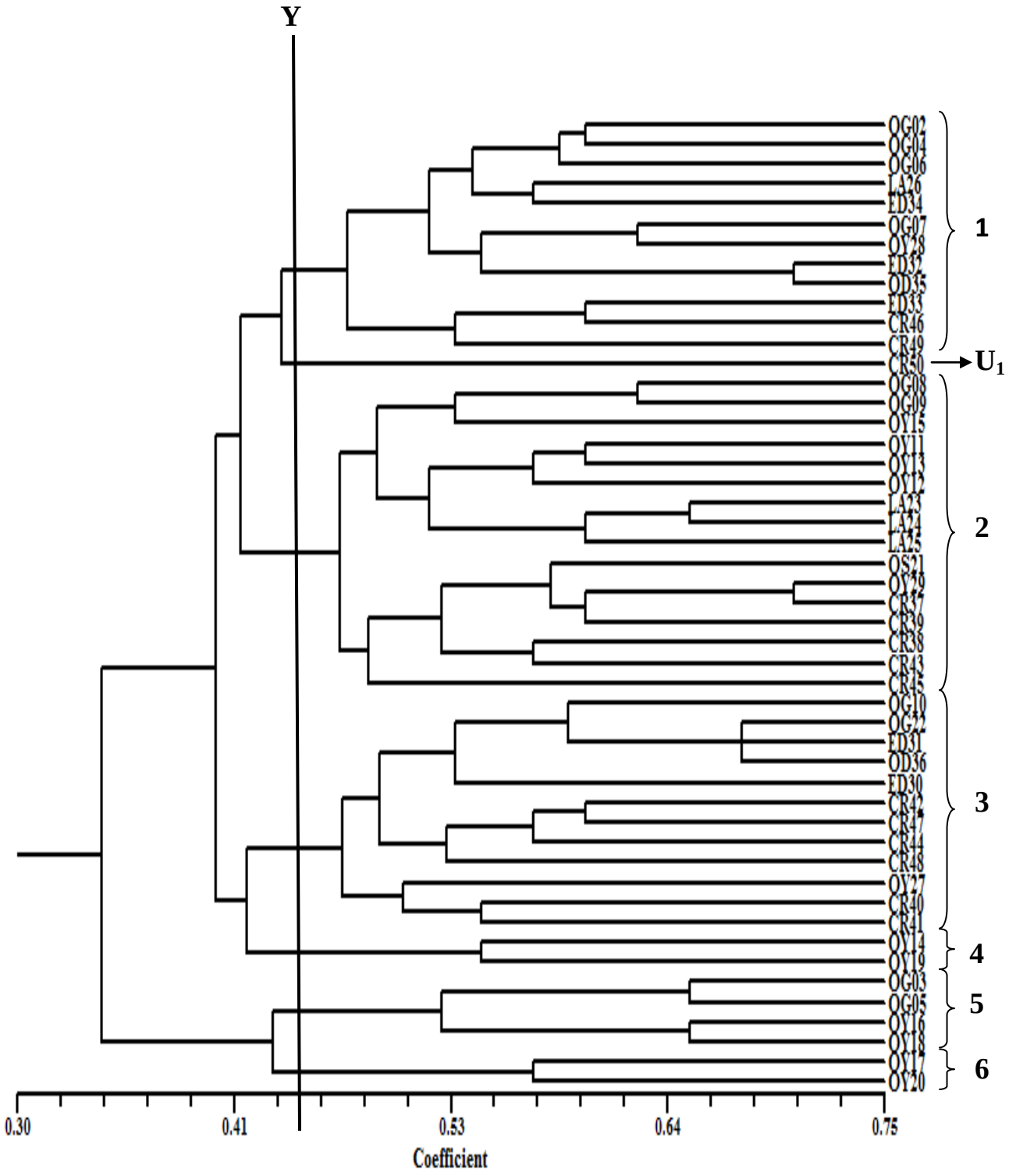


Figure 4b: Dendrogram of the Combined Morphological Data (Both Qualitative and Quantitative).

Legend: Y represents truncated line at a Co-efficient of Similarity of 0.43 (i.e. 43%); Six major clusters and one Ungrouped (U_1) sample were equally distinguishable at that Similarity Coefficient.

At a truncated line of about 51% (i.e. a co-efficient of 0.51), still in the dendrogram of the combined data, 11 major clusters and 2 ungrouped (i.e. samples CR50 and CR45 respectively) were formed (**Figure 4c**).

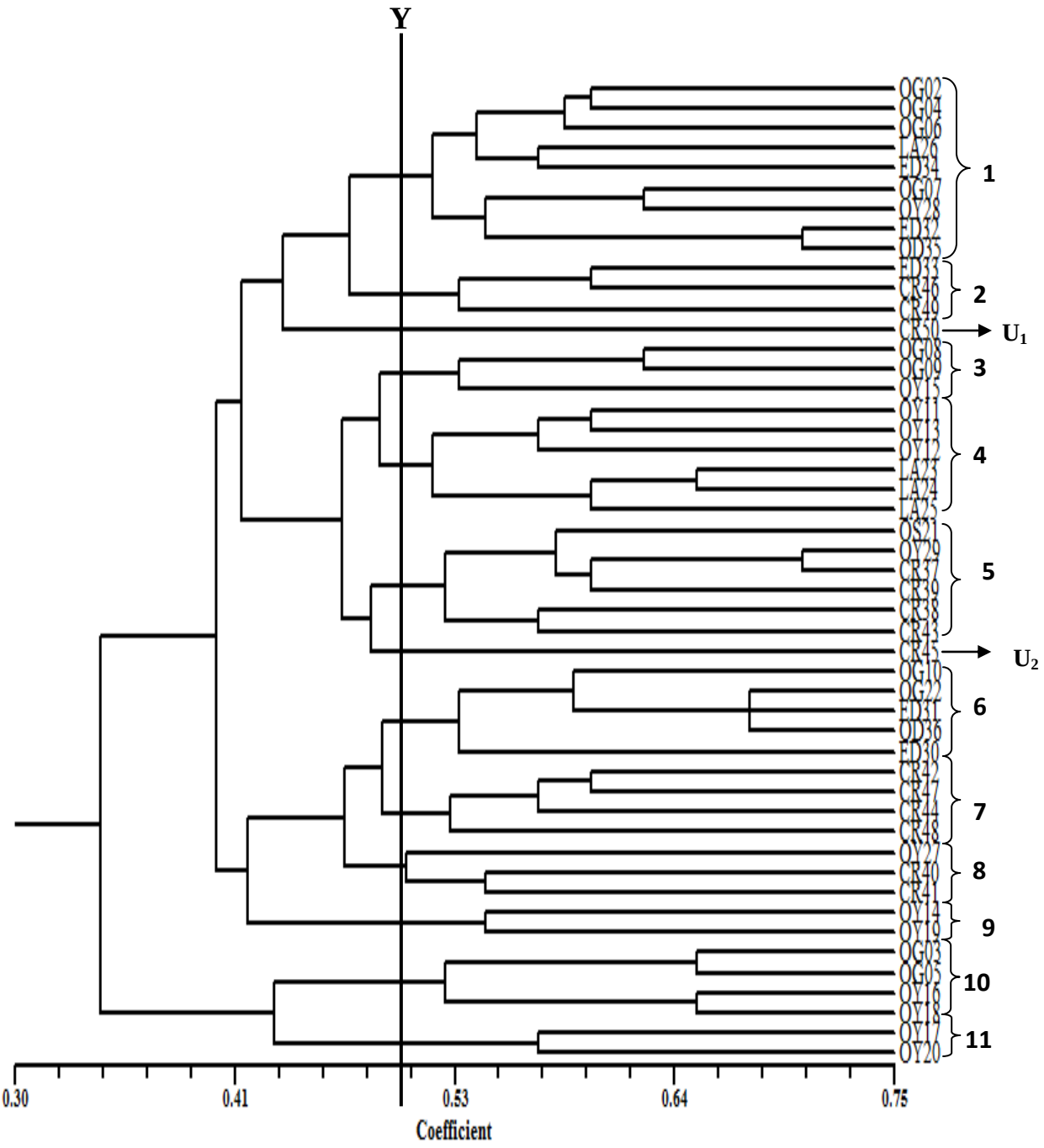


Figure 4c: Dendrogram of the Combined Morphological Data at a Similarity Coefficient of 0.51 (or about 51 %.)

Legend: Y represents truncated line at a Co-efficient of Similarity of 0.51 (or about 51 %.); Eleven Clusters and 2 Ungrouped (U_1 and U_2) samples were equally distinguishable at that Similarity Coefficient.

The scale of the dendrogram of combined data was between 0.34 and 0.75 with a mean value of 0.55. The dendrogram generated from the combined data (**figures 4b and c**) was more informative as more characters are involved and clearly showed the phenetic relationship among the samples used in this study. The grouping pattern as shown in **Figure 4c** revealed that Cluster 1 was the largest group with nine samples represented by three of *S. dasyphyllum* and six of *S. macrocarpon* while clusters 9 and 11 have the lowest with two samples each. All samples of *S. dasyphyllum* and *S. macrocarpon* are grouped together in Cluster 1. All *S. nigrum* and *S. scabrum* are grouped together in Cluster 10; Cluster 11 contained exclusively all *S. aethiopicum* samples. Cluster 4 contained exclusively most *S. gilo* samples; Cluster 5 is made up of most samples of *S. torvum* and two samples of *S. indicum*. Remaining accessions of *S. torvum* and *S. indicum* occurred in other clusters. Most of *S. melongena* samples occurred exclusively in Clusters 6, 7 and 8. However, samples CR50 (*S. macranthum*) and CR45 (*S. gilo*) remained ungrouped.

4.6 Principal Component Analysis of Morphological Data

The clustering pattern of accessions obtained in the dendrogram was further investigated through Principal Component Analysis (PCA) to determine the relationship among plant traits and accessions. Accessions were compared through ordination analysis and projected on two dimensional matrix plots to show the relationship among the 49 eggplant accessions studied (**Figure 5a**). Four major groups (A, B, C and D) are formed while two accessions (numbered 8 and 49), corresponding to OG09 (*S. erianthum*) and CR50 (*S. macranthum*) separated out from any cluster. Cluster C happens to be the largest with eighteen samples while the smallest is Cluster B with four samples. Clusters A and D have eleven and fourteen samples respectively.

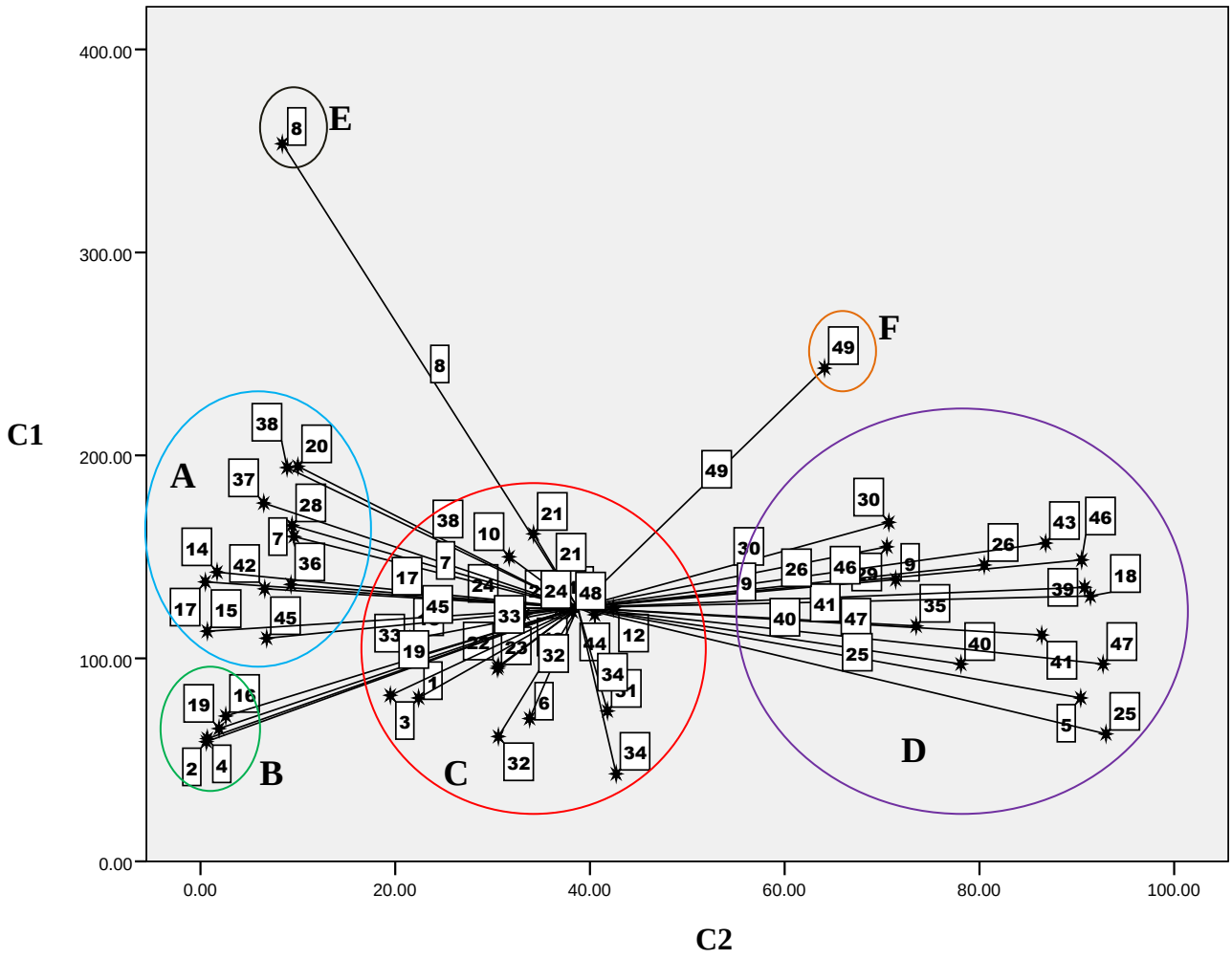


Figure 5a: Relationship among 49 *Solanum* accessions studied based on Principal Components Analysis using Data for Eleven Quantitative Morphological Traits.

Legend: A, B, C and D represent the four clusters; E and F are the ungrouped samples.

The bivariate matrix plot was also projected on a three dimensional scale to display the similarity or dissimilarity distance between different accessions in space (**Figure 5b**). Two major clusters (X and Y) are formed this time around. The larger been X with forty four samples distantly followed by Y with three samples. Again, samples OG09 and CR50 (numbered 8 and 49 respectively) separated out and remained ungrouped. Sample CR50 has earlier been observed to be ungrouped in the results on dendrogram (figures 4b and c).

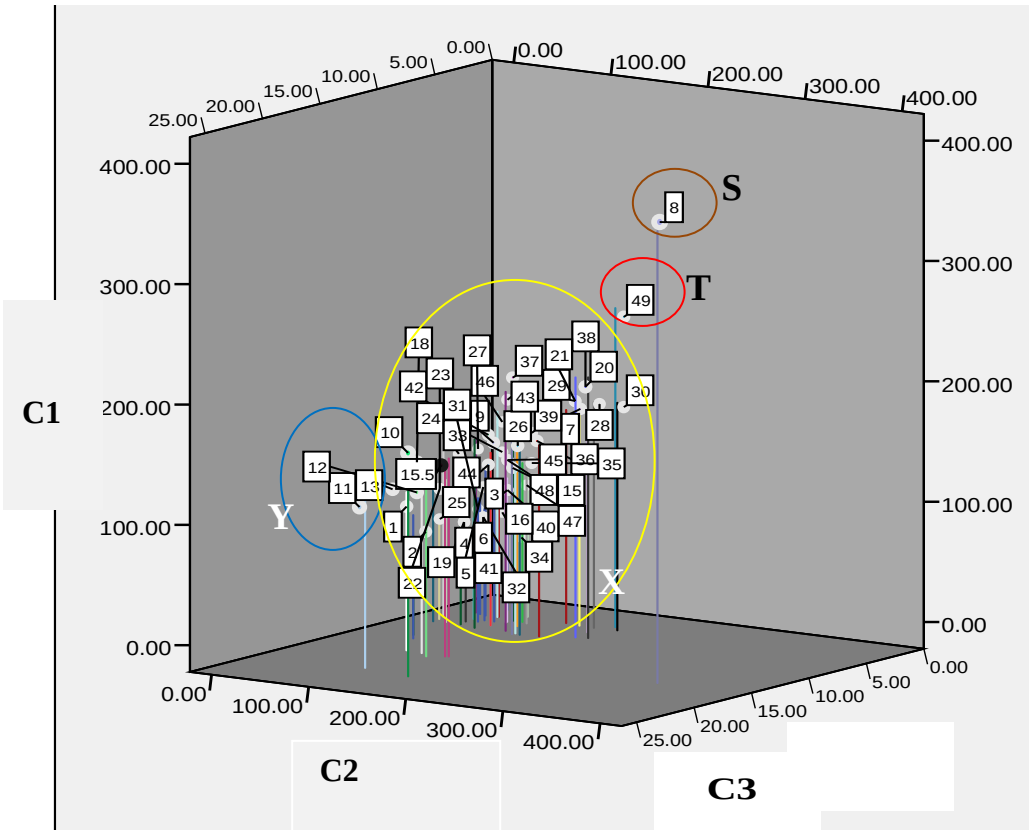


Figure 5b: Projection of the 49 *Solanum* Accessions Morphologically Characterized on a Three Dimensional Basis.

Legend: Illustration of the Dis (similarity) distance among them in space and hence their clustering into distinct groups of similar accessions using the three major Components C1, C2 and C3; X and Y are the 2 major clusters formed while S and T are the ungrouped samples.

NOTE: In **figures 5a** and **b**, 1 represents OG02, 2 represents OG03, etc. See **Table 10** for further clarification.

Table 10: Serial number and respective sample I.D with name used for Principal Component Analysis.

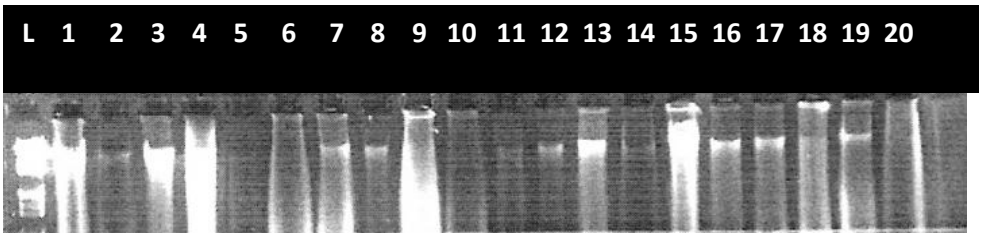
S/N	Sample I.D No.	Name	S/N	Sample I.D No.	Name
1	OG02	*Solanum dasyphyllum*	14	OY15	*S.incanum* L.(Green,small fruit)
2	OG03	*S. nigrum*	15	OY16	*S. scabrum*
3	OG04	*S. dasyphyllum*	16	OY17	*S. aethiopicum*
4	OG05	*S. nigrum*	17	OY18	*S. scabrum*
5	OG06	*S. macrocarpon* (White fruit)	18	OY19	*S. melongena* (White fruit)
6	OG07	*S. macrocarpon* (Green fruit)	19	OY20	*S. aethiopicum*
7	OG08	*S. torvum*	20	OS21	*S. torvum*
8	OG09	*S. erianthum*	21	OG22	*S. melongena* (Green fruit)
9	OG10	*S. melongena* (Green fruit)	22	LA23	*S. gilo* Raddi (Green egg-shaped fruit)
10	OY11	*S. gilo* Raddi (White fruit)	23	LA24	*S. gilo* Raddi (Green round fruit)
11	OY12	*S. gilo* Raddi (White fruit)	24	LA25	*S. gilo* Raddi (Green round fruit with greenish purple stem)
12	OY13	*S. gilo* Raddi (White fruit)	25	LA26	*S. macrocarpon* (Green fruit)
13	OY14	*S. gilo* Raddi (White fruit)	26	OY27	*S. melongena* (Purple fruit)

TABLE 10 (contd.): Serial number and respective sample I.D with name used for Principal Component Analysis.

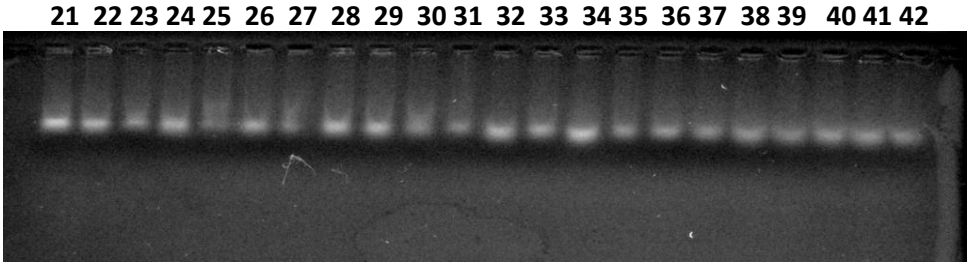
S/N	Sample I.D No.	Name	S/N	Sample I.D No.	Name
27	OY28	*S. macrocarpon* (Brown fruit)	39	CR40	*S. melongena* (long & white fruits; thorny)
28	OY29	*S. torvum*	40	CR41	*S. melongena* (long & purple fruits, no thorns, white flowers)
29	ED30	*S. melongena* (Purple fruit)	41	CR42	*S. melongena?* (long&purple fruits,thorny,white flowers)
30	ED31	*S. melongena* (Green fruit)	42	CR43	*S.indicum* subsp.*distichum* var. *distichum*
31	ED32	*S. macrocarpon* (Green fruit)	43	CR44	*S.melongena* (Round,purple white striped fruits, no thorns)
32	ED33	*S. gilo* Raddi (Green fruit)	44	CR45	*S. gilo* (Green,white striped fruits, & no thorns)
33	ED34	*S. dasyphyllum*	45	CR46	*S. indicum* subsp.*distichum* var. *distichum*
34	OD35	*S. macrocarpon* (Green fruit)	46	CR47	*S. melongena* (Purplish-white fruits with depression at the top&no thorns)
35	OD36	*S. melongena* (Green fruit)	47	CR48	*S. melongena* (Long purple fruits with white patches at the head & no thorns)
36	CR37	*S. torvum*			
37	CR38	*S. indicum* subsp. *distichum* var. *distichum*	48	CR49	*S. gilo* (Pure white fruits & no thorns)
38	CR39	*S. torvum*	49	CR50	*S. macranthum*

4.7 Molecular Characterization

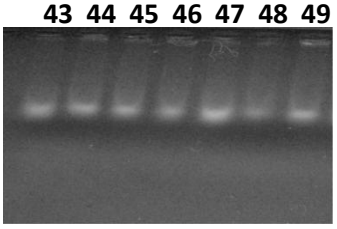
The quality of extracted DNA samples was determined on a 1% agarose gel electrophoresis before and after cleaning. The resulting electrophorogram of both the unclean samples and from 20 randomly selected clean samples are as shown in **Plate 20**:



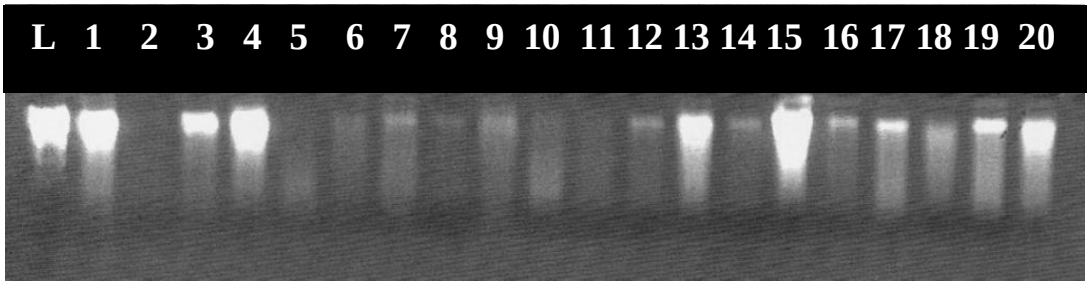
A



B



C



D

Plate 20: Electrophorogram of Extracted DNA Samples

Legend: (A- C) Before Cleaning (D) After Cleaning

4.7.1 Spectrophotometry

Spectrophotometer readings revealed the concentration of the DNA samples to range from 1 to 220ng/µl. The results (**Appendix 8**) showed that the DNA from most samples was good and highly concentrated. This could be because DNA was extracted from both fresh and freshly dried part of the samples collected. The purity of the DNA samples was equally measured at 260nm (**Figure 6**) and 280nm (**Figure 7**) wavelengths and the relative absorbance ratio ($A_{260/280}$) was equally determined. The ratio ranged from 1.06 to 1.91 (**Figure 8)**.

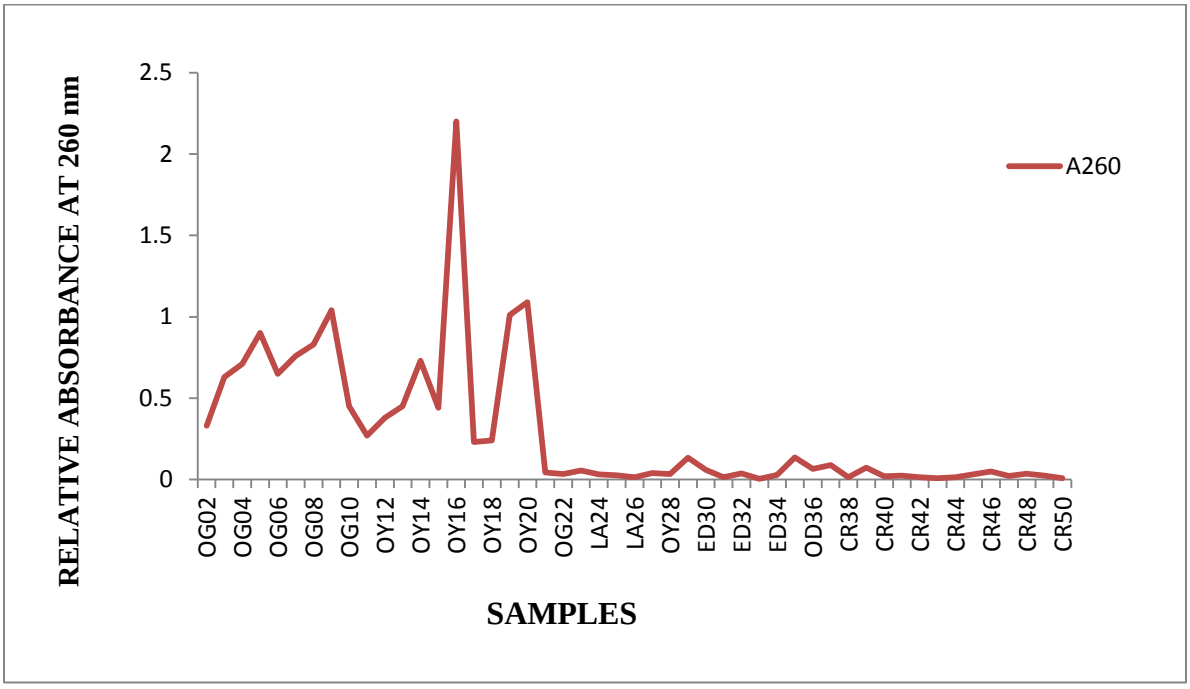


Figure 6: Relative absorbance of DNA samples of eggplant *Solanum* accessions at 260nm.

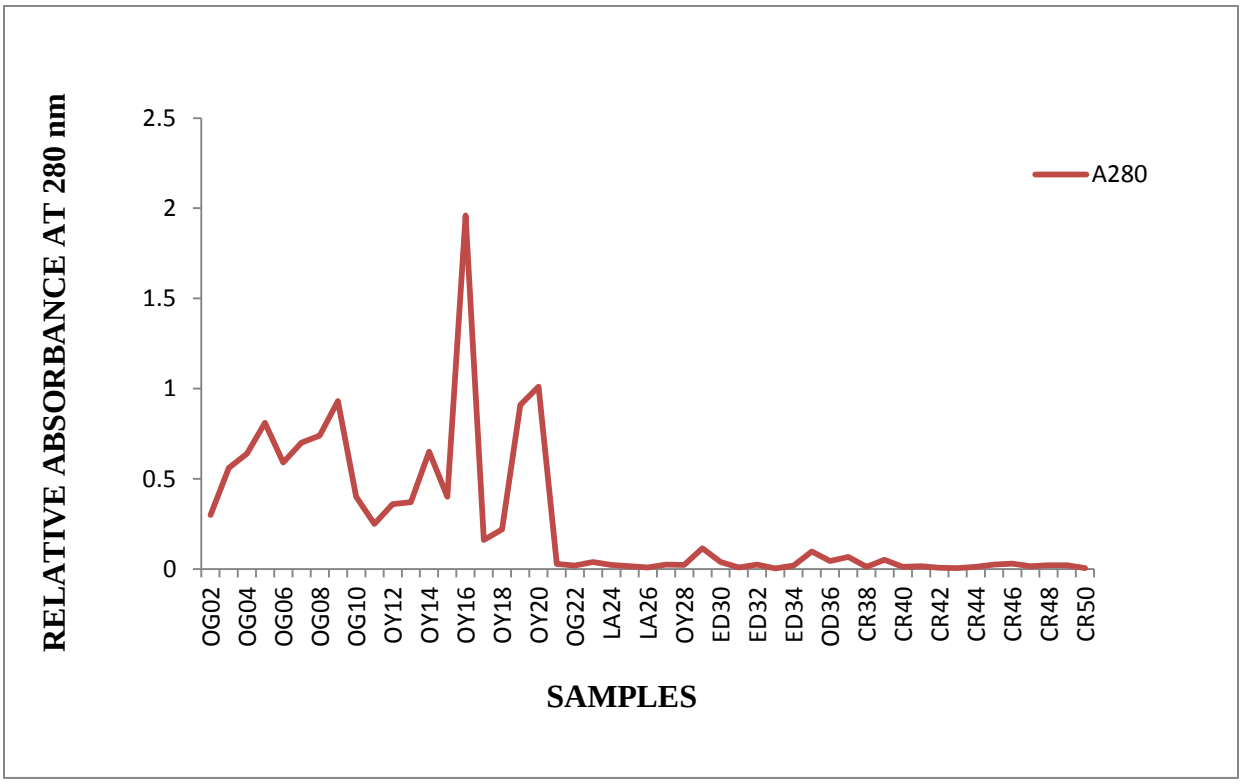


Figure 7: Relative absorbance of DNA samples of eggplant *Solanum* accessions at 280nm

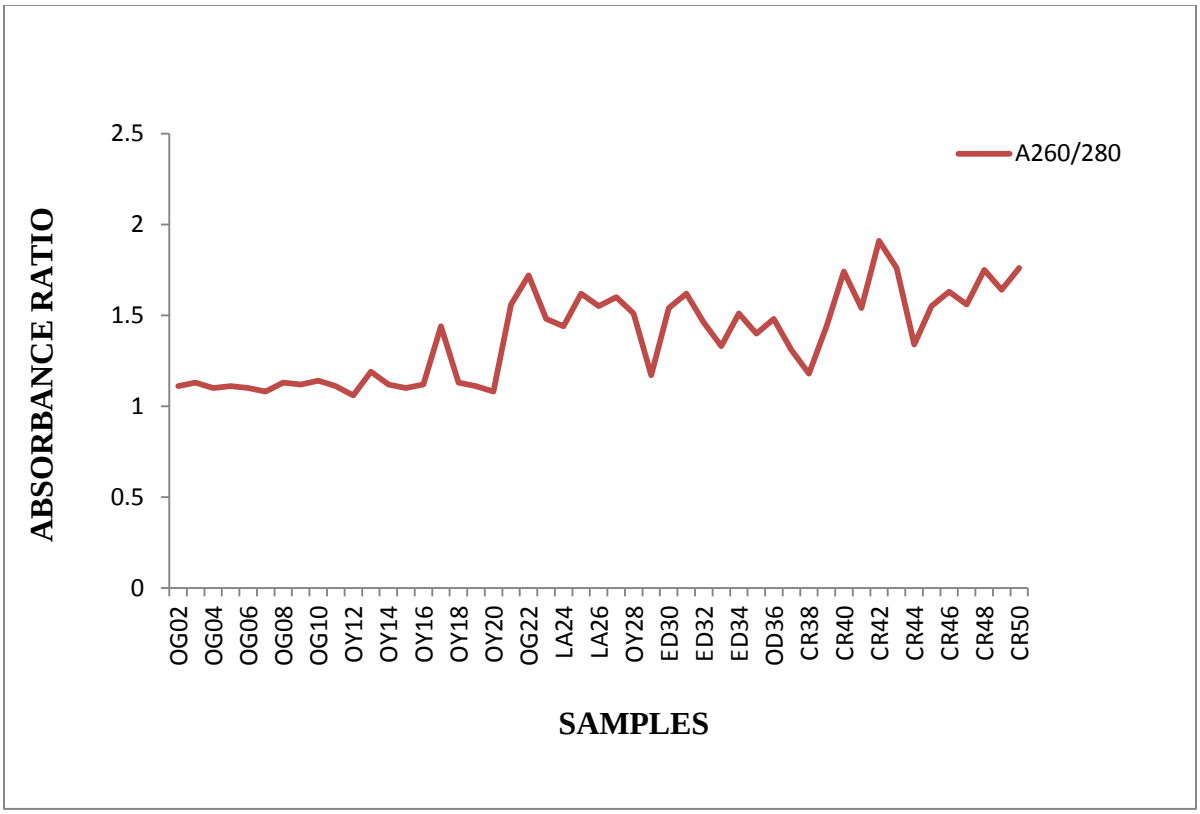


Figure 8: Relative absorbance Ratio ($A_{260/280}$) of DNA samples of eggplant *Solanum* accessions.

4.7.2 Polymerase Chain Reaction (PCR)

Amplification of the internal transcribed spacer (ITS) region of nuclear ribosomal DNA was carried out on all the samples. The amplified ITS gene included ITS2 – ITS5 and ITS3 – ITS4 regions. Agarose gel electrophoresis of the PCR products using different primers (**Table 11**) showed DNA with distinct bands. The electrophorogram of both regions are shown in **Plate 21** and **Plate 22** respectively.

Table 11: Different Primers used for PCR and their Sequences (according to White *et al*., 1990; Taberlet *et al*., 1991; and Baldwin *et al*., 1995).

Sequenced Region	Primer used	Primer sequence (5′- 3′)
ITS2 – ITS5	ITSleu1	(5'-GTC CAC TGA ACC TTA TCA TTT AG-3')
ITS3 – ITS4	ITS4	(5'- TCC TCC GCT TAT TGA TAT GC-3')
TrnF (between trnl C- trnl D)	UniC	(5'-CGAAATCGGTAGACGCTACG-3')
TrnL (between trnl E- trnl F)	UniF	(5'-ATTTGAACTGGTGACACGAG-3')

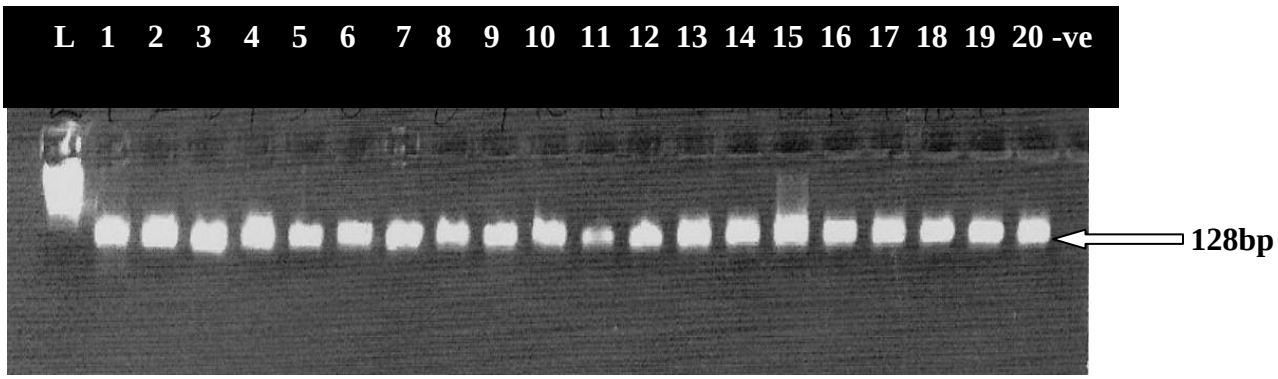


Plate 21: Electrophorogram of ITS2 – ITS5 amplification

Legend: L = 100bp DNA Ladder; -ve = Negative control; 1 – 20 represents amplicon from selected samples.

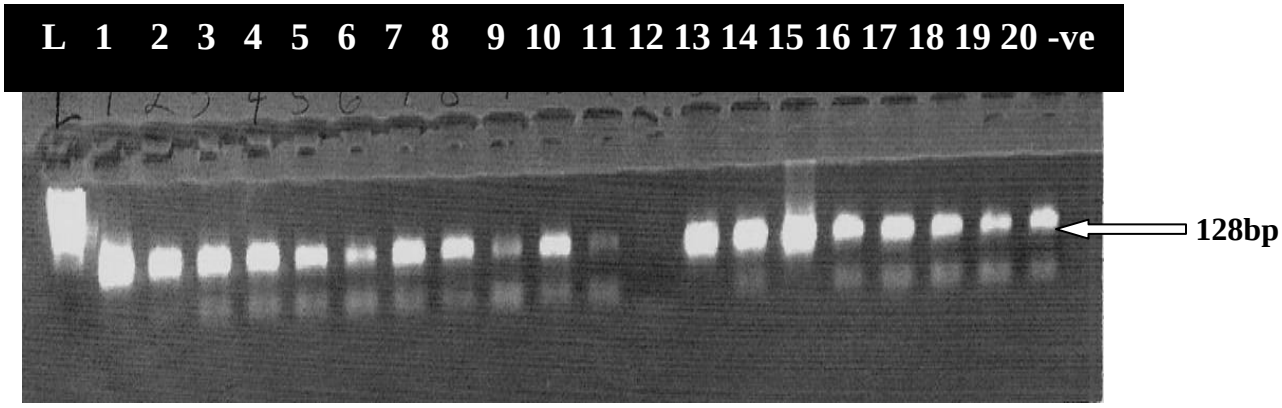


Plate 22: Electrophorogram of ITS3 – ITS4 amplification

Legend: L = 100bp DNA Ladder; -ve = Negative control; 1 – 20 represents amplicon from selected samples.

Samples 11 and 12 in the above electrophorogram (**Plate 22**) did not show clearly. They were re-cleaned and run again. **Plate 23** below shows their electrophorogram.

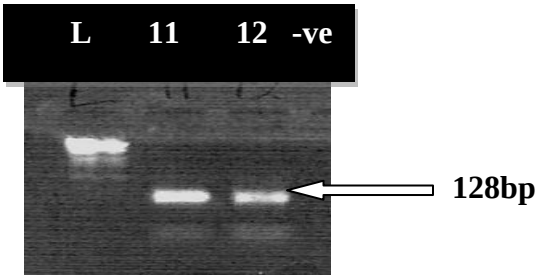


Plate 23: Electrophorogram of re-cleaned ITS3 – ITS4 amplification of Samples 11 and 12

Legend: L = 100bp DNA Ladder; -ve = Negative control

The non-coding cpDNA regions trnL-trnF spacer plus trnL intron are adjacent and are amplified separately. Primers used for these amplifications are also shown in **Table 11**. Note that trnL includes region between trnl C- trnl D and trnF includes region between trnl E – trnl F. The electrophorogram of both regions are shown in **Plates 24** and **Plate 25** respectively.

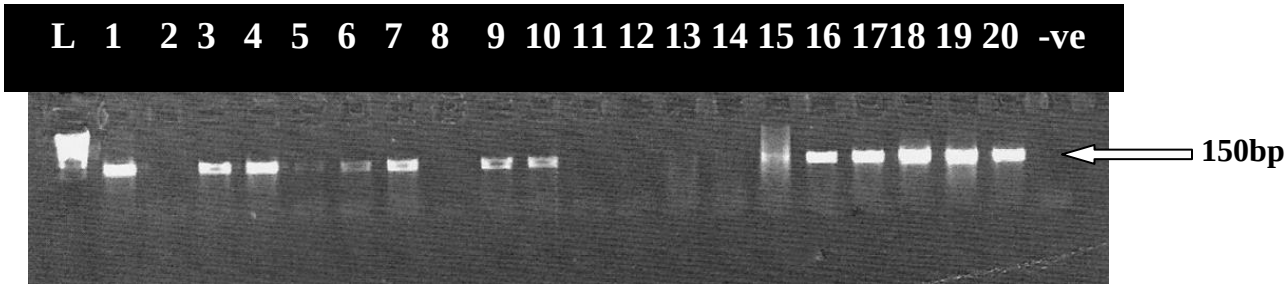


Plate 24: Electrophorogram of trnl C-trnl D Amplification

Legend: L = 100bp DNA Ladder; -ve = Negative Control; 1 – 20 represents amplicon from selected samples.

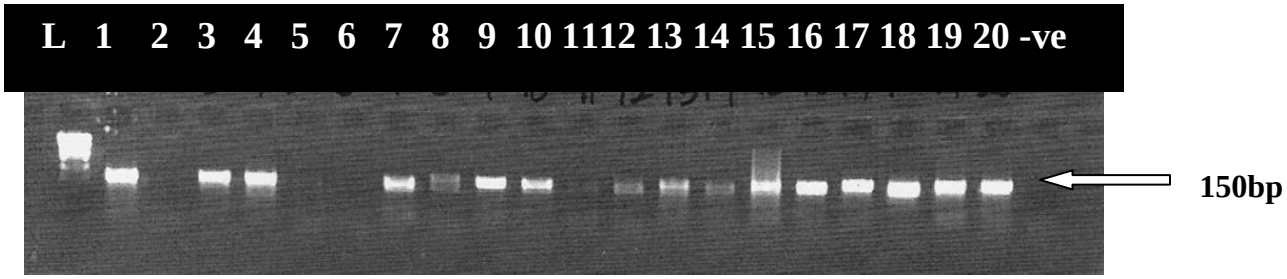


Plate 25: Electrophorogram of trnl E-trnl F Amplification

Legend: L = 100bp DNA Ladder; -ve = Negative Control); 1 – 20 represents amplicon from selected samples.

4.7.3 DNA Sequencing of both ITS and trnL-trnF spacer plus trnl intron regions.

The products of amplification of these regions (i.e. the amplicon) obtained for all samples were sequenced using the primers shown in **Table 12.**

Table 12: List of Primers used for Sequencing (according to White *et al*., 1990; Taberlet *et al*., 1991; and Baldwin *et al*., 1995).

Sequenced Region	Primer used	Primer sequence (5′- 3′)
ITS	ITS4	(5'- TCC TCC GCT TAT TGA TAT GC-3')
	ITS5HP	5'-GGA AGG AGA AGT CGT AAC AAG G-3').
trnL-trnF	UniC	(5'-CGAAATCGGTAGACGCTACG-3')
	UniF	(5'-ATTTGAACTGGTGACACGAG-3')

4.7.4 Basic Local Alignment Search Tool (BLAST) Results.

The result obtained by subjecting the nucleotide sequence obtained from the three plastid (non-coding trnL intron and intergenic spacer trnL-trnF) regions to BLAST is as shown in **Table 13**.

Table 13: BLAST Results and Percentage Maximum Identity

Sample NO.	Sample ID	Sample Name	% Query Coverage		% Maximum Identity	
			trnL	trnF	trnL	trnF
1	CR46	*S. indicum* subsp. *distichum* var. *distichum*	100	100	100 *S. aethiopicum*	88 *S. torvum*
2	CR47	*S. melongena*	100	76	100 *S. aethiopicum*	99 *S. melongena*
3	CR38	*S. indicum* subsp. *distichum* var. *distichum*	100	100	100 *S. aethiopicum*	98 *S. torvum*
4	CR50	*S. macranthum*	99	100	95 *S. crinitum*	98 *S. torvum*
5	OS21	*S. torvum*	99	-	99 *S. torvum*	-
6	ED31	*S. melongena*	99	-	99 *S. aethiopicum*	-
7	ED32	*S. macrocarpon*	100	99	99 *S. macrocarpon*	99 *S. torvum*
8	OY29	*S .torvum*	100	99	99 *S. dasyphyllum*	99 *S. torvum*
9	ED30	*S. melongena*	100	89	99 *S. melongena*	99 *S. melongena*
10	OY27	*S. melongena*	100	89	100 *S. melongena*	99 *S. melongena*
11	CR37	*S .torvum*	100	36	98 *S. anguivi*	100 *S. torvum*
12	CR39	*S .torvum*	-	100	-	97 *S. torvum*

Table 13 (Contd.): BLAST Results and Percentage Maximum Identity.

Sample NO.	Sample ID	Sample Name	% Query Coverage		% Maximum Identity	
			trnL	trnF	trnL	trnF
13	OY17	*S. aethiopicum*	-	89	-	99 *S. aethiopicum*
14	OY19	*S. melongena*	94	-	99 *S. melongena*	-
15	OG04	*S. dasyphyllum*	99	89	99 *S. dasyphyllum*	99 *S. dasyphyllum*
16	OY18	*S. scabrum*	99	77	99 *S. nigrum*	97 *S. scabrum*
17	LA24	*S. gilo*	100	77	99 *S. aethiopicum*	97 *S. scabrum*
18	OG22	*S. melongena*	100	97	99 *S. aethiopicum*	96 *S. aethiopicum*
19	LA23	*S. gilo*	99	98	96 *S. aethiopicum*	92 *S. aethiopicum*
20	LA25	*S. gilo*	99	89	99 *S. aethiopicum*	99 *S. aethiopicum*

4.7.5 Phylogenetic estimation using Maximum Likelihood (PhyML)

Nucleotide sequences generated from Sequencing of ITS region and trnL-trnF spacer plus trnl intron were analyzed with Seaview4 software to draw the phylogenetic tree using the Maximum Likelihood (PhyML) option of the software. The resulting phylogenetic tree is shown in **Figure 9**.

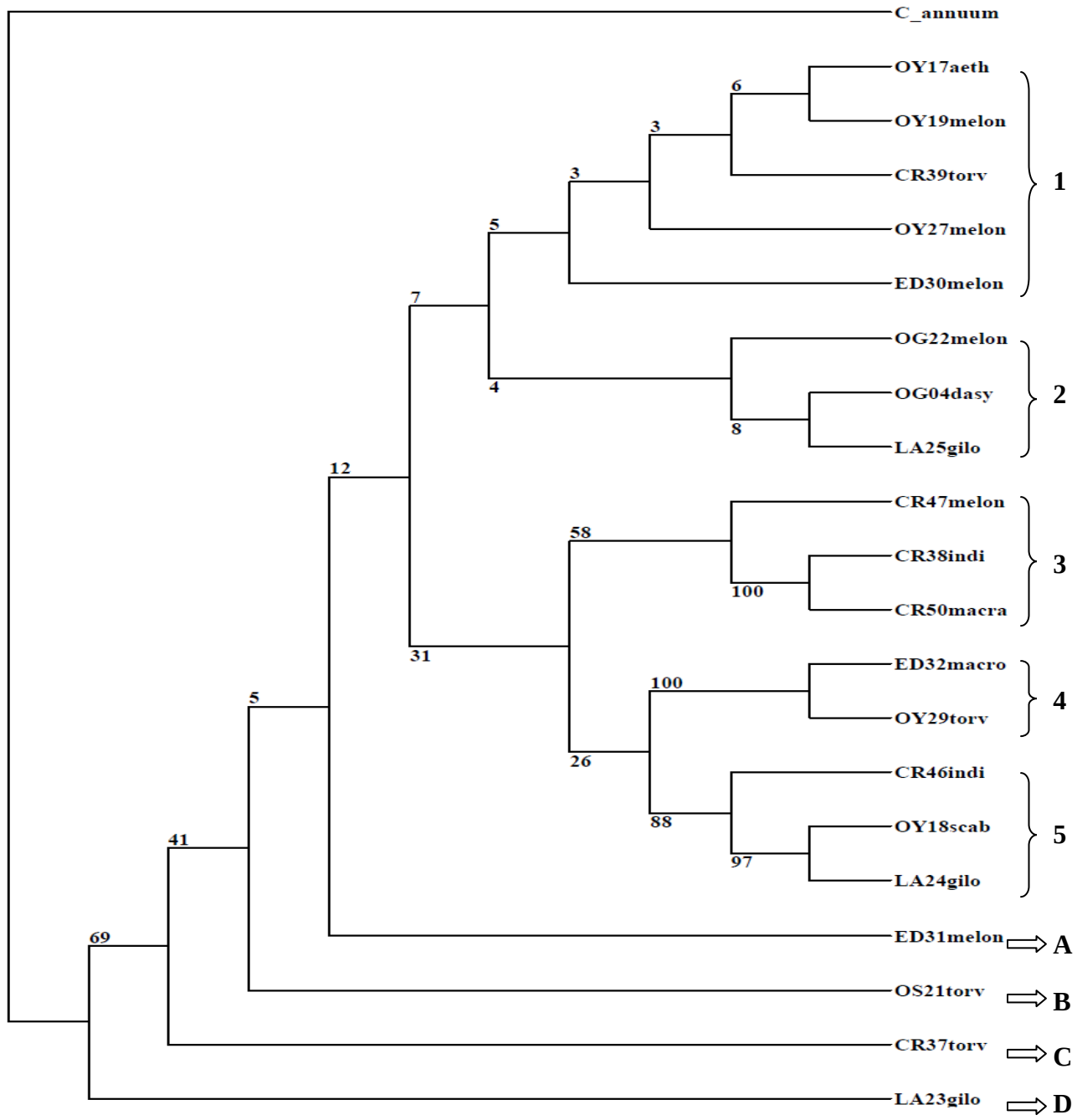


Figure 9: Phylogenetic Relationships within eggplant *Solanum* using *Capsicum annuum* L. as the outgroup.

Legend: Bootstrap supports are indicated above branches; Five clusters and four ungrouped (A, B, C and D) samples are also noticeable.

The figure shows five major clusters (1 to 5) and four ungrouped [A to D, corresponding to *S. gilo* (LA23), *S. torvum* (OS21 and CR37) and *S. melongena* (ED31) respectively]. Cluster 1 contains highest number with five accessions while Cluster 4 is the lowest with two samples. Each of the remaining three clusters contained three accessions respectively. The highest bootstrap value of 100 was observed between *S. macrocarpon* and *S. torvum*, and between *S. macranthum* and *S. indicum*. This was closely followed by a value of 97 between *S. scabrum* (OY18) and *S. gilo* (LA24). However, low bootstrap values were observed among those found in clusters 1 and 2.

CHAPTER FIVE

5.0 DISCUSSION OF RESULTS

The ability to characterize morphological diversity is indispensable for effective management and sustainable use of eggplant genetic resources in breeding programmes. Primary characterization involves measuring simple plant characters that can be easily recorded through visual observations at different plant growth stages (leaf area, fruit shape, size and colour, plant prickliness and hairiness). Secondary characterization, as said earlier, deals with more complicated morphological traits of agronomic importance such as pest and disease resistance, yield potential, biochemical properties. Primary characterization was employed in this study to measure morphological plant characters at flowering and/or fruiting stage during which further growth has been temporarily suspended. Morphological descriptors used here for the collection and measurement followed IPGRI/FAO (1996) (now known as Bioversity International) standard.

A simple classification of eggplant collections into different cultivar groups is necessary for promoting their use in crop improvement and conservation. According to Kumar *et al*., (2008), one way to promote use is to develop a simple classification system based on key morphological characters such as fruit shape and colour that will uncover the pattern of variation in eggplant landraces. In many crops, simple and useful systems of classification have been developed that rely on only a few simply inherited and easily observable traits (Riley *et al*., 1996). This would allow curators to focus on characterizing a greater portion of their collections for these key traits.

A wide range of variation for most descriptors studied was found in the collection studied. The results obtained in the present investigation revealed wide morphological diversity among the 49 samples for almost all the thirty seven quantitative and qualitative morphological descriptors used. Inflorescence, leaf and fruit characters vary considerably among all the samples. This is in

agreement with findings elsewhere (e.g. Prohens *et al*., 2005). The grouping together in **Figure 4c** of *S. gilo* and *S. melongena* in Cluster 9 clearly indicates closeness between *S. melongena* and *S. gilo. S. macrocarpon* and *S. dasyphyllum* were grouped with each other in Cluster 1, an indication that they are only distantly related to cultivated type (*S. melongena*) and are more closely related to each other. This is in contrast to the findings of Naujeer (2009) who observed *S. macrocarpon* accessions occurring in the same cluster with *S. melongena* indicating their close genetic affinities and phenotypic relationship. *S. indicum* is grouped with *S. gilo* in Cluster 2 and also clustered with *S. torvum* in Cluster 5. This is an indication of relatedness of *S. gilo* with *S. indicum* and *S. torvum* whereas *S. indicum* and *S. torvum* are distantly related. The occurrence of all accessions belonging to *S. nigrum* and *S. scabrum* in Cluster 10 shows a very close relatedness between them. One accession of *S. torvum* also occurs in the same cluster with *S. erianthum* and *S. incanum* (Cluster 3), indicating closeness among them.

It could be observed that *S. gilo* is the only species that showed close relatedness with cultivated *S. melongena* in this study. All others are only distantly related (if at all) with it. The occurrence of all accessions except one of *S. melongena* in three clusters (Clusters 6, 7 and 8) shows that they all belong to the same species irrespective of the geographic location of collection. The same could equally be said of most accessions of *S. gilo* occurring exclusively in Cluster 4. The observed *S. macranthum* not grouped with any of the other accessions is a clear indication of its non-relatedness or its distantly relatedness with all others. The reason for one accession of *S. gilo* (CR45) not grouped with either other *S. gilo* accessions or any other accession is not yet known. It may even be a different species that was wrongly identified as *S. gilo*.

The results obtained in this study revealed wide morphological diversity among the 49 accessions for all the thirty seven quantitative and qualitative morphological descriptors analyzed. This result

showed the diversity among different groups and within the same species. Plant's stem/branches, leaf, inflorescence/ flower and fruit characters vary considerably among accessions. Although the 13 accessions of *S. melongena* studied are characterized by wide diversity in stem/branches, leaf, inflorescence/ flower and fruit characters, their overall close morphology allows them to be grouped together in distinct clusters (Clusters 6, 7 and 8 in **Figure 4c**). This observation agrees with the report of Furini and Wunder (2004).

The level of polymorphism observed in the present study is high when both qualitative and quantitative data were combined, indicating a wide and diverse genetic base. The correlation coefficient 0.75 for the highest similarity between genotypes and the least 0.34 displayed a good separation from a conserved region of the genome. These results are in agreement with those earlier obtained by other workers (e.g. Furini and Wunder, 2004; Singh *et al*., 2006; Sifau *et al*., 2009). However, the results differ from those workers who studied variation among the cultivated and weedy taxa of *S. melongena* by allozymes (Isshiki *et al*., 1994) and RAPD (Hassan and Lester, 1990; Karihaloo and Gottlieb, 1995) analyses. These authors observed little genetic variability among the genotypes studied and suggested the existence of a very small gene pool from which the cultivated forms arose.

Interestingly, collections originating from various parts of the study area did not form well-defined distinct groups and were interspersed with each other. This clustering together of accessions from different areas of collection as observed in this study implies no association between morphological variation and the geographic origin of accessions. This supports the findings of Singh *et al*., 2006, who equally observed no association between RAPD pattern and the geographic origin of accessions. According to Furini and Wunder (2004), diverse geographic origin of two

accessions may not necessarily reflect in genetically diverse plant materials although these parameters are central in genetic diversity studies. Frary *et al*.(2003) had explained this observation by the fact that the phenotypes for certain traits (leaf and fruit characters) are controlled by a limited number of genes with major effects on phenotypic traits and their quantitative trait loci are conserved during domestication and plant evolution.

PCA multivariate analysis reveals that fruit traits, both qualitative (shape, size and colour) and quantitative (length, width and mean fresh weight having coefficient of variation 68.11%, 53.03% and 82.75% respectively) with a cumulative total variation of 67.96% most effectively discriminated between *S. melongena* and their related species. These traits can be used as important marker traits in the classification of *S. melongena* and other *Solanum* accessions found in Nigeria into specific cultivar groups. This finding corroborates the work of Kumar *et al*., (2008), who stated that fruit colour, size and shape are the most distinctive characters that vary between the cultivated *Solanum* species and their wild types. Samples OG09 (*S. erianthum*) and CR50 (*S. macranthum*) which are both trees in nature and labelled 8 and 49 respectively in **figures 5a** and **b** (**Table 10**) occur separately without clustering with any other samples. This had earlier been observed in figure **4b** thus confirming their distantly relatedness to *S. melongena* and other *Solanum* samples.

However, morphological diversity observed in all the accessions is not restricted to fruit characters only. Other vegetative, inflorescence/ flower and fruit traits such as leaf blade lobing, flower colour, leaf blade length, leaf blade width, plant height, stem length and internode length are equally identified as useful marker traits that also distinguished between eggplant accessions and their related species and wild types. As discussed earlier, a high level of morphological diversity in

vegetative, inflorescence and fruit traits was manifested in the 13 samples of *S. melongena* and related species characterized. Naujeer (2009) citing Portis *et al*., (2006) who worked on Italian pepper landraces suggests that independent selections pressures across a number of crop generations to a restricted gene pool can result in genetic divergence within a particular landrace. This could also explain the phenotypic variation manifested in plant characters such as fruit traits among individuals within the different accessions studied. Mace *et al*., (1999) reported a high degree of morphological plasticity in *S. melongena* and their related species which may have further contributed in the differentiation process and accumulation of variation in eggplant landraces and traditional varieties.

The method of plant collection and duration of drying the material is important for the survival of the DNA (Savolainen *et al*., 1995), the shorter the duration of drying of plant specimen the better the quality of the DNA obtained therefrom. Samples used in this study were collected fresh from the field and dried gradually with silica gel over a short period of time. This accounted for the high quality of the DNA obtained from most samples recorded in this study. More than half of the DNA samples had relative absorbance ratio of 1.30 – 1.90, which was good enough to proceed to Polymerase Chain Reaction (PCR) after cleaning.

The combination of several markers from both nuclear and plastid genomes as well as coding and non-coding regions improved the resolution of phylogenetic relationships within the family. Several mutations occurring in flanking regions of widely used plastid and nuclear regions such as matK (Harrington *et al*., 2005) and ITS (Edwards and Gadek, 2001) made the amplification of molecular markers in *Solanum* difficult. This further added to the challenge the genus is facing at the taxonomic level. These mutations complicate the compilation of multilocus data sets without missing any data. Maximizing taxa and markers representation to provide a reliable phylogenetic

hypothesis inferred from nuclear and plastid genomes is required to propose a new classification for eggplant family (Weese and Bohs, 2007). The inclusion of missing data was widely recognized as a major drawback in phylogenetic analyses during the early 90s (Wiens and Reeder, 1995). However, simulations and empirical analyses as reported by Kumar *et al*., (2001), Wiens (2003) and Phillipe *et al*., (2004) have shown that taxa comprising high levels of missing data could be accurately placed in phylogenies. Moreover, adding incomplete taxa to a phylogenetic analysis was even shown to improve the accuracy of a given topology, e.g. by subdividing misleading long branches (Wiens, 2005, 2006).

Molecular analysis in this study was based on a combination of one nuclear (ITS region- ITS1, 5.8S, ITS2) and two plastid (non-coding trnL intron and intergenic spacer trnL-trnF) markers. Coding plastid regions have proven to be useful in addressing phylogenetic relationships at higher taxonomic levels (Clayton, 2007; Muellner *et al*., 2007), whereas non-coding regions (introns and intergenic spacers) were shown to be more useful at lower taxonomic ranks (Baldwin, 1992; Soltis and Soltis, 1997). The combination of ITS and trnL-trnF spacer plus trnl intron gene regions resulted in a well resolved phylogenetic hypothesis, with results strongly suggesting that most of the *Solanum* accessions in this study are monophyletic (i.e. with common ancestor). This finding is in agreement with the findings of Olmstead and Palmer (1997); and Levin *et al*., (2005), who also found out that majority of *Solanum* taxa comprise a monophyletic lineage, though some taxa are polyphyletic, but in sharp contrast with Weese and Bohs (2007) who, based on their findings, suggested that most traditionally recognized *Solanum* subgenera are not monophyletic. Of the three gene regions, trnL-trnF spacer was especially useful for phylogenetic inference, with both a high percentage of parsimony-informative sites as well as a low level of homoplasy. This is however, in contrast with the findings of Levin *et al*., (2005) who found the granule-bound starch synthase gene (GBSSI) or waxy as being useful for phylogenetic inference in *Solanum* taxa. The

observed highest bootstrap value of 100 between *S. macrocarpon* (ED32) and *S. torvum* (OY29), and between *S. macranthum* (CR50, which did not group with any accession when morphological data was analyzed) and *S. indicum* (CR38), followed closely by a value of 97 between *S. scabrum* (OY18) and *S. gilo* (LA24) is an indication of a close relationship among those species concerned and possibility of having a common ancestor. The observed lower bootstrap values in clusters 1 and 2 showed that, though members may have a common ancestor, each one has diverged out and evolved in such a way that they share little or no resemblance with each other. It should be noted as well that *S. torvum* separated out from *S. melongena*, and even where they grouped together they have a very low bootstrap value of 3 (the lowest), an indication of distant relatedness. This corroborates the discovery of Mace *et al*., (1999); Furini and Wunder, (2004) and Isshiki *et al*., (2008) who also found out that *S. torvum* section Torva separated out from section Melongena and other related *Solanum* species.

There are situations whereby results from molecular study disagree with that of morphological analysis. For example, molecular analysis revealed a high bootstrap value between *S. macranthum* and *S. indicum*, which shows that they are closely related but in morphological characters, *S. macranthum* remained ungrouped, meaning that, it is distantly related to all other accessions. Other cases of disagreement that were equally observed are those between *S. scabrum* and *S. gilo* on one hand and *S. torvum* and *S. macrocarpon* on the other. These samples did not group with each other in morphological analysis but grouped together in molecular analysis with high bootstrap values of 97% and 100% respectively. Therefore, molecular analysis was able to show that they are related which was not possible with morphological analysis. However, there were other cases whereby observations in morphological analysis were equally made in molecular analysis. That is, results obtained using morphological parameters were confirmed by molecular analysis. For instance, *S. torvum* separated out from *S. melongena* in both morphological and molecular analyses. *S. dasyphyllum* also separated out from *S. gilo* in morphological analysis but grouped together with a

very low bootstrap value of 8% in molecular analysis. The implication in both cases is that they are distantly related.

The result obtained from BLAST confirmed some earlier authenticated names of samples based on morphological characters on one hand, and disagreed with some previous nomenclature on the other. For example, as shown in **Table 13**, names of samples labelled OG04, OY17, OY19, OS21, OY27, ED30, ED32, and CR39 were molecularly confirmed based on sequenced data. Similarly, molecular sequenced data was able to correctly identify both samples labelled OG22 and CR47 as *S. aethiopicum* which were earlier wrongly identified both as *S. melongena* based on their morphology. However, more sequence data especially from genes (e.g. GBSSI, matK, rbcL) that are known for phylogenetic inference are needed for some samples before their names based on morphological data could be molecularly confirmed. For example, names of samples OY18, LA24, ED31, CR38, CR46 and CR50 could not be confirmed molecularly because sequenced regions in them (ITS and trnL-trnF) are not informative enough to authenticate them. In most previous studies, *S. aethiopicum* was mostly used as synonymous to *S. gilo* (e. g. Sękara *et al*., 2007). That could be the probable reason why sequenced regions of samples LA23 and LA25 which are both *S. gilo* aligned well with sequenced data of *S. aethiopicum* obtained from NCBI data base.

The combination of molecular markers and morphological characterization as used in this study has greatly helped in cultivar identification and clarification of the phylogenetic affinities of the large and complex genus *Solanum*. As a result, the combination was able to solve the taxonomic problem of the genus to a large extent as found in Southern Nigeria and by extension in the country as a whole. This high level of intra and inter specific variations displayed within eggplant accessions and between its relatives as reported in this study could be effectively used in genetic improvement of cultivated eggplant varieties as well as *in-situ* and *ex-situ* conservation.

CHAPTER SIX

6.0 SUMMARY OF FINDINGS/CONCLUSIONS.

This is presented below:

OBJECTIVES	SUMMARY OF FINDINGS
(1) To explore the variation in germplasm of different species of vegetable *Solanum* in Southern Nigeria, with emphasis on the collections, identification, documentation and preservation of all voucher specimens in secure repositories.	(1) The results obtained in the present investigation revealed wide morphological variation among the samples studied for almost all the thirty seven morphological descriptors studied. These samples having been identified and authenticated were kept at both LUH and FHI. However, no association between morphological variation and the geographic origin of accessions was observed.
(2) To determine the level of genetic diversity among individuals of different and same species using both morphological and DNA-based markers.	(2) The level of polymorphism (both inter- and intra-specific genetic variability) observed in the present study is high, indicating a wide and diverse genetic base. This was confirmed by sequences from trnL-trnF gene region.

(3) To establish phylogenetic relationships among the taxa studied using both morphological and molecular data obtained**.**	(3) Phylogenetic analysis based on combined morphological characters with molecular data from trnL intron, trnL spacer and ITS sequences provides strong support for the monophyly of the genus. This was made possible when *Capsicum* was included as an outgroup (although relationships among subgenus are still weakly supported).
(4) To identify agronomic marker traits useful for classification of the taxa using Principal Component Analysis.	(4)Principal Component Analysis (PCA) multivariate analysis revealed that fruit traits (shape, size and colour) with a cumulative total variation of 67.96% most effectively discriminated between *S. melongena* and their related species. These traits can be used as important agronomic traits in the classification of *S. melongena* and other vegetable *Solanum* accessions found in Nigeria into specific cultivar groups.

6.1 Contributions to Knowledge

The study has contributed to knowledge in the following ways:

1. The study discovered two additional *Solanum* species which hitherto not listed among members found in Nigeria e.g. *S. macranthum* and *S. incanum*; the authenticated voucher specimens of all collected vegetable *Solanum* have been deposited at both the University of Lagos Herbarium (LUH) and Forestry Herbarium Ibadan (FHI) for reference purposes – a major contribution to germplasm conservation.
2. The thesis produced the first report on the position of Nigerian species of eggplant using molecular data and identified fruit traits (such as shape, size and colour), leaf and inflorescence/ flower as useful agronomic traits that distinguished between eggplant accessions and their related species – a major contribution to Plant Systematics.
3. The study provided species database of vegetable *Solanum* in Nigeria and by extension in Africa with emphasis on variation patterns – a major contribution to global biodiversity information system.
4. The genomic DNA samples which were produced have been deposited at the DNA Bank of the Royal Botanic Gardens Kew, London – this serves as background material for the conservation of *Solanum* in Nigeria.

REFERENCES

Allard, J. (1996). Eggplant in Japan. *PHM Revue Horticultural* **374**: 55-56.

Amar, Z. (2000). Agricultural Produce in the Land of Israel in the Middle Ages. Yad Izhak Ben-Zvi, P, Jerusalem. 377pp.

Anderson, G. J., and Symon, D. E. (1989). Functional Dioecy and Andromonoecy in *Solanum*. *Evolution* **43**: 204-219.

Aubert, S., Daunay, M. C. and Pochard, E. (1989). Steroidal Saponins of Eggplant (*Solanum melongena* L.). Interest in Food Analysis Methodology, Localization in the Fruit. *Agronomy* **9**: 641-651.

Ayad, W. G., Hodgkin, T., Jaradat, A. and Rao, V. R. (1995). Molecular Genetic Techniques for Plant Genetic Resources. Report of an IPGRI Workshop, International Plant Genetic Resources Institute, Rome, Italy. 256pp.

Baksh, S., (1979). Cytogenetic Studies of the F1 Hybrid of *Solanum incanum* L. x *S. melongena* L. Variety ‘‘Giant of Benares’’ *Euphytica* **28**: 793–800.

Baldwin, B. G. (1992). Phylogenetic Utility of the Internal Transcribed Spacers of Nuclear Ribosomal DNA in Plants: an Example from the Compositae. *Molecular Phylogenetics and Evolution* **1**: 3- 16.

Baldwin, B. G., Sanderson, M. J., Porter, J. M., Wojciechowski, M. F., Campbell, C. S. and Donoghue, M. J. (1995). The ITS region of Nuclear Ribosomal DNA: a Valuable Source of Evidence on Angiosperm Phylogeny. *Annals of the Missouri Botanical Garden* **82**: 247– 277.

Behera, T.K., P. Sharma, B.K. Singh, G. Kumar, R. Kumar, T. Mohapatra, and N.K. Singh. (2006). Assessment of genetic diversity and species relationships in eggplant (*Solanum melongena* L.) using STMS markers. *Scientia Horticulturae* **107**: 352-357.

Bell, A. D., and Dines, T. D. (1995). Branching Patterns in the Solanaceae. In: Hoch, P. C. and Stephenson, A. G. (Eds.). Experimental and Molecular Approaches to Plant Biosystematics. Missouri Botanical Garden, St. Louis, MO. 271pp.

Bennett, M. D. and Leitch, I. J. (2004). Plant DNA C-Values Database (release 3.0, Dec 2004). http://www.rbgkew.org.uk/cval/ homepage.html. Assessed 10th June 2010.

Bharathan, G., Goliber, T. E., Moore, C., Kessler, S., Pham, T. and Sinha, N. R. (2002). Homologies in Leaf Form Inferred from KNOXI Gene Expression during Development. *Science* **296**: 1858- 1860.

Bohs, L. (2001). Revision of *Solanum* Section *Cyphomandropsis* (Solanaceae). *Systematic Botany Monographs* **61**:1–85.

Bohs, L. (2005). Major Clades in Solanum Based on ndhF Sequence Data. In: Keating, R. C., Hollowell, V. C. and Croat, T. B. (Eds.). A festschrift for William G. D'Arcy: the legacy of a taxonomist. *Monographs in Systematic Botany from the Missouri Botanical Garden*, St. Louis, MO **104:** 27-49.

Bohs, L. and Olmstead, R. G. (1997). Phylogenic Relationship in *Solanum* (Solanaceae) Based on ndhF Sequences. *Systematic Botany* **22:**5–17.

Bohs, L. and Olmstead, R. G. (1999). *Solanum* Phylogeny Inferred from Chloroplast DNA Sequence Data. In: Nee, M., Symon, D. E., Lester, R.N. and Jessop, J. P. (Eds.). *Solanaceae IV: Advances in Biology and Utilization*. Royal Botanical Gardens, Kew. 545pp.

Bohs, L. and Olmstead, R. G. (2001). A Reassessment of Normania and Triguera (Solanaceae). *Plant Systematics and Evolution* **228:**33–48.

Bremer, B., Bremer, K., Heidari, N., Erixon, P., Olmstead, R. G., Anderberg, A. A., Källersjö, M., and Barkhordarian, E. (2003). Phylogenetics of Asterids based on 3 Coding and 3 non-coding Chloroplast DNA Markers and the Utility of Non-coding DNA at Higher Taxonomic Levels. *Molecular Phylogenetics and Evolution* **24**:274-301.

Brown, K. S., Jr. (1987). Chemistry at the Solanaceae/Ithomiinae Interface. *Annals of Missouri Botanical Garden* **74**: 359-397.

Buchmann, S. L. (1983). Buzz Pollination in Angiosperms. In: Jones, C. E. and Little, R. J. (Eds.). Handbook of Experimental Pollination Biology. New York: Van Nostrand Reinhold Co., Inc. 233pp.

Bukenya, Z. R. and Carasco, J. F. (1995). *Solanum* (Solanaceae) in Uganda. *Bothalia* (**25**)**1**:43–59.

Bukenya, Z. R., and Carasco, J. F. (1999). Ethnobotanical aspects of *Solanum* L. (Solanaceae) in Uganda. In: Nee, M., Symon, D. E., Lester, R. N. and Jessop, J. P. (Eds.), Solanaceae IV: Advances in Biology and Utilization. Royal Botanical Gardens, Kew. 545pp.

Cao, G., Sofic, E. and Prior, R. L. (1996). Antioxidant Capacity of Tea and Common Vegetables. *Journal of Agricultural and Food Chemistry* **44:** 3426–3431.

Child, A. and Lester, R. N. (2001). Synopsis of the Genus *Solanum* L. and its Infrageneric Taxa. In: van der Berg, R. G., Barendse, G. W. M, van der Weerden, W. M., Mariani, C. (Eds.). Solanaceae V: advances in taxonomy and utilization. Nijmegen University Press, The Netherlands.155pp.

Chowdhury, B. (1976). Vegetables. National Book Trust. 4th edn. New Delhi, India. 206pp.

Chowdhury, B. (1995). Eggplant. In: Smartt, J. and Simmonds, N.W. (Eds.). *Evolution in Crop Plants*. 528 pp.

Cipollini, M. L., Bohs, L. A., Mink, K., Paulk, E., and Böhning-Gaese, K. (2002) Patterns of Secondary Compounds within Fleshy Fruits: Ecology and Phylogeny. In: Levey, D. J., Silva, W. R. and Galetti, M. (Eds.). *Seed Dispersal and Frugivory: Ecology, Evolution and Conservation*. CABI Publishing, Wallingford, Oxfordshire, UK. 178pp.

Clayton, J. W. (2007). Molecular Phylogeny of the Tree-of-Heaven Family (Simaroubaceae) Based on Chloroplast and Nuclear Markers. *International Journal of Plant Science* **168**:1325 – 1339.

Collonnier, C., Fock, I., Kashyap, V., Rotino, G.L., Daunay, M.C., Lian, Y., Mariska,I.K., Rajam, M.V., Servaes, A., Ducreux, G., and Sihachakr, D. (2001). Applications of Biotechnology in Eggplant. *Plant Cell, Tissue and Organ Culture* **65:**91-107.

D'Arcy, W.G. (1972). Solanaceae Studies II: Typification of Subdivisions of *Solanum*. *Annals of the Missouri Botanical Garden* **59:** 262-278.

D'Arcy, W. G. (1979). The Classification of the Solanaceae. In: Hawkes, J. G., Lester, R. N. and Skelding, A. D. (Eds.). *The Biology and Taxonomy of the Solanaceae*. Academic Press, London. 738pp.

D'Arcy, W. G. (1991). The Solanaceae Since 1976, with a Review of its Biogeography. In: Hawkes, J. G., Lester, R. N., Nee, M., Estrada, N. (Eds), *Solanaceae III: Taxonomy, Chemistry and Evolution*. Royal Botanic Gardens, Kew, Richmond, UK. 653pp.

Daly, D. E., Cameron, K. M., Stephenson, D. W. (2001). Plant Systematics in the Age of Genomics. *Plant Physiology* **127:** 1328–1333.

Daunay, M. C. and Lester, R. N. (1988). The Usefulness of Taxonomy for Solanaceae Breeders, with Special Reference to the Genus *Solanum* and to *Solanum melongena* L. (Eggplant). *Capsicum Newsletter* **7:** 70–79.

Daunay, M. C., Lester, R. N. and Laterrot, H. (1991). The Use of Wild Species for the Genetic Improvement of Brinjal Eggplant (*Solanum melongena*) and Tomato (*Lycopersicon esculentum*). In: Hawkes, J. G., Lester, R. N., Nee, M., Estrada, N. (Eds.). *Solanaceae III: Taxonomy, Chemistry and Evolution.* Royal Botanic Gardens, Kew, Richmond, UK. 653pp.

Daunay, M. C., Dalmon, A., and Lester, R. N. (1999). Management of a Collection of *Solanum* Species for Eggplant (*Solanum melongena*) Breeding Purposes. In: Nee, M., Symon, D. E., Lester, R. N. and Jessop, J. P. (Eds.). *Solanaceae IV: Advances in Biology and Utilization*. Royal Botanic Gardens, Kew. 545pp.

Daunay, M. C., Lester, R. N. and Ano, G. (2001a). Cultivated Eggplants in Tropical Plant Breeding. 2nd Edition, Oxford University Press, Oxford. 225pp.

Daunay, M. C., Lester, R. N., Gebhardt, C., Hennart, J. W., Jahn, M., Frary, A. and Doganlar, S. (2001b). Genetic Resources of Eggplant (*Solanum melongena* L.) and Allied Species: a New Challenge for Molecular Geneticists and Eggplant Breeders. In: van den Berg, R. G., Barendse, G. W. M., van der Weerden, G. M., Mariani, C. (Eds.). *Solanaceae V: Advances in Taxonomy and Utilization*. Nijmegen University Press, Nijmegen, The Netherlands. 354pp.

Daunay, M. C., Aubert, S., Frary, A., Doganlar, S., Lester, R. N., Barendse, G., van der Weerden, G., Hennart, J. W., Haanstra, J., Dauphin, F. and Jullian, E. (2004). Eggplant (*Solanum melongena*) Fruit Colour: Pigments, Measurements and Genetics. In: *Proceedings of the 12th Eucarpia Meeting on Genetics and Breeding of Capsicum and Eggplant*. Noordwijkerhout, The Netherlands. 126pp.

Daunay, M. C., Janick, J., and Laterrot, H. (2007). Iconography of the Solanaceae from Antiquity to the 17th Century: A Rich Source of Information on Genetic Diversity and Uses. *Chronica Horticulturae* **47(3)**: 16 -22.

Deb, D. B. (1989). *Solanum melongena, S. incanum* Versus *S. insanum* (Solanaceae). *Taxon* **38:** 138–139.

Doganlar, S., Frary, A., Daunay, M. C., Lester, R. N. and Tanksley, S. D. (2002a). A Comparative Genetic Linkage Map of Eggplant *(Solanum melongena)* and Its Implication for Genome Evolution in the Solanaceae. *Genetics* **161:**1697-1711.

Doganlar, S., Frary, A., Daunay, M. C., Lester, R. N. and Tanksley, S. D. (2002b). Conservation of Gene Function in the Solanaceae as Revealed by Comparative Mapping of Domestication Traits in Eggplant. *Genetics* **161:**1713- 1726.

Donzella, G., Spena, A. and Rotino, G. L. (2000). Transgenic Parthenocarpic Eggplants: Superior Germplasm for Increased Winter Production. *Molecular Breeding* **6(1)**: 79-86.

Doyle, J. J. and Doyle, J. L. (1990). Isolation of Plant DNA from Fresh Tissue. *Focus* **12**: 13- 15.

Driskell, A. C., Ane, C., Burleigh, J. G., McMahon, M. M., O'Meara, B. C. and Sanderson, M. J. (2004). Prospects for Building the Tree of Life from Large Sequence Databases. *Science* **306**: 1172 – 1174.

Drummond, B. A., III. (1986). Coevolution of Ithomiine Butterflies and Solanaceous Plants. In: D'Arcy, W. G. (ed.). *Solanaceae: Biology and Systematics*. Columbia University Press, New York. 372pp.

Drummond, B. A., III, and Brown, K. S. Jr. (1987). Ithomiinae (Lepidoptera: Nymphalidae): Summary of Known Larval Food Plants. *Annals of the Missouri Botanical Garden* **74**: 341-358.

Edwards, K. J. and Gadek, P. A. (2001). Evolution and Biogeography of *Alectryon* (Sapindaceae). *Molecular Phylogenetics and Evolution* **20:** 14 – 26.

Encyclopaedia Iranica. (1996). Encyclopædia Iranica, XV/3. Edited by Ehsan Yarshater Columbia University Center for Iranian Studies, New York. http://www.iranicaonline.org/articles. Assessed January 13th 2011.

Fay, M. F., Olmstead, R. G. and Richardson, J. E. (1998). Molecular Data Support the Inclusion of *Duckeodendron cestroides* in Solanaceae. *Kew Bulletin* **53**: 203–212.

Food and Agricultural Organization (FAO). (1996). Global Plan of Action for the Conservation and Sustainable Utilization of Plant Genetic Resources for Food and Agriculture. FAO, Rome. 63p.

Food and Agricultural Organization (FAO) (2003) Statistics http://apps.fao.org/faostat. Assessed July 2010.

Fory, P.A., I. Sánchez, A. Bohórquez, H. Ramírez, C.I. Medina, and M. Lobo. (2010). Genetic Variability of the Colombian Collection of Lulo (*Solanum quitoense* Lam.) and Related Species of Section Lasiocarpa. *Revista Facultad Nacional de Agronomía, Medellín* **63(2)**: 5465-5476.

Frary, A., Doganlar, S., Daunay, M. C. and Tanksley, S. D. (2003). QTL Analysis of MorphologicalTraits in Eggplant and Implications for Conservation of Gene Function during Evolution of Solanaceous Species. *Theory of Applied Genetics* **107**: 359 – 370.

Frary, A., Doganlar, S. and Daunay, M.C. (2007). Eggplant. *Genome Mapping and Molecular Breeding in Plants* **5:**231-257.

Furini, A. and Wunder, J. (2004). Analysis of Eggplant (*Solanum melongena*)-Related Germplasm: Morphological and AFLP Data Contribute to Phylogenetic Interpretations and Germplasm Utilization. *Theoretical and Applied Genetics* **108**: 197–208.

Gaston, K. J. and May, R. M. (1992). Taxonomy of Taxonomists. *Nature* **356**: 281–282.

Gbile, Z. O. (1987). Nigerian *Solanum S*pecies. *Nigerian Field* **52**: 19-26.

Gbile, Z .O. and Adesina, S. K. (1988). Nigerian *Solanum* Species of Economic Importance. *Annals of the Missouri Botanical Garden* **75(3):** 862-865.

Gebhardt, S.E. and Thomas, R.G. (2002). Nutritive Value of Foods. *Home Garden Bull (USDA)* **72:**80–81.

Gepts, P. (1993). The Use of Molecular and Biochemical Markers in Crop Evolution Studies**.** *Evolution Biology* **27:** 51–94.

Hammer K., Knupffer, H., Laghetti, G., Perrino, P. (1992). *Seeds from the Past: a Catalogue of the Crop Germplasm of South Italy and Sicily*. Germplasm Institute of C.N.R, Bari, Italy. 273pp.

Hampl, V., Pavlieek, A., Flegr, J. (2001). Construction and Bootstrap Analysis of DNA Fingerprinting-based Phylogenetic Trees with the Freeware Program FreeTree:

Application to Trichomonad Parasites. *International Journal of Systematic and Evolutionary Microbiology* **51:**731–735.

Harrington, M. G., Edwards, K. J., Johnson, S. A., Chase, M.W. and Gadek, P. A. (2005). Phylogenetic Inferences in Sapindaceae sensu lato Using Plastid matK and rbcL DNA Sequences. *Systematic Botany* 30:266 – 382.

Hassan, S. M. Z. and Lester, R. N. (1990a). Crossability Relationships and Invitro Germination of F1 Hybrids between *Solanum melongena* L. and *S. panduriforme* E. Meyer (*S. incanum* L. *sensu ampl*). *SABRAO Journal of Breeding and Genetics* **22:**65–72.

Hassan, S. M. Z. and Lester, R. N. (1990b). Comparative Micromorphology of the Seed Surface of *Solanum melongena* L. (Eggplant) and Allied Species. *Pertanika* **13:**1–8.

Hawkes, J. G., Lester, R. N. and Skelding, A. D. (1979). *The Biology and Taxonomy of the Solanaceae*. Academic Press, London. 738pp.

Herrera-Arellano, A., Jiménez-Ferrer, E., Vega-Pimentel, A.M., Martínez-Rivera, M.L., Hernández- Hernández, M., Zamilpa, A. and Tortoriello, J. (2004). Clinical and Mycological Evaluation of Therapeutic Effectiveness of *Solanum chrysotrichum* Standardized Extract on Patients with *Pityriasis capitis* (dandruff). A Double Blind and Randomized Clinical Trial Controlled with Ketoconazole. *Planta Medica* **70 (6):** 483-488.

Hunziker, A. T. (1979.) South American Solanaceae: a Synoptic Survey. In: Hawkes, J. G., Lester, R. N. and Skelding, A. D. (Eds.). *The Biology and Taxonomy of the Solanaceae*. Academic Press, London. 738pp.

Hunziker, A. T. (2001). *Genera Solanacearum*. A. R. G. Ganter Publishing Co., Ruggell. 120pp.

Hyde, M. A. and Wursten, B. (2010). Flora of Zimbabwe: Cultivated Plants: Genus Page: *Solanum*. http://www.zimbabweflora.co.zw/cult/genus.php?genus_id=1249, assessed 8 March 2011 at 21.30 GMT.

IBPGR. (1990). Descriptors for Eggplant. Bioversity International (formerly known as International Board for Plant Genetic Resources), Rome, Italy.

International Solanaceae Genome Project (SOL). Genomics of the Genus *Solanum* – Solanaceae source. http://www.sgn.cornell.edu/solanaceae-project/. Assessed 31st March 2011.

Isshiki, S., Okubo, H., Fujieda, K. (1994). Phylogeny of Eggplant and Related *Solanum* Species Constructed by Allozyme Variation. *Scientia Horticulturae* **59**: 171–176.

Isshiki, S., N. Iwataa and M. M. R. Khana. (2008). ISSR variations in eggplant (*Solanum melongena* L.) and related *Solanum* species. *Scientia Horticulturae* **117**: 186-190.

Jacoby, A., Labuschagne, M. T., Viljoen, C. D. (2003). Genetic Relationship between Southern African *Solanum retroflexum* Dun and Other Related Species Measured by Morphological and DNA Markers. *Euphytica* **132:**109–113.

Jarvis, C. (2007). Order out of Chaos, Linnaean Plant Names and their Types. (1st edn.). The Linnaean Society of London in Association with Natural History Museum, London, UK. 658pp.

Kalloo, G. (1993). Eggplant, *Solanum melongena* L. In Kalloo, G., Berg, B. O. (Eds.). Genetic Improvement of Vegetable Crops. Pergamon Press, Oxford, UK. 675pp.

Karihaloo, J. L. and Gottlieb, L. D. (1995). Allozyme Variation in the Eggplant, *Solanum melongena* L. (Solanaceae). *Theoretical and Applied Genetics* **90:** 578–583.

Karihaloo, J. L., and Rai, M. (1995). Significance of Morphological Variability in *Solanum insanum* L. *(sensu lato*). *Plant Genetic Resources Newsletter* **103**:24–26.

Karihaloo, J. L., Brauner, S. and Gottlieb, L. D. (1995). Random Amplified Polymorphic DNA Variation in the Eggplant *Solanum melongena* L. *Theoretical and Applied Genetics* **90**:767–770.

Karihaloo, J. L., Kaur, M. and Singh, S. (2002). Seed Protein Diversity in *Solanum melongena* L. and its Wild and Weedy Relatives. *Genetic Resources and Crop Evolution* **49:** 533–539.

Kashyap, V., Kumar, S. Collonier, C., Fusari, F., Haicour, R., Rotino, G. L. Sihachak, R. D. and Rajam, M. V. (2003). Biotechnology of Eggplant. *Scientia Horticulturae* **97** (1): 1-25.

Kaur, M., Singh, S. and Karihaloo, J. L. (2004). Diversity of Enzyme Electrophoretic Patterns in the Eggplant Complex. *Journal of Plant Biochemistry and Biotechnology* **13:**69-72.

Kelchner, S. A. (2000). The Evolution of Non-coding Chloroplast DNA and its Application in Plant Systematics. *Annals of the Missouri Botanical Garden* **87**: 482–498.

Khan, R. (1979). *Solanum melongena* and Its Ancestral Forms in *The Biology and Taxonomy of the Solanaceae*, edited by J. G. Hawkes, R. N. Lester and A. D. Skelding. Academic Press, London. 636pp.

Kitching, I. J., Forey, P. L., Humphries, C. J., Williams, D. M. (1998). Cladistics: The Theory and Practice of Parsimony Analysis, 2nd edn., Oxford University Press, Oxford. 205pp.

Knapp, S. (1986) Reproductive Biology of *Solanum* Section Geminata in a Costa Rican Cloud Forest. In: D'Arcy, W. G. (ed.), Solanaceae: Biology and Systematics. Columbia University Press, New York. 372pp.

Knapp, S. (1989). A Revision of the *Solanum nitidum* Group (Section Holophylla pro parte): Solanaceae. *Bulletin of the British Museum* (*Natural History*), *Botany* **19:**63–112.

Knapp, S. (1991). A Revision of the *Solanum* Sessile Species Group (Section Geminata pro parte: Solanaceae). *Botanical Journal of Linnaean Society* **105:**179–210.

Knapp, S. (2002a). Floral Diversity and Evolution in the Solanaceae. In: Cronk, Q.C.B., Bateman, R.M., Hawkins, J.A. (Eds). *Developmental Genetics and Plant Evolution*. Taylor & Francis, London.365pp.

Knapp, S. (2002b). *Solanum* Section *Geminata* (Solanaceae). *Flora Neotropica Monograph* **84**:1–405.

Knapp, S., Persson, V., and Blackmore, S. (1998). Pollen Morphology and Functional Dioecy in *Solanum* (Solanaceae). *Plant Systematics and Evolution* **210**: 113-139.

Knapp, S., Bohs, L., Nee, M. and Spooner, D. M. (2004). Solanaceae: a Model for Linking Genomics with Biodiversity. *Comparative and Functional Genomics* **5:**285–291.

Kumar, G., Meena, B. L., Kar, R., Tiwari, S. K., Gangopadhyay, K. K., Bisht, I. S. and Mahajan, R. K. (2008). Morphological Diversity in Brinjal (*Solanum melongena* L.) Germplasm Accessions. *NLAB Plant Genetic Resources: Characterization and Utilization* **6**: 232-236.

Kumar, S., Tamura, K., Jakobsen, I.B. and Nei, M. (2001). MEGA 2.0: Molecular Evolutionary Genetics Analysis Software. *Bioinformatics* **17:** 1244–1245.

Lester, R. N. (1997). Typification of Nineteen Names of African *Solanum* Species Described by A. Richard and others, including *S. campylacanthum* and *S. panduriforme. Botanical Journal of the Linnaean Society* **125:**273-293.

Lester, R .N. (1998). Genetic Resources of Capsicum and Eggplants. 10th EUCARPIA Meeting on Genetics and Breeding of Capsicum and Eggplant). 90 pp.

Lester, R. N. and Hassan, S. M. Z. (1990). The Distinction between *Solanum incanum* L. and *Solanum insanum* L. (Solanaceae) *Taxon* **39**: 521–523.

Lester, R. N. and Hassan, S. M. Z. (1991). Origin and Domestication of the Brinjal Eggplant, *Solanum melongena,* from *S. incanum*, in Africa and Asia. In: Hawkes, J.G., Lester, R.N., Nee, M., Estrada-R, N. (Eds.), Solanaceae III: Taxonomy, Chemistry, Evolution. The Royal Botanic Gardens, Kew, Richmond, UK. 500 pp.

Lester, R. N. and J. G. Hawkes. (2001). Solanaceae. In: Hanelt, P. and Institute of Plant Genetics and Crop Plant Research (Eds.), Mansfeld's Encyclopedia of Agricultural and Horticultural Crops. Springer-Verlag, Berlin. 1856pp.

Lester, R. N. and Daunay, M. C. (2003). Diversity of African Vegetable *Solanum* Species and its Implications for a Better Understanding of Plant Domestication. In: Knuppfler, H., Ochsmann, J. (eds.), Rudolf Mansfield and Plant Genetic Resources. *Gatersleben (DEU)* **22**: 137-152.

Lester, R. N., Francesco-Ortega, J. and Al-Ase, M. (1999). Convergent Evolution of Heterandry (Unequal Stamens) in *Solanum*, Proved by Spermoderm SEM In: M. Nee, D. E. Symon, R. N. Lester and J.P. Jessop (Eds.), *Solanaceae IV: Advances in Biology and Utilization*. Royal Botanic Gardens, Kew. 545pp.

Levin, R. A, Watson, K. and Bohs, L. (2005). A Four-gene Study of Evolutionary Relationships in *Solanum* Section Acanthophora. *American Journal of Botany* **92(4):**603–612.

Levin, R. A., Myers, N. R. and Bohs, L. (2006). Phylogenetic Relationships among the "Spiny *Solanums*" (*Solanum* Subgenus *Leptostemonum*, Solanaceae). *American Journal of Botany* **93(1)**:157-169.

Mabberley, D. J. (2008). The Mabberley Plant Book; a Portable Dictionary of Plants, their Classification and Uses. Cambridge University Press, Cambridge, U.K. 368pp.

Mace, E. S., Lester, R. N. and Gebhardt, C. G., (1999). AFLP Analysis of Genetic Relationships among the Cultivated Eggplant, *Solanum melongena* L., and Wild Relatives (Solanaceae). *Theoretical and Applied Genetics* **99:** 626–633.

Maddison, W. P., and Maddison, D. R. (2000). MacClade 4: Analysis of Phylogeny and Character Evolution. Sinauer, Sunderland, Massachusetts, USA.

Martin, F. W. and Rhodes, A. M. (1979). Subspecific Grouping of Eggplant Cultivars. *Euphytica* **28**: 367 – 383.

Miller, J. T. and Spooner, D. M. (1999). Collapse of Species Boundaries in the Wild Potato *Solanum brevicaule* Complex (Solanaceae sect. Petota): Molecular Data. *Plant Systematics and Evolution* **214**:103–130.

Mueller, L. A., Solow, T. H., Taylor, N., Skwarecki, B., Buels, R., Binns, J., Lin, C., Wright, M. H., Ahrens, R., Wang, Y., Herbst, E. V., Keyder, E. R., Menda, N., Zamir, D. and Tanksley, S. D. (2005). The SOL Genomics Network: A Comparative Resource for Solanaceae Biology and Beyond. *Plant Physiology* **138** :1310-1317.

Muellner, A. N., Vassiliades, D. D. and Renner, S. S. (2007). Placing Biebersteiniaceae, a Herbaceous Clade of Sapindales, in a Temporal and Geographic Context. *Plant Systematics and Evolution* **266:** 233 – 252.

Naujeer, H. B. (2009). Morphological Diversity in Eggplant (*Solanum melongena* L.), their Related Species and Wild Types Conserved at the National Gene Bank in Mauritius. A Master's Thesis submitted to the International Master Programme at the Swedish Biodiversity Centre, Uppsala. 74pp.

Nee, M. (1999). Synopsis of *Solanum* in the New World. In: Nee, M., Symon, D. E., Lester, R.N., Jessop, J.P. (Eds) Solanaceae IV: advances in biology and utilization. Royal Botanical Gardens, Kew, 545pp.

Nee, M., Bohs, L., and Knapp, S. (2006). New species of *Solanum* and *Capsicum* (Solanaceae) from Bolivia, with Clarification of Nomenclature in some Bolivian *Solanum*. *Brittonia* **58**: 322-356.

Neigel, J.E. and Avise, A.C. (1986). Phylogenetic Relationship of Mitochondrial DNA under Various Models of Speciation. In: Karlin, S., Nevo, E. (Eds). Evolutionary Processes and Theory. Academic Press, New York. 534pp.

Ng, N.Q. and Padulosi, J. (1997). Origin, Taxonomy and Morphology of *Vigna unguiculata* (L.) walp In: Singh, B.B., Mohan, D.R., Raj, O., Dashiel, K.E. and Jachai, L.E.N. (eds). *Advances in Cowpea Research*. Hong –Kong. 76 pp.

Obute, G. C., Benjamin, C. N. and Okoli, B. E. (2006). Cytogenetic Studies on some Nigerian Species of *Solanum* L. (Solanaceae). *African Journal of Biotechnology* **5 (9):**689-692.

Ogundipe, O. T. and Daramola, S. O. (1998). Epidermal Studies in some *Solanum* (Solanaceae). *Boletim da Sociedada Broteriana Ser* **4 (69)**: 101 – 112.

Ogundipe, O. T., Ajayi, G. O. and Adeyemi, T. O. (2009). Phytoanatomical and Antimicrobial Studies on *Gomphrena celosioides* Mart. (Amaranthaceae). *Hamdard Medicus* **51(3):**146 – 156.

Ogunkanmi, L. A. (2006). *Genetic Diversity in Cowpea and its Wild Relatives.* PhD Thesis Department of Botany and Microbiology, University of Lagos, Lagos. 158pp.

Okoli, B. E. (1988). Cytotaxonomic Studies of Five West African Species of *Solanum* L. (Solanaceae). *Feddes Repertorium* **99(5-6)**: 183-187.

Olmstead, R. G. and Palmer J. D (1992). Chloroplast DNA Phylogeny of the Solanaceae: Subfamilial Relationships and Character Evolution. *Annals of the Missouri Botanical Garden* **79**: 346–360.

Olmstead, R. G, and Sweere, J. A. (1994). Combining Data in Phylogenetic Systematics: an Empirical Approach using Three Molecular Datasets in the Solanaceae. *Systematic Biology* **43**: 467–481.

Olmsted, R. G. and Palmer. J. D. (1997). Implications for the Phenology, Classification and Biogeography of *Solanum* from cpDNA Restriction Site Variation. *Systematic Botany* **22**:19-29.

Olmstead, R .G., Sweere, J. A., Spangler, R. E., Bohs, L. and Palmer, J. D. (1999). Phylogeny and Provisional Classification of the Solanaceae Based on Chloroplast DNA. In: Nee, M., Symon, D. E., Lester, R. N. and Jessop, J. P. (Eds.). *Solanaceae IV: Advances in Biology and Utilization*. Royal Botanic Gardens, Kew. 545pp.

Oyelana, O. A. and Ugborogho, R. E. (2008). Phenotypic Variation of F1 and F2 Populations from Three Species of *Solanum* L. (Solanaceae). *African Journal of Biotechnology* **7 (14)**:2359-2367.

Pearce, K. and Lester, R. N. (1979). Chemotaxonomy of the Cultivated Eggplant – A New Look at the Taxonomic Relationships of *Solanum melongena* L. In: J. G. Hawkes, N. Lester and A. D. Skelding (Eds), *Linnean Society Symposium Series (7)*. Academic Press, London, UK. 738pp.

Pellissier, M., Haymes, K.M. and Williams, S.M. (1992). Parentage Analysis Using RAPD PCR. *Nucleic Acids Research* **20**:5493

Phillipe, H., Snell, E. A., Bapteste, E., Lopez, P., Holland, P. W. H. and Casane, D. (2004). Phylogenomics of Eukaryotes: Impact of Missing Data on Large Alignments. *Molecular Biology of Evolution* **21:**1740 – 1752.

Polignano, G., Uggenti, P., Bisignano, V. and Gatta, C. D. (2009). Genetic Divergence Analysis in Eggplant (*Solanum melongena* L.) and Allied Species. *Genetic Resources* and *Crop Evolution* **9**: 1- 11.

Portis, E., Nervo, G., Cavallanti, F., Barchi, L. and Lanteri, S. (2006). Multivariate Analysis of Genetic Relationships between Italian Pepper Landraces. *Crop Science* **46**: 2517 – 2525.

Prohens, J., Blanca, J. M. and Nuez, F. (2005). Morphological and Molecular Variation in a Collection of Eggplants from Secondary Centre of Diversity: Implications for Conservation and Breeding. *Journal of American Society of Horticultural Science* **130(1)**: 54-63.

Quattrocchi, U. (2000). CRC World Dictionary of Plant Names. 4 R-Z. Taylor and Francis US. 3058pp.

Rambaut, A. (1996). Se-Al: Sequence Alignment Editor. Available at http://evolve.zoo. ox.ac.uk/. Department of Zoology, University of Oxford, Oxford, UK. Assessed November 2010.

Richman, A. D., and Kohn, J. R. (2000). Evolutionary Genetics of Self-incompatibility in the Solanaceae. *Plant Molecular Biology* **42**: 169-179.

Riley, K., Ramanatha, R. V., Zhou, M. D., Quek, P. (1996). Characterization and Evaluation of Plant Genetic Resources-Present Status and Future Challenges. In: Plant Genetic Resources: Characterization and Evaluation, NIAR, Ibaraki, Japan. 130 pp.

Rohlf, F. J. (1996). NTSYS-pc: Numerical Taxonomy and Multivariate Analysis System, version 2.02j. Exeter Software. Setauket, New York.

Ronquist, F., van der Mark, P. and Huelsenbeck, J. P. (2009). Bayesian Phylogenetic Analysis Using MR BAYES. In: Philippe, L., Marco, S. and Anne-Mieke, V. (Eds.), *The Phylogenetic Handbook: A Practical Approach to Phylogenetic Analysis and Hypothesis Testing. 2nd edition*. Cambridge University Press, UK. 226pp.

Sakata, Y. (1992). Taxonomic Relationships between *Solanum melongena* (Eggplant), *S.incanum* and *S. marginatum*, Based on Chloroplast DNA. *Proceedings of the 8th Meeting on Genetics and Breeding of Capsicum and Eggplant* **8**: 278-282.

Sakata, Y. and Lester, R. N. (1994). Chloroplast DNA Diversity in Eggplant (*Solanum melongena*) and its Related Species *S. incanum* and *S. marginatum*. *Euphytica* **80**: 1–4.

Sakata, Y. and Lester, R. N. (1997). Chloroplast DNA Diversity in Brinjal Eggplant (*Solanum melongena* L.) and Related Species. *Euphytica* **97**: 295–301.

Sakata, Y., Nishio, T. and Matthews, P. J. (1991). Chloroplast DNA Analysis of Eggplant (*Solanum melongena*) and Related Species for their Taxonomic Affinity. *Euphytica* **55**: 21–26.

Samuels, B. J. (1996). *Solanum incanum sensu lato* (Solanaceae): Taxonomy, Phylogeny and Distribution. Ph. D thesis, University of Birmingham, UK. 109pp.

Sanger, F., Air, G. M. and Barrell, B. G. (1977). Nucleotide Sequence of Bacteriophage phi X174 DNA. *Nature* **265** (5596): 687–95.

Savolainen, V., Cuenoud, P., Spichiger, R., Martinez, M. D. P., Crevecoeur, M. and Manen, J. F. (1995). The Use of Herbarium Specimens in DNA Phylogenetics: Evaluation and Improvement. *Plant Systematics and Evolution* **197**: 87 – 98**.**

Sazima, M., Vogel, S., Cocucci, A. and Hauser, G. (1993). The Perfume Flowers of Cyphomandra (Solanaceae): Pollination by Euglossine Bees, Bellows Mechanism, Osmophores, and Volatiles. *Plant Systematics and Evolution* **187**: 51-88.

Scippers, R. R. (2000). African Indigenous Vegetables: an Overview of the Cultivated Species. Natural Resources Institute, Chatham, UK. 274pp.

Seithe, A. and Anderson, G. J. (1982). Hair Morphology and the Relationships of Species in *Solanum* Sect. Basarthun. *Plant. Sits. Evol*. **139**: 229–256.

Sękara, A., S. Cebula, and E. Kunicki. (2007). Cultivated Eggplants – Origin, Breeding Objectives and Genetic Resources, a Review. *Folia Horticulturae Annals* **19(1):**97-114.

Sifau, M. O., Ogunkanmi, L. A., Oboh, B. O., and Ogundipe, O.T. (2009). Phenotypic Characterization of Edible Vegetable *Solanum* (Solanaceae) in South-Western Nigeria. *Proceedings of the 33rd Annual Conference of the Genetics Society of Nigeria* **33**: 98-103.

Silva, M. E., Santos, R. C., O'Leary, M. C. and Santos, R. S. (1999). Effect of Aubergine (*Solanum melongena*) on Serum and Hepatic Cholesterol and Triglycerides in Rats. *Brazilian Archives of Biology and Technology* **42**(3): 339-342.

Singh, A.K., Singh, M., Singh, A.K., Singh, R., Kumar, S., and Kalloo, G. (2006). Genetic Diversity within the Genus *Solanum* (Solanaceae) as Revealed by RAPD Markers. *Current Science* **90(5)**:711-716.

Sinha, N. (1997). Simple and Compound Leaves: Reduction or Multiplication? *Trends in Plant Science* **2**: 396-402.

Soltis, D. E. and Soltis, P. S. (1997). Phylogenetic Relationships among Saxifragaceae sensu lato: A Comparison of Topologies Based on 18S rDNA and rbcL Sequences. *American Journal of Botany* **84**: 504 – 522.

Soltis, D. E. and Soltis, P. S. (2003). The Role of Phylogenetics in Comparative Genetics. *Plant Physiology* **132**: 1790–1800.

Spooner, D. M., Tivang, J., Nienhuis, J., Miller, J. T., Douches, D. S. and Contreras, M. A. (1996). Comparison of Four Molecular Markers in Measuring Relationships among the Wild Potato Relatives *Solanum* Section Etuberosum (subg. Potatoe). *Theoretical and Applied Genetics* **92(5**):532–540

Spooner, D. M., Ugarte, M. L. and Skroch, P. W. (1997). Species Boundaries and Interrelationships of Two Closely Related Sympatric Diploid Wild Potato Species, *Solanum astleyi* and *S.boliviense*, Based on RAPDs. *Theoretical and Applied Genetics* **92(5–6)**:764–771

Stedje, B. and Bukenya-Ziraba, R. (2003). RAPD Variation in *Solanum anguivi* Lam. and *S. aethiopicum* L. (Solanaceae) in Uganda. *Euphytica* **131**: 293–297.

Stommel, J. R. and Whitaker, B. D. (2003). Phenolic Acid Content and Composition of Eggplant Fruit in a Germplasm Core Subset. *Journal of American Society of Horticultural Science* **128**: 704 -710.

Stone, J. L. (2002). Molecular Mechanisms Underlying the Breakdown of Gametophytic Self-incompatibility. *Quarterly Review of Biology* **77**: 17-32.

Swofford, D., Olsen, G., Waddel, P. and Hillis, D. (1996). Phylogenetic Inference. In Hillis, D., Moritz, C., Mable, B. (Eds.). *Molecular Systematics.* Sinauer, Sunderland, MA. 168pp.

Symon, D. E. (1979.) Sex Forms in *Solanum* (Solanaceae) and the Role of Pollen Collecting Insects. In: J. G. Hawkes, R. N. Lester, and A. D. Skelding (Eds.), *The biology and taxonomy of the Solanaceae*. Academic Press, London. 738pp.

Taberlet, P., Gielly, L., Pautou G. and Bouvet, J. (1991). Universal Primers for Amplification of Three Non-coding Regions of Chloroplast DNA. *Plant Molecular Biology* **17**: 1105–1109.

Takahata, N. and Nei, M. (1985). Gene Genealogy and Variance of Interpopulational Nucleotide Difference. *Genetics* **110**:325–344.

Tohme, J., Orlando-Gonzalez, D., Beebe, S. and Duque, M. C. (1996). AFLP Analysis of Gene Pools of a Wild Bean Core Collection. *Crop Science* **36**:1375–1384.

Van de Zande, L. and Bijlsma, R. (1995). Limitations of the RAPD Technique in Phylogeny Reconstruction in Drosophila. *Journal of Evolutionary Biology* **8**:645–656

Van den Berg, R., Bryan, G., Del Rio, A. and Spooner, D. M. (2002). Reduction of Species in the Wild Potato *Solanum* Section Petota Series Longipedicellata: AFLP, RAPD and Chloroplast SSR Data. *Theoretical and Applied Genetics* **105(8):**1109–1114.

Weese, T. L. and Bohs, L. (2007). A Three Gene Phylogeny of the Genus *Solanum* (Solanaceae). *Systematic Botany* **32**:445–463.

Welsh, J. and McClelland, M. (1990). Fingerprinting Genomes Using PCR with Arbitrary Primers. *Nucleic Acids Research* **18**: 7213–7218.

Whalen, M. D. (1984). Conspectus of the Species Group in *Solanum* subgenus *Leptostemonum*. *Gentes Herbarum* **12**: 179–282.

Whalen, M. D. and Costich, D. E. (1986). Andromonoecy in *Solanum*. In: W. G. D'Arcy (ed.), *Solanaceae: Biology and Systematics*. Columbia University Press, New York. 372pp.

White, T. J., Bruns, T., Lee, S. and Taylor, J. (1990). Amplification and Direct Sequencing of Fungal Ribosomal RNA Genes for Phylogenetics. In: M. Innis, D. Gelfand, J. Sninsky, and T. White [eds.], PCR Protocols: A Guide to Methods and Applications. Academic Press, San Diego, California, USA. 322pp.

Wiens, J. J. (2003). Missing Data, Incomplete Taxa and Phylogenetic Accuracy. *Systematic Biology* **52**: 528 - 538.

Wiens, J. J. (2005). Can Incomplete Data Rescue Phylogenetic Analyses from Long-Branch Attraction? *Systematic Biology* **54**: 731 - 742.

Wiens, J. J. (2006). Missing Data and the Design of Phylogenetic Analyses. *Journal of Biomedical Information* **39**: 34 - 42.

Wiens, J. J. and Reeder, T. W. (1995). Combining Data Sets with Different Numbers of Taxa for Phylogenetic Analysis. *Systematic Biology* **44**: 548 - 558.

Williams, J. K. F., Kubelik, A. R., Livak, K. G., Rafalski, J. A. and Tingey, S. V. (1990). DNA Polymorphisms Amplified by Arbitrary Primers are Useful as Genetic Markers. *Nucleic Acids Research* **18**:6531–6535.

APPENDICES

Appendix 1: *Solanum* Distribution in West Tropical Africa and Nigeria (International Solanaceae Genome Project (SOL).

Names associated with West Tropical Africa	Names associated with Nigeria
Solanum aculeastrum Dunal	*Solanum aculeastrum* Dunal
Solanum aculeatissimum Jacq.	*Solanum aculeatissimum* Jacq.
Solanum aethiopicum L.	*Solanum americanum* Mill.
Solanum americanum Mill.	*Solanum anguivi* Lam.
Solanum anguivi Lam.	*Solanum chrysotrichum* Schltdl.
Solanum anomalum Thonn.	*Solanum dasyphyllum* Schumach. & Thonn.
Solanum betaceum Cav.	*Solanum distichum* Schumach. & Thonn.
Solanum capsicoides All.	*Solanum erianthum* D.Don
Solanum cerasiferum Dunal	*Solanum lycopersicum* L.
Solanum chrysotrichum Schltdl.	*Solanum macrocarpon* L.
Solanum dasyphyllum Schumach. & Thonn.	*Solanum mauritianum* Scop.
Solanum distichum Schumach. & Thonn.	*Solanum terminale* subsp. *inconstans* (C.H.Wright)
Solanum erianthum D.Don	Heine
Solanum forskalii Dunal	*Solanum torvum* Sw.
Solanum incanum L.	
Solanum lichtensteinii Willd.	
Solanum lycopersicum L.	
Solanum macrocarpon L.	
Solanum mauritianum Scop.	
Solanum melongena L.	
Solanum subinerme Jacq.	
Solanum terminale subsp. inconstans (C.H.Wright)	
Heine	
Solanum terminale Forssk.	
Solanum torvum Sw.	

Appendix 2: Modified IPGRI (1990) Eggplant descriptor list showing both qualitative and quantitative traits used in this study.

1.0 Vegetative characters

PLANT'S STEM/BRANCHES

Name:

Identification Number:

Location: (1) Latitude

(2) Longitude

(3) Elevation (metres above sea level)

Photograph No.:

Zone:

Town/Village:

Date: (DDMMYYYY)

Time:

Plant Growth Habit: (3) Upright

(5) Intermediate

(7) Prostrate

****Plant's Height (cm)**

Plant Size: Visual estimation of the whole plot

(3) Small

(5) Intermediate

(7) Large

****Stem Length at the first inflorescence (cm):** Measured as the total length of the stem, from the stem base to the point of insertion of the first inflorescence in the stem, at the time of flowering of the first inflorescence.

Vigour of the Plant: (3) Weak (5) Intermediate (7) Strong

Stem Pubescence Density: (0) Glabrous

(3) Sparse

(5) Intermediate

(7) Dense

Stem Colour: (1) Green

(2) Greenish with purple spots

(3) Greenish purple

(4) Purple

(5) Dark purple

****Internode Length (cm):** Measured at a middle distance between the top and the base of the main stem.

LEAVES

Type of Leaves: (1) Simple

(2) Compound

****Petiole length (cm):**

Petiole Colour: (1) Green

(2) Greenish with purple spots
(3) Greenish purple
(4) Purple
(5) Dark purple

Foliage Density: (3) Sparse

(5) Intermediate

(7) Dense

Leaf Attitude: (1) Semi-erect

(2) Horizontal
(3) Dropping

****Leaf Blade length (cm):** (3) Short (~10cm)

(5) Intermediate (~20cm)

(7) Long (~30cm)

****Leaf Blade width (cm):** (3) Narrow (~5cm)

(5) Intermediate (~10cm)

(7) Wide (~15cm)

***Leaf Blade length/width ratio:**

Position of the widest part of the leaf blade:

(1) Base

(3) Bottom 1/3

(5) Middle

(7) Top 1/3

Leaf shape: Indicate the shape of the leaf for accessions with simple leaves and of the terminal leaflet for accessions with compound leaves.

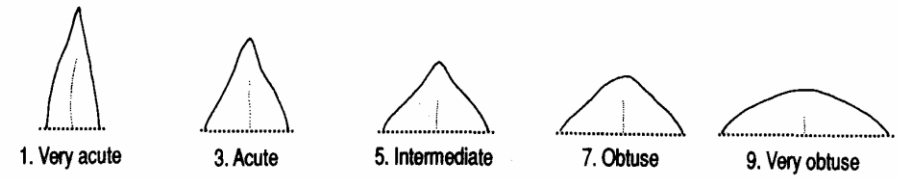
(1)	Elongated	(5)	Cordiform
(2)	Lanceolate	(6)	Elliptic
(3)	Ovate	(7)	Rounded
(4)	Obovate		

Leaf apex shape: Indicate the shape of the apex of the leaf for accessions with simple leaves and of the terminal leaflet for accessions with compound leaves.

(1)	Acute
(2)	Intermediate
(3)	Obtuse

Leaf Blade tip angle

1	Very acute	(< 15°)
3	Acute	(~ 45°)
5	Intermediate	(~ 75°)
7	Obtuse	(~110°)
9	Very obtuse	(>160°)



Leaf Colour: Observed on the adaxial (upper) surface:

(1)	Light green	(4)	Greenish purple
(2)	Green	(5)	Purple
(3)	Dark green		

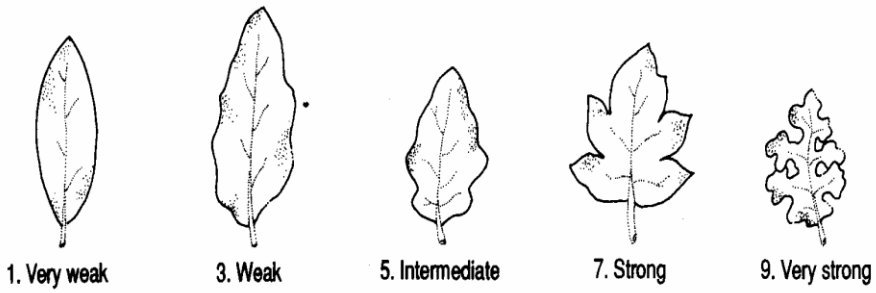
Anthocyanin colouration of leaf veins: (3) Green

(5) Main veins purple and the rest green

(7) Purple

Leaf Blade lobing

1 Very weak
3 Weak
5 Intermediate
7 Strong
9 Very strong



Leaf hairiness type: Observed on abaxial (under) side

(1) Glabrous (no hairs)
(2) Puberulent (Very minutely downy hairs)
(3) Velutinous (short dense soft hairs)
(4) Pilose (fine soft hairs)
(5) Hirsute (coarse stiff hairs)

Leaf Surface Attitude:

(3) Flat
(5) Intermediate
(7) Very Convex

2.0 Reproductive characters

INFLORESCENCE/FLOWER AND FRUITS

Inflorescence Type: (1) Generally uniparous (one axis at each branching, as a cyme)

(2) Both partly uniparous, partly multiparous

(3) Generally multiparous (more than one axis at each branching, as a cyme)

Corolla colour: 1 White

2 Stripped (white >75% and purple <25%)

3 Stripped (white 50–75% and purple 25–50%)

4 Stripped (white 25–50% and purple 50–75%)

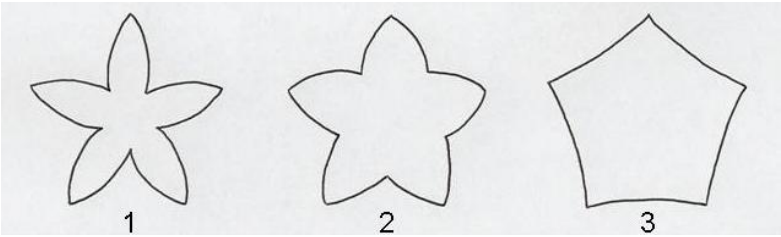
5 Stripped (white < 25% and purple >75%)

6 Purple

Corolla Type: (1) Stellate

(2) Intermediate/semi-stellate

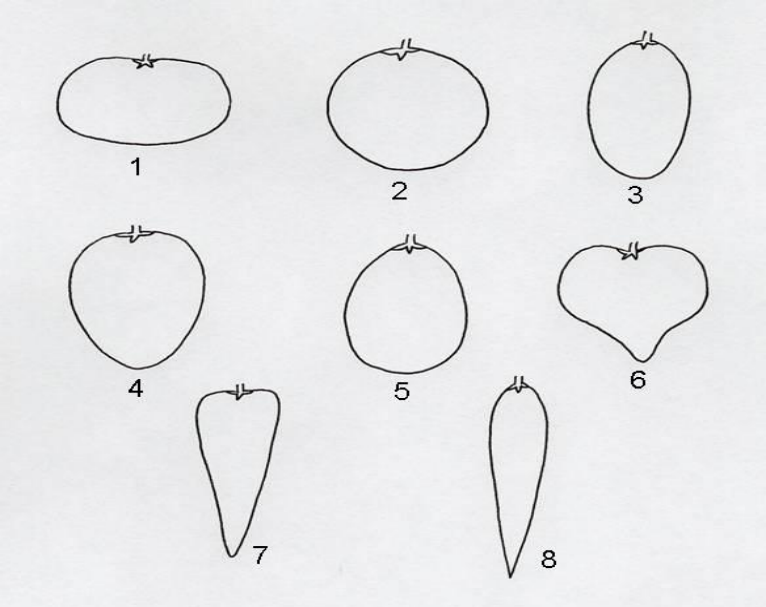
(3) Rotate



***Mean fresh Fruit Weight (g)**: Records would be taken of fruits of at least 5 different plans on the 1st truss at maturity stage. Average of 10 fruits from different plants

Predominant Fruit Shape:

1	Flattened	6	Cordiform
2	Rounded	7	Conical
3	Ellipsoid	8	Elongate
4	Ovate	99	Other (specify in "Notes")
5	Obovate		



Fruit predominant colour at commercial ripeness:

1	Dark green	5	Golden yellow
2	Light green	6	Orange yellow
3	Milk white	7	Lilac
4	Pale yellow	8	Purple
		9	Purple black

Fruit secondary colour at commercial ripeness:

0	Absent	5	Golden yellow
1	Dark green	6	Orange yellow
2	Light green	7	Lilac
3	Milk white	8	Purple
4	Pale yellow	9	Purple black

Fruit flesh colour:

1	Dark green	5	Golden yellow
2	Light green	6	Orange yellow
3	White	7	Orange
4	Pale yellow	8	Salmon

Number of fruits per inflorescence: Mean number of fruits for the first three trusses

Fruit size uniformity: (3) low

(5) Intermediate

(7) High

****Fruit length (cm):**

****Fruit width (cm):**

***Fruit length/width ratio:**

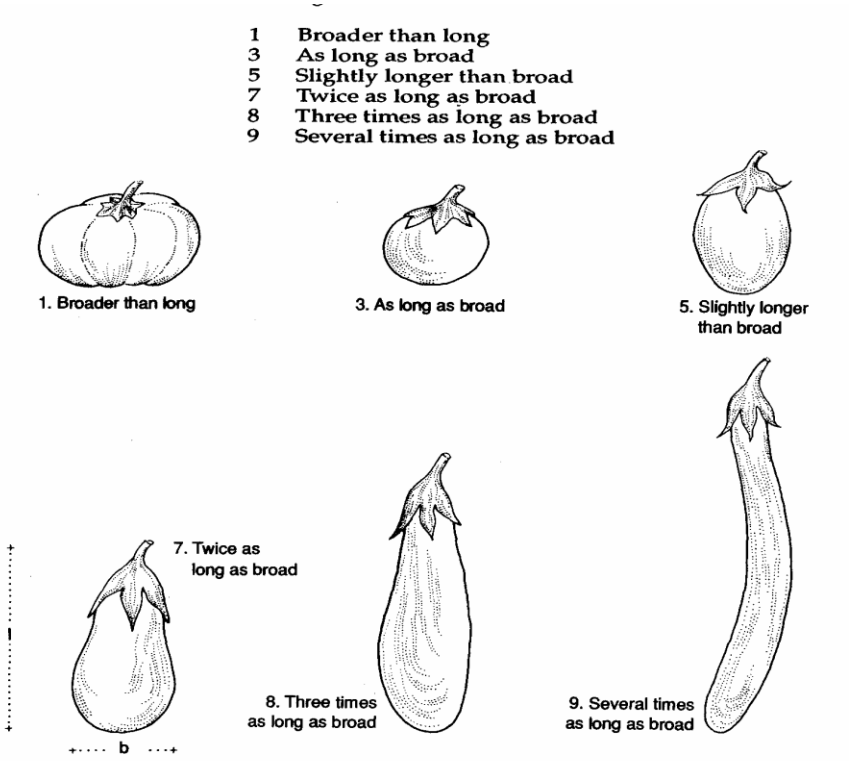
Position of the widest part of the fruit:

(3) Less than ¼ way from base to tip

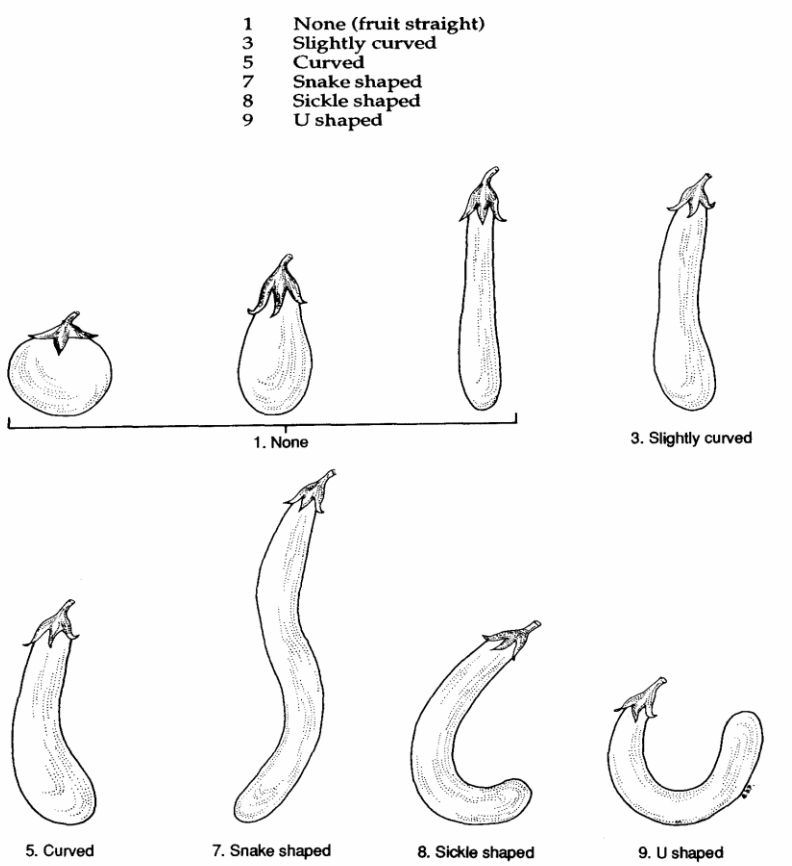
(5) Between ¼ and ½ way from base to tip

(7) More than ½ way from base to tip

Fruit Length/ Breadth Ratio



Fruit curvature



NOTE: ** = Quantitative traits

Courtesy: **International Plant Genetic Resources Institute (IPGRI, now known as Bioversity International), Rome, Italy.**

Appendix 3: Samples and places of collection

Sample I.D No.	Longitude	Latitude	Place of collection	Zone/ State
OG02	E003^{0}13'40.3"	N06^{0}59'26.8"	Wasinmi	SW/Ogun
OG03	E003^{0}13'34"	N06^{0}59'26.7"	Wasinmi	SW/Ogun
OG04	E003^{0}07'29"	N07^{0}06'41.5"	Joga orile	SW/Ogun
OG05	E003^{0}07'23.3"	N07^{0}06'42.3"	Joga orile	SW/Ogun
OG06	E003^{0}03'09.2"	N07^{0}02'03.1"	Abulemaria	SW/Ogun
OG07	E003^{0}03'09.3"	N07^{0}02'03.5"	Abulemaria	SW/Ogun
OG08	E003^{0}01'42.1"	N07^{0}00'38.8"	Wasimi- Imasai	SW/Ogun
OG09	E003^{0}01'38.4"	N07^{0}00'45.8"	Wasimi- Imasai	SW/Ogun
OG10	E003^{0}01'37.6"	N07^{0}00'45.11"	Wasinmi-Imasai	SW/Ogun
OY11	E003^{0}42'35.9"	N08^{0}49'05.8"	Igboho	SW/Oyo
OY12	E003^{0}42'35.4"	N08^{0}49'05.5"	Igboho	SW/Oyo
OY13	E003^{0}42'35.2"	N08^{0}49'05.3"	Igboho	SW/Oyo
OY14	E003^{0}42'35.6"	N08^{0}49'04.9"	Igboho	SW/Oyo
OY15	E003^{0}42'35.8"	N08^{0}49'05.2"	Igboho	SW/Oyo
OY16	E003^{0}42'35.7"	N08^{0}49'05.6"	Igboho	SW/Oyo
OY17	E003^{0}46'04.0"	N08^{0}50'42.8"	Igboho	SW/Oyo
OY18	E003^{0}45'55.7"	N08^{0}50'39.9"	Igboho	SW/Oyo
OY19	E003^{0}47'17.3"	N08^{0}49'51.1"	Igbope	SW/Oyo

Appendix 3 (Contd.): Samples and places of collection

Sample I.D No.	Longitude	Latitude	Place of collection	Zone/ State
OY20	E003^{0}47'16.1"	N08^{0}49'55.1"	Igboho	SW/Oyo
OS21	E003^{0}59'43.5"	N07^{0}55'20.1"	Iwo	SW/Osun
OG22	E003^{0}52'12.3"	N07^{0}47'12.3"	J3 Camp, Ijebu Ode	SW/Ogun
LA23	E003^{0}24'15.2"	N06^{0}31'25.3"	Bariga, Lagos	SW/Lagos
LA24	E003^{0}35'12.3"	N06^{0}49'12.3"	Agbowa- Ikosi	SW/Lagos
LA25	E003^{0}35'20.5"	N06^{0}49'14.3"	Agbowa- Ikosi	SW/Lagos
LA26	E003^{0}34'56.3"	N06^{0}47'15.5"	Agbowa- Ikosi	SW/Lagos
OY27	E003^{0}59'33.5"	N07^{0}23'20.1"	Alakia, Ibadan	SW/Oyo
OY28	E003^{0}57'35.0"	N07^{0}27'01.0"	Papa village, Akobo, Ibadan	SW/Oyo
OY29	E003^{0}59'34.2"	N07^{0}23'21.8"	Alakia, Ibadan	SW/Oyo
ED30	E005^{0}59'59.9"	N06^{0}05'29.2"	Ugo town	SE/Edo
ED31	E006^{0}00'01.5"	N06^{0}05'28.9"	Ugo town	SE/Edo
ED32	E005^{0}59'10"	N06^{0}00'0.9"	Ugo town	SE/Edo
ED33	E005^{0}59'00"	N06^{0}00'0.9"	Ugo town	SE/Edo
ED34	E006^{0}00'01.9"	N06^{0}05'27.2"	Ugo town	SE/Edo
OD35	E004^{0}52'12.3"	N06^{0}47'12.3"	Ore town	SW/Ondo
OD36	E004^{0}52'18.6"	N06^{0}47'12.3"	Ore town	SW/Ondo
CR37	E008^{0}20'03.0"	N05^0 20'06.0"	Okomita, Akamkpa (Calabar-Ikom Road)	SS/Cross-River
CR38	E008^{0}25'33.3"	N05^0 54'5.5"	Edondon Village, Obubra LGA.	SS/Cross-River

Appendix 3 (Contd.): Samples and places of collection

Sample I.D No.	Longitude	Latitude	Place of collection	Zone/ State
CR39	E008^{0}25$'$34.3$''$	N05^0 51$'$48.9$''$	Edondon Village, Obubra LGA.	SS/Cross-River
CR40	E008^{0}19$'$24.7$''$	N05^0 21$'$05.3$''$	Eng-Huat Rubber Estate Hq.Camp, Akamkpa	SS/Cross-River
CR41	E008^{0}19$'$34.5$''$	N05^0 22$'$06.4$''$	Eng-Huat Rubber Estate Hq.Camp, Akamkpa	SS/Cross-River
CR42	E008^{0}23$'$38.2$''$	N05^0 20$'$52.6$''$	Obung Village	SS/Cross-River
CR43	E008^{0}23$'$37.1$''$	N05^0 20$'$52.1$''$	Obung Village	SS/Cross-River
CR44	E008^{0}23$'$38.5$''$	N05^0 20$'$52.1$''$	Obung Village	SS/Cross-River
CR45	E008^{0}23$'$39.9$''$	N05^0 20$'$52.1$''$	Obung Village	SS/Cross-River
CR46	E008^{0}23$'$39.0$''$	N05^0 20$'$46.9$''$	Obung Village	SS/Cross-River
CR47	E008^{0}23$'$39.0$''$	N05^0 20$'$45.5$''$	Obung Village	SS/Cross-River
CR48	E008^{0}23$'$38.4$''$	N05^0 20$'$45.4$''$	Obung Village	SS/Cross-River
CR49	E008^{0}23$'$39.1$''$	N05^0 20$'$45.6$''$	Obung Village	SS/Cross-River
CR50	E008^{0}20$'$59.3$''$	N04^0 59$'$22.7$''$	Ediba-Holycan Junction, Off Marian Road, Calabar	SS/Cross-River

Appendix 4: Total Variance Explained by Principal Axis Factoring using Extraction Method

Factor	Initial Eigen values		
	Total	% of Variance	Cumulative %
1	3.292	29.925	29.925
2	2.797	25.429	55.354
3	2.017	18.341	73.694
4	0.971	8.824	82.518
5	0.926	8.421	90.940
6	0.355	3.226	94.165
7	0.309	2.809	96.974
8	0.211	1.920	98.894
9	0.091	0.825	99.718
10	0.020	0.183	99.902
11	0.011	0.098	100.000

Appendix 5: Pair-wise Analysis Data combined for both Qualitative and Quantitative (Morphology) for NTSYS.

1	49	37																																	
OG02	5	3	5	1	1	1	5	1	5	2	3	2	7	3	4	5	1	2	1	1	2	6	3	5	5	1	80.5	41.9	12.3	0.0	19.6	12.0	3.7	4.4	22.4	1.6	0.9
OG03	5	3	0	1	1	1	7	2	3	2	3	2	1	3	1	3	3	1	1	2	2	1	2	7	5	1	59.0	32.6	11.5	3.3	11.3	7.0	1.1	1.2	0.6	1.6	0.9
OG04	3	3	3	1	1	1	5	1	3	2	3	2	7	3	3	5	1	2	1	1	1	6	3	5	5	1	81.8	52.0	6.1	0	31.5	17.7	3.0	3.5	19.5	1.8	0.9
OG05	5	3	0	1	1	1	5	2	3	2	3	2	1	3	1	5	3	1	1	2	2	1	2	7	5	1	60.5	36.5	7.7	3.3	7.6	6.3	1.2	1.2	0.7	1.2	1.0
OG06	3	3	0	1	1	1	5	1	5	2	3	2	7	3	1	7	1	2	3	1	3	6	3	5	5	1	80.5	41.2	6.6	0.0	30.6	19.5	3.6	7.3	90.4	1.6	0.5
OG07	5	3	0	1	1	1	5	2	5	2	3	2	3	3	1	5	1	2	1	1	1	6	3	5	5	1	70.4	42.0	6.1	0.0	12.3	6.8	3.1	4.5	33.8	1.8	0.7
OG08	3	3	5	1	1	1	3	2	5	2	3	1	5	3	2	5	3	1	1	2	2	6	2	7	5	1	160.0	145.6	5.9	7.2	22.2	15.5	1.1	1.2	9.6	1.4	1.0
OG09	3	3	3	1	1	1	7	2	5	2	3	1	1	3	2	5	3	1	1	2	2	6	2	7	5	1	353.5	331.2	15.5	5.5	21.9	12.5	1.2	1.0	8.4	1.6	1.2
OG10	3	5	3	1	1	1	5	2	5	3	3	2	5	3	2	5	1	2	1	5	2	6	3	5	7	1	138.8	98.6	8.5	5.5	19.4	10.6	6.3	4.8	71.4	1.8	1.3
OY11	3	7	5	1	1	1	7	2	5	2	3	2	7	3	2	5	1	1	1	4	3	6	3	7	5	1	150.0	140.8	21.1	7.6	22.9	16.2	5.9	4.2	31.7	1.4	1.4
OY12	3	7	5	1	1	1	5	2	5	2	3	1	7	3	1	5	1	1	1	2	3	6	3	5	5	1	124.8	88.2	20.4	6.1	25.9	19.0	4.3	4.4	36.9	1.4	1.0
OY13	3	5	5	1	1	1	7	2	5	2	3	1	5	3	2	5	1	1	1	2	3	6	3	7	5	1	124.9	99.0	16.0	5.6	29.7	21.9	4.3	4.9	42.4	1.4	0.9
OY14	5	5	5	3	3	1	5	2	5	2	3	1	5	5	2	5	1	1	1	5	3	6	3	5	5	1	109.4	69.3	15.2	5.4	20.3	12.3	5.5	4.0	21.5	1.7	1.4
OY15	3	7	5	3	3	1	5	2	5	2	3	1	7	3	2	5	3	1	1	2	2	6	2	7	7	1	142.5	115.1	15.4	4.4	20.9	20.5	1.6	1.6	1.7	1.0	1.0
OY16	5	3	0	1	1	1	5	2	5	2	3	1	1	3	1	3	3	1	1	1	1	8	2	7	5	1	113.4	98.6	8.2	5.3	6.4	5.6	1.2	1.3	0.7	1.0	0.8
OY17	5	5	3	1	1	1	5	2	5	2	2	2	3	3	1	3	2	1	1	1	2	6	3	7	7	1	71.6	42.5	4.9	6.2	11.2	7.4	1.5	2.3	2.6	1.5	0.7
OY18	3	3	0	1	1	1	5	2	5	2	2	1	1	3	1	3	3	1	1	1	1	8	2	7	5	1	137.7	112.6	15.5	4.4	10.3	7.1	1.1	1.2	0.5	1.5	0.9
OY19	5	5	5	1	1	1	5	2	5	2	3	1	5	3	2	5	1	2	3	5	3	6	3	5	7	1	130.7	87.3	15.4	6.5	15.1	10.7	9.0	5.2	91.4	1.4	1.7
OY20	5	5	3	1	1	1	5	2	5	2	3	1	3	3	1	5	1	1	1	1	2	6	3	7	5	1	65.4	32.1	6.9	6.0	11.5	7.9	1.5	1.7	1.9	1.5	0.9
OS21	3	3	5	1	1	1	3	2	5	2	3	1	5	3	2	3	1	2	1	2	2	6	3	7	5	1	194.6	161.7	7.2	6.0	21.9	18.2	1.2	1.2	10.0	1.2	1.1
OG22	3	5	5	1	1	1	7	2	5	3	5	2	5	3	2	5	1	2	3	3	2	6	3	5	7	1	161.3	122.0	4.6	1.4	10.8	7.9	8.2	3.8	34.2	1.4	2.2
LA23	3	5	3	1	1	1	7	2	5	3	3	1	5	3	2	5	1	1	1	2	2	6	3	7	5	1	96.1	19.9	7.6	4.3	17.7	10.5	3.3	3.5	30.6	1.7	0.9
LA24	3	5	3	1	1	1	7	2	5	3	3	1	5	3	2	5	1	1	1	5	2	6	3	5	5	1	95.1	9.2	7.8	4.2	18.7	11.0	3.3	3.2	30.5	1.7	1.1
LA25	3	5	3	3	3	1	7	2	5	3	3	1	7	3	2	5	1	1	1	2	2	6	3	5	5	1	124.5	3.2	6.4	2.6	16.2	10.9	2.9	3.1	31.1	1.6	0.9

Appendix 5 (Contd.): Pair-wise Analysis Data combined for both Qualitative and Quantitative (Morphology) for NTSYS.

LA26	3	3	0	1	1	1	5	1	5	3	3	2	5	3	1	5	1	2	2	1	2	6	3	5	5	1	62.8	7.1	7.1	1.0	18.9	10.4	3.4	3.7	93.0	1.8	0.9
OY27	5	3	5	1	1	1	7	2	5	3	5	2	5	3	2	5	1	2	3	3	9	6	3	7	5	1	146.0	105.5	8.4	4.2	15.0	9.2	7.2	5.0	80.5	1.7	1.5
OY28	5	3	0	1	1	1	5	2	5	3	7	2	3	3	1	5	1	2	1	2	1	6	3	5	5	1	121.7	15.2	4.6	1.1	28.9	15.5	2.3	2.4	33.3	1.9	1.0
OY29	3	5	5	2	1	1	5	2	5	3	3	1	7	3	2	3	1	2	1	2	2	6	3	7	5	1	165.5	138.0	4.7	7.5	25.6	23.9	1.3	1.2	9.4	1.1	1.0
ED30	3	5	5	3	3	1	7	2	5	3	7	2	5	5	2	5	1	2	2	5	8	6	3	5	7	1	155.0	106.0	4.4	6.2	21.6	16.4	6.3	3.9	70.5	1.3	1.7
ED31	3	5	5	1	1	1	7	2	5	3	5	2	5	3	2	5	1	2	2	5	2	6	3	5	7	1	167.0	161.0	4.6	5.0	17.2	11.8	4.7	2.8	70.7	1.5	1.7
ED32	3	3	0	1	1	1	7	1	5	3	5	2	5	3	1	5	1	2	1	2	1	6	3	5	5	1	74.1	22.1	4.8	0.0	28.8	15.6	4.2	5.6	41.8	1.9	0.8
ED33	3	3	5	1	1	1	5	2	5	3	3	2	5	3	2	5	1	1	1	3	2	6	3	5	5	1	61.5	30.9	6.1	2.2	10.9	6.7	7.3	7.2	30.6	1.7	1.0
ED34	3	3	7	1	1	1	5	2	5	3	3	2	7	3	3	5	1	2	2	2	2	6	3	5	5	1	100.0	30.0	5.0	0.0	29.2	17.0	3.5	3.2	21.5	1.7	1.1
OD35	3	5	0	1	1	1	7	1	5	3	5	2	5	3	1	5	1	2	1	2	1	6	3	5	5	1	43.1	30.1	3.1	0.0	20.8	11.7	4.2	5.5	42.7	1.8	0.8
OD36	3	5	5	1	1	1	7	2	5	3	5	2	5	3	2	5	1	2	3	5	2	6	3	5	7	1	116.0	79.5	5.6	4.8	18.6	13.9	8.7	5.0	73.5	1.3	1.8
CR37	3	5	5	2	1	1	5	2	5	3	3	1	7	3	2	3	1	2	1	2	2	6	3	7	5	1	136.4	123.0	8.2	6.2	18.5	15.6	1.2	1.2	9.3	1.2	1.0
CR38	3	3	5	1	1	1	3	2	3	3	3	1	3	3	2	5	3	2	1	2	2	6	3	7	5	1	176.5	85.0	7.9	1.1	4.9	2.8	0.5	0.5	6.5	1.8	1.1
CR39	3	5	5	1	1	1	7	2	5	3	5	1	7	3	2	5	3	2	1	2	2	6	3	7	5	1	194.0	151.0	7.4	6.3	22.4	19.5	1.3	1.2	8.9	1.2	1.1
CR40	3	3	3	1	1	1	5	2	5	3	5	1	7	3	2	5	1	2	2	8	3	6	3	7	5	1	135.0	80.4	6.5	3.4	13.1	9.1	11.8	4.2	90.8	1.5	2.9
CR41	5	3	3	1	1	1	5	2	5	3	5	1	5	3	2	5	1	2	2	3	8	6	3	5	7	1	97.2	61.7	5.8	3.8	12.7	9.1	5.1	3.3	78.1	1.4	1.6
CR42	3	3	3	1	3	1	5	2	3	3	5	2	5	5	2	5	1	2	2	5	8	6	4	5	7	1	111.5	60.2	7.1	2.7	11.7	7.5	7.8	3.3	86.4	1.6	2.4
CR43	3	3	5	1	1	1	3	2	5	3	3	2	3	3	2	3	3	2	1	2	1	6	3	5	5	1	134.3	55.5	8.1	3.3	9.3	5.1	0.8	0.8	6.6	1.9	1.0
CR44	3	3	5	1	1	1	3	2	5	3	5	2	7	5	2	5	1	2	2	5	8	6	4	5	7	1	156.8	62.9	6.0	4.3	12.3	10.2	7.7	4.4	86.8	1.2	1.8
CR45	3	3	5	1	1	1	3	2	5	3	5	1	3	3	2	3	1	2	1	3	8	6	4	3	5	1	121.5	60.1	7.1	2.0	8.7	5.6	3.1	2.3	40.5	1.6	1.4
CR46	3	3	5	1	1	1	5	2	5	3	3	2	5	2	2	3	1	1	1	2	1	6	3	5	5	1	109.9	72.7	5.4	0.9	5.6	3.2	1.0	0.8	6.8	1.8	1.2
CR47	3	3	3	1	1	1	5	2	5	3	5	2	5	3	2	5	1	2	2	5	8	6	4	7	7	1	148.5	42.2	5.0	5.3	19.5	14.6	5.6	4.4	90.5	1.4	1.3
CR48	5	5	3	2	1	1	5	2	5	3	5	2	7	5	2	5	1	2	1	5	8	6	4	5	7	1	97.2	62.5	4.1	4.3	13.4	9.6	8.0	3.9	92.7	1.4	2.1
CR49	3	3	3	1	1	1	5	2	5	3	7	2	5	3	2	5	1	2	1	3	3	6	4	5	5	1	123.0	63.0	5.4	3.3	15.2	11.0	4.1	3.0	38.2	1.4	1.4
CR50	3	7	7	1	1	1	7	2	5	3	3	2	9	3	3	5	3	2	5	2	2	6	3	5	5	1	243.0	150.7	3.9	7.1	25.0	19.7	3.8	3.3	64.1	1.3	1.6

Appendix 6A: Qualitative Morphological Characters.

PLANT'S ID	PGH	PS	SPD	SC	PC	TOL	FD	LA	PWPLB
OG02	Intermediate	Small	Intermediate	Green	Green	Simple	Intermediate	Semi erect	Middle
OG03	Intermediate	Small	Glabrous	Green	Green	Simple	Dense	Horizontal	Bottom1/3
OG04	Upright	Small	Sparse	Green	Green	Simple	Intermediate	Semi erect	Bottom1/3
OG05	Intermediate	Small	Glabrous	Green	Green	Simple	Intermediate	Horizontal	Bottom1/3
OG06	Upright	Small	Glabrous	Green	Green	Simple	Intermediate	Semi erect	Middle
OG07	Intermediate	Small	Glabrous	Green	Green	Simple	Intermediate	Horizontal	Middle
OG08	Upright	Small	Intermediate	Green	Green	Simple	Sparse	Horizontal	Middle
OG09	Upright	Small	Sparse	Green	Green	Simple	Dense	Horizontal	Middle
OG10	Upright	Intermediate	Sparse	Green	Green	Simple	Intermediate	Horizontal	Middle
OY11	Upright	Large	Intermediate	Green	Green	Simple	Dense	Horizontal	Middle
OY12	Upright	Large	Intermediate	Green	Green	Simple	Intermediate	Horizontal	Middle
OY13	Upright	Intermediate	Intermediate	Green	Green	Simple	Dense	Horizontal	Middle
OY14	Intermediate	Intermediate	Intermediate	Green purple	Green purple	Simple	Intermediate	Horizontal	Middle
OY15	Upright	Large	Intermediate	Green purple	Green purple	Simple	Intermediate	Horizontal	Middle
OY16	Intermediate	Small	Glabrous	Green	Green	Simple	Intermediate	Horizontal	Middle
OY17	Intermediate	Intermediate	Sparse	Green	Green	Simple	Intermediate	Horizontal	Middle
OY18	Upright	Small	Glabrous	Green	Green	Simple	Intermediate	Horizontal	Middle
OY19	Intermediate	Intermediate	Intermediate	Green	Green	Simple	Intermediate	Horizontal	Middle
OY20	Intermediate	Intermediate	Sparse	Green	Green	Simple	Intermediate	Horizontal	Middle

KEY: **PGH = Plant Growth Habit; SC = Stem Colour; PS = Plant Size; PC = Petiole colour; TOL = Type of leaf; LA = leaf attitude; PWPLB = Position of widest part of the leaf blade; SPD = Stem pubescence density; FD = Foliage density.**

Appendix 6A (Contd.): Qualitative Morphological Characters.

PLANT'S ID	PGH	PS	SPD	SC	PC	TOL	FD	LA	PWPLB
OS21	Upright	Small	Intermediate	Green	Green	Simple	Sparse	Horizontal	Middle
OG22	Upright	Intermediate	Intermediate	Green	Green	Simple	Dense	Horizontal	Middle
LA23	Upright	Intermediate	Sparse	Green	Green	Simple	Dense	Horizontal	Middle
LA24	Upright	Intermediate	Sparse	Green	Green	Simple	Dense	Horizontal	Middle
LA25	Upright	Intermediate	Sparse	Green purple	Green purple	Simple	Dense	Horizontal	Middle
LA26	Upright	Small	Glabrous	Green	Green	Simple	Intermediate	Semi erect	Middle
OY27	Intermediate	Small	Intermediate	Green	Green	Simple	Dense	Horizontal	Middle
OY28	Intermediate	Small	Glabrous	Green	Green	Simple	Intermediate	Horizontal	Middle
OY29	Upright	Intermediate	Intermediate	Green with purple spots	Green	Simple	Intermediate	Horizontal	Middle
ED30	Upright	Intermediate	Intermediate	Green purple	Green purple	Simple	Dense	Horizontal	Middle
ED31	Upright	Intermediate	Intermediate	Green	Green	Simple	Dense	Horizontal	Middle
ED32	Upright	Small	Glabrous	Green	Green	Simple	Dense	Semi erect	Middle
ED33	Upright	Small	Intermediate	Green	Green	Simple	Intermediate	Horizontal	Middle
ED34	Upright	Small	Dense	Green	Green	Simple	Intermediate	Horizontal	Middle
OD35	Upright	Intermediate	Glabrous	Green	Green	Simple	Dense	Semi erect	Middle
OD36	Upright	Intermediate	Intermediate	Green	Green	Simple	Dense	Horizontal	Middle
CR37	Upright	Intermediate	Intermediate	Green with purple spots	Green	Simple	Intermediate	Horizontal	Middle
CR38	Upright	Small	Intermediate	Green	Green	Simple	Sparse	Horizontal	Bottom 1/3
CR39	Upright	Intermediate	Intermediate	Green	Green	Simple	Dense	Horizontal	Middle
CR40	Upright	Small	Sparse	Green	Green	Simple	Intermediate	Horizontal	Middle

KEY: **PGH = Plant Growth Habit; SC = Stem Colour; PS = Plant Size; PC = Petiole colour; TOL = Type of leaf; LA = leaf attitude; PWPLB = Position of widest part of the leaf blade; SPD = Stem pubescence density; FD = Foliage density.**

Appendix 6A (Contd.): Qualitative Morphological Characters.

PLANT'S ID	PGH	PS	SPD	SC	PC	TOL	FD	LA	PWPLB
CR41	Intermediate	Small	Sparse	Green	Green	Simple	Intermediate	Horizontal	Middle
CR42	Upright	Small	Sparse	Green	Green purple	Simple	Intermediate	Horizontal	Bottom 1/3
CR43	Upright	Small	Intermediate	Green	Green	Simple	Sparse	Horizontal	Middle
CR44	Upright	Small	Intermediate	Green	Green	Simple	Sparse	Horizontal	Middle
CR45	Upright	Small	Intermediate	Green	Green	Simple	Sparse	Horizontal	Middle
CR46	Upright	Small	Intermediate	Green	Green	Simple	Intermediate	Horizontal	Middle
CR47	Upright	Small	Sparse	Green	Green	Simple	Intermediate	Horizontal	Middle
CR48	Intermediate	Intermediate	Sparse	Green with purple spots	Green	Simple	Intermediate	Horizontal	Middle
CR49	Upright	Small	Sparse	Green	Green	Simple	Intermediate	Horizontal	Middle
CR50	Upright	Large	Dense	Green	Green	Simple	Dense	Horizontal	Middle

KEY: **PGH = Plant Growth Habit; SC = Stem Colour; PS = Plant Size; PC = Petiole colour; TOL = Type of leaf; LA = leaf attitude; PWPLB = Position of widest part of the leaf blade; SPD = Stem pubescence density; FD = Foliage density.**

Appendix 6B: Qualitative Morphological Characters.

PLANT'S ID	LS	LAS	LCAS	LBLo	ACLV	LHTAS	LSA	IT
OG02	Lanceolate	Acute	Green	Strong	Green	Pilose	Intermediate	Generally uniparous
OG03	Lanceolate	Acute	Green	Very weak	Green	Glabrous	Flat	Generally multiparous
OG04	Lanceolate	Acute	Green	Strong	Green	Velutinous	Intermediate	Generally uniparous
OG05	Lanceolate	Acute	Green	Very weak	Green	Glabrous	Intermediate	Generally multiparous
OG06	Lanceolate	Acute	Green	Strong	Green	Glabrous	Very convex	Generally uniparous
OG07	Lanceolate	Acute	Green	Weak	Green	Glabrous	Intermediate	Generally uniparous
OG08	Lanceolate	Acute	Light green	Intermediate	Green	Puberulent	Intermediate	Generally multiparous
OG09	Lanceolate	Acute	Light green	Very weak	Green	Puberulent	Intermediate	Generally multiparous
OG10	Ovate	Acute	Green	Intermediate	Green	Puberulent	Intermediate	Generally uniparous
OY11	Lanceolate	Acute	Green	Strong	Green	Puberulent	Intermediate	Generally uniparous
OY12	Lanceolate	Acute	Light green	Strong	Green	glabrous	Intermediate	Generally uniparous
OY13	Lanceolate	Acute	Light green	Intermediate	Green	Puberulent	Intermediate	Generally uniparous
OY14	Lanceolate	Acute	Light green	Intermediate	Main vein purple rest green	Puberulent	Intermediate	Generally uniparous
OY15	Lanceolate	Acute	Light green	Strong	Green	Puberulent	Intermediate	Generally multiparous

KEY: **LBLo = Leaf Blade Lobing; LAS = Leaf Apex Shape; LHTAS = Leaf Hairiness type on Abaxial Side; ACLV = Anthocyanin Colouration of Leaf Veins; LS = Leaf shape; LCAS = Leaf colour on the adaxial surface; IT= Inflorescence type; LSA = leaf surface attitude.**

Appendix 6B (Contd.): Qualitative Morphological Characters.

PLANT'S ID	LS	LAS	LCAS	LBLO	ACLV	LHTAS	LSA	IT
OY16	Lanceolate	Acute	Light green	Very weak	Green	Glabrous	Flat	Generally multiparous
OY17	Lanceolate	Very acute	Green	Weak	Green	Glabrous	Flat	Partly uni- & multiparous
OY18	Lanceolate	Very acute	Light green	Very weak	Green	Glabrous	Flat	Generally multiparous
OY19	Lanceolate	Acute	Light green	Intermediate	Green	Puberulent	Intermediate	Generally uniparous
OY20	Lanceolate	Acute	Light green	Weak	Green	Glabrous	Intermediate	Generally uniparous
OS21	Lanceolate	Acute	Light green	Intermediate	Green	Puberulent	Flat	Generally uniparous
OG22	Ovate	Intermediate	Green	Intermediate	Green	Puberulent	Intermediate	Generally uniparous
LA23	Ovate	Acute	Light green	Intermediate	Green	Puberulent	Intermediate	Generally uniparous
LA24	Ovate	Acute	Light green	Intermediate	Green	Puberulent	Intermediate	Generally uniparous
LA25	Ovate	Acute	Light green	Strong	Green	Puberulent	Intermediate	Generally uniparous
LA26	Ovate	Acute	Green	Intermediate	Green	Glabrous	Intermediate	Generally uniparous
OY27	Ovate	Intermediate	Green	Intermediate	Green	Puberulent	Intermediate	Generally uniparous
OY28	Ovate	Obtuse	Green	Weak	Green	Glabrous	Intermediate	Generally uniparous
OY29	Ovate	Acute	Light green	Strong	Green	Puberulent	Flat	Generally uniparous
ED30	Ovate	Obtuse	Green	Intermediate	Main vein purple rest green	Puberulent	Intermediate	Generally uniparous

KEY: **LBLo = Leaf Blade Lobing; LAS = Leaf Apex Shape; LHTAS = Leaf Hairiness type on Abaxial Side; ACLV = Anthocyanin Colouration of Leaf Veins; LS = Leaf shape; LCAS = Leaf colour on the adaxial surface; IT= Inflorescence type; LSA = leaf surface attitude.**

Appendix 6B (Contd.): Qualitative Morphological Characters.

PLANT'S ID	LS	LAS	LCAS	LBLO	ACLV	LHTAS	LSA	IT
ED31	Ovate	Intermediate	Green	Intermediate	Green	Puberulent	Intermediate	Generally uniparous
ED32	Ovate	Intermediate	Green	Intermediate	Green	Glabrous	Intermediate	Generally uniparous
ED33	Ovate	Acute	Green	Intermediate	Green	Puberulent	Intermediate	Generally uniparous
ED34	Ovate	Acute	Green	Strong	Green	Velutinous	Intermediate	Generally uniparous
OD35	Ovate	Intermediate	Green	Intermediate	Green	Glabrous	Intermediate	Generally uniparous
OD36	Ovate	Intermediate	Green	Intermediate	Green	Puberulent	Intermediate	Generally uniparous
CR37	Ovate	Acute	Light green	Strong	Green	Puberulent	Flat	Generally uniparous
CR38	Ovate	Acute	Light green	Weak	Green	Puberulent	Intermediate	Generally multiparous
CR39	Ovate	Intermediate	Light green	Strong	Green	Puberulent	Intermediate	Generally multiparous
CR40	Ovate	Intermediate	Light green	Strong	Green	Puberulent	Intermediate	Generally uniparous
CR41	Ovate	Intermediate	Light green	Intermediate	Green	Puberulent	Intermediate	Generally uniparous
CR42	Ovate	Intermediate	Green	Intermediate	Main vein purple rest green	Puberulent	Intermediate	Generally uniparous
CR43	Ovate	Acute	Green	Weak	Green	Puberulent	Flat	Generally multiparous
CR44	Ovate	Intermediate	Green	Strong	Main vein purple rest green	Puberulent	Intermediate	Generally uniparous
CR45	Ovate	Intermediate	Light green	Weak	Green	Puberulent	Flat	Generally uniparous

KEY: **LBLo = Leaf Blade Lobing; LAS = Leaf Apex Shape; LHTAS = Leaf Hairiness type on Abaxial Side; ACLV = Anthocyanin Colouration of Leaf Veins; LS = Leaf shape; LCAS = Leaf colour on the adaxial surface; IT= Inflorescence type; LSA = leaf surface attitude.**

Appendix 6B (Contd.): Qualitative Morphological Characters.

PLANT'S ID	LS	LAS	LCAS	LBLO	ACLV	LHTAS	LSA	IT
CR46	Ovate	Acute	Green	Intermediate	Green	Puberulent	Flat	Generally uniparous
CR47	Ovate	Intermediate	Green	Intermediate	Green	Puberulent	Intermediate	Generally uniparous
CR48	Ovate	Intermediate	Green	Strong	Main vein purple rest green	Puberulent	Intermediate	Generally uniparous
CR49	Ovate	Obtuse	Green	Intermediate	Green	Puberulent	Intermediate	Generally uniparous
CR50	Ovate	Acute	Green	Very strong	Green	Velutinous	Intermediate	Generally multiparous

KEY: **LBLo = Leaf Blade Lobing; LAS = Leaf Apex Shape; LHTAS = Leaf Hairiness type on Abaxial Side; ACLV = Anthocyanin Colouration of Leaf Veins; LS = Leaf shape; LCAS = Leaf colour on the adaxial surface; IT= Inflorescence type; LSA = leaf surface attitude.**

Appendix 6C: Qualitative Morphological Characters.

PLANT'S ID	CT	CC	PFS	FPCCR	FSCCR	FFC	FSU	PWF	FC
OG02	Semi stellate	White	Flattened	Light green	Orange yellow	White	Intermediate	Btw 1/4 and 1/2	None
OG03	Stellate	White	Rounded	Light green	Dark green	Light green	High	Btw 1/4 and 1/2	None
OG04	Semi stellate	White	Flattened	Dark green	Orange yellow	White	Intermediate	Btw 1/4 and 1/2	None
OG05	Stellate	White	Rounded	Light green	Dark green	Light green	High	Btw 1/4 and 1/2	None
OG06	Semi stellate	Equal white and purple	Flattened	Milk white	Orange yellow	White	Intermediate	Btw 1/4 and 1/2	None
OG07	Semi stellate	White	Flattened	Dark green	Orange yellow	White	Intermediate	Btw 1/4 and 1/2	None
OG08	Stellate	White	Rounded	Light green	Orange yellow	Light green	High	Btw 1/4 and 1/2	None
OG09	Stellate	White	Rounded	Light green	Orange yellow	Light green	High	Btw 1/4 and 1/2	None
OG10	Semi stellate	White	Obovate	Light green	Orange yellow	White	Intermediate	More than 1/2	None
OY11	Stellate	White	Ovate	Milk white	Orange yellow	White	High	Btw 1/4 and 1/2	None
OY12	Stellate	White	Rounded	Milk white	Orange yellow	White	Intermediate	Btw 1/4 and 1/2	None
OY13	Stellate	White	Rounded	Milk white	Orange yellow	White	High	Btw 1/4 and 1/2	None
OY14	Stellate	White	Obovate	Milk white	Orange yellow	White	Intermediate	Btw 1/4 and 1/2	None
OY15	Stellate	White	Obovate	Light green	Orange yellow	Light green	High	More than 1/2	None

KEY: **PFS = Predominant fruit shape; FPCCR = Fruit predominant colour at commercial ripeness; FSCCR = Fruit secondary colour at commercial ripeness; FFC = Fruits flesh colour; FSU = Fruit size uniformity; PWF = Position of widest part of the fruit; CC = Corolla colour; CT = Corolla type; FC = Fruit curvature.**

Appendix 6C (Contd.): Qualitative Morphological Characters.

PLANT'S ID	CT	CC	PFS	FPCCR	FSCCR	FFC	FSU	PWF	FC
OY16	Stellate	White	Flattened	Dark green	Purple	Light green	High	Btw 1/4 and 1/2	None
OY17	Stellate	White	Flattened	Light green	Orange yellow	White	High	More than 1/2	None
OY18	Stellate	White	Flattened	Dark green	Purple	Light green	High	Btw 1/4 and 1/2	None
OY19	Semi stellate	Equal white and purple	Obovate	Milk white	Orange yellow	White	Intermediate	More than 1/2	None
OY20	Stellate	White	Flattened	Light green	Orange yellow	White	High	Btw 1/4 and 1/2	None
OS21	Semi stellate	White	Rounded	Light green	Orange yellow	White	High	Btw 1/4 and 1/2	None
OG22	Semi stellate	Equal white and purple	Ellipsoid	Light green	Orange yellow	White	Intermediate	More than 1/2	None
LA23	Stellate	White	Rounded	Light green	Orange yellow	White	High	Btw 1/4 and 1/2	None
LA24	Stellate	White	Obovate	Light green	Orange yellow	White	Intermediate	Btw 1/4 and 1/2	None
LA25	Stellate	White	Rounded	Light green	Orange yellow	White	Intermediate	Btw 1/4 and 1/2	None
LA26	Semi stellate	More white than purple	Flattened	Light green	Orange yellow	White	Intermediate	Btw 1/4 and 1/2	None
OY27	Semi stellate	Equal white and purple	Ellipsoid	Purple black	Orange yellow	White	High	Btw 1/4 and 1/2	None
OY28	Semi stellate	White	Rounded	Dark green	Orange yellow	White	Intermediate	Btw 1/4 and 1/2	None
OY29	Semi stellate	White	Rounded	Light green	Orange yellow	White	High	Btw 1/4 and 1/2	None
ED30	Semi stellate	More white than purple	Obovate	Purple	Orange yellow	White	Intermediate	More than 1/2	None

KEY: **PFS = Predominant fruit shape; FPCCR = Fruit predominant colour at commercial ripeness; FSCCR = Fruit secondary colour at commercial ripeness; FFC = Fruits flesh colour; FSU = Fruit size uniformity; PWF = Position of widest part of the fruit; CC = Corolla colour; CT = Corolla type; FC = Fruit curvature.**

Appendix 6C (Contd.): Qualitative Morphological Characters.

PLANT'S ID	CT	CC	PFS	FPCCR	FSCCR	FFC	FSU	PWF	FC
ED31	Semi stellate	More white than purple	Obovate	Light green	Orange yellow	White	Intermediate	More than 1/2	None
ED32	Semi stellate	White	Rounded	Dark green	Orange yellow	White	Intermediate	Btw 1/4 and 1/2	None
ED33	Stellate	White	Ellipsoid	Light green	Orange yellow	White	Intermediate	Btw 1/4 and 1/2	None
ED34	Semi stellate	More white than purple	Rounded	Light green	Orange yellow	White	Intermediate	Btw 1/4 and 1/2	None
OD35	Semi stellate	White	Rounded	Dark green	Orange yellow	White	Intermediate	Btw 1/4 and 1/2	None
OD36	Semi stellate	Equal white and purple	Obovate	Light green	Orange yellow	White	Intermediate	More than 1/2	None
CR37	Semi stellate	White	Rounded	Light green	Orange yellow	White	High	Btw 1/4 and 1/2	None
CR38	Semi stellate	White	Rounded	Light green	Orange yellow	White	High	Btw 1/4 and 1/2	None
CR39	Semi stellate	White	Rounded	Light green	Orange yellow	White	High	Btw 1/4 and 1/2	None
CR40	Semi stellate	More white than purple	Elongate	Milk white	Orange yellow	White	High	Btw 1/4 and 1/2	None
CR41	Semi stellate	More white than purple	Ellipsoid	Purple	Orange yellow	White	Intermediate	More than 1/2	None
CR42	Semi stellate	More white than purple	Obovate	Purple	Orange yellow	Pale yellow	Intermediate	More than 1/2	None
CR43	Semi stellate	White	Rounded	Dark green	Orange yellow	white	Intermediate	Btw 1/4 and 1/2	None
CR44	Semi stellate	More white than purple	Obovate	Purple	Orange yellow	Pale yellow	Intermediate	More than 1/2	None
CR45	Semi stellate	White	Ellipsoid	Purple	Orange yellow	Pale yellow	Low	Btw 1/4 and 1/2	None

KEY: **PFS = Predominant fruit shape; FPCCR = Fruit predominant colour at commercial ripeness; FSCCR = Fruit secondary colour at commercial ripeness; FFC = Fruits flesh colour; FSU = Fruit size uniformity; PWF = Position of widest part of the fruit; CC = Corolla colour; CT = Corolla type; FC = Fruit curvature.**

Appendix 6C (Contd.): Qualitative Morphological Characters.

PLANT'S ID	CT	CC	PFS	FPCCR	FSCCR	FFC	FSU	PWF	FC
CR46	Stellate	White	Rounded	Dark green	Orange yellow	White	Intermediate	Btw 1/4 and 1/2	None
CR47	Semi stellate	More white than purple	Obovate	Purple	Orange yellow	Pale yellow	High	More than 1/2	None
CR48	Semi stellate	White	Obovate	Purple	Orange yellow	Pale yellow	Intermediate	More than 1/2	None
CR49	Semi stellate	White	Ellipsoid	Milk white	Orange yellow	Pale yellow	Intermediate	Btw 1/4 and 1/2	None
CR50	Semi stellate	More purple than white	Rounded	Light green	Orange yellow	white	Intermediate	Btw 1/4 and 1/2	None

KEY: **PFS = Predominant fruit shape; FPCCR = Fruit predominant colour at commercial ripeness; FSCCR = Fruit secondary colour at commercial ripeness; FFC = Fruits flesh colour; FSU = Fruit size uniformity; PWF = Position of widest part of the fruit; CC = Corolla colour; CT = Corolla type; FC = Fruit curvature.**

Appendix 7: Mean Values for Quantitative Morphological Characters

PLANT'S ID	PHF (cm)	SLFI (cm)	IL (cm)	PL (cm)	LBL (cm)	LBW (cm)	FL (cm)	FW (cm)	MFFW (g)	LBL/LBW	FL/FW
OG02	80.5	41.9	12.3	0.0	19.6	12.0	3.7	4.4	22.4	1.6	0.9
OG03	59.0	32.6	11.5	3.3	11.3	7.0	1.1	1.2	0.6	1.6	0.9
OG04	81.8	52.0	6.1	0	31.5	17.7	3.0	3.5	19.5	1.8	0.9
OG05	60.5	36.5	7.7	3.3	7.6	6.3	1.2	1.2	0.7	1.2	1.0
OG06	80.5	41.2	6.6	0.0	30.6	19.5	3.6	7.3	90.4	1.6	0.5
OG07	70.4	42.0	6.1	0.0	12.3	6.8	3.1	4.5	33.8	1.8	0.7
OG08	160.0	145.6	5.9	7.2	22.2	15.5	1.1	1.2	9.6	1.4	1.0
OG09	353.5	331.2	15.5	5.5	21.9	12.5	1.2	1.0	8.4	1.6	1.2
OG10	138.8	98.6	8.5	5.5	19.4	10.6	6.3	4.8	71.4	1.8	1.3
OY11	150.0	140.8	21.1	7.6	22.9	16.2	5.9	4.2	31.7	1.4	1.4
OY12	124.8	88.2	20.4	6.1	25.9	19.0	4.3	4.4	36.9	1.4	1.0
OY13	124.9	99.0	16.0	5.6	29.7	21.9	4.3	4.9	42.4	1.4	0.9
OY14	109.4	69.3	15.2	5.4	20.3	12.3	5.5	4.0	21.5	1.7	1.4
OY15	142.5	115.1	15.4	4.4	20.9	20.5	1.6	1.6	1.7	1.0	1.0
OY16	113.4	98.6	8.2	5.3	6.4	5.6	1.2	1.3	0.7	1.0	0.8
OY17	71.6	42.5	4.9	6.2	11.2	7.4	1.5	2.3	2.6	1.5	0.7
OY18	137.7	112.6	15.5	4.4	10.3	7.1	1.1	1.2	0.5	1.5	0.9
OY19	130.7	87.3	15.4	6.5	15.1	10.7	9.0	5.2	91.4	1.4	1.7
OY20	65.4	32.1	6.9	6.0	11.5	7.9	1.5	1.7	1.9	1.5	0.9
OS21	194.6	161.7	7.2	6.0	21.9	18.2	1.2	1.2	10.0	1.2	1.1
OG22	161.3	122.0	4.6	1.4	10.8	7.9	8.2	3.8	34.2	1.4	2.2
LA23	96.1	19.9	7.6	4.3	17.7	10.5	3.3	3.5	30.6	1.7	0.9
LA24	95.1	9.2	7.8	4.2	18.7	11.0	3.3	3.2	30.5	1.7	1.1
LA25	124.5	3.2	6.4	2.6	16.2	10.9	2.9	3.1	31.1	1.6	0.9
LA26	62.8	7.1	7.1	1.0	18.9	10.4	3.4	3.7	93.0	1.8	0.9
OY27	146.0	105.5	8.4	4.2	15.0	9.2	7.2	5.0	80.5	1.7	1.5

KEY: **PHF= Plant height at Flowering; SLFI= Stem length at 1st Inflorescence; IL= Internode length; PL= Petiole length; LBL=Leaf blade length; LBW= Leaf width length; FL=Fruit length; FW= Fruit width; MFFW= Mean Fresh Fruit weight.**

Appendix 7 (Contd.): Mean Values for Quantitative Morphological Characters

PLANT'S ID	PHF (cm)	SLFI (cm)	IL (cm)	PL (cm)	LBL (cm)	LBW (cm)	FL (cm)	FW (cm)	MFFW (g)	LBL/LBW	FL/FW
OY28	121.7	15.2	4.6	1.1	28.9	15.5	2.3	2.4	33.3	1.9	1.0
OY29	165.5	138.0	4.7	7.5	25.6	23.9	1.3	1.2	9.4	1.1	1.0
ED30	155.0	106.0	4.4	6.2	21.6	16.4	6.3	3.9	70.5	1.3	1.7
ED31	167.0	161.0	4.6	5.0	17.2	11.8	4.7	2.8	70.7	1.5	1.7
ED32	74.1	22.1	4.8	0.0	28.8	15.6	4.2	5.6	41.8	1.9	0.8
ED33	61.5	30.9	6.1	2.2	10.9	6.7	7.3	7.2	30.6	1.7	1.0
ED34	100.0	30.0	5.0	0.0	29.2	17.0	3.5	3.2	21.5	1.7	1.1
OD35	43.1	30.1	3.1	0.0	20.8	11.7	4.2	5.5	42.7	1.8	0.8
OD36	116.0	79.5	5.6	4.8	18.6	13.9	8.7	5.0	73.5	1.3	1.8
CR37	136.4	123.0	8.2	6.2	18.5	15.6	1.2	1.2	9.3	1.2	1.0
CR38	176.5	85.0	7.9	1.1	4.9	2.8	0.5	0.5	6.5	1.8	1.1
CR39	194.0	151.0	7.4	6.3	22.4	19.5	1.3	1.2	8.9	1.2	1.1
CR40	135.0	80.4	6.5	3.4	13.1	9.1	11.8	4.2	90.8	1.5	2.9
CR41	97.2	61.7	5.8	3.8	12.7	9.1	5.1	3.3	78.1	1.4	1.6
CR42	111.5	60.2	7.1	2.7	11.7	7.5	7.8	3.3	86.4	1.6	2.4
CR43	134.3	55.5	8.1	3.3	9.3	5.1	0.8	0.8	6.6	1.9	1.0
CR44	156.8	62.9	6.0	4.3	12.3	10.2	7.7	4.4	86.8	1.2	1.8
CR45	121.5	60.1	7.1	2.0	8.7	5.6	3.1	2.3	40.5	1.6	1.4
CR46	109.9	72.7	5.4	0.9	5.6	3.2	1.0	0.8	6.8	1.8	1.2
CR47	148.5	42.2	5.0	5.3	19.5	14.6	5.6	4.4	90.5	1.4	1.3
CR48	97.2	62.5	4.1	4.3	13.4	9.6	8.0	3.9	92.7	1.4	2.1
CR49	123.0	63.0	5.4	3.3	15.2	11.0	4.1	3.0	38.2	1.4	1.4
CR50	243.0	150.7	3.9	7.1	25.0	19.7	3.8	3.3	64.1	1.3	1.6

KEY: **PHF= Plant height at Flowering; SLFI= Stem length at 1st Inflorescence; IL= Internode length; PL= Petiole length; LBL=Leaf blade length; LBW= Leaf width length; FL=Fruit length; FW= Fruit width; MFFW= Mean Fresh Fruit weight.**

Appendix 8: Spectrophotometric result of DNA Quantification.

S/N	SAMPLE I.D	SPECTROPHOTOMETRIC READINGS				
		CONC. ng/ µl	A_{230}	A_{260}	A_{280}	$A_{260/280}$
1	OG02	33	0.45	0.33	0.30	1.11
2	OG03	63	0.83	0.63	0.56	1.13
3	OG04	71	0.93	0.71	0.64	1.10
4	OG05	90	1.18	0.90	0.81	1.11
5	OG06	65	0.82	0.65	0.59	1.10
6	OG07	76	0.87	0.76	0.70	1.08
7	OG08	83	1.09	0.83	0.74	1.13
8	OG09	104	1.39	1.04	0.93	1.12
9	OG10	45	0.54	0.45	0.40	1.14
10	OY11	27	0.34	0.27	0.25	1.11
11	OY12	38	0.50	0.38	0.36	1.06
12	OY13	45	0.52	0.45	0.37	1.19
13	OY14	73	0.91	0.73	0.65	1.12
14	OY15	44	0.57	0.44	0.40	1.10
15	OY16	220	-	2.20	1.96	1.12
16	OY17	23	0.23	0.23	0.16	1.44
17	OY18	24	0.32	0.24	0.22	1.13
18	OY19	101	1.28	1.01	0.91	1.11
19	OY20	109	1.40	1.09	1.01	1.08
20	OS21	4	0.068	0.044	0.028	1.56
21	OG22	3	0.022	0.033	0.019	1.72
22	LA23	6	0.061	0.056	0.038	1.48
23	LA24	3	0.063	0.032	0.022	1.44
24	LA25	2	0.036	0.025	0.015	1.62
25	LA26	1	0.021	0.014	0.009	1.55
26	OY27	4	0.109	0.040	0.025	1.60
27	OY28	3	0.068	0.033	0.022	1.51

Appendix 8 (Contd.): Spectrophotometric result of DNA Quantification.

S/N	SAMPLE I.D	SPECTROPHOTOMETRIC READINGS				
		CONC. ng/ µl	A_{230}	A_{260}	A_{280}	$A_{260/280}$
29	ED30	6	0.053	0.059	0.038	1.54
30	ED31	1	0.041	0.013	0.008	1.62
31	ED32	4	0.105	0.037	0.025	1.46
32	ED33	1	0.010	0.004	0.003	1.33
33	ED34	3	0.062	0.027	0.018	1.51
34	OD35	14	0.165	0.136	0.097	1.40
35	OD36	7	0.125	0.065	0.044	1.48
36	CR37	9	0.089	0.088	0.067	1.31
37	CR38	1	0.027	0.013	0.011	1.18
38	CR39	7	0.141	0.073	0.051	1.44
39	CR40	2	0.045	0.019	0.011	1.74
40	CR41	2	0.025	0.023	0.015	1.54
41	CR42	1	0.054	0.013	0.007	1.91
42	CR43	20	0.006	0.008	0.005	1.76
43	CR44	26	0.015	0.014	0.012	1.34
44	CR45	46.6	0.032	0.031	0.025	1.55
45	CR46	26	0.025	0.049	0.030	1.63
46	CR47	56.7	0.018	0.022	0.015	1.56
47	CR48	58	0.025	0.035	0.020	1.75
48	CR49	27	0.020	0.024	0.020	1.64
49	CR50	25.7	0.006	0.008	0.005	1.76