MORPHOLOGICAL CHARACTERIZATION AND DIVERGENCE ANALYSIS OF OKRA (*Abelmoschus esculentus (*L.) Moench*)* COLLECTIONS AT GAMBELLA, SOUTH WESTERN ETHIOPIA

M.Sc. THESIS

BY:

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MARCH, 2013

JIMMA UNIVERSITY

MORPHOLOGICAL CHARACTERIZATION AND DIVERGENCE ANALYSIS OF OKRA (*Abelmoschus esculentus (*L.*)* Moench*)* COLLECTIONS AT GAMBELLA, SOUTH WESTERN ETHIOPIA

M.Sc Thesis

Submitted to the School of Graduate Studies Jimma University, College of Agriculture and Veterinary Medicine

In Partial Fulfillment of the Requirements for the Degree of Master of Science in Horticulture (Vegetable Science)

By Mihretu Yonas

March, 2013

Jimma University

Jimma University College of Agriculture and Veterinary Medicine Department of Horticulture and Plant science

Thesis Submission for External Defense Request Form (F-07)

Name of Student: Mihretu Yonas ID No. 05565/03

Program of study: Horticulture (Vegetable science)

Title: Morphological Characterization and Divergence Analysis of Okra (*Abelmoschus esculentus (*L.)Moench*)* Collections at Gambella, South Western Ethiopia

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I dedicate this thesis to my family for nursing me with affections and love and their dedicated partnership for success in my life.

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BIOGRAPHICAL SKETCH

Mihretu Yonas Berkessa, was born on December 31 1986 at Mettu town, Ilubabor Zone of Oromia Regional State. He attended his elementary and junior secondary schools at Mettu Petros primary school and secondary at Mettu high school and preparatory school at Jimma high school. Following the completion of his preparatory education, he joined Jimma University College of Agriculture and Veterinary Medicine in 2004 and graduated with BSc. Degree in Horticulture in July, 2007. After graduation, he was employed by the Gambella Agricultural Research Institute in Plant Science Department and has been working as assistant researcher, until he joined the graduate studies program of Jimma University College of Agriculture and Veterinary Medicine a Master of Science degree in Horticulture (Vegetable Science).

ACKNOWLEDGEMENTS

All praises and thanks are to Almighty GOD, who is the entire source of all knowledge and kinds of wisdom endowed to mankind. His abundant blessing that flourished my thoughts and fulfilled my ambitions and my modest efforts in the form of this write up and made this material contribution towards the deep ocean of scientific knowledge already existing.

Conducting of this thesis research from project proposal, field work, and to the final write up of the thesis could have not been fruitful if it were not for a generous assistance of individuals and institutions. In the first place, I am profoundly indebted to my major advisor Mr Weyessa Garedew (Ass. Prof) and co-adviser Mr. Adugna Debela (Ass. Prof). for their encouragement, willingness to supervise my research and their valuable comments from early stage of proposing the research to the final thesis research results write up, which helped me in the completion of the study. So I would like to extend my deepest gratitude to them for their continuous technical support and commitment throughout my research. Without the encouragement, insight and professional expertise of my advisors, the completion of this work would not have been possible.

My special thanks go to Gambella Agricultural Research Institute for sponsoring my graduate study. I would like also to extend my special gratitude to Mrs Kassaye Tolessa (MSc.) for support given during my research work. I would like also to express my sincere gratitude to my parents; father, mother, for their dedication in bringing me up and for their strong support throughout my life and my academic career. I would like to express my deepest gratefulness for the support, for her understanding in the difficult times we had, and love that I received from my dear fiancé Seblewengel Wasihun and all her families, they have been the source of my strength and encouragement. I consider this academic achievement as a success to her as well. Finally, I owe an enormous thank to my fantastic friends. Your support and encouragement has been valuable; and I am forever grateful.

LIST OF ACRONYMS AND ABBREVIATIONS

CL	Cluster
GA	Genetic Advance
GAM%	Genetic Advance Under Percent Mean
GCV	Genotypic Coefficients of Variation
GV	Genetic Variance
H^2	Heritability in Broad Sense
IBC	Institute of Biodiversity Conservation
IPGRI	International Plant Genetic Resource Institute
GARI	Gambella Agricultural Research Institute
PCA	Principal Component Analysis
PCV	Phenotypic Coefficients of Variation
PV	Phenotypic Variance
SAS	Statistical Analysis System

TABLE OF CONTENTS

LIST OF TABLES

LIST OF TABLE IN THE APPENDIX

LIST OF FIGURES IN THE APPENDIX

Appendix Figure Page

MORPHOLOGICAL CHARACTERIZATION AND DIVERGENCE ANALYSIS OF OKRA (*Abelmoschus esculentus* (L.) Moench) COLLECTIONS AT GAMBELLA, SOUTH WESTERN ETHIOPIA

ABSTRACT

Okra(Abelmoschus esculentus (L.) Moench) is an economically important vegetable crop grown in tropical and sub-tropical parts of the world which belongs to family Malvaceae. It is apparently originated in Ethiopia. The objectives of the study were to characterize accessions based on qualitative and quantitative morphological traits and determine the range of diversity among the accessions with the ultimate goal of providing a basis for varietal improvement and conservation. Twenty five okra accessions were planted in 2011/ 2012 at Gambella in randomized complete block design with three replications. Data on eight qualitative and 20 quantitative traits were collected and subjected to various statistical analyses. Cluster analysis based on qualitative characters indicated the formation of four clusters and existence of wide variability based on their vegetative, shape, pigmentation, growth and fruit characteristics. The analysis of variance showed significant differences ($p<0.01$) among the accessions for all characters measured. Estimate of phenotypic and genotypic coefficients of variation also showed the presence of variability among the accessions for the majority of the character. Relatively high phenotypic (137.25, 118.10, and 92.08%) and genotypic coefficients of variation (106.58, 89.79 and 88.89%) were observed for days to maturity, number of primary branch and fruit length respectively. High heritability (96.76% and 96.50%) coupled with high genetic advance as percent of mean (106.32% and 97.25%) were recorded for internodes length and plant height respectively. Correlation study between various quantitative characters highlighted significant association among characters fruit yield was positive and highly significant genotypic correlation with fruit length (r= 0.74), average fruit weight (r= 0.62), fruit diameter (r= 0.61), seed per pod (r= 0.56), hundred seed weight (r= 0.68), internodes number (r=0.89, plant height (r= 0.58), and number of pod per plant (r=0.66). Path coefficient analysis at genotypic level revealed that internodes number had highly positive direct effect on fruit yield (p = 6.90) followed by average fruit weight (p = 6.89) which had positively genotypic correlation with yield. Cluster and distance analysis of quantitative characters based on multivariate analysis pointed out the existence of five divergent groups. The maximum distance was observed between cluster II and I (2846) while the minimum was between I and III (213.64). Principal component analysis indicated that six principal components explained about 83% of the total variation. Differentiation of germplasm into different cluster was because of cumulative effect of number of characters. Accession like GM7, GM9 and GH13 from Gambella collection AS4 and AS11 from Assosa collection are recommended if they promoted for the next breeding work as they are high yielder accessions compared to the others. The present study indicated a considerable amount of variability for the majority of the qualitative and quantitative characters in okra for exploitation. However, it is recommended that the experiment should be repeated at more location and years with more collections to confirm the obtained results.

1. INTRODUCTION

Okra (*Abelmoschus esculentus* (L.) Moench) is an economically important vegetable crop grown in tropical and sub-tropical parts of the world which belongs to family Malvaceae. It is apparently originated in Ethiopia, the mountainous or plateau area of Eritrea, and the eastern, higher part of the Anglo-Egyptian Sudan (Aladele *et al.*, 2008). It is widely distributed from Africa to Asia, in Southern Europea, the Mediterranean and all of the America (Oyelade *et al*., 2003).

Okra, commonly known as "lady finger", is primarily suitable for cultivation as a garden crop as well as on large commercial farms. The crop grows well in hot weather, especially in the regions with warm nights (>20 ^{0}C) (Ndunguru and Rajabu, 2004). It is sensitive to frost, water logging and drought conditions. The total area and production under okra in the world is reported to be 838.15 thousand hectare and 5,389.4 thousand tons respectively. It is mainly grown in India, Nigeria, Sudan, Pakistan, Ghana, Egypt, Benin, Saudi Arabia, Mexico and Cameroon. Largest production area is in India followed by Nigeria. Highest productivity is reported from Egypt (15.71 tons/ha) followed by Saudi Arabia (11.53tons/ha) (Jayakumar, 2002). Although there is no complete record on production area and productivity of the crop under Ethiopian condition, it has high diversity in some parts of the country particularly in the Southwestern low lands (550 to 650 m asl) region (Leipzig, 1996). For generation, farmers in Gambella and Asossa have been cultivating for its fruit and leaf to use as a food and medicine of different diseases (Uman Utow and Usman kedir, 2011; Personal communication).

Okra is a multipurpose crop due to its various uses of the fresh leaves, buds, flowers, pods and stems, pods and seeds (Schippers, 2000). It is mainly grown for its young immature fruits and consumed as a vegetable, raw, cooked or fried. Okra has a prominent position among fruit vegetables due to its multiple virtues like high nutritive and medicinal value, ease of cultivation, wide adaptability, year round cultivation, and export potential and bountiful returns.

It is a common ingredient of soups and sauces. The fruits can be conserved by drying or pickling. The leaves are sometimes used as spinach or cattle feed, the fibers from the stem for cord, the mucilage's for medical and industrial purposes, and the seeds as a substitute for coffee (Jayakumar, 2002). This vegetable crop provides an important source of vitamins and minerals (Lamont, 1999) have also reported significant levels of carbohydrate, potassium and magnesium. The seeds of okra are reported to contain between 15% and 26% protein and over 14% edible oil content (Kumar *et al.,* 2010). Okra is a repository of valuable nutrients nearly half of which is soluble fiber in the form of gums and pectins. Soluble fiber helps to lower serum cholesterol, reducing the risk of heart disease. The other half is insoluble fiber which helps to keep the intestinal tract healthy, decreasing the risk of some forms of cancer, especially colorectal cancer. Nearly 10% of the recommended levels of vitamin B6 and folic acid are also present in a half cup of cooked okra (FAO, 2004).

Genetic resources provide basic material for selection and improvement through breeding to ensure food security. Conservation and utilization of plant genetic resources are important components of ex-situ collections. Morpho-agronomical characters and diversity analysis of crops is also very essential in order to pave way for genetic improvement. It is first step in any crop improvement program (De Vicente *et al.,* 2005). Adequate characterization for agronomic and morphological traits is necessary to facilitate utilization of germplasm by breeders. Characterization of genetic resources therefore refers to the process by which accessions are identified, differentiated or distinguished according to their character. It provides information on diversity within and between crop collections. This enables the identification of unique accessions essential for curators of gene banks (Ren *et al.,* 1995).

Moreover, information obtained on genetic relatedness among genetic resources of crop plants is useful, both for breeding and for the purposes of germplasm conservation. The main advantage of conducting morphological characterization, among others is that published descriptor lists are readily available for most major crop species including okra (Hoogendijk and Williams, 2001). Morphological characterization is therefore a highly recommended first step that should be undertaken before more in-depth biochemical or molecular studies are employed in any diversity studies. (Tatineni *et al.*, 1996) proposed that a large number of

polymorphic markers are required to measure genetic relationships and genetic diversity in a reliable manner, which is relatively adequate in okra (Hoogendijk and Williams, 2001).

In applied plant breeding, however, in cases where the origin of lines is not known, selection of parents based on the geographical diversity alone may not be enough. The concept of genetic distance has been vital importance in many situations and more so in differentiating well defined populations. Genetic distance is the extent of gene difference between cultivars as measured by allelic frequency at a sample locus. Genetic divergence of two cultivars was defined as a function of their ancestry, geographic separation and adaptation to different environment.

Despite its multi-directional importance and utility, there are no improved varieties for cultivation by okra farmers in Ethiopia. Okra has been collected and maintained for long period of time by Institute of Biodiversity Conservation (IBC) and other research center like Gambella Agricultural Institute (GARI) but they are not yet characterized and their variability is not known. Moreover, there has not yet been any previous reported attempt by breeders at improving the crop in terms of developing core collections for higher yield and quality. The accessions under cultivation, over the years in the various regions across the country are landraces. However, some of these landraces are associated with challenges such as high susceptibility to diseases, pests and nematode and less productive (Sinnadurai, 1992). Therefore, it is imperative to develop improved varieties for further production and export of this vegetable crop. To develop improved varieties of crops, characterization and morphological diversity study was necessitated as an important first step, among others. So, with a view to generate database pertaining to some of the morph-agronomic traits, this study was undertaken with the following objectives:

General objective

- To characterize morphologically and analyze genetic variability among okra accessions.

Specific objectives

- Characterize okra accessions based on qualitative and quantitative morphological traits.
- Determine the range of diversity among the accessions with the ultimate goal of providing a basis for varietal improvement and conservation.

2. LITERATURE REVIEW

2.1 Origin and Distribution

Okra (*Abelmoschus esculentus* (L.) Moench) is a warm-season annual herbaceous vegetable crop grown primarily for immature fruits used in soups and stews. Okra apparently originated in Ethiopia, the mountainous or plateau area of Eritrea, and the eastern, higher part of the Anglo-Egyptian Sudan. Okra was distributed from Ethiopia to North Africa, the eastern Mediterranean, Arabia, and India. Although it has been commonly cultivated in Egypt for many hundreds of years, no sign of it has ever been found in any of the ancient monuments or relics of old Egypt (Aladele *et al.*, 2008).

The word okra is borrowed from a West African language, probably from Igbo or Akan (Weerasekar, 2006). The generic name Abelmoschus is derived from Arabic 'abu-l-mosk' (father of musk) in allusion to the smell of the seeds whereas the specific epithet means 'musk smelling'. It is also known by different name in different country. Latin binomial names for okra are *Abelmoschus esculentus* and *Hibiscus esculentus* (Kumar *et al.*, 2010), and it is commonly known as bhindi in India, krajiab kheaw in Thailand, okra plant, ochro, okoro, quimgombo, gombo, kopi arab, kacang bendi and bhindi in South East Asia. However, in Middle East it is known as bamia, bamya or bamieh and gumbo in Southern USA, and lady's finger in England (Ndunguru and Rajabu, 2004). On the other hand, in Portuguese and Angola, okra is known as quiabo, and as quimbombo in Cuba, gombo commun, gombo, gumbo in France, mbamia and mbinda in Sweden, and in Japan as okura. Lastly, it is also found in Taiwan, where it is called qiu kui (Siemonsma and Kouame, 2000). In Ethiopia the crop also has different name in different regions like 'Amula' in Gambella and 'Qenqes' or 'Sharma ' in Asosa (Uman Utow and Usman kedir, 2011 Personal communication).

2.2 Botany and Taxonomy

Okra was previously included in the genus Hibiscus. Later, it was designated to *Abelmoschus*, which is distinguished from the genus Hibiscus by the characteristics of the calyx: spatulate, with five short teeth, connate to the corolla and caduceus after flowering (Daniel, 2011). Although about fifty species have been described, eight are most widely accepted. There is significant variation in the chromosome numbers and ploidy levels in *Abelmoschus*. The

lowest chromosome number known is 2n = 56 for *A. angulosus* and the highest are close to 200 for *A. caillei*. Even within *A. esculentus*, chromosome numbers 2n = 72, 108, 120, 132 and 144 are in regular series of polyploids with n = 12 (Ikram *et al*., 2010).

Stem of okra is semi woody and sometimes pigmented with a green or reddish tinges color. It is Erect, variable in branching, with many short branches that are attached to thick semi woody stem. The stem attains height from 0.5- 2.5 m. The leaves are alternate up to 30 cm in length, rough, hairy, heart-shaped or 3-5 palmately lobed with serrated margins and linear-oblong or triangular lobes (Rai and Yadav, 2005). *Abelmoschus* species have hermaphrodite flowers which is self fertile, axialary, solitary, on short pedicle, 5mm in diameter; epicalyx of up to 10 bracteoles: calyx of 5 sepal, hairy, joined, 2-3 cm in length: corolla yellow, 5 petal, with dark red or purple base, 5-7 in length; stigmas red or dark stemen purple, 5-9; stamen numerous, un titled at base; ovary superior. They are self-compatible and show variable levels of cross-fertilization. So the breeding system is intermediate between obligate and facultative autogamy. However, the observed variability in out crossing, partly due to differences in local ecology - insects (nature, mobility and density - is correlated with all pollen arrivals (Hamon and Koechlin, 1991). The flower of okra is open at dawn, remains open all morning and closes in the middle of the afternoon. It is wilted in the evening and the petals fall the next day.

Fruit of this plant is a hairy capsule, 6-8 cm long, variable in color, white , dark green or red, ridged, round and short or pointed and pyramidal, 10-25 cm, 2-3 cm in diameter with upward pointing hairs in young stage. Okra seeds are dicotyledonous and they vary in shape; roundness, kidney or spherical with epigeal germination (Hamon and Koechlin, 1991). On average 30-80 seeds per pod , four to five millimeter in diameter, dark green to grey – black , rounded, with conspicuous caruncle ; approximately 20seed/g. The plants form a deeply penetrating taproot with dense shallow feeder roots reaching out in all direction in the upper 45cm of the soil.

2.3 Nutritional Potential

2.3.1 Food values

Almost all parts of okra plant are consumed, like fresh okra fruits are used as vegetable, roots and stem are used for clearing the cane juice and leaves and stems are used for making fiber and ropes. Okra seeds containing good quality edible oil and high protein are used to complement other protein sources. Okra has a flavor similar to asparagus and eggplant, and it's popular in much of the world, including Africa, the Middle East, Greece, Turkey, India, the Caribbean, South America and the Southern United States. It's an excellent source of fiber and pods were commonly used as source of food. It is one of plants that are known for its mucilaginous quality. Due to its mucilage it is used as soup binder which gives soup its desired slimy consistency. It is also use as binder and adhesives (Schippers, 2000). Not only okra pods were used in an okra plant but also leaves and stem of okra were often used as source of fibers and ropes.

Okra is the only vegetable crop of importance in the Malvaceae family, cotton is the most important economic crop in this family. It is used by different people in different ways. Agbo *et al.* (2008) reported that the immature pods are used as boiled vegetables; they are eaten fresh, canned or frozen. Avallone *et al.* (2008) reported that they are dried and used as soup thickeners, or used in stews. Further he also reported that the stem and mature pods produce fiber, which is used in paper making and for textile purposes. Potassium, sodium, magnesium and calcium are the principal elements in pods, which contain about 17% seeds. Presence of Fe, Zn, Mn and Ni also has been reported (Jayakumar, 2002). Fresh pods are low in calories (20 per 100 g), practically no fat, high in fiber, and have several valuable nutrients, including about 30% of the recommended levels of vitamin C (16 to 29 mg), 10 to 20% of folate (46 to 88 mg) and about 5% of vitamin A (14 to 20 RAE) (Schneeman, 1998). Both pod skin and seeds are excellent source of zinc (80 mg/g) (Cook *et al.,* 2000). Okra seed is mainly composed of oligomeric catechins (2.5 mg/g of seeds) and flavonol derivatives (3.4 mg/g of seeds), while the mesocarp is mainly composed of hydroxycinnamic and quercetin derivatives (0.2 and 0.3 mg/g of skins). Pods and seeds are rich in phenolic compounds with important biological properties like quartering derivatives, catechin oligomers and hydroxycinnamic derivatives (Brown *et al.,* 1999). These properties, along with the high content of

carbohydrates, proteins, glycol-protein, and other dietary elements enhance the importance of this food stuff in the human diet (Schneeman, 1998). Fresh okra pods are the most important vegetable source of viscous fiber, an important dietary component to lower cholesterol (Ahmed, 2003). Seven-days-old fresh okra pods have the highest concentration of nutrients (Agbo *et al.*, 2008).

2.3.2 Health value

Vegetables are important protective food for the maintenance of health and prevention of disease (Rai and Yadav, 2005). They contain valuable food ingredients, which can be successfully utilized to build up and repair the body. Okra contains vegetable mucilage that provides valuable service to the regeneration of small and large intestine flora. It improves fertile soil in the intestine, so that the micro organism can best flourish. Okra is top in the list of anti cancer vegetable (Ahmed, 2003). It also strengthens the mucous membrane and immune system. Culture of red blood cell and formation is activated and sleeping memory cells are brought into speed. The mineral content keeps blood pressure and circulation. The high mineral content keeps stable blood pressure and circulation. The plant has been used medicinally in treatment of several disorders, anti-cancer, anti-microbial and hypoglycemic activities of the plant have been reported. Okra mucilage binds cholesterol and bile acid carrying toxins dumped into it by filtering liver, thus might act as a hepato protective agent (Schneeman, 1998).

2.4. Agronomic Characteristics

2.4.1 Soil and climate

Okra can be grown in a wide range of soils. Well drained sandy to clay soils supplied with enough organic matter are good for okra cultivation. However, loose, friable and well-manure loam soils having the pH range 6.0 to 6.8 are the best. Alkaline, saline soils and soils with poor drainage are not good for this crop. Being a warm season crop it is susceptible to cold and frost. It thrives well during warm, moist season although it grows fairly well in the hottest summer. Okra is a tropical crop. Its optimal temperature for germination, growth and fruit setting is between 25^{o}C and 30^{o}C. The seeds do not germinate below 17°C. Okra flowers drop at 42°C day temperature (Amjad *et al.*, 2001). Uniform day and night temperature levels

are preferred by okra, wide difference between day and night temperatures reduces the seed yield considerably.

2.4.2 Land preparation, manure and fertilizer

Land should be thoroughly prepared by deep ploughing, harrowing, laddering etc. Manure and fertilizer of okra should be given as per availability of nutrients in the soil which is determined by soil testing. Manures and fertilizers are applied at the rate of 15 ton compost, 150 kg Urea, 100 kg TSP and 100 kg MP per hectare. The entire amount of compost, TSP and half of both urea and MP are applied during land preparation while the rest of the urea and MP are applied at three equal installments at 30,45 and 60 days of sowing (Jayakumar, 2002).

2.4.3 Seed sowing

Seed should be sown during mid June-Mid July or Mid February-mid March. Soil temperatures between 27-30°C help in quick and better seedling emergence (Schneeman, 1998). The percentage of seed germination of okra is relatively low, due to occurrence of hard seed in this plant (Luis *et al*., 2010). Seeds should be soaked in clean water for 24 hours before sowing. Seeds to be sown in lines and in small hills spacing of 60 cm between rows and 45cm between plants are to be maintained. Three seeds per hole should be sowed at the depth of three to five cm and the seedlings had thinned to one plant per stand two weeks after emergence (Rai and Yadav, 2005).

2.4.4 Irrigation

Okra is considered to grow well under drought conditions, although plant has shown reduction in yield under drought stress (Ahmad, 2003). Although okra is a drought tolerant plant, the availability of water has significant impact on okra production. It has been found that there was a linear relationship between okra production and the amount of water supplied (Batra *et al*., 1983). Okra is furrow irrigated crop throughout the growing season. If the soil has good moisture at planting, the young seedlings will grow 3 to 5 inches before irrigation is needed. The crop should be irrigated at an interval of 4-5 days in summer and whenever required in rainy season. However, if the rains are satisfactory no irrigation is required in rainy season crop. Heavy early irrigation tend to cool the soil and slow plant growth. The

plants should not be water stressed for maximum yields. During the harvest period, every other row is irrigated leaving a dry furrow for pickers to walk (Lamont, 1999).

2.4. 5 Weeds, Disease and insect pests

Okra is harvested over a long period of time and full season weed control is important. Where mechanical cultivation is necessary, it should be shallow and done only as often as required to control weeds. Organic mulches help control weeds and conserve moisture. First weeding may be done 15 to 20 days after seed sowing. Total three to four weeding are recommended up to the harvesting time (Dhall *et al.,* 2003).

This crop is attacked by a large number of fungi, bacteria, viruses, nematodes, mycoplasma and insects which are responsible for tremendous reduction in its yield and quality. The insects found on okra vary from year to year, but various beetles (flea, Japanese, blister and cucumber beetles) and worms (mostly corn earworm) are generally the most common. Inspect plants frequently and treat with an approved product if needed. The more serious okra disease pests include root knot nematode, Southern stem blight and wilt (Ahmed, 2003). A combination of crop rotation and good soil management is important for controlling these diseases. Foliage blights may occur, but generally they do not reach a level that requires treatment. Blossom blight can be serious during persistent rainy periods (Holmes and Kemble, 2009).

2.4. 6. Harvesting

The pods are ready for harvesting in about 45- 60days after seed sowings, depending up on varieties and season. Only tender fruit should be picked. Fruits are harvested 4 to 6 days after the flower has opened, and the fruits are not fibrous (fruits 2 to 4 inches long). The most widely used measure of harvest maturity is pod length and diameter. Typically, okra should be harvested when the pods are 7.5 cm to 12.5 cm long (3 in to 5 in). However, there may be a strong market demand for smaller sized okra and harvest stage should be adjusted accordingly. Due to the rapid rate of growth and development, okra should be harvested every other day to ensure pods remain within the marketable size range. Mature fruits should be

removed and discarded as they reduce the plant growth and decrease yield. Harvesting is recommended at least every other day for size and quality (Adeniji and Peter, 2005).

2.5 Characterization

Plants are living things that have morphological, structural, and functional characteristics that enables them to adapt to the habitat where they are established, interacting with changing environmental conditions. Furthermore, they have internal information system that coordinate and control all the processes pertaining to life maintenance, so that they succeed in sustaining a certain degree of permanence across space and time. Important information is believed to exit in plant genome and to express itself as morphological, structural, or functional attributes. It contained in germplasm, which therefore becomes the holder of a species entire sum of hereditary characteristics. However, it should be emphasized that, to use it germplasm should be understood in detail that is the type of attributes it possesses should be determined. The process of gaining such understanding is knows as germplasm characterization (Upadhyaya *et al*., 2008).

The word ‘characterize’ is also a synonym of ‘distinguish’, which is to mark as separate or different, or to separate into kinds, classes or categories. Thus, characterization of genetic resources refers to the process by which accessions are identified or differentiated. This identification may in broad terms, refer to any difference in the appearance or make up of an accession (Vicente *et al.*, 2005). Adequate characterization for agronomic and morphological traits is necessary to facilitate utilization of germplasm by breeders. To achieve this, germplasm accessions of all crops are characterized for morphological and agronomic traits in batches over the years. Germplasm characterization is the recording of distinctly identifiable characteristics which are heritable. This needs to be distinguished from preliminary evaluation, which is the recording of a limited number of agronomic traits considered to be important in crop improvement.

Germplasm characterization is carried out in precision fields by spaced planting under adequate agronomic conditions and plant protection. For each accession several morpho-agronomic traits are recorded using the descriptors developed in collaboration with IPGRI

(International Plant Genetic Resources Institute) (Upadhyaya *et al*., 2008). Information about a germplasm accession is essential if collections are to be effectively conserved, catalogued, and retrieved from gene banks (De Vicente *et al*., 2005). The major objectives of germplasm characterization are describing accessions, establish their diagnostic characteristics and identify duplicates. Also it helps to classify groups of accessions using sound criteria. On the other hands germplasm characterization enables once in identify accessions with desired agronomic traits and select entries for more precise evaluation and develop interrelationships between, or among traits and between geographic groups of cultivars by estimating the extent of variation in the collection.

Standard characterization and evaluation of accessions may be routinely carried out by using different methods, including traditional practices such as the use of descriptor lists of morphological characters. They may also involve evaluation of agronomic performance under various environmental conditions. In contrast, genetic characterization refers to the description of attributes that follow a Mendelian inheritance or that involve specific DNA sequences. In this context, the application of biochemical assays such as those that detect differences between isozymes or protein profiles, the application of molecular markers and the identification of particular sequences through diverse genomic approaches all qualify as genetic characterization methods (Rubenstein and Heisey, 2003).

Traits required for characterization are generally highly heritable ones which are expressed, within acceptable limits of deviation, over a range of agro-climatic conditions. This is essential because these traits are expected to help us identify an accession and may be used to monitor the identity of an accession over a number of regenerations. These generally include a number of morphological, botanical features, with little ambiguity and which can be observed easily (Sawadogo *et al.,* 2006). Characters such as leaf shape, flower color, seed coat (testa) color fall into this group. Despite the ease with which these could be recorded, there is a need to define the exact (growth stage) time to make the observation and method of recording so that it can be easily understood by the user community and other evaluators. Thus, characterization is primarily the responsibility of the genebank curator (Woolfe *et al.*, 1977)

and helps to describe the diversity in collections and assists the curator to manage these collections effectively.

2.6. Phenotypic and Genotypic Diversity

Crop diversity is the variance in genetic and phenotypic characteristics of plants used in agriculture. Crops may vary in seed size, branching pattern, in height, flower color, fruiting time, or flavor. They may also vary in less obvious characteristics such as their response to heat, cold or drought, or their ability to resist specific diseases and pests (Singh *et al.,* 2006). It is possible to discover variation in almost every conceivable trait, including nutritional qualities, preparation and cooking techniques, and of course how a crop tastes. If a trait cannot be found in the crop itself, it can often be found in a wild relative of the crop; a plant that has similar species that have not been farmed or used in agriculture, but exist in the wild (Bisht *et al*., 1995).

Phenotypic variation is the variation of the physical traits, or phenotypic characters of the organism, such as differences in anatomical, physiological, biochemical, or behavioral characteristics. Local environmental conditions can alter phenotypic characters (Thormann *et al*., 1994). According to Sawadogo *et al*. (2006), okra is characterized by diversity based on form and color of fruits and stems. Omonhinmin and Osawaru (2005) have reported wide morphological variation among accessions of okra, particularly in *A. caillei* (West African) types. There are reports of diversity studies in okra that used morphological markers (Karp *et al.,* 1997).

Genetic variation includes genetic differences between species and within species. It can be divided into inter-population diversity and intra-population diversity, and further into the diversity within an individual expressed by differences between alleles in the two chromosomes of diploid organisms (degree of individual heterozygosity) (Hazare and Basu, 2000). The existing relationships between traits are generally determined by the genotypic, phenotypic and environmental correlations. The genotypic and phenotypic co-efficient of variations (GCV and PCV) are the measures of variability among the genotypes under study. The genotypic co-efficient of variation (GCV) measuring the range of genetic variability for

different plant characters helps to compare this variability and phenotypic co-efficient of variation (PCV) indicated the interaction effect of environment on these traits (Weerasekar, 2006).

Adeoluwa and Kehinde (2001) evaluated 35 accessions of West African Okra (*Abelmoschus caillei*) and reported that: The accessions demonstrated showed wide variability for all characters evaluated. Variation was expressed in all qualitative traits studied except in leaf and petal color. Phenotypic variances were generally higher than their respective genotypic variances thus revealing the role of environmental factors. High PCV and High GCV were observed for pod yield per plant and peduncle length, respectively.

The genetic variability, correlation and path coefficients analyses were studied in 22 diverse accessions of okra by Mehta *et al.* (2006) for fruit yield and its component traits during summer 2003 and they stated that the values of PCV were higher than that of GCV values for all the seven traits indicating influence of environmental effects in expression of these traits. Dhall *et al.* (2003) observed high genotypic coefficient of variation and high phenotypic coefficient of variation for plant height, total yield per plant, marketable yield per plant, and number of fruits per plant and virus incidence. On the other hand Bendale *et al.* (2003) examined thirty okra accessions for first flowering node, pod length, pod weight, plant height, nodes per plant, intermodal length, number of branches per plant, seeds per pod, hundred seed weight, number of pods per plant and yield per plant. The phenotypic coefficient of variation for all the characters was higher than genotypic coefficient of variation. Number of branches per plant, yield per plant and number of pods per plant showed high genotypic coefficient of variation and high phenotypic coefficient of variation.

2.7 Heritability (broad sense) and Genetic Advance

The main aim of breeding program is to increase the yield (Singh, 2007). Yield is the end product of action and interaction of the vital activities of the plants throughout the life cycle. Therefore, for the improvement of crop yields by breeding is determined by the amount of heritable or genetic variation to that of non-heritable portion. Together with heritability, genetic advance gives estimates of realizable gain at a specific intensity of selection which is

an important tool in plant breeding (Haq *et al*., 2008). Therefore, heritability estimates along with genetic advance are normally more helpful in laying emphasis in selection for yield and yield components. However, the extent of improvement of a character would be dependent not only on heritability but also on the amount of variability in the population, where selection is to be made (Shahid *et al*., 2002).

Heritability is the transmissibility of characters from parents to offspring. In broad sense it is the ratio of genotypic variance to total phenotypic variance in percentage (Asish *et al.,* 2008). Heritability estimates provides information about the extent to which a particular character can be transmitted to the successive generations. On the other hand, heritability expressed as percentage of additive component of variance is referred as narrow sense heritability (Mittal and Sethi, 2004)

If heritability is 100% then phenotypic performance would be perfect indication of genotypic value (Johnson *et al*., 1955). The principal uses of heritability estimates are: to determine the relative importance of genetic effect which could be transferred from parent to offspring, to determine which selection method would be most successful to improve the character and to predict gain from selection (Poehlman and Slepper, 1995).

Knowledge of heritability of a trait thus guides a plant breeder to predict behavior of succeeding generations and helps to predict the response to selection (Shahid *et al*., 2002). It is a parameter of a population. With regard to the population, it is a constant not a variable. It is not observable but can be estimated from data. Because data are generated by a random process, the estimated heritability is a variable not a constant. In other words, different samples will generate different estimates of the heritability (Weerasekar, 2006).

Genetic advance (GA) is the improvement over the base population that can be potentially achieved from selection. It is a function of the heritability of the trait, the amount of phenotypic variation and the selection differential that the breeder uses. When high heritability is accompanied with high genetic advance, it indicates additive gene effects and selection may be effective. When low heritability accompanied with low genetic advance, it

indicates predominance of environmental effects and selection would be ineffective. High heritability with low genetic advance indicates the importance of non-additive gene effects while low heritability with high genetic advance indicates the importance of additive gene effects (Haq *et al*., 2008). Mehta *et al.* (2006), reported from his study the GCV, heritability and genetic advance as percentage of mean were higher for fruit yield, average fruit weight, plant height and fruit length in okra which might be attributed to additive gene action resulting their inheritance. Genetic Variability and Correlation Studies in Okra (*Abelmoschus escutentus* (L) Moench.) by Krushna *et al.* (2007) indicated that the estimate of high heritability (broad séance) accompanied with high-expected genetic advance for fruit yield per plant and plant height indicating the presence of additive gene action in the expression of these traits. The estimates of heritability (broad sense) were of high magnitude for green fruit yield per plant, plant height at harvest, days to maturity and number of internodes per plant indicating the major role of genotype and ultimately less environmental influence.

Paiva *et al.* (1998) conducted an experiment in 11 okra cultivars and estimated high heritability for fruit length, diameter, fruit weight, plant height and number of branches per plant. On the other study Dhall *et al*. (2003) recorded that characters like fruit length, plant height, number of fruits per plant and exhibited high heritability along with high genetic advance indicating the dominant gene action. Dhankar and Dhankar (2002) also reported that fruit yield, number of fruits per plant and plant height showed moderate to high heritability in study of two years. The genetic advance was found medium to low for all the traits which indicates that there is limited scope for improvement through selection procedures. Jaiprakashnarayan *et al.* (2004) observed high heritability coupled with high genetic advance for plant height , internodes length, number of nodes on main stem in okra. Indurani and Veerargavathatham (2003) noticed high heritability coupled with high genetic advance for characters such as plant height at first flower bud appearance, number of fruits per plant and yield per planting in okra. Relatively similar result was obtained by Singh *et al.* (2007).

2.8 Correlation

The knowledge of association of plant characters as determined by the correlation coefficient is helpful for selection of desirable characters under a breeding program. Thus measurements of correlation coefficient between characters are a matter of considerable importance in selection indices and also permit the prediction of correlated response (Weerasekar, 2006). The correlation between two traits refers to a situation where the two traits vary with each other, either positively or negatively, within a breeding population. Correlation could be due to genetic or environmental causes. The result of correlation is of great value in determining the most effective procedures for selection of superior genotypes for improvement (Mehta *et al.,* 2006).

According to Singh and Sharma (2012) knowledge of the correlation among traits serves as a guide to prevent the elimination of some useful traits at the expense of other desirable traits during selection. For example, where desirable traits are known to be negatively correlated, caution during selection would be needed to ensure a good balance of desirable traits in improved cultivars. In addition knowing correlation among traits in a breeding population serves in facilitating indirect selection for fruit yield through selection for yield components. To evaluate relationships, correlation analyses are used such that the values of two characters are analyzed on a paired basis, results of which may be either positive or negative. Ariyo (1990) has established that in developing a variety it would be difficult to exercise simultaneous selection for major yield characters when these characters are negatively correlated, but when they are positively associated, component breeding would be very effective.

The phenotypic correlation measures the degree of association of two variables and is determined by genetic and environmental factors. The environmental correlation is mainly responsible for the association of traits of low heritability. The genotypic correlation on the other hand, which represents the genetic portion of the phenotypic correlation, is the only one of inheritable nature and therefore, used to orient breeding programs (Falconer, 1996). According to Bello *et al.* (2006) the genotypic correlation coefficients on okra evaluation showed more significantly relationship between the pairs of characters than the phenotypic

correlations. This suggests that the characters are more related genotypically than phenotypically. On the other hand Singh and Singh (2006) observed high genotypic and phenotypic coefficient of variation for number of branches per plant, fruit yield per plant, plant height and fruit lengthin okra. Singh *et al.* (2007) observed high magnitude of genotypic coefficient of variation and phenotypic coefficient of variation for number of branches per plant, plant height, number of fruits per plant and fruit yield in which phenotypic coefficient of variation was higher than corresponding genotypic coefficient of variation. Gandhi *et al.* (2001) reported that the dry fruit yield was highly and significantly dependent on number of nodes per plant, intermodal length, number of fruits per plant and seed yield per plant.

On other study on okra Niranjan and Mishra (2003) observed that fruit yield was positively and significantly correlated with edibility period of fruits, number of fruits per plant, fruit length, number of seeds per fruit, fruit weight, plant height and number of branches per plant at both genotypic and phenotypic levels. Associations were significant at the genotypic levels only between edibility period of fruit and number of branches per plant. Jaiprakashnarayan and Mulge (2004) noticed that total yield per plant was positively and significantly correlated with number of fruits per plant, average fruit weight, number of nodes on main stem, fruitlength, plant height at 60 and 100 days after sowing and number of leaves at 45 and 100 days, but negatively and significantly correlated with number of locules per fruit, number of nodes at first flowering and first fruiting.

2.9 Path Analysis

It is often observed that certain quantitative characters of economic importance are associated with one another. Yield is considered to be dependent on several of its component traits. In such cases, the knowledge on association between such characters is quite helpful to plant breeders to formulate their selection strategies. (Pradip *et al.,* 2010) The study of simple correlation does not provide an exact picture of relative importance of direct and indirect influence of each of the component character towards the direct character. So, this can be overcome by following path coefficient analysis technique by further partitioning the correlation coefficient into direct and indirect effects. Assuming yield as a contribution of several characters which are correlated among themselves and to the yield, the concept of path

coefficient analysis was originally developed by Wright in 1921, but the technique was first used for plant selection by Dewey and Lu in 1959. Path analysis is simply standardized partial regression coefficient, which splits the correlation coefficients into the measures of direct and indirect effects of a set of independent variables on the dependent variable (Mehta *et al.*, 2006).

Character association and path are pre-requisites for improvement of any crop (Crawford, 1990). The estimate of path co-efficient analysis is important for better understanding of the crop. The major advantage of path analysis is that, it permits the partitioning of the correlation coefficient into its components, one component being the path coefficient that measures the direct effect of a predictor variable upon its response variable; the second component being the indirect effect(s) of a predictor variable on the response variable through another predictor variable. In agriculture, path analysis has been used by plant breeders to assist in identifying traits that are useful as selection criteria to improve crop yield (Surek and Beser, 2003). Mehta *et al.* (2006) shows Path coefficients on okra revealed that fruit girth had the maximum direct effect followed by fruit length towards fruit yield. Thus, the fruit yield in okra can be improved by selecting for higher fruit length, fruit girth and average fruit weight simultaneously. On the other study Nasit *et al.* (2009) stated that number of fruits per plant had maximum indirect contribution via fruit length and fruit weight in building strong positive association with yield.

2.10 Cluster Analysis

Clustering is the process of grouping (or clustering) objects in categories based on their common attributes or relationships. It is very useful because it allows one to visualize similarities among taxa by the levels at which they are grouped together (Crawford, 1990). To measure distance among clusters, a number of methods are available and varies according to the way in which “closest” is defined at each stage of merging groups (Hoogendijk and Williams, 2001). The first method is hierarchical clustering which is away to investigate grouping in one’s data simultaneously over a variety of scales, by creating a cluster tree. The tree is not a single set of clusters but rather a multi-level hierarchy where clusters at one level are joined to clusters at the next higher level. This allows one to decide what level or scale of

clustering is most appropriate in an application (Hastie *et al.*, 2009). On other hands, hierarchical cluster analysis highlights the nature of relationship between any types of samples described by any type of descriptors. It could serve as a basis for selection of parental types that could result to superior hybrids. The second one is non hierarchical clustering**:** These methods generally operate on units by varieties matrix and seek to partition the units into a specified number of groups to optimize some criterion. The most common criterion used is maximizing between groups sum of squares which is equivalent to minimizing within group sum of square. This method assigns each individual to a unique group by comparing it with the initial classes so that its positioning is the most appropriate.

Thirupathi *et al.* (2012) using genetic divergence analysis of indigenous and exotic collections of okra (*Abelmoschus esculentus* (L.) Moench), categorized 100 genotypes in to 11 different clusters. Patel *et al.* (2006) also grouped 26 genotypes of okra into six clusters using multivariate analysis.

2.11 Divergence (D^2 statistics)

The success of breeding program depends to a large measure on the degree of genetic divergence. Genetic diversity is of paramount importance for heterosis and it is an important factor for any heritable improvement. Divergence analysis generates valuable information on the nature and degree of genetic diversity, which is useful for selecting desirable lines from germplasm for successful breeding program (Pachiyappan and Saravanan, 2012; Patel, 2006). Selection of lines based on individual attribute may not be as advantageous as the one based on a number of important traits collectively. Multivariate analysis provides valuable information on the extent of genetic diversity present in the germplasm. Mahalanobis D^2 statistics is a unique tool for quantifying degree of divergence between biological populations at genetic level. Mahalanobis D^2 statistics is based on multivariate analysis and serves to be a good index of genetic diversity.

Multivariate analysis following Mahalanobis D^2 statistics revealed rich genetic diversity for various growth, earliness and yield associated traits in the germplasm offering a great scope for improvement of okra (Ghai *et al*., 2005; Singh *et al*., 2007; Bendale *et al*., 2003). The

existing diversity has been exploited in various breeding programs, which resulted in the development and release of a good number of varieties in okra in different countries in the world. According to Thirupathi *et al.* (2012) genetic divergence analysis following Mahalanobis D^2 statistics, revealed considerable genetic diversity among 100 genotypes of okra (*Abelmoschus esculentus* (L.) Moench) for all the seventeen quantitative characters which pertaining to the growth, earliness and yield (Hastie *et al.*, 2009).

2.12 Principal Component Analysis

Principal component analysis (PCA) is a mathematical algorithm that reduces the dimensionality of the data while retaining most of the variation in the data set. It accomplishes this reduction by identifying directions, called principal components, along which the variation in the data is maximal. By using a few components, each sample can be represented by relatively few numbers instead of by values for thousands of variables. **(**Markus, 2008**).**

According to Bisht *et al.* (2008) report, using principal component suggested that days to flowering, plant height, and various fruit characteristics were important components of variability and contributed significantly to the total variation observed in okra. On the other study by Nwangburuka *et al*. (2011), the mean contributions of plant height, days to flowering, branches per plant, fresh pod width, mature pod width, fresh pod length, pod weight per plant, pod per plant, seeds per pod, and seed weight per plant were relatively high in the principal axes confirming the major contributions of these traits to seed yield in okra.

3. MATERIALS AND METHODS

3.1. Description of Experimental Site

The experiment was conducted at Gambella which is located 777 Km from Addis Ababa in south west. The experimental site was located at the confluence of the Baro river and has a latitude of 8°15′ N and longitude of 34°35′ E with an elevation of 526 meters above sea level. Gambella is situated in the sub-humid hot to warm agro ecological zone that is well known for okra cultivation by the farmers. The experiment was conducted on alluvial soil type with a pH of 6.5. Based on ten years (2001 to 2010) meteorological data, the average annual rainfall of the study area was 1020.5 mm with a monthly mean range of 46.4 mm to 114.7 mm per year and the average annual minimum and maximum temperatures are 20.1^{o}C and 35.7^{o}C, respectively (National Meteorology Agency Gambella Branch, 2012) (Appendix Table 1, 2 and 3). The study area is characterized by a uni modal rainfall pattern which occurs from early April and extends to the end of November. On the time of the experiment the temperature was 37^{o}C with erratic type of rain fall distribution.

3.2 Experimental Materials and Design

Twenty-five okra accessions which have been collected by Gambella Agricultural Research Institute from Gambella and Asosa regions were used for the study (Table 1). The accessions were arranged in a randomized complete block design (RCBD) with three replications making a total of 75 plots. The total land area of the experiment was 520.8 m^2. Each plot was 2 m × 3.2 m (6.4 m^2), with 1 m between blocks and 0.5 m between plots. Seed sowing is done on March 20011. The spacing between plants was 45cm and between rows was 60 cm. Sawing was done in May 2011. Three seeds per hole had sowed at the depth of three to five cm and the seedlings had thinned to one plant per stand two weeks after emergence. Each plot has 5 rows and 4 plants per row making a total of 20 plants per plot. Furrow irrigation was used to control moisture stress as supplementary when there is no rain. Weeding commenced at two weeks after sowing and subsequent weeding was carried out at 7 and 12 weeks. Mankozeb at the rate of 1 L /ha was sprayed subsequently at the interval of three weeks to the completion of harvest.

Table1. *Abelmoschus esculentus* accessions and their area of collection

No	Accession name	Zone	Woreda	Local name
1	GM- 1	Agnuak	Bonga	Amula
2	GM- 2	Agnuak	Bonga	Amula
3	GM-3	Agnuak	Bonga	Amula
4	GM-4	Agnuak	Chobokir	Amula
5	GM-5	Agnuak	Chobokir	Amula
6	GM-6	Agnuak	Chobokir	Amula
7	GM-7	Agnuak	Chobokir	Amula
8	GM-8	Agnuak	Pinkyo	Amula
9	GM-9	Agnuak	Tagni	Amula
10	GM-11	Agnuak	Jawe	Amula
11	GM-12	Agnuak	Abol	Amula
12	GM-12	Nuwer	Lare	Amula
13	GM-13	Agnuak	Eley	Amula
14	GM-14	Nuwer	Itang	Amula
15	AS-1	Assosa	Abrahamo	Qenqes
16	AS- 2	Assosa	Abrahamo	Sharma
17	AS-3	Assosa	Furfur	Sharma
18	AS-4	Assosa	Furfur	Bamiya
19	AS-5	Assosa	Furfur	Sharma
20	AS-6	Assosa	Furfur	Qenqes
21	AS-7	Assosa	Afamagale	Qenqes
22	AS-8	Assosa	Kuldadine	Qenqes
23	AS-9	Assosa	Surqole	Qenqes
24	AS- 10	Assosa	Bambasi	Qenqes
25	AS- 11	Assosa	Bambasi	Sharma

Source of seed : Gambella Agriculture Inistitute

3.3 Data Collection

Both quantitative and qualitative traits were recorded from nine randomly selected plants from the three middle rows using International Plant Genetic Resources Institute (IPGRI, 1991) descriptor list for okra species were used to record and describe both the quantitative and qualitative traits. Based on the descriptor the following quantitative and qualitative traits were recorded.

3.3.1 Quantitative traits

1. Days to emergence - number of days from sowing to 50 % seedling emergence was recorded.

2. Days to 50 percent flowering - The number of days taken from the date of sowing to the day on which 50 percent of the population flowered was recorded.

3. Leaf length (cm) – was measured form the attachment of the base of the leaf and petiole to the tip of the leaf using ruler.

4. Leaf width (cm) – were measured at the widest part of the leaf using ruler.

5. Internodes length (cm) - The length of the internodes between the fifth and sixth node was measured at time of maturity before harvest.

6. Stem diameter (mm) - Stem diameter at the basal region of plants was measured using vernier calipers at full maturity stage.

7. Peduncle length (cm) - Pedicel length of the five fruits from nine plants prior to picking was measured at fully matured stage.

8. Number of epicalyxes - The number of epicalyxes flowers of five samples from nine plant at flowering stage from each plot was counted.

9. Number of primary branches per stem - The total number of primary branches per plant was counted at final picking and average of nine plants was taken.

10. Days to maturity - The number of days from sowing to the date of first harvest was recorded from nine sample plants selected from central rows.

11. Plant height (cm) - The height of the plant from the ground level to the tip was measured for nine sample plants and average was computed at the time of final harvest**.**

12. Fruit length (cm) - The length of five tender fruits from nine plants was measured from the base of calyx to the tip of the fruit.

13. Fruit diameter (mm) - Diameter of the five tender fruits from nine plants was measured with the help of a vernier caliper at the center of the fruit.

14. Average fruit weight (g) - Each of five tender fruits from nine plants after harvest was weighed using sensitive balance.

15. Number of pod per plant- fruits of individual plants from central rows for each plot at each harvest was recorded.

16. Number of internodes- The total number of internodes per plant was counted at final picking and average of nine plants was taken.

17. Number of ridges on fruit - Determined by counting the number of ridges in ten randomly selected pods from total harvest of the plot.

18. Number of seeds per pod - Fully matured and dried ten fruits per nine plants were harvested and the number of seeds per fruit was recorded.

19. Hundred seed weight (g) - 100 seed weight of dried seeds from total harvest of the plot was recorded using sensitive balance.

20. Yield per plot (kg) - Weight of tender fruits from each plot harvest was recorded and finally totaled.

3.3.2 Qualitative traits

1. Plant habit – this was identified whether the plant 1) Densely branched at apex (DBA), 2) Densely branched base (DBB) 3) branched all over (DBO)

2. Flower color - Red coloration of petal base was identified whether it is 1) inside only or 2) both side.

3. Leaf color – is assessed from that of lamina and ribs 1) Totally green, 2) green with red vein.

4. Leaf Petiole color – 1) Green, 2) Red above but green below part of the petiol, 3) Red on both sides

5. Pod color – Main color of the pod will be observed at harvesting stage. 1) Green 2) red

6. Stem color – 1) Green, 2) Green with red pach 3) red or purple.

Color chart was used for all color identification of pod, leaf, stem and petiol. (http://w3schools.com/html/html_colorfull.asp).

7. Shape of leaf- 2) heart-shaped 3) broadly ovate 4) star shaped (palmately lobed) 7) palmately lobed with serrated margins and linear-oblong or triangular lobes.

8. Position of fruit on main stem- The position of fruits on the main stem of the accessions was five distinct variations; **1**) Erect, 2) Intermediate, 3) Horizontal, 4) Slightly falling, 5)Totally falling.

3.4 Data Analysis

3.4.1 Analysis of variance (ANOVA)

Data of quantitative characters were subjected to analysis of variance (ANOVA) using SAS version 9.2 (SAS, 2008) to examine the presence of statistically significant differences among accessions for the characters measured. Assumption of ANOVA was checked. Divergence analysis (D^2), Path coefficient analysis, phenotypic coefficient of variation (PCV), genotypic coefficient of variation (GCV), broad-sense heritability (h^2) and expected genetic advance as percentage to mean (GAM) was carried out by Genres software (GenRese, 1994) Least Significant Difference (LSD) was employed to identify genotypes that are significantly different from each other.

The model for RCBD design:

$$Y_{ijk}=\mu +\tau_i+ +\beta_j + \delta_{ij} + \varepsilon_{ijk}$$

Where $Y_{ijk\ the\ j^{th}}$ observation of i^{th} treatment

Where

μ is the population mean δ_{ij} replicate, is the sampling error, and

τ_i is the treatment effect of the i^{th} treatment, ε_{ijk} is the random error

β_j is the rep effect of the j^{th}, replicate,

3.4.2. Estimation of Genotypic and Phenotypic Coefficient of Variation

The variability of each quantitative trait was estimated by simple measures such as mean, range, standard deviation, phenotypic and genotypic variances, and coefficients of variation. The phenotypic and genotypic coefficients of variation were computed using the formula suggested by Burton and de Vane (1953) as follows.

$$\text{Genotpic variance} = \frac{MS_t - MS_e}{\bar{X}}$$

Where, $\sigma^2 g$ = genotypic variance

MSt = mean square of treatment

MSe = mean square of error

r = number of replications

$$\text{Phenotipic variance}(\sigma 2p) = \delta_g^2 + \delta_e^2$$

Where, $\sigma^2 p$ = Phenotypic variance

$\sigma^2 g$ = Genotypic variance

$\sigma^2 e$ = Environmental variance

$$\text{Phenotypic coefficient of variation (PCV)} = \frac{\delta_p^2}{\bar{x}} * 100$$

$$\text{Genotipic coefficient of variation (GCV)} = \frac{\delta_g^2}{\bar{x}} * 100$$

$\bar{x} =$ Population mean of the character being evaluate

3.4.3. Estimation of heritability and genetic advance

Heritability in broad sense

Broad sense heritability values were estimated based on the formula of Falconer and Mackay (1996) as follows:

$$H^2 = \frac{\delta_g^2}{\delta_p^2} * 100$$

Where, H^2 = heritability in the broad sense.

$\sigma^2 p$ = Phenotypic variance

$\sigma^2 g$ = Genotypic variance

Expected genetic advance under selection (GA)

Genetic advance in absolute unit (GA) and percent of the mean (GAM), assuming selection of superior 5% of the genotypes was estimated in accordance with the methods illustrated by Johnson *et al*. (1955) as :

$$GA = K * \delta^2 * h^2$$

Where, GA= Genetic advance

δ^2= the phenotypic standard deviation on mean basis;

H^2 = heritability in broad sense

k = the standardized selection differential at 5% selection intensity (K = 2.063).

Genetic advance as percent of mean

Genetic advance as percent of mean was estimated following the procedure of Johnson *et al.* (1955).

$$GAM = \frac{GA}{\bar{X}} * 100$$

Where, GAM= Genetic advance percent of the mean

GA= Genetic advance

$\bar{x}$ = Population mean of the character being evaluate

3.4.4. Phenotypic and genotypic correction coefficient analysis

Phenotypic correlation (r_p), the observable correlation between two variables, which includes both genotypes and environmental components between two variables, were estimated using the formula suggested by Johnson *et al*. (1955) and Singh and Chaudhury (1987).

$$rp = \frac{Pcovxy}{\sqrt{(Vpx.Vpy)}}$$

$$rg = \frac{Gcovxy}{\sqrt{(Vgx.Vgy)}}$$

Where, r_p = Phenotypic correlation coefficient

r_g = Genetoypic correlation coefficient

$Pcov_{xy}$ = Phenotypic covariance between variables x and y

$Gcov_{xy}$ = Genotypic covariance between variables x and y

V_px= Phenotypic variance for variable x

$V_g x$ = Genotypic variance for variable x

$V_p y$ = Phenotypic variance for variable y

$V_g y$= Genotypic variance for variable

3.4.5. Path coefficient analysis

The direct and indirect effect of yield related traits on yield per plot were worked out through path coefficient analysis. The analysis was made following the method suggested by Dewey and Lu (1959). The formula was as follows.

$$Rij = Pij + \Sigma rikpk$$

Where,

rij = Mutual association between the independent character (i) and dependent character, grain yield (j) as measured by the correlation coefficients.

Pij = Components of direct effects of the independent character (i) as measured by the path Coefficients and

Σrikpk j = summation of components of indirect effect of a given independent character (i) on a given dependent character (j) via all other independent characters (k).

The contribution of the remaining unknown factor was measured as the residual factor (PR), which is calculated as: $\sqrt{1 - R2}$ Where: - R2 =Σpij rij. The magnitude of P_R indicates how best the causal factors account for the variability of the dependent factor (Singh & Chaudhary, 1987). That is, if PR value is small (for instance, nearly zero), the dependent character considered is fully explained by the variability in the independent characters, where as higher PR value indicates that some other factors which have not been considered, need to be included in the analysis to account fully the variation in the dependent character .

3.4.6. Cluster analysis

Clustering was performed for 20 quantitative and 8 quantitative characters using the proc cluster procedure of SAS version 9.2 (SAS institute, 2008) by employing the method of average linkage clustering of hierarchical clustering called Un-weighted pair group methods with arithmetic average (UPGMA). The numbers of clusters were determined by following the approach suggested by Copper and Miligan (1988) by looking in to three statics namely Pseudo F, Pseudo t^2 and cubic clustering criteria.

3.4.7. Genetic divergence

Genetic divergences between clusters were calculated using the generalized Mahalanobis's D^2 statistics (Mahalanobis, 1936) using the equation: Squared distance (D_2) for each pair of genotype combinations was computed using the following formula:

$$D_{ij}^2 = (X_i - X_j)S^{-1}(X_i - X_j)$$

Where, D^2_{ij} = the square distance between any two genotypes i and j,

X_i and X_j = the vectors for the values for genotype i_{th} and j_{th} genotypes, and

S^{-1} = the inverse of pooled variance covariance matrix.

The D^2 values obtained for pairs of clusters were tested for significance at 0.05level of significance against the tabulated values of for p degrees of freedom, where p is the number of variables considered (Singh and Chaudhary, 1987).

3.4.8. Principal component analysis

Principal component analysis was performed using correlation matrix by employing SAS procedure (SAS, 2008

The principal components were derived as follows, suppose X^T = X1, Xp is a p dimensional random variable with mean μ and co - variance matrix ∑. Then

$Y1= a_{1j}x_{1+} a_{2j}x_{2+}$ ------- $a_{pj}x_{p} = a^T_j x$ Yj= Y1+Y2+Yp are principal components

$a_j^T = a_{1j}$……apJ are vector constants (Eigen vectors) $a_j^T a_j = \sum a^2_{kj}=1$

Var (yi) =Var ($a_1^T x$)= a ti $\sum a_1$

Where, X is a character trait, A is a coefficient (Eigen vector), Y is principal component, P is number of characters and J is number principal component.

4. RESULTS AND DISCUSSIONS

4.1 Morphological characterization based on qualitative characters

4.1.1. Frequency distribution based on qualitative characters

4.1.1.1 Growth characteristics

In the present study the frequency distribution of growth characters were recorded. Accordingly, 28%, 32% and 40 % of the accessions were recorded for branching position at main stem (general growth appearance), for densely branched at apex (DBA), densely branched at the base (DBB) and densely branched all over the main stem (DBO) respectively (Table 2). On the other hand, there were differences observed on the arrangement of position of fruit on main stem which means 32% showed erect, 48% intermediate, 20% of the accessions shows slightly falling arrangement respectively (Table2).

4.1.1.2 Leaf shape characteristics

Out of the four phenotypic classes of this character, largely palmate lobed, broadly ovate and star shaped leaf were found predominant position among the okra accessions (Table2). Of the test accessions, 28% produced leaves with the shape scored largely palmelate lobed shape, 36% had leaves with the shape broadly ovate and 36% of the accessions had leaves with star shaped.

4.1.1.3 Pigmentation characteristics

The position of red color on the deepest part of flower petal of the accessions was also assessed and 36 % of them showed red coloration at both sides and 64% showed only inside part (Table 2). Moreover, the results of the study showed two distinct leaf colors. The dominant leaf color was green (72 %) and some accession (28%) had green leaf with red veins. The results of the study depicted two distinct stem colors; green and green with red patch. In all, 84% of the accessions had their stems tinged green and a small (16%) of the accessions were tinged green with red patch. Petiole color showed two distinct colors that are green and green-with-red veins petiole colors representing 72% and 28% respectively among the okra accessions. Furthermore, the variability in qualitative characters exhibited by okra collection point out that, in the studied germplasm, a good possibility exists of finding a range

of desirable traits to meet demands for specific attributes requested by researchers, farmers and consumers.

Previously Sekyere *et al.* (2011) reported Branching position-at-main-stem , Fruit color, Fruit pubescence, fruit shape (form) and fruit position on the stem showed variability in his study of Characterization of okra (*Abelmoschus spp.* L.) germplasm based on morphological characters in Ghana. In view of the above, it may be reasonable to state the above stated morphological characters could be considered as the possible morphological markers for selection of okra.

Table 2. Frequency distribution of eight qualitative characters among accessions of okra studied at Gambella in 2011/2012

Qualitative character	Code	Frequency (%)
Leaf color	1- Green	28
	2- Green with red vein	72
Leaf shape	2 – hearted (cordate)	0
	3 – broadly ovate	36
	4- star shaped (palmate lobed)	36
	7-largely palmate lobed	28
Flower Red coloration	1- inside only	64
	2- both side	36
Petiol color	1- green	72
	2- red above and green bellow	28
	3- red	0
Stem color	1- green	84
	2- green with red patch	16
	3- red or purple	0
Fruit color	1- yellowish green	40
	2- green	60
	3- green with red patch	0
Branch habit	1-Densely branched at apex (DBA)	28
	2- Densely Branched Base (DBB)	32
	3- Branched all over (DBO)	40
Position of fruit on the main stem	1- Erect,	32
	2- Intermediate	48
	3- Horizontal	0
	4- Slightly falling	20
	5- Totally falling	0

4.1. 2. Cluster analysis using qualitative traits

Grouping of accessions based on their homogeneity is crucial**.** In the present study, this approach was adopted and the accessions of okra were clustered into four distinct groups based on eight qualitative characters (Table 3, Appendix figure2).The characters used for clustering were branching habit, leaf color, leaf petiole color, pod color, stem color, leaf shape, red coloration of petal base and position of fruit on the main stem. In similar study Sekyere *et al.* (2011) grouped 25 accessions into four main sub-cluster groups of okra accessions at a coefficient of 0.63. Accessions were put into cluster groups based on certain qualities unique to them.

Table 3. Clusters of *Abelmoschus esculentus* accessions based on qualitative characters studied at Gambela in 2011/2012

Cluster	Number of accessions	Accessions name	Major characteristics
I	2	GM2, and GM5	LC= Green with red vein; LS= broadly ovate; PTC= red above with green below; FC= green; SC= Green with red patch; RCL = both side; BH= densely branched at apex and PF= intermediate.
II	16	GM1, GM6, GM9, GM11, GM12, GM13, GM14, AS1, AS2, AS3, AS5, AS6, AS7, AS8, AS9 and AS10	LC=; green; LS= broadly ovate and palmate lobed; PTC= green; FC green and yellowish green fruit; SC= green; RCL = both sides and inside only; BH=densely Branched Base, Branched at apex and Branched all over the main stem and PF= erect and intermediate.
III	5	GM3, GM7, GM10, AS4 and AS11	LC= green; LS= largely palmate lobed; PTC= green; FC= green; SC= green; RCL = both sides and inside only; BH=Densely branched at apex and Branched all over the main stem; PF= slightly falling.
IV	2	GM4and GM8	LC= green with red vein; LS= largely palmate lobed; PTC= green; FC= red above with green below petiole; SC= green with red patch; RCL = both side; BH= Densely branched all over the main stem, and PF= intermediate.

LC= leaf color; LS= leaf shape; PTC= petiole color; FC= fruit color; SC=Stem color; RCL = flower red coloration; BH= branching habit and PF= position of fruit on the main stem.

The number of accessions belonging to each cluster varied from two in clusters I and IV to 16 in cluster II. The two accessions grouped under cluster I were both from Gambella collection. They typically possess Green with red vein leaf, red above with green below petiole, green with red patch stem, green fruit, the red coloration in the deepest part of the flower petal showed both side, broadly ovate leaf, Densely Branched at apex stem and had intermediate type of fruit position on the main stem. Cluster II was the largest and consisted of 16 accessions, seven from Gambella, and nine from Assosa collections. Accessions grouped under this cluster have predominantly green in leaf, stem and petiole color, green and yellowish green fruit, the red coloration in the deepest part of flower petal showed both sides and inside only, broadly ovate and palmate lobed leaf, Densely Branched Base, Branched at apex and Branched all over the main stem and had both erect and intermediate type of fruit position on the main stem.

Likewise, Cluster III which comprised 5 accessions, three from Gambella, two from Asossa collections. Accessions falling in to this cluster were characterized by green leaf, stem, petiole and fruit, the red coloration in the deepest part of the flower petal showed inside only as well as both side, largely palmate lobed leaf shape, Densely branched at apex and Branched all over the main stem and had slightly falling type of fruit position on the main stem and had slightly falling type of fruit position on the main stem. Two accessions categorized under cluster IV which are from Gambella collections had Green with red vein leaf, red above with green below petiole, green with red patch stem, green fruit, the red coloration in the deepest part of flower petal showed both side, densely branched all over the main stem and had intermediate type of fruit position on the main stem. In general, the study based on qualitative traits confirmed the presence of considerable diversity, which can be exploited in the genetic improvement and germplasm management of the crop.

4.2. Analysis of Variance (ANOVA)

The result of analysis of variance (ANOVA) of 20 quantitative characters for the 25 accessions showed bellow (Table4, Appendix Table 4). Mean square values of all morphological attributes showed highly significant differences ($p<0.01$) in all the traits**.**

Table 4. Analysis of variance for 20 quantitative characters of *Abelmoschus esculentus* accessions studied at Gambella in 2011/2012

SV	DF	DEM	DFF	DM	LL	LW	STD	INL	NBR	PDL	FL
Replication	2	0.28	0.04	0.57	2.97	0.84	4.41	0.65	0.28	0.01	0.74
Accessions	24	1.50**	66.77**	258.25**	46.01**	48.57**	52.76**	11.35**	35.02**	3.95**	284.63**
Error	48	0.29	65.10	1.37	1.20	5.32	2.39	0.51	0.29	0.09	3.36

Continued

SV	DF	FW	FD	NRG	SPP	HSW	NEP	INN	PH	NPP	FY/P
Replication	2	6.47	7.5	0.09	26.45	0.33	0.05	3.77	523.69	4.12	4.74
Accessions	24	4532.83**	362.88**	0.38**	390.49**	1.80**	0.47**	69.73**	7315.08**	80.23**	59.55**
Error	48	12.55	4.22	0.07	22.52	0.43	0.13	5.46	626.44	6.03	2.99

*Significant 0.05 probability level; **= highly significant at 0.01 level of probability level.

DF= degree of freedom; DEM= Days to emergence; DFF= Days to 50% flowering; DM=Days maturity; LL= Leaf length (cm); LW= Leaf width (cm); STD= Stem diameter (mm); INL= Internodes length (cm); NBR= number of primary branch; PDL= Peduncle length (cm); FL= Fruit length (cm); FW= Average fruit weight (g); FD= Fruit diameter (mm); NRG= Number of ridge; SPP= Seed per pod; SW=hundred seed weight (g); NEP= Number of epicalyxes; INN= Internodes number; PH= Plant height (m); NPP= Number of pod per plant; FY/P= Fruit yield per plot (Kg)

4.3. Mean and Range

Estimated range, mean, and standard error of the mean are presented in Tables 5. Wide range of variation in character studied was observed. The highest value was almost two times of the minimum value for number of seed per pod, three fold for number of pod per plants, fivefold for fruit diameter and six fold for fruit yield per plot. The highest fruit yield (18kg/plot) was recorded from accession AS11, while low yield of (3 kg/plot) was obtained from AS3 which is found in the third qualitative and fourth quantitative clustered groups

Table 5 Means, standard errors and ranges for 20 quantitative characters in okra studied at Gambella in 2011/2012

Quantitative character	Mean	SE	Range
Days to emergency	5.56	± 0.5	4-7
Days to 50% flowering	55.72	±1.0	49-64
Days to maturity	90.51	±1.1	82-106
Leaf length	30.68	±1.1	24-41
Leaf width	39.29	±2.3	35-43
Stem diameter	30.73	±1.5	22-39
Internodes length	6.28	±0.2	4-12
Number of primary branch	5.37	±0.7	2-13
Peduncle length	4.34	±0.2	22-42
Fruit length	24.89	±1.4	11-45
Average fruit weight	73.10	±1.4	12- 150
Fruit diameter	25.11	±1.2	9-45
Number of ridge	7.78	±0.2	7-8
Seed per pod	98.98	±1.6	79-114
100 seed weight	59.43	±0.4	5-8
Number of epicalyxes	9.31	±0.1	9-10
Internodes number	26.39	±0.6	17-36
Plant height	66.14	±7.4	42-96
Number of pod per plant	20.94	±0.5	11-30
Fruit yield (kg/plot)	8.86	±1.4	3-18

4.4. Estimates of Genetic Parameters

4.4.1 Estimates of variance components

Generally characters or agronomic traits are highly influenced by the environment. In this study, characters were reacting in different way i.e. the magnitude of response was quite different as it was measured by the phenotypic coefficient of variation. According to Deshmukh *et al*. (1986) PCV and GCV values greater than 20% are regarded as high, whereas values less than 10% are considered to be low and values between 10% and 20% to be medium.

In this typical study the highest GCV was recorded for the quantitative traits like days to maturity (89.79), stem diameter (21.08), internodes length (52.47), number of primary branches (106.58), peduncle length (34.61), fruit length (65.6), average fruit weight (88.89), fruit diameter (74.25) plant height (48.04), number of pod per plant (42.34) and fruit yield (44.35)(Table 6). On the other hand, the highest phenotypic coefficients of variation (PCV) was recorded for parameter like days to emergence (32.07), days to maturity (137.25), leaf length (27.69), leaf width (23.24) internodes length (53.33), number of primary branch (118.09), peduncle length (62.45), fruit length (69.62), fruit width (92.07), seed per plant (22.36), hundreds seed weight, inter node number (31.09), plant height (48.91), number of pod per plants (46.12) and for the fruit yield(49.55). This view is also in agreement with the observation of Bendale *et al.* (2003) who observed number of branches per plant showed both high genotypic and phenotypic coefficient of variation and with finding of Adeoluwa and Kehinde (2001) who reported high PCV and high GCV value for pod yield per plant and peduncle length, respectively. This implied that, selection for these characters would be more effective as compared to those having lower PCV and GCV values.

The lowest GCV were recorded for character like number of ridge (5.987) and number of epicalyxes (5.06), where as the lowest PCV were recorded for only number of epicalyxes (8.89) indicating a narrow range of variability for these characters and restricting the scope for selection for the crop improvement. Characters showed medium GCV values are days to emergence (14.87), days to fifty percent flowering (10.40), leaf length (18.71) and leaf width (13.63). As to the medium PCV value, characters like days to fifty percent flowering (17.79) and number of ridge. (11.92),

In this study, wide variations between PCV and GCV were observed in most of the characters like days to emergency, days to maturity, leaf length, leaf width, peduncle length, stem diameter, number primary of branch. (Table 6), indicating high pressure of the environment on these characters and less effect of genetic factors.

In general, the study confirmed that the okra accessions collected from Gambella and Asossa and used in the current study were phenotypically and genotypically diverse. This indicates the existence of large diversity in okra for quantitative characters implying the need for further collection of germplasm from the untouched geographical areas of the country to broaden the genetic base for future breeding program.

4.4.2 Estimates of heritability and expected genetic advance

Heritability estimates ranged from 21.49 for days to emergency to 96.76 for internodes length. Maximum heritability was obtained from internodes length followed by plant height, inter node number, average fruit weight and fruit diameter. Therefore, the above characters may respond effectively to selection pressure. On the other hand, days to emergence, number of ridge, peduncle length, number of epicalyxes and number of seed per pod had relatively low heritability (Table 6) indicating limited possibility of improvement of these characters via selection. The other parameters showed moderate heritability estimates. Singh (1990) observed that if heritability of a character is very high around 80% or more, selection for such a character should be fairly easy. This is because there would be a close correspondence between the genotype and phenotype due to relatively small contribution of the environment to the phenotype. But for characters with low heritability, say less than 40%, selection may be considerably difficult or virtually impracticable due to the masking effect of the environment on the characteristics of germplasm. In support to the findings of this investigation, Paiva *et al.* (1998) reported high heritability for fruit length, diameter, fruit weight, plant height and number of branches per plant. On the other study Dhall *et al*. (2001) recorded that characters like fruit length, plant height and number of fruits per plant exhibited high heritability.

The values of genetic advance for different characters of okra accessions in this particular were different. Genetic advance as per cent mean was categorized as high (≥20%), moderate (10-20%) and low (0-10%) (Johnson *et al.*, 1955) These values are also expressed as percentage of the accession mean for each character so that comparisons could be made among various characters, which had different units of measurement. Accordingly, the result indicated that the progress that could be expected from selection of accessions ranged from 5.94 % for number of epicalyxes to 198.15% for number of primary branches (Table 6). High heritability coupled with high genetic advance is an important instrument for ensuing selection of the best individuals. In this study, high heritability along with high genetic advance as percent of the mean was obtained for internodes length, number of primary branch, number of pod per plant, fruit length, average fruit weight, fruit diameter, plant height and fruit yield (Table 6).

High GCV along with high heritability and high genetic advance will provide better information than single parameters alone (Ikram *et al.*, 2010). Hence, in this study, number of branch, fruit length, average fruit weight, fruit diameter, plant height and fruit yield exhibited high genotypic coefficients of variation, high heritability together with high genetic advance as percent of means. These results indicate that these characters are controlled by additive gene action; phenotypic selection for the improvement of these characters may be effective.

Table 6. Estimates of components of variance, PCV, GCV, heritability and genetic advance for 20 quantitative characters in okra studied at Gambela in 2011/2012

Quantitative characters	$\delta^2 g$	$\delta^2 p$	GCV	PCV	H^2 (%)	GA	GA (%)
Days to emergence	0.68	3.17	14.87	32.07	21.49	0.79	14.20
Days to 50% flowering	45.07	110.17	11.38	17.79	40.91	8.85	14.99
Days to maturity	8452.59	19748.28	89.79	137.25	42.80	123.91	121.01
Leaf length	32.93	72.17	18.71	27.69	45.63	7.99	26.03
Leaf width	28.53	82.97	13.63	23.24	34.39	6.45	16.46
Stem diameter	41.90	74.31	21.08	28.08	56.39	10.01	32.62
Internodes length	10.92	11.28	52.47	53.34	96.76	6.70	106.32
Number of primary branch	32.04	39.34	106.58	118.10	81.45	10.52	198.15
Peduncle length	2.26	7.35	34.61	62.45	30.72	1.72	39.52
Fruit length	261.24	294.27	65.60	69.62	88.78	31.37	127.32
Average Fruit weight	4120.54	4421.47	88.89	92.08	93.19	127.66	176.77
Fruit diameter	337.46	368.14	74.25	77.55	91.67	36.23	146.43
Number of ridge	0.22	0.86	5.99	11.92	25.24	0.48	6.20
Seed per pod	373.73	493.38	19.46	22.36	75.75	34.66	34.89
100 seed weight	1.28	1.94	16.68	20.58	65.66	1.89	27.84
Number of epicalyxes	0.23	0.69	5.07	8.89	32.45	0.56	5.94
Internodes number	70.52	74.81	31.90	32.86	94.27	16.80	63.81
Plant height	6152.09	6375.03	48.04	48.91	96.50	158.73	97.23
Number of pod per plant	78.03	92.61	42.34	46.12	84.25	16.70	80.05
Fruit yield	51.02	58.81	44.35	49.55	86.77	13.71	161.85

4.5. Association among Characters

Estimates of phenotypic and genotypic coefficients between each pair of characters are presented in Table7. The magnitudes of genotypic correlation coefficients for most of the characters were higher than their corresponding phenotypic correlation coefficients, except few cases, which indicate the presence of inherent or genetic association among various characters.

4.5.1 Genotypic correlation

Fruit yield was positive and highly significant genotypic correlation with fruit length (r= 0.74), average fruit weight (r= 0.62), fruit diameter (r= 0.61), seed per pod (r= 0.56), hundred seed weight (r= 0.68), internodes number (r=0.89,, and number of pod per plant (r=0.66). It is of interest to note that the significant positive correlation coefficients estimated at genotypic level were also found significant and positive at phenotypic level. Moreover, the significantly higher magnitudes of positive genotypic correlation than the corresponding phenotypic correlation in respect to some of the characters suggest that these characters were genetically controlled. The above parameters can be improved simultaneously.

The present study is agreed with the results reported by Niranjan and Mishra (2003) who observed fruit yield was positively and significantly correlated with number of fruits per plant, fruit length, number of seeds per fruit, average fruit weight, plant height and number of branches per plant at both genotypic and phenotypic levels and also agreed with finding of Singh *et al.* (2007) who reported high magnitude of genotypic and phenotypic coefficient of variation for number of branches per plant, plant height, number of fruits per plant and fruit yield. Therefore, frrom the correlation analysis average fruit weight, fruit diameter, seed per pod, number of pod per plant, internodes number, plant height, and hundred seed weight, were found to be important yield components.

The interdependency of other characters on each other's was also recorded. Fruit length exhibited a positive and significant genotypic correlation with fruit diameter, seed per pod, average fruit weight, number of epicalyxes and peduncle length (Table 7). In contrary it has

negative significant correlation with, days to 50% flowering and number of primary branch. Average fruit weight had negatively and significant genotypic correlation with days to 50% flowering and number of branch. It showed positively genotypic correlation with peduncle length, seed per pod, fruit length and weight. Fruit diameter had positive and significant genotypic correlation with peduncle length, seed per pod, fruit length, average fruit weight, number of epicalyxes and number of seed per pod. It showed negatively genotypic correlation with days to 50% flowering and number of branch. Internodes number exhibited a positive and significant genotypic correlation with number of pod per plant, days to 50% flowering, leaf width, stem diameter, number of branches, number of epicalyxes and plant height. However, it is negatively but significantly correlated hundred seed weight (Table 7).

Height of the plant exhibited genotypic positive correlation with that of days to 50% flowering, internodes length, internodes number and number of epicalyxes. On the other hands, it showed negative genotypic correlation with some characters like days to maturity, leaf length, fruit length, average fruit weight, fruit diameter, seed per pod and hundred seed weight insignificantly. For this reason we can improve this parameters simultaneously.

In general, high genotypic correlation suggests that selection directed to one character directly affects the other. Conversely, low or non-correlation suggests independence of association that would be possible to select independently for the two characteristics for diverse directions. In this study, genotypic correlations were higher than the phenotypic correlations. This may be possibly due to the masking action of genes on the influence of environment in the expression of characters indicating the association is largely due to genetic reson, which agrees with the findings of Bello *et al*. (2006).

Table 7.Genotypic Correlation coefficient among 20 traits in 25 *Abelmoschus esculentus* accessions studied at Gmbella in 2011/2012

Quantitative characters	DEM	DFF	DM	LL	LW	STD	INL	NBR	PDL	FL	FW	FD	NRG	SPP	HSW	NEP	INN	PH	NPP	FY/P
DEM	1	0.17	0.15	-0.46*	0.02	0.19	0.15	0.24	0.27	-0.16	-0.12	-0.07	-0.15	-0.10	-0.14	0.90**	-0.01	0.03	0.12	0.17
DFF		1	0.38	-0.21	0.49*	0.42*	-0.07	0.6**	0.63**	0.44**	-0.42*	-0.31	0.26	-0.23	-0.16	0.74**	0.53**	0.45*	0.35	-0.18
DM			1	-0.07	0.57**	0.40*	-0.28	0.70**	0.33	-0.34	-0.33	-0.44*	0.22	-0.37	-0.05	-0.50**	0.19	-0.17	0.20	-0.16
LL				1	-0.07	-0.23	-0.05	-0.36	-0.07	0.32	0.30	0.19	0.33	0.096	0.40*	-0.70**	-0.29	-0.13	-0.35	-0.09
LW					1	0.59**	-0.07	0.64**	0.45*	-0.25	-0.25	-0.29	0.31	-0.28	-0.21	0.65**	0.43*	0.18	0.45*	-0.13
STD						1	-0.06	0.47*	0.51**	-0.25	-0.16	-0.23	0.18	-0.25	-0.37	0.73**	0.61**	0.29	0.59**	0.21
INL							1	-0.13	0.25	-0.11	-0.11	-0.06	0.09	-0.04	0.09	0.33	-0.31	0.66**	-0.36	-0.03
NBR								1	0.49**	-0.50**	-0.52**	-0.43*	0.04	-0.43*	-0.35	0.61**	0.50**	0.18	0.49*	-0.27
PDL									1	0.51**	0.53**	0.47*	0.17	0.64**	0.35	-0.18	0.02	0.19	-0.08	-0.20
FL										1	0.93**	0.92**	0.07	0.80**	0.28	0.68**	-0.32	-0.23	-0.28	0.74**
FW											1	0.85**	-0.03	0.79**	0.35	0.22	-0.34	-0.25	-0.30	0.62**
FD												1	0.02	0.78**	0.05	0.77**	-0.28	-0.12	-0.30	0.61**
NRG													1	-0.04	0.20	0.87**	0.103	0.20	-0.03	0.58**
SPP														1	0.43*	0.86**	-0.31	-0.14	-0.30	0.56**
HSW															1	-0.21	-0.53**	-0.20	-0.60**	0.68**
NEP																1	0.32	0.84**	0.76**	0.21
INN																	1	0.41**	0.93**	0.89**
PH																		1	0.17	-0.58**
NPP																			1	0.66**
FY/P																				1

*Significant 0.05 (r=0.39) probability level; **= highly significant at 0.01 (r=0.50) level of probability level.

DEM= Days to emergency; DFF= Days to 50% flowering; DM= Days to maturity; LL= Leaf length (cm); LW= Leaf width (cm); ST= Stem diameter (mm); INL= Internodes length (cm); NBR= number primary of branch; PDL= Peduncle length (cm); FL= Fruit length (cm). FW= Average fruit weight (g); FD= Fruit diameter (mm); NRG= Number of ridge; SPP= Seed per pod; SW=hundred seed weight (g); NEP= Number of epicalyxes; INN= Internodes number; PH= Plant height (m); NPP= Number of pod per plant; FY/P= Fruit yield per plot (Kg)

4.5.2 Phenotypic Correlation

Like that of genotypic correlation fruit length (r= 0.52), average fruit weight (r= 0.57), fruit diameter (r= 0.52), seed per pod (r= 0.44), number of epicalyxes (r= 0.72), internodes number (r=0.73) and number of pod per plant (r=0.63) had highly significant positive phenotypic correlation with fruit yield. Similar finding has been reported by Jaiprakashnarayan and Mulge (2004) who noticed that total yield per plant was positively and significantly correlated with number of fruits per plant, average fruit weight, number of nodes on main stem, fruit length, plant height at 60 and 100 days after sowing and number of leaves at 45 and 100 days, On the other study Dhankar and Dhankar (2002). Bendale *et al*. (2003) examined 30 okra genotypes and found that pod length, pod weight, plant height, nodes per plant and number of pods per plant were positively correlated with the yield observed that fruit yield was significantly and positively correlated with the number of fruits and branches per plant. The investigation indicated that possibilities of developing early, short and high yielding cultivars by exploiting aforesaid associations.

Fruit length exhibited a positive and significant phenotypic correlation with peduncle length, fruit diameter and seed per pod and it had negatively and significant phenotypic correlation with number of branch (Table5).Average fruit weight had positive and highly significant phenotypic correlation with peduncle length, fruit length and seed per pod and negative and significant phenotypic correlation with number of primary branch. Internodes number had positively and significant phenotypic correlation with days to 50% flowering, number of primary branch stem diameter, plant height and number of pod per plant. So while improving inter nodes number for improving yield we can simultaneously improve these parameters together. And it showed negatively significant phenotypic correlation with hundred seed weight. This indicates that while improving internodes number we have to consider that we are decreasing number of seeds per pod. Fruit length exhibited negatively and significant phenotypic correlation with number of branch and had positively and significant phenotypic correlation with average fruit weight, peduncle length and number of seed per pod.

On the other hands, Plant height showed a positively and significantly correlation with intermodal length and days to 50% flowering. It exhibited a negatively and insignificant phenotypic correlation with day to emergence, days to maturity, average fruit weight, fruit length, fruit diameter, hundred seed weight and seed per pod. Number of pod had positively and significantly phenotypic correlation with stem diameter and internodes number. It shows negatively and significantly correlation with hundred seed weight (Table 8).

Table 8. Phenotypic Correlation coefficient among 20 traits in 25 *Abelmoschus esculentus* accessions studied at Gambella in 2011/2012

Quantitative character	DEM	DFF	DM	LL	LW	ST	INL	NBR	PDL	FL	FW	FD	NRG	SPP	HSW	NEP	INN	PH	NPP	FY/P
DEM	1	0.10	0.09	-0.40	0.07	0.09	0.08	0.21	-0.12	-0.14	-0.07	-0.08	-0.10	-0.06	-0.18	0.09	0.02	-0.02	0.09	0.07
DFF		1	0.32	-0.20	0.33	0.35	-0.04	0.54**	-0.06	-0.36	-0.39	-0.26	0.28	-0.20	-0.04	0.10	0.46*	0.39*	0.32	-0.13
DM			1	-0.10	0.39*	0.32	-0.24	0.63**	-0.45*	-0.29	-0.29	-0.36	0.20	-0.32	-0.09	-0.01	0.14	-0.13	0.27	-0.26
LL				1	-0.03	-0.20	-0.03	-0.36	0.32	0.31	0.29	0.18	0.31	0.07	0.26	-0.20	-0.25	-0.13	-0.30	-0.08
LW					1	0.49*	-0.12	0.47*	-0.19	-0.18	-0.21	-0.24	0.21	-0.28	-0.17	0.13	0.31	0.07	0.29	-0.05
ST						1	-0.04	0.40*	0.09	-0.23	-0.15	-0.21	0.15	-0.23	-0.16	-0.04	0.49*	0.27	0.49*	0.08
INL							1	-0.13	0.10	-0.12	-0.10	-0.05	0.07	-0.05	0.12	0.02	-0.29	0.67**	-0.32	-0.27
NBR								1	-0.41*	-0.49*	-0.51**	-0.41*	0.08	-0.39	-0.24	0.20	0.44*	0.16	0.44*	-0.21
PDL									1	0.48*	0.51**	0.43*	0.15	0.56**	0.22	-0.03	0.02	0.18	-0.07	0.28
FL										1	0.90**	0.89**	0.04	0.72**	0.16	0.02	-0.27	-0.21	-0.24	0.52**
FW											1	0.83**	0.09	0.73**	0.22	0.04	-0.31	-0.24	-0.28	0.57**
FD												1	-0.12	0.70**	0.06	0.11	-0.27	-0.11	-0.28	0.52*
NRG													1	-0.09	0.03	-0.18	0.13	0.17	0.02	-0.14
SPP														1	0.32	0.12	-0.27	-0.12	-0.25	0.44*
HSW															1	-0.20	-0.42*	-0.09	-0.41*	-0.16
NEP																1	0.09	0.08	0.20	0.72**
INN																	1	0.39*	0.91**	0.73**
PH																		1	0.22	-0.54**
NPP																			1	0.63**
FY/P																				1

*Significant 0.05 (r=0.39) probability level; **= highly significant at 0.01 (r=0.50) level of probability level.

DEM= Days to emergency; DFF= Days to 50% flowering; DM= Days to maturity; LL= Leaf length (cm); LW= Leaf width (cm); ST= Stem diameter (mm); INL= Internodes length (cm); NBR= number primary of branch; PDL= Peduncle length (cm); FL= Fruit length (cm). FW= Average fruit weight (g); FD= Fruit diameter (mm); NRG= Number of ridge; SPP= Seed per pod; SW=hundred seed weight (g); NEP= Number of epicalyxes; INN= Internodes number; PH= Plant height (m); NPP= Number of pod per plant; FY/P= Fruit yield per plot (Kg)

4.6. Path Coefficient Analysis

Path coefficient analysis (Table 9) at genotypic level revealed that internodes number had highly positive direct effect on fruit yield (p = 6.91) followed by average fruit weight (p = 6.71) which had positive genotypic correlation with yield. However, as internodes number becomes higher, it has a negative impact on the plant height and internodes length. But it has a positive impact on fruit length and fruit diameter which could be a cause for the high correlation coefficient that existed between fruit yield and internodes number (r g=0.89). Hence, while undertaking selection for fruit yield, one has to consider these two yield components in okra because when selecting plants with higher number of fruit, they may produce short internodes and plant height.

On the other hand, days to emergence, first flowering, leaf length, internodes length, number of primary branch, number of ridge, seed per pod, number of epicycles and plant height have negative direct effect on fruit yield. Though the direct effect of plant height on fruit yield was negative (p = -4.39), its correlation coefficient was negative (rg = -0.58) as it has high positive indirect effect internodes number, internodes length and fruit length. Likewise, even if internodes length the maximum negative direct effect (p = -5.08) on fruit yield, its correlation was positive with plant height because of its high positive indirect effect on number of pod per plant, average fruit weight and hundred seed weigh.

The residual effect (h = 0.25) is relatively high indicating that the trait considered in this analysis failed to sufficiently explain the variation in *Abelmoschus esculentus* yield. This suggests that more yield components should be considered to account for the variation in *Abelmoschus esculentus* yield.

Table 9. Genotypic direct (bold and underlined) and indirect effects of some characters on fruit yield of okra accessions studied at Gambella in 2011/2012.

Quantitative characters	DEM	DFF	D*M*	LL	LW	STD	INL	NBR	PDL	FL	FW	FD	NRG	SPP	HSW	NEP	INN	PH	NPP	rg
DEM	**-0.40**	0.06	0.25	0.05	0.01	0.04	0.77	-0.42	-0.02	0.89	0.09	-0.48	0.10	0.03	-0.22	-0.05	-0.12	-0.15	-0.26	0.17
DFF	-0.07	**0.33**	0.06	0.02	0.27	0.09	-0.20	-1.55	-0.30	2.33	0.30	-1.91	-0.16	0.08	-0.25	-0.06	3.57	-1.99	-0.73	-0.18
DM	-0.06	0.13	**1.65**	0.00	0.32	0.09	-1.94	-1.19	-0.08	1.83	0.24	-2.80	-0.14	0.13	-0.07	0.03	1.38	0.77	-0.43	-0.16
LL	0.19	-0.27	-0.99	**-0.76**	-0.04	-0.05	-0.08	0.62	0.05	-1.73	-0.22	1.24	-0.10	-0.03	0.66	0.94	-1.05	0.80	0.73	-0.09
LW	-0.01	0.16	0.94	0.59	**0.55**	0.13	-0.06	-1.09	-0.03	1.33	0.18	-1.87	-1.19	0.40	-0.04	-0.07	1.66	-0.80	-0.93	-0.13
STD	-0.08	0.14	0.67	0.18	0.33	**0.22**	-0.31	-0.80	0.02	1.34	0.12	-1.47	-0.11	0.09	-1.58	-0.04	4.03	-1.27	-1.23	0.21
INL	1.19	-0.24	-0.07	-0.31	-0.04	-0.01	**-5.08**	0.23	0.02	0.61	0.08	-0.43	-0.06	0.02	0.15	-1.92	-2.18	-2.91	0.75	-0.03
NBR	-0.10	0.20	1.16	0.03	0.36	0.10	-0.70	**-1.69**	-0.07	1.66	0.37	-2.74	-0.03	0.15	-0.54	-0.08	3.48	-0.81	-1.02	-0.27
PDL	0.05	-0.08	-0.82	-0.26	-0.12	0.02	0.55	0.74	**0.15**	-2.68	-0.38	2.97	-0.11	-0.22	0.55	0.10	0.01	-0.87	0.18	-0.20
FL	0.07	-0.15	-0.57	0.04	-0.14	-0.05	-0.59	0.85	0.08	**5.25**	-0.66	5.79	-0.04	-0.27	0.45	-0.04	-0.22	0.92	0.58	0.74
FW	0.05	-0.14	-0.55	-0.02	-0.94	-0.55	-1.57	0.89	0.09	-4.92	**6.71**	5.38	0.02	-0.26	0.05	-0.01	-5.37	1.13	0.63	0.62
FD	0.03	-0.10	-0.74	-0.16	-0.16	-0.05	-0.35	0.64	0.09	-4.85	-0.61	**6.27**	0.01	-0.26	0.83	-0.04	-0.96	0.40	0.64	0.61
NRG	0.06	0.09	0.38	0.03	0.17	0.04	0.47	-0.08	0.07	-0.37	0.02	0.01	**0.60**	0.02	0.32	0.10	0.71	-0.90	0.06	0.58
SPP	0.04	-0.08	-0.62	-0.72	-0.16	-0.06	-0.25	0.77	0.09	-3.23	-0.56	4.98	0.03	**-0.33**	0.90	-0.05	-2.19	0.62	0.65	-0.16
HSW	0.06	-0.05	-0.08	0.00	-0.12	-0.08	0.50	0.60	0.06	-1.52	-0.25	0.70	-0.12	-0.14	**1.54**	0.07	-3.61	1.92	1.23	0.68
NEP	0.37	0.37	-0.83	0.29	0.61	0.15	1.70	-2.46	-0.03	-3.58	-0.16	4.03	1.02	-0.29	-1.87	**-0.05**	9.00	-3.70	-3.63	0.21
INN	0.01	0.19	0.33	0.82	0.24	0.13	-1.99	-0.89	0.09	1.69	0.30	-1.90	-0.52	0.11	-0.82	-0.07	**6.91**	-1.82	-1.92	0.89
PH	-0.01	0.15	-0.39	0.08	0.10	0.06	3.03	-0.40	0.03	1.22	0.18	-0.90	-1.52	0.05	-0.32	-0.05	2.86	**-4.39**	-0.36	-0.58
NPP	-0.05	0.12	0.30	0.67	0.25	0.13	1.86	-0.84	-0.14	1.41	0.28	-1.91	0.02	0.11	-0.92	-0.10	6.05	-0.80	**-2.05**	0.66

Residual effect= 0.25

*Significant 0.05(r=0.39) probability level; **= highly significant at 0.01(r=0.50) level of probability level.

DEM= Days to emergency; DFF= Days to 50% flowering; DM=days to maturity; LL= Leaf length (cm); LW= Leaf width (cm); ST= Stem diameter (mm); INL= Internodes length (cm); NBR= number of branch; PDL= Peduncle length (cm); FL= Fruit length (cm). FW= average fruit weight (g); FD= Fruit diameter (mm); NRG= Number of ridge; SPP= Seed per pod; SW=hundred seed weight (g); NEP= Number of epicalyxes; INN= Internodes number; PH= Plant height (m); NPP= Number of pod per plant.

4.7. Cluster Analysis of Quantitative Characters

Cluster analysis based on quantitative characters grouped 25 germplasm accessions in to five distinct clusters (Table 10, (Appendix figure1) in which the first cluster consisted of 13 accession (52%), the second cluster consisted of 6 accessions (24%), the third clusters consisted 3 accessions (12%), the fourth cluster contained of 2 accession (8%) and the fifth cluster consisted of only one accession (4%) from the total accessions. The distribution pattern of genotypes into five clusters confirmed the existence of diversity among the genotypes.

Table 10. Clusters of okra accessions based on quantitative characters studied at Gambella in 2011/2012.

Cluster	Number of accessions	Accessions name
I	13	GM1, GM3, GM4, GM5, GM6, GM8, GM10, GM11, GM12, GM13, GM14, AS8, AS9.
II	6	AS1, AS2, AS3, AS5, AS6, AS7
III	3	GM2, GM7, GM9
IV	2	AS4, AS11
V	1	AS10

The mean value of the 20 quantitative characters in each cluster is presented in Table 11. Cluster I consisted of 13 genotypes having the characteristic of relatively narrow leaf width (37.56 mm) and high number of epicalyxes (9.44). Cluster II could be characterized by relatively late fruit maturing (104.22) and had relatively long leave length (42.21 cm) and wider stem diameter (32.70 mm). It also known by having the short fruit length (12.30 cm), peduncle length (3.02 cm) and plant height (45.27 cm) with small average fruit weight , number of seed per pod (84.72 number) and fruit yield per plot (3.95 kg) (Table 11).

Cluster III consisted of three genotype. The shortest time to germinate (5.22), and relatively had narrow stem diameter (29.38 mm). These cluster also had longer peduncle (5.21cm). Additionally it had fruits with high number of ridge (7.67), with small number of internodes (23.03). Cluster IV had two genotype having the characteristic of small internodes length

(4.45 cm) with long plant height (95.14 cm), heavier average fruit weight (148.32 g). On the other hand it has relatively high number of seed per pod (107.933) and yield per plot (95.14 kg) The last cluster V had only one genotype having the characteristics of early harvesting date (84.00), short leaf length (24.21cm) however it had wider leaf (44.07 cm) and fruit width (45.10 mm). On the other hand, it is known by high number of primary branches (10.47), internodes number (29.65), and seed per pod (113.13.) as well as number of pod per plant (29.65).

Table 11. Cluster means for 20 quantitative characters of okra accessions studied at Gambella in 2011/2012.

Quantitative characters	Cluster 1	Cluster 2	Cluster 3	Cluster 4	Cluster 5
Days to emergence	5.41	6.06	5.22	5.33	6.00
Days to 50% flowering	54.05	62.33	52.67	52.50	53.33
Days to maturity	85.44	104.22	84.00	93.67	84.00
Leaf length (cm)	31.09	29.28	33.10	31.76	24.21
Leaf width (cm)	37.56	42.21	38.06	40.67	44.07
Stem diameter (mm)	30.20	32.70	29.38	29.70	31.91
Internodes length (cm)	6.56	5.65	7.47	4.45	6.53
Number of branch	3.78	9.31	2.49	5.73	10.47
Peduncle length (cm)	4.75	3.02	5.21	4.45	4.20
Fruit length (cm)	25.39	12.30	28.81	43.43	45.07
Average fruit weight	79.48	22.31	113.81	148.32	22.37
Fruit diameter (mm)	26.86	9.73	29.15	43.81	45.10
Number of ridge	7.73	7.94	7.67	7.87	7.67
Seed per pod	100.85	84.72	105.00	112.87	113.13
100 seed weight	6.64	6.11	6.89	7.17	6.00
Number of epicalyxes	9.44	9.06	9.33	9.17	9.33
Internodes number	26.14	28.77	23.03	24.23	29.65
Plant height (cm)	68.82	45.27	80.61	95.14	55.05
Number of pod per plant	20.08	24.16	16.91	18.56	29.65
Fruit yield (kg/plot)	9.47	3.95	11.01	17.55	6.65

Based on cluster means, it is evident from the data (Table 11) that germplasm accessions falling in cluster IV and V showed higher performance for the characters of interest viz., internodes number, Internodes length, fruit length, average fruit weight and plant height. Furthermore, most of these characters also had positive genotypic association with fruit yield per plot except internodes length (Table 7). Hence, their potential as parents in heterotic breeding work seems possible. On the other hand, cluster II, which is consisted of 5 germplasm accessions was the least in performance for the majority of quantitative characters studied (Table 8). For example, all of the accessions grouped under this cluster gave the least fruit yield and average fruit weight. The result also pointed out that the importance of accessions in cluster II for their exploitation in fruit yield improvement appeared limited in view of their poor performance for the majority of the characters of interest. This indicated that different clusters have different breeding values that enable breeders to improve different traits and parental selection should be made based on the relative merits of each cluster for each trait depending on the objective of the breeding program.

4. 8. Divergence Analysis

The squared distance indicated in Table 12. Test of significance show significance between all cluster distances. The minimum squared distance was between clusters I and III (213.64) followed by cluster III and IV (410.38). Maximum squared distance was between cluster II and IV (2846) followed by cluster I and II (2513) and cluster II and IV (1698.0). Generally this study revealed that germplasm accessions included in this study are highly divergent. Thirupathi *et al.* (2012) revealed considerable genetic diversity among 100 genotypes of okra (*Abelmoschus esculentus* (L.) Moench) were grouped into 11 distinct clusters for all the seventeen quantitative characters which pertaining to the growth, earliness and yield.

According to Ghaderi *et al.* (1984) increasing parental distance implies a great number of contrasting alleles at the desired loci, and then to the extent that these loci recombine in the F2 and F3 generation following a cross of distantly related parents, the greater will be the opportunities for the effective selection for yield factors. Minimum inter cluster distance was observed between clusters I and III (213.64) indicating that genotypes in these clusters were

not genetically diverse or there was little genetic diversity between these clusters. This signifies that crossing of genotypes from these two clusters might not give higher hetrotic value in F1 and narrow range of variability in the segregating F2 population. Maximum genetic recombination is expected from the hybridization of the parents selected from divergent cluster groups. Therefore, maximum recombination and segregation of the progenies is expected from crosses involving parents selected from cluster II and IV followed by cluster I and II and cluster II and IV. however the breeder must specify his objectives in order to make best use of the characters where the characters are divergent.

Thus, it can be concluded that selection of genotypes from the most divergent clusters may exhibit a high heterosis besides fruit yield. Therefore, hybridization between the genetically diverse parents in further breeding programs may produce large variability and better recombinants in the segregating generations.

Table 12 Average inter cluster (off diagonal) D^2 values among three clusters in *Abelmoschus esculentus* accessions studied at Gambella in 2011/2012.

Clasture	I	II	III	IV	V
I		2513**	213.64**	1034**	931.53**
II			1698**	2846**	1421**
III				410.38**	1638**
IV					867.58**
V					

**: significant X^2 =28.87 and X^2 = 34.81 at 5 and 1% probability level

4.9. Principal Component Analysis

The principal component analysis (Table 13) revealed that six principal components PC1, PC2, PC3, PC4, PCA5 and PCA6 with Eigen value 10.65, 30.4, 2.41, 1.7, 1.62 and 1.32 respectively, have accounted for 83% of the total variation. The first two principal components PC1 and PC2 with values of 43%, and 12.9% respectively, contributed more to the total variation. According to Chahal and Gosal (2002), characters with largest absolute values closer to unity with in the first principal component influence the clustering more than those with lower absolute values closer to zero. Therefore, in this study, differentiation of the genotypes in to different cluster was because quantitative traits such as days to fifty percent flowering, stem diameter, number of seed per pod, number of pod per plants, internodes length, inter nodes number and plant height.

Agronomic characters having relatively higher value in the first principal component (PC1) were days to 50% flowering, fruit length, average fruit weight, fruit diameter, seed per pod, hundred seed weight, and number of seed per pod and plant height had more contribution to the total diversity and they were responsible for the differentiation of the six clusters. Characters like stem diameter, internodes number, peduncle length had contributed a lot for principal component (PC2); peduncle length, internodes length, number of primary branch, days to maturity , plant height and number of epicalyxes had contributed in the third principal component (PC3); days to emergence, days to maturity, leaf length, leaf width, stem diameter, internodes length and number of ridge the fourth principal component (PC4); leaf length, leaf width, stem diameter and number of epicalyxes had contributed in the fifth principal component (PC5); days to emergence, leaf width, internodes length, number of primary branch and number of ridge in the six principal component(PC6) were the major contributors to each principal components (PC).

Generally quantitative traits such as days to fifty percent flowering, stem diameter, number of seed per pod, , number of pod per plants, internodes length, inter nodes number and plant height contributed to the variation in two PCs out of the six PCs. This result further confirmed the presence of genetic diversity for use in improvement program of okra.

Table 13. Eigenvectors and Eigen values of the first six principal components (PCs) for characters of *Abelmoschus esculentus* accessions studied at Gambella in 2011/2012.

Quantitative characters	PC1	PC2	PC3	PC4	PC5	PC6
Days to emergence	0.14	0.12	-0.09	-0.31	0.17	0.31
Days to 50% flowering	0.27	0.00	-0.05	0.16	0.05	-0.09
Days to maturity	0.21	-0.02	-0.25	0.28	0.16	0.14
Leaf length	-0.10	-0.09	0.08	0.53	-0.33	0.00
Leaf width	0.14	0.21	-0.09	0.30	0.26	0.29
Stem diameter	0.13	0.36	0.08	0.12	0.29	-0.19
Internodes length	0.01	-0.15	0.50	-0.14	0.11	0.25
Number of branch	0.21	0.18	-0.16	0.03	0.19	0.25
Peduncle length	-0.17	0.26	0.33	0.00	-0.17	-0.05
Fruit length	-0.25	0.18	-0.05	-0.03	0.08	0.21
Fruit weight	-0.27	0.10	0.02	0.16	-0.02	-0.04
Fruit diameter	-0.25	0.17	-0.01	-0.05	0.15	0.17
Number of ridge	0.09	0.10	0.20	0.39	-0.01	0.37
Seed per pod	-0.25	0.14	0.01	-0.08	0.14	0.11
100 seed weight	-0.28	0.12	0.03	0.11	0.01	-0.01
Number of epicalyxes	-0.05	0.06	0.45	-0.09	0.45	-0.10
Internodes number	0.15	0.42	-0.08	-0.04	-0.16	-0.23
Plant height	-0.28	0.16	0.01	0.11	0.01	-0.03
Number of pod per plant	0.16	0.39	-0.16	-0.11	-0.08	-0.16
Fruit yield (kg/plot)	-0.21	0.31	-0.01	0.15	0.04	-0.22
Eigen value	10.65	3.04	2.41	1.70	1.62	1.32
Difference	7.61	0.62	0.72	0.08	0.30	0.41
Proportion	0.43	0.12	0.10	0.07	0.06	0.05
Cumulative	0.43	0.55	0.64	0.71	0.78	0.83

5. SUMMARY AND CONCLUSION

This study was conducted at Gambella during the 2011/2012 main cropping season. The main objectives of the study were to characterize morphologically and analyze genetic variability among okra accession. Specifically this research aim were to characterize okra accessions based on qualitative and quantitative morphological traits and determine the range of diversity among the accessions with the ultimate goal of providing a basis for varietal improvement and conservation.

Toward this effort, a total of 25 *Abelmoschus esculentus* (L) Moench accessions were used. International Plant Genetic Resources Institute (IPGRI, 1990) descriptor list for okra species were adapted to record and describe both the quantitative and qualitative traits. Cluster analysis based on morphological qualitative characters revealed four distinct groups. The characters used for clustering were branching habit, leaf color, leaf petiole color, pod color, stem color, leaf shape, red coloration of petal base and position of fruit on the main stem.

The analysis of variance for quantitative characters showed highly significant difference among the accessions for all parameters recorded. The range and mean performance showed the presence of considerable amount of variability among the germplasm accessions. For instance, for fruit yield (3-18), plant height (42-96 cm), fruit length (11-45 cm), average fruit weight (12-150 g), fruit diameter (9-45) and seed per pod (79-11).

The analysis of phenotypic (PCV) and genotypic (GCV) coefficients of variation also showed the presence of variation among the accessions. In general, the estimates of PCV values were higher than the corresponding GCV values for all the characters studied. PCV ranged from 8.893 for number of epicalyxes to 118.01 numbers of primary branches whereas, GCV ranged from 5.07 for number of epicalyxes to 106.63 for number of primary branches. Among the various quantitative characters, relatively higher PCV and GCV were observed for number of primary branches (118.10 and 106.58), average fruit weight (92.08 and 88.89) and days to maturity (137.25 and 89.79). It may therefore be important to point out that characters with

relatively higher PCV and GCV values should be given due attention for an effective selection in yield improvement of *A. esculentus.*

Fruit length, average fruit weight, fruit diameter, seed per pod, number of epicalyxes, internodes number, plant height, and number of pod per plant had positive and highly significant phenotypic and genotypic correlations with fruit yield. Negative phenotypic correlation were recorded for day to maturity, leaf length and width, internodes length, number of primary branch and number of ridges insignificantly. On the other hand, fruit yield had significantly negative genotypic correlation with plant height. The interdependency of other characters on each other's was also recorded for both genotypic and phenotypic correlation.

Path coefficient analysis at genotypic level revealed that internodes number had highly positive direct effect on fruit yield, followed by fruit weight which had positively genotypic correlation with yield.

Hierarchical clustering analysis, based on quantitative characters, grouped 25 okra accessions into five major clusters. Each cluster group exhibited wide range in the extreme values and peculiarity for most of the quantitative characters, which would offer broad opportunities for using the accessions in the future breeding programs to develop varieties suitable for different purposes.

The minimum squared distance was between clusters I and III (213.64) followed by cluster III and IV (410.38). Maximum squared distance was between cluster II and IV (2846) followed by cluster I and II (2513) and cluster II and IV (1698.0).). Generally this study revealed that germplasm accessions included in this study are highly divergent. The principal component analysis revealed that six principal components PC1, PC2, PC3, PC4, PC and PC 6 with Eigen value 10.65, 30.4, 2.41, 1.7, 1.62 and 1.32 respectively, have accounted for 83% of the total variation. The first two principal components PC1 and PC2 with cumulative values of 55%, contributed more to the total variation. In general, it appeared that there was no reasonable degree of correspondence between cluster groups formed by qualitative and quantitative approaches.

In conclusion, the present study exhibited the presence of genetic diversity for several morphological qualitative and quantitative characters among okra germplasm accessions. The existence of genetic diversity is potential resource for improvement of the crop through selection and hybridization. Therefore, the observed variability for very important traits in okra such as for yield should be exploited in order to improve the productivity of this valuable crop. Therefore, Accession like GM7, GM9 and GH13 from Gambella collection AS4 and AS11 from Assosa collection are recommended if they promoted for the next breeding work as they are high yielder accessions compared to the others.

Generally in this study, okra accessions for morphological characterization and divergence analysis were grown in single environment during one season. However, it is apparent that morphological characters are usually affected by environmental factors (Tessier, 1991). This is particularly true of quantitative characters in which plant breeders are most interested (Singh, 1997). Therefore, the result of the present investigation may vary with location and season. That means, further study with due emphasis on quantitative characters is required to determine the presence and magnitude of genotype-environment interaction in okra growing regions of Ethiopia. Furthermore, as a high morphological variation between cultivars is not a guarantee for a high genetic variation (Kreike, 2004), molecular or biochemical studies need to be considered as complementary to morphological studies. Since simple selection of superior types among the existing cultivars could result in identification of promising lines, okra cultivars from other geographical areas of Ethiopia need to be collected so as to broaden the base of existing breeding program.

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7. APPENDICES

Appendix Table 1: Mean monthly total rainfall (mm) of the study area, 2002 – 2012.

year	Jan.	Feb.	March	April	May	June	July	August	Sept.	Oct.	Nov.	Dec.	mean	total
2002	0.00	0.00	15.60	116.70	305.40	62.60	195.70	235.60	195.60	147.60	58.60	1.50	120.51	1455.41
2003	2.50	3.50	19.70	113.80	153.40	132.20	310.60	196.40	118.80	63.10	35.30	6.50	104.34	1260.14
2004	0.00	0.50	36.50	32.50	104.50	104.50	238.40	59.40	180.50	60.50	35.50	0.00	76.99	929.79
2005	10.20	4.20	20.20	40.80	128.30	205.70	131.40	246.10	103.60	87.50	47.30	0.00	92.56	1117.86
2006	1.90	54.70	46.80	43.00	212.50	88.00	145.10	268.70	271.10	222.10	16.20	6.00	124.23	1500.33
2007	0.00	0.00	60.80	106.10	145.80	112.30	179.80	78.90	142.00	164.30	24.20	0.00	91.56	1105.76
2008	0.00	0.00	12.30	45.80	103.40	51.70	218.80	161.80	73.30	107.90	21.10	15.30	73.25	884.65
2009	0.00	0.00	0.60	26.50	98.00	71.60	152.20	125.10	42.70	38.00	2.10	0.00	50.27	607.07
2010	0.00	5.80	2.50	2.80	85.60	138.70	276.20	199.00	238.30	129.60	49.40	0.00	101.83	1229.73
2011	0.00	6.20	3.00	0.00	215.50	136.60	144.80	226.30	253.70	97.50	24.80	0.00	100.06	1208.46
2012	0.00	6.00	3.20	0.60	225.00	179.30	150.90	250.60	279.60	150.00	28.60	0.00	112.68	1386.48
Mean	1.33	7.35	20.11	48.05	161.58	116.65	194.90	186.17	172.65	115.28	31.19	2.66	85.62	942.15

Source: National Meteorology Agency Gambella Branch, 2012

Appendix Table 2: Monthly average maximum temperature (^{O}C) of the study area, 2002 – 2012.

Year	JAN	FEB	MAR	APR	MAY	JUN	JUL	AUG	SEP	OCT	NOV	DEC	Mean
2002	40.9	42.2	40.1	38.7	36.3	34.9	33.4	31.2	32.6	34.3	36.4	40.6	36.8
2003	39.3	40.6	40.4	37.9	33.3	31.9	30.5	31.4	31.8	35.0	35.4	36.6	35.3
2004	39.1	40.6	39.4	40.1	38.4	32.0	30.5	32.9	33.5	36.0	36.9	36.8	36.4
2005	39.3	42.0	40.6	40.0	36.2	31.8	33.1	31.6	31.4	33.4	35.0	37.9	36.0
2006	38.5	40.6	41.3	42.1	36.1	35.4	33.8	30.0	29.1	34.0	36.8	40.7	36.5
2007	39.6	28.2	38.9	37.8	33.9	32.7	30.9	30.3	29.8	31.6	37.6	36.1	34.0
2008	39.2	40.8	41.2	40.6	39.2	36.0	31.7	30.6	33.5	36.3	36.6	38.9	37.1
2009	34.8	37.4	39.5	36.8	34.0	32.1	31.6	23.7	21.7	24.3	33.6	29.3	31.6
2010	36.2	38.2	37.9	39.2	39.0	34.5	33.5	33.7	33.6	33.6	34.1	33.3	35.6
2011	36.5	41.0	41.6	41.8	41.5	40.6	36.1	35.9	33.2	33.9	35.2	37.7	37.9
2012	36.9	42.3	42.2	41.6	41.8	41.2	37.2	36.5	32.4	35.6	36.8	39.7	38.7
Mean	41.7	43.0	43.9	43.3	40.6	37.9	35.9	34.4	34.0	36.5	39.1	40.4	39.2

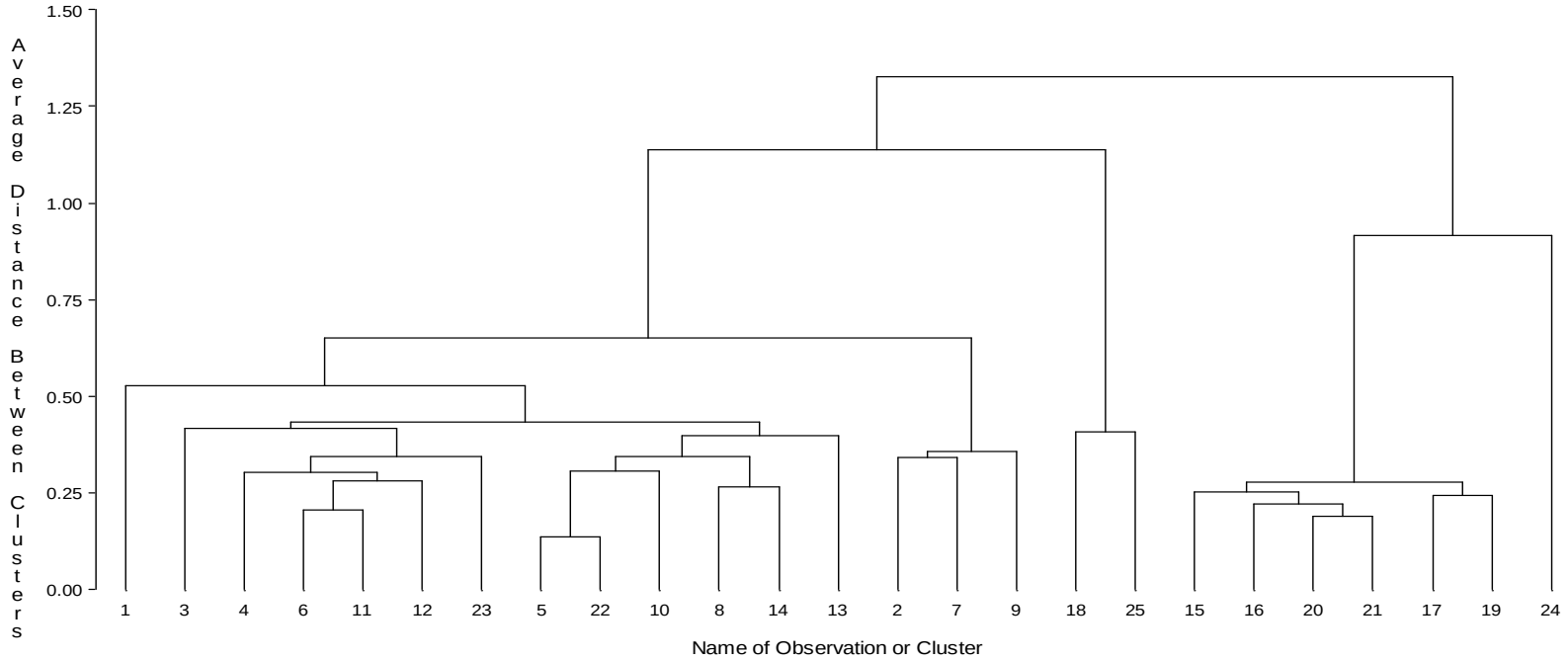
Appendix Table 3: Monthly average minimum temperature (^{o}C) of the study area, 2002– 2012

Year	JAN	FEB	MAR	APR	MAY	JUN	JUL	AUG	SEP	OCT	NOV	DEC	Mean
2002	20.1	22.7	22.5	21.5	23.3	22.5	23.5	23.6	23.4	22.5	24.7	24.9	17.16667
2003	19.5	22.8	24.7	23.4	21.1	21	20.1	19.9	20.2	19.4	18.5	17.4	15.16667
2004	15.4	16.3	20.6	21.4	20.2	17.3	17.4	17.9	17.9	17.9	17.7	15.6	13.075
2005	14.9	17	20.4	20.2	20.1	20.1	20	20.1	20.1	20.6	21	19.4	14.44167
2006	19	19.8	20.1	21.9	20.9	20.4	20	19.7	19.8	20.9	19.6	19.5	15.06667
2007	19.2	19.9	20.2	21.6	22	20	15.3	14.5	15	14.6	14.8	15.2	12.925
2008	19.8	22	23.5	20.1	13.3	12.4	18.3	20.1	20	19.8	17.6	19.8	14.15833
2009	21.6	21.7	21.6	21.3	19.3	18.9	18.5	18.5	19.2	19.9	20.9	20.5	15.15
2010	21.4	23.1	24.7	26	23.8	22.4	21.9	21.4	20.7	20.8	20	19.5	16.375
2011	18.2	20.6	22.8	24.6	20.5	20.4	22.3	20.2	22.2	20.2	20.8	20.7	15.66667
2012	18.9	22.6	25	22.6	20.5	21.6	23.3	18	22.9	20.7	26.6	22	16.35833
Mean	189.1	205.9	221.1	222	204.5	195.4	197.3	195.9	198.5	196.6	195.6	192.5	167.0167

Source: National Meteorology Agency Gambella Branch, 2012

Appendix Table 2. Means for 25 accessions studied at Gambella studied at Gambella in 2011/2012.

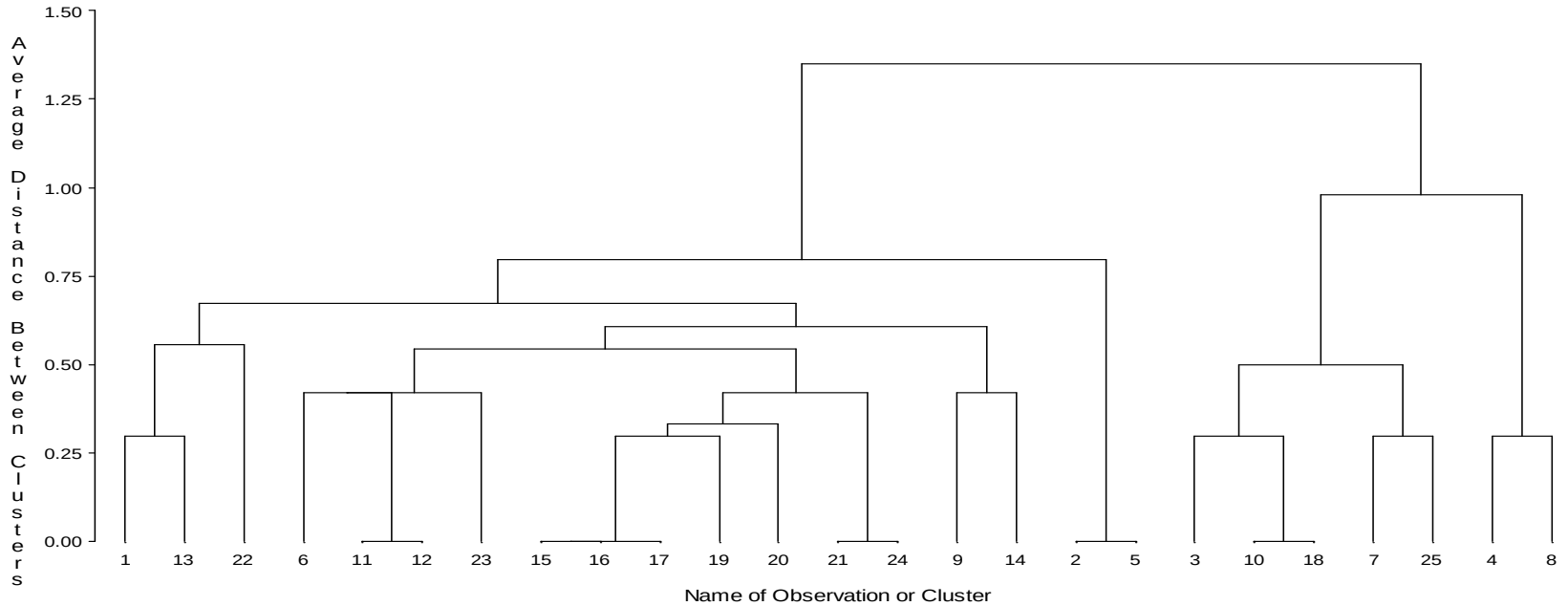
Acce.	DEM	DFF	DM	LL	LW	STD	INL	NBR	PDL	FL	FW	FD	NRG	SPP	HSW	NEP	INN	PH	NPP	FY/P
1	5.3	52.7	83.7	25.7	35.2	27.2	11.6	2.1	3.7	21.5	58.6	24.1	8.1	93.3	7.7	10	20.1	57.3	14.1	6
2	5	52.3	83.3	33.1	38.9	29.4	11.7	2.5	5.3	31.3	106.6	32.4	8	108.9	6.7	10	17.5	77.6	11.1	6.5
3	4.7	50.3	83.3	29.8	36.2	32.3	4.3	2.3	3.3	27	94.6	32	7.7	90.3	5.7	9	28.6	71.2	23.2	10.5
4	5	56	82	31.4	41.2	27.4	6.4	5.1	5.9	25.2	64.7	25.2	8	95.8	5.3	9	34.3	64.9	26.7	7
5	5.3	52	83.7	29.4	35.3	31.4	5.2	3.1	5.2	30.1	87.9	30.4	7	107.6	7	9.7	26.5	74	20.5	10.2
6	5.7	54	82.3	31	36.9	36.5	5.6	5.1	5.9	24.1	77.5	25.1	7.3	100.4	6.3	10	26.5	68.1	20.5	10.9
7	5.3	53	82.7	40.8	40.4	30.3	6.3	2.7	6	29.7	118.1	29.7	8	96.2	7.3	9	24.9	79.8	17.9	12.7
8	6	53.3	83.7	33.7	35.3	24.4	7.4	3.1	3.5	22.5	83.4	22.2	7.3	100	8	8.7	16.8	66.7	10.9	5.5
9	5.3	52.7	86	25.4	34.8	28.4	4.3	2.3	4.3	25.4	116.7	25.4	7	109.9	6.7	9	26.7	84.4	21.7	13.8
10	5	52	93.7	37.5	34.7	22.5	4.3	1.9	4.1	32.4	84.5	35.8	8	110.7	6.7	9	20.9	72	16.2	8.2
11	5	53.3	84.7	33.3	38.5	33.6	7.8	2.8	4.4	27	75.9	22.3	8	93.8	6	9.3	32.9	67.5	26.2	11.9
12	5.3	54	87	33	36.8	25	7.7	4.1	3.8	24.2	74.8	23.7	7	89.8	6.3	9.3	23.5	62.7	17.5	8.5
13	4.7	60.7	87	31.7	37.7	34.3	6.8	4.6	6.3	20.9	84.4	20.9	8	114.1	7.7	9.3	31.1	76.5	24.5	13.7
14	5.3	54.3	84	28.5	42.7	27.3	5.1	2.6	4.7	24.9	77.3	24.9	8	110.9	7	9.7	21.3	69.8	16.3	6.2
15	7	61.3	104.3	28.8	46.3	36.7	5.9	13.2	3	13.5	24.1	9.6	8	86.7	6.3	9	36.5	49.1	29.8	4.3
16	4.3	63	104.3	34.8	47	33.9	4.6	8.2	2.9	12.5	26.5	8.7	8	78.7	6	9.3	27.4	44.2	20.4	4.6
17	5.7	63.7	105	28.4	34.6	26.8	5.9	12.5	3	10.5	22.9	10.5	8	85.7	6.3	9	26.6	45.1	19.6	2.7
18	5.3	52.7	82.7	31.2	35.5	25.7	3.7	2.1	4.5	45.3	146.4	45	7.7	112.4	7	9	24.7	94.5	19	16.7
19	6	61.7	101.7	27.6	39.6	30.8	5.9	9.2	3.3	13.6	12.4	9	8	86.7	7	9	26.8	42	26.8	5.2
20	7	63	105.7	24.7	44.6	33	6.3	8.5	2.6	10.8	25.8	10.7	7.7	85	6	9	28.3	46.4	21.3	3.3
21	6.3	61.3	104.3	31.4	41.2	34.9	5.2	4.3	3.3	12.8	22.1	9.7	8	85.7	5	9	27.1	44.9	27.1	3.6
22	6.3	49	83.7	31.2	36.4	32	6.3	4.7	6.6	29.6	90	31.2	8	112.2	7.7	9.7	27.5	76.6	20.6	11.1
23	6.7	61	92	27.9	41.5	38.5	6.9	7.9	4.4	20.7	79.5	31.5	8	92.3	5	10	29.8	67.2	23.8	13.4
24	6	53.3	84	24.2	44.1	31.9	6.5	10.5	4.2	45.1	22.4	45.1	7.7	113.1	6	9.3	29.7	55.1	29.7	6.6
25	5.3	52.3	104.7	32.3	45.9	33.7	5.2	9.4	4.4	41.5	150.2	42.6	8	113.3	7.3	9.3	23.8	95.8	18.1	18.4
Grand mean	5.6	56	90.4	30.7	39.3	30.7	6.3	5.4	4.3	24.9	73.1	25.1	7.8	99	6.7	9.3	26.4	166.8	20.9	8.7
CV%	9.8	7.6	5.3	3.6	5.9	5	11.4	10.1	6.9	7.4	4.8	8.2	11.6	15.8	9.9	4	8.9	15	11.7	20
LSD5%	0.89	1.7	1.92	1.8	3.78	2.53	1.17	0.89	0.49	3.01	5.81	3.37	0.46	7.79	1.07	0.6	3.83	41	4.03	2.83



Appindix Figure 1

Dendrogram depicting the genetic relationship of okra germplasm based on 20 quantitative characters evaluated at Gambella in 2011/12.

NB. The numbers representation is as indicated in Table 1



Appendix Figure 2. Dendrogram depicting the genetic relationship of okra germplasm based on eight qualitative characters evaluated at Gambella in 2011/12

NB. The numbers representation is as indicated in Table 1