

EVALUATION OF F3 SEGREGATING COWPEA (*Vigna unguiculata*) POPULATION
DEVELOPED FROM INSECT-MEDIATED CROSSES

by

KHOLOFELO CAROLINE MOLOTO

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Supervisor: PROF J.A.N ASIWE

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DECLARATION

I, Kholofelo Caroline Moloto hereby declare that this mini-dissertation which I submit for the requirement of the degree Master of Science in Agriculture (Agronomy) at the University of Limpopo is my original work (unless stated otherwise) in design and execution; that is it has not been submitted at this University or any other University by me or anyone else.

Moloto K.C. (Ms)

Date

DEDICATION

To my beloved siblings Alfred and Gillian Moloto; I have led the way, follow and prosper; the sky is the only limit. My partner Dennis Sejaphala, we have done it yet again daddy. To the loving souls of my grandparents Maria "Moratiwa" and George "Oom G" Malatji who died during my quest to be educated; I always look up to you, you will always be loved.

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Jesus, my redeemer, my saviour, my healer, my provider, thank you for your faithfulness to the cross, thank you for not letting my foot slip, for delivering me from the claws of the enemy and laying a table for me before my enemies. My purpose here on earth is to worship and praise you father, thus receive my praises for this wonderful work which would not have been possible if you were not with me in this journey. Indeed, Luke 18:27. Amen.

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To my immediate family, thank you for standing by me, especially my little brother Alfred, thank you for your sleepless nights of prayer, here are the results. My little sister Gillian, thank you for your helping hand in the field and during the shelling process. My mother, Rebecca Moloto; the role you played since my conception till now is remarkable, thank you for nurturing me, supporting me, praying for me and above all for loving me unconditionally. To my father, Lawrence Moloto thank you for teaching me to be independent.

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ABTRACT

Erratic rainfall and insect infestation are some of the factors that limit cowpea production in Limpopo (Asiwe, 2009). Improved cowpea varieties available for dryland production in Limpopo do not meet the farmer's needs. Specific trait crosses are needed to be made with adapted varieties for the purpose of developing high yielding, pest tolerant and adapted varieties. Varieties with desired specific traits were planted in isolation plots for outcrossing by insects. F1 lines derived from parental lines that were half sib seeds were harvested and planted as F2, and seeds obtained from F2 generation were used to develop F3 segregating population at University of Limpopo Experiential Farm, Syferkuil, and planted along with parents. This F3 generation forms the current evaluation on which this report is based. Data collected included; aphid severity (measured on a 9-point scale), number of days to 90 % maturity, grain yield, 100 seed weight and number of genotypes selected for advancement. Results showed significant variation among progeny over parents therefore expressing transgressive segregation. Genotypes derived from pedigree TX08-30-5 were five while pedigree IT98K-205-8 and IT97K-499-35 had four genotypes each that expressed early maturity (80-90 days) which are desirable for evading drought. Twenty-three genotypes from pedigree GEC and three from pedigree IT97K-499-35 obtained grain yield (>101 g/plot and >161 g/plot) that was better than their parents. Only one genotype derived from pedigree TX08-30-1 expressed high resistance to aphids better than the parent with a mean aphid score range of 1-2. Four genotypes from pedigree TX08-30-5 obtained bigger 100 seed weight (>18.6 g) compared to parent. These progeny also proved to be high yielders as they expressed heterosis for all yield attributes than their parents. Eighty-two genotypes were selected for advancement. Significant variations were observed among progeny giving opportunity to make selections. In most cases, genotypes outperformed their parents which indicate positive heterosis. The 82 promising genotypes selected will be advanced for further selections from multi location testing for stability and adaptation.

Key words: *Vigna unguiculata*, high yielding, insect pest resistance and segregation

CHAPTER 1

GENERAL INTRODUCTION

1.1 Background of the study

Cowpea (*Vigna unguiculata* (L.) Walp) is an important staple food in most African homesteads, consumed as leafy vegetable, dry seeds, young and immature seeds (IITA, 2014). It is a cheap source of protein, as it contains about 13-25% of crude protein essential for human diet (Ayo-vaughan *et al.*, 2013). Cowpea residues are used as a source of fodder for animal feed (Moalafi *et al.*, 2010). Its canopy covers the ground reducing risks of erosion, avoiding heating of the soil by solar radiation thereby conserving moisture through prevention of surface water evaporation, runoff and promoting infiltration (Fatokun *et al.*, 2002).

Cowpea fixes atmospheric nitrogen and makes it available for plant use (IITA, 2014). Therefore, cowpea can be used in rotations or in intercropping with cereals to enhance soil fertility and thereby reducing cost of commercial nitrogen fertilizers (DAFF, 1997). Furthermore, cowpea residues can be incorporated into the soil to add organic matter to the soil hence increasing the soil porosity, permeability and making it to be friable (IITA, 2014).

Several abiotic and biotic factors limit the productivity of cowpea in Limpopo Province thereby reducing yield (Asiwe, 2009b). Genetic improvement can be used to enhance production. Several improved varieties have been introduced and released in South Africa with low adaptation. Thus, there is a need for incorporation of specific traits into well-adapted varieties. In normal essence, genetic improvement is done through hand or artificial pollination. However, hand pollination success rate is low (0.5-50 %), there is high rate of abscission of manipulated flowers and it requires low temperatures, high humidity and well managed greenhouses (Rachie *et al.*, 1975).

In cases like this, breeders resort to outcrossing for varietal development. Due to lack of conducive facilities for hand pollination, parents with desirable traits were planted in isolation plot for recombination by insects. This study was conducted to develop high yielding varieties with combined resistance in Limpopo province using insect pollinated varieties. Cowpea pollens are known to be too heavy and sticky to

be dispersed by wind, except by insects (Belane *et al.*, 2011). Planting multi-trait parental lines in isolation for recombination is a veritable technique for developing cowpea lines where there are no facilities for pair-wise hand pollination. Insect mediated pollinations become a way out to generate segregating populations of half-sib genotypes.

1.2 Problem statement

Low rainfall with poor distribution (drought prone area), climate change and pests are some of the factors that limit agricultural production in Limpopo Province despite the fact that it is a noted province for crop production in South Africa (Asiwe, 2009b). High yielding and insect pest resistant cowpea varieties suitable for dryland production in Limpopo Province have been introduced from international agricultural centres (IITA, 2014). However, these varieties cannot meet the needs of farmers (Asiwe, 2009a) as only few of them that are well adapted. Development of genotypes with specific traits is essential but with adaptation background. Therefore; specific trait crosses need to be made with adapted varieties for the purpose of developing high yielding cowpea genotypes with combined resistance through insect mediated crosses.

1.3 Rationale of the study

Well adapted and improved cowpea varieties in South Africa are few and farmers are limited with varietal choices coupled with low yield (Asiwe, 2012). It is important that well adapted broad based varieties be developed to avoid relying on exotic varieties. This will increase the availability of varieties that the farmers can grow as well as giving them a choice of varieties to improve their productivity (Singh, 2014). The cultivation of the varieties will in the long term improve the protein consumption of poor rural communities, since cowpea is rich in protein (13-25 %). In addition, it will generate income for South Africa as the varieties will be commercialised and produced largely for export. This will eventually contribute to the national gross domestic product (GDP) in the agriculture sector.

1.4 Purpose of the study

1.4.1 Aim

The aim of the study was to evaluate F3 segregating population for high yielding combined resistant cowpea genotypes in Limpopo Province.

1.4.2 Objectives

The objectives of the study were to:

- i develop F1 hybrid population through insect mediated crosses.
- ii evaluate F3 segregating populations for high yielding and insect pest resistance.

1.5 Hypothesis

- i F3 segregating populations do not differ with their parents.

CHAPTER 2

LITERATURE REVIEW

2.1 Cowpea

2.1.1 Origin, characteristics and importance of cowpea

Cowpea (*Vigna unguiculata* (L.) Walp.) is a dicotyledonous plant from family Fabaceae previously named leguminoseae, order Fabaceae (Saraih, 2010), subfamily Faboideae, tribe Phaseoleae, sub-tribe Phaseolinae, genus *Vigna* and section *Catiang* (Saraih, 2010). It is a diploid species that contains 22 chromosomes ($2n = 2x = 22$) (Mukherjee, 1968). According to Ladeinde (1977), cowpea originated from Asia but its wild species shows the centre of origin is Africa. Abandonon (2004) and Ajeigbe *et al.* (2005) found that all evidence of cowpea's origination pointed back to Africa. This was substantiated by Timko and Singh (2008) who suggested that cowpea might have originated from both Asia and Africa. However, they found that there was no evidence of wild cowpea species in Asia which led them to conclude that cowpea originated in Africa and was only diversified through birds. Worldwide, cowpea is called in various names; in English speaking parts of Africa it is called "Cowpea", in Senegal "Seub" and "Niao", in Nigeria "Wake" or "Ewa", in Sudan "Luba hilo" and in United State it is called "Black-eyed peas or Southern peas (Timko and Singh, 2008).

Cowpea is characterised by erect, semi erect, climbing and trailing growth habits depending on the genotype (Kabululu, 2008; Timko *et al.*, 2007). It is an annual herbaceous crop with a taproot that bears lateral branches with nodules that are responsible for nitrogen fixation (Gomez, 2004). Cowpea flowers are large with straight keels and they come in distinct colours (white, cream, purple, yellow and blue) and open early in the morning and close before noon (Gomez, 2004). Flowers are borne on racemose inflorescence at the end of peduncles and each peduncle carries two to three pods, but can carry four or more depending on the genotype and the environmental conditions (Timko and Singh, 2008). By nature, cowpea is dominantly a self-pollinated plant. However, more than 5 % rates of outcrossing by bumble bees and wild bees have been recorded, thus breeders need to take care and control outcrossing especially where it is not needed to avoid off-types (Timko *et al.*, 2007). Normally, breeding lines in nurseries are planted in separated plots to

avoid cross pollination by these bumble bees while other breeders prefer it in the greenhouse.

The leaves are dark green, shinier, trifoliate and alternating each other (Gomez, 2004) and are shaped lanceolate, linear and ovate (Saraih, 2010). The pod color varies from light green during early stages of maturity to yellow, light brown, pink and purple (Saraih, 2010), while the pod shape is cylindrical and may be curved or straight (Timko *et al.*, 2007). The seeds are round and kidney shaped with a smooth, rough and wrinkled coat that comes in various colours viz; white, cream, red, speckled, brown and black (Saraih, 2010). Cowpea is of epigeal germination, its cotyledon emerges above the ground during germination (Timko *et al.*, 2007).

Cowpea is one of the most important grain legume crops for human consumption (IITA, 2014). It is a great source of cheap protein, minerals and vitamins essential in human diets (Ayo-Vaughan *et al.*, 2013). Cowpea is relatively low in fat and its protein consists of amino acids with high levels of folic acid, fibre for digestive system and vitamin B for prevention of spinal cord defects in unborn babies (Timko *et al.*, 2007). Almost every part of cowpea is consumed; leaves, green pods, green peas and dried grains can all contribute to a delicious meal (Timko and Singh, 2008). Shelled green peas are canned and frozen while others are enjoyed fresh. The seeds are either processed into flour, boiled, dried or dry roasted (Martins, 2014). Cowpea haulms are dried and stored as fodder for winter feeds (Gomez, 2004) this is an added advantage for cowpea growers as they can either sell the fodder to livestock farmers or use them if they have their own livestock (Fatokun, not dated).

According to IITA (2013), cowpea is reported to be a highly economical crop as it can fix atmospheric nitrogen and making it readily available to the plant through a symbiosis relationship with a soil borne nodulating bacteria in its root nodules. This grants cowpea a great chance of surviving in poor soils with low levels of nitrogen, phosphorus and organic matter (Singh *et al.*, 2003). Its ability to fix nitrogen is also an added advantage to farmers as they can save cost of buying nitrogen fertilizers. IITA (2013) further explained that cowpea leaf litters ameliorate the soil by adding organic matter to the soil when decomposed, thus cowpea is desired in intercropping systems (DAFF, 1997). Furthermore, cowpea residues can be ploughed in and incorporated into the soil after harvesting to add organic matter to the soil. Crop

residues also acts as a mat to prevent soil crusting by covering the soil surface and preventing direct effect of rain onto the soil. Cowpea residues help reduce erosion by increasing the soil water contact time thereby increasing infiltration and reducing runoff speed. For the above mention attributes that cowpea possesses; it is now widely called a dual or multi-purpose crop.

2.1.2 Cowpea types

Cowpea plant types can be classified as fodder, grain and dual purpose. Grain types and dual types mature early (takes up to 60 days) and they are tolerant to harsh conditions such as high temperature, drought and poor soil while the fodder types are indeterminate (takes up to 150 days to mature) depending on photoperiod (Timko and Singh, 2008).

i Fodder type

Fodder cowpea type is cultivated for animal feed. Fresh or conserved vines and leaves are used as hay or silage for animal feeding (Feedipedia, 2018). When cowpea is grown for hay, growers cut it when 25 % of the pods have coloured. However, hay quality declines with crop maturity. Under dry-land conditions cowpea yield ranges from 500 kg/ha to 4000 kg/ha dry matter and up to 8000 kg/ha dry matter in irrigated areas (Oushy, 2008). Forage cowpea contains as much as 21.56 % of crude protein. They usually mature late with least tendency to produce grains.

ii Grain type

Grain type cowpea is cultivated for grain or seed. Early maturing varieties of cowpea are often used for grain because shorter growing period escapes drought.

iii Dual purpose

Dual purpose types serve two purposes. They provide both grain and fodder which can be used in different cropping systems (Feedipedia, 2018). They have been genetically modified to contain both grain and fodder type traits. Some also have added benefit of being insect pest and disease resistant. Grain yield from dual purpose cowpea ranges from 362 kg/ha to 907 kg/ha. They are usually intermediate in their maturity. Some of the varieties can out-yield grain types.

2.1.3 Growth habit

Cowpea plant growth habit is characterised as erect, semi-erect, acute erect, trailing, intermediate, semi prostrate and crawling type (IBPGR, 1983). However growth habit is sometimes influenced by the photo-period and growth conditions (Timko *et al.*, 2007). The erect cowpeas are determinate and early maturing type with small leaves and seeds while the spreading and climbing types are indeterminate with both early and late-maturing traits, large and small seeds being produced (Kabululu, 2008).

i Erect

Erect types grow upright at a narrow angle to the ground and they facilitate farm operations like weeding, insecticide application and harvesting through machinery as they allow machinery to pass through easily (Abadassi, 2015). They also do not require much intra-row spacing and grow up to 80 cm (Feedipedia, 2018).

ii Semi-erect

This type is partially erect. They are between upright and spreading type. They grow upright and bend over to trail on the ground if not supported. The branches grow perpendicular to the main stem, but do not touch the ground.

iii Acute erect

Acute erect type's branches form acute angles with the main stem (IBPGR, 1983).

iv Trailing

Their stems trail or coil around plants or any structure as they grow upwards and they grow up to 2 m (Feedipedia, 2018)

v Intermediate

The branches in intermediate growth type touch the ground (IBPGR, 1983)

vi Semi prostrate

The main stem reaches 20 or more centimetres

vii Crawling or spreading

They are prostrate, spreading along the ground or lying flat on the ground. Their width is greater than their height. They suppress weeds as they have tendencies of covering the ground, therefore less weedicide application is needed (Abadassi, 2015).

2.1.4 World and national production

Worldwide, an area of about 14.5 million hectares is cultivated to cowpea, which accounts to 4.5 million metric tons of harvest (Saraih, 2010). Nigeria, Niger, Burkina Faso, Senegal, Ghana, Cameroon and Mali are regarded as the top cowpea producers in the world, with about 50 % of the world cowpea grain being produced in Nigeria (Chiulele, 2010). After Nigeria, Niger is the second world cowpea producing country at 650 000 tons annual yield (Timko and Singh, 2008). Asia, Oceania, Middle East, Southern Europe, Southern USA, Central USA and South America are also considered as notable cowpea producers (Chiulele, 2010).

In South Africa, cowpea is produced in Limpopo, Mpumalanga, North West and Kwa-Zulu Natal provinces (DAFF, 2011). Information on the total amount of hectares farmed on cowpea is not available. According to Asiwe (2009b); for the past thirty years a radical reduction in efficient research of cowpea cultivation has been noted in South Africa. Cowpea production in South Africa is relatively low as compared to other staple crops such as maize, groundnut and wheat. This is mainly because it is less consumed or preferred as compared to other crops. Other reasons include; lack of funding from government to support production, lack of interest by researchers in promoting and improving production and lack of improved quality seed; thus, South Africa's cowpea production level is regarded as subsistence (Asiwe, 2009b).

Cowpea is widely planted under dryland conditions by small-scale farmers as it is drought tolerant (DAFF, 1997). However, for germination adequate water is required for moisture imbibition. According to Mathews (2012), cowpea requires a range of 20-30 °C day temperatures and 18-24 °C night temperatures for seed germination, pod set and dry matter production and it is prone to frost damage as it does not tolerate temperatures below 15 °C. He further explained that cowpea requires well drained, loamy soils with a pH range between 5.0 and 6.5, at a soil planting depth of

2-5 cm (not too deep and not too shallow). Sandy loam or lighter clay soils are ideal for cowpea production. However, due to mycorrhizal symbiosis and nitrogen fixation processes cowpea is tolerant to low fertility soil situations (Saraih, 2010). Cowpea spacing includes; 75-100 cm inter row and 7-10 cm intra-row spacing for bush type varieties and 30-60 cm for vining type (DAFF, 1997).

2.1.5 Cowpea production constraints

Despite cowpea being a hardy crop (drought tolerant and adaptation to poor soils) than other crops, its growth and development is still constrained by several biotic and abiotic stresses that consequently limit yield (Asare, 2012). Major cowpea plant abiotic stresses include; drought (due to erratic rainfall), low and high temperatures and low soil fertility while biotic stresses includes; diseases, nematodes, weeds and insect pests (Asare, 2012; Saraih, 2010).

i Drought

Drought is the major constraint of cowpea production, especially during seed germination and flower setting which could constitute to a tremendous yield loss or total crop failure. Cowpea requires 760-1520 mm of rainfall during its growth period (Davis *et al.*, 1991). Asiwe *et al.* (2005) reported that in most developing countries, low and poorly distributed rainfall is the major cause of drought. There are two types of drought, which are intermitted and terminal drought, based on the time of its occurrence. Chiulele (2010) explained intermitted drought as drought that occurs anytime during the crop's vegetative stage, while terminal drought is one that occurs at the end of the crop's growth stage, affecting the reproductive stage (flowering and seed development). Intermitted drought results in low biomass accumulation due to loss of leaves and reduction in leaf area. Drought during flowering and grain filling results in low number of pods and seed weight. As a drought coping mechanism, cowpea reduces their leaf growth, roll, curls their leaves, and closes their stomata to reduce transpiration losses (Davis *et al.*, 1991).

ii Temperature

Responses of plants to temperature differ across different crop species and for cowpea, 20-35 °C is ideal for the crop to thrive. Extreme temperatures during reproduction will result in less viable pollen, low fertilization and no pod formation

that will further reduce crop yield (Hatfield and Prueger, 2015). Hatfield and Prueger (2015) further stated that minimum temperatures reduce plant biomass accumulation and hence yield. High temperatures results in high respiration which further results in high plant water demand.

iii Soil

Cowpea thrives well in a wide range of soil texture such as heavy clay (with good drainage and well aeration) and sandy soils (Valenzuela and Smith, 2002). However, Valenzuela and Smith (2002) added that cowpea cannot withstand waterlogged condition. Furthermore, unfertile soil with low unfavourable microbial activities, poor soil physical structure and low water holding capacity impairs growth of cowpea (IITA, 1982).

iv Diseases

Cowpea is attacked by at least 35 diseases; depending on agro-ecological area and the stage of plant growth (Masenya, 2016). Major problematic diseases that affect cowpea are; fungal, viral and parasitic diseases.

- Fungal disease

Fungal diseases include; fusarium Wilt, *Pythium* stem rot, Powdery mildew, *Colletotrichum* brown blotch (Magloire, 2005). Fusarium wilt (*Fusarium oxysporum* *vr. tracheiphilum*) results in yellowing of lower leaves, stunted growth and wilting (Oyewale and Bamaiyi, 2013). Infected stem shows brick red tissue when split (N2Africa, 2014). *Pythium* stem rot is caused by *Pythium aphaniderinatum*. It is identified as grey-green water soaked girdle stem while in high humid areas the stem base appears as white cottony mycelia (Singh and Allen, not dated). Powdery mildew causal agent is *Erysiphe polygoni* and it thrives well in dry and shady conditions. Mycelium forms white but turns greyish patches on leaves that scatters and spreads to other plant parts (Singh and Allen, not dated). *Colletotrichum* brown blotch is caused by *Colletotrichum capsici* and *C. truncatum* species. They are mostly found in the tropics and sub-tropics. They form purplish brown discoloration of pods and all the plant parts followed by stem cracking. Infected pods bear black fruiting bodies (Magloire, 2005). Growing resistant varieties can control fungal diseases. Crop rotation reduces the fungal inoculum while certified disease free

seeds can also prevent disease outbreak. Farmers must practise sanitation, which is; tilling land and removing dead plant debris that might be inoculum (Singh and Allen, nd). Fungicides can be used to control fungal diseases. The following are commercially available fungicides for cowpea; Maxim, Maxim-XL, Mefenoxam, Metalaxyl, Mycostop, Ridomil-Gold and Thiram.

- Viral diseases

There are approximately 8 viral diseases which affect cowpea that includes; Cowpea aphid-borne mosaic virus (CABMV), Cowpea golden mosaic virus (CGMV), and Cowpea mosaic virus (CPMV). Cowpea aphid borne mosaic virus (CABMV) is transmitted by aphids. It produce light dark green mosaic patterns on leaves that results in thickening of the affected parts and thereof stunted growth and failure to produce normal pods or no pods if the virus was transmitted in the early stage of crop growth (N2Africa, 2014). Cowpea golden mosaic virus is spread by white flies (*Bemisia spp.*). Leaves of infected plants appear distorted, blistered and yellow in colour (Singh and Allen, not dated). Cowpea mosaic virus is transmitted by beetles, grasshoppers, thrips and several weeds. Its symptoms include; green mottles, severe mosaic, leaf distortion and blistering (Singh and Allen, not dated). Viral diseases can be controlled by sanitation (cleaning tools and hands after touching infected plants, tilling and removal of weeds and dead plant debris), planting certified disease free planting and using virus resistant varieties. Pesticides, sticky cards and pheromones can be used to reduce virus vectors.

- Bacterial diseases

Bacterial pathogens include; Bacterial pustule and Bacterial blight, (Singh *et al.*, 1983). Bacterial pustule (spot) is caused by *Xanthomonas spp.* Its symptoms includes tiny dark water soaked dots under leaves surfaces that enlarges into bigger circular dots (Singh and Allen, not dated) and dark brown necrotic spots on the upper surface of the leaf (Singh *et al.*, 1983). The leaves eventually die off when heavily infected. In young germinating seedlings, bacterial blight turns the seedlings brown-red and they eventually die (Oyewale and Bamaiyi, 2013). Leaves develop round irregular spots that are brown in colour with chloric halos. The spots later advance to the stem and pods resulting in stem breakage and shrivelled seeds (Singh and Allen, not dated). Bacterial diseases can be controlled by pruning infected leaves, using

resistant cultivars, growing windbreaks that will hinder dispersal of inoculum and crop rotation to reduce initial inoculum. Tillage practices may be used to get rid of plant debris. Avoid using sprinkler irrigation as it will create moist conditions that are favourable for bacterial survival. Proper spacing must be used as dense spacing increase humidity which favours bacterial growth.

v Nematodes

Nematodes are a major problem in most cowpea grown areas. The most destructive root knot nematode species that causes damage and yield loss are reported to be *Meloidogyne incognita*, *M. arenaria* and *M. javanica* (Claudius-Cole *et al.*, 2010). High populations of nematodes prevent and stop nodulation while their low populations affect nodulation by slowing it down (Claudius-Cole *et al.*, 2010). Furthermore, nematode damage results in nutrient deficiency and therefore other physiological dysfunctions as it hampers plant water and nutrient uptake (Muthamia and Kimani, 2016).

The use of nematicide to control nematodes is discouraged as they are detrimental to the environment (Muthamia and Kumani, 2016) and they have been banned in some countries. Muthamia and Kumani (2016) further explained that the use of nematode resistant varieties is the best option one can use to control nematodes. They further identified variety KVu 27-1 as the most resistant variety that support low number of *M. incognita* species and that suppresses root galls (Muthamia and Kumani, 2016).

vi Weeds

Weeds offer a good refuge for pests and they reduce yield and growth of crops (N2Africa). The most prevalent weed in cowpea is parasitic weed (*Striga*) commonly known as witchweed (Singh and Allen, not dated). *Striga* has two species namely; *Striga gesneriodes* pre-dominant in Sudan and West Africa and *Alectra vogelii* pre-dominant in Guinea, Sudan, Central Africa, parts of Eastern and Southern Africa (Timko and Singh, 2008). Weeds pose competition of nutrients and water with the main crop, harbouring diseases and insects that damage the main crop (Masenya, 2016). Infestation results in veinal chlorosis and withering of leaves, furthermore, it result in reduced growth, dwarfing and early senescence (Singh and Allen, not

dated). Example of other weeds that affects cowpea includes; Prickly pear (*Opuntia spp.*), St. Johnswort (*Hypericum perforatum*), Alligator weed (*Alternanthera philoxeroides*), Itch grass (*Rottboellia cochinchinensis*) and Milk weed (*Euphorbia heterphylla*)

Weeds can be controlled either manually or chemically. Manual weeding includes using hand hoe or rouging out weeds by hand. Manual weeding must be done 2 weeks after planting and 4-5 weeks after planting (N2Africa). However, manual weeding is labour intensive and encourages more weeds to come out. Chemical use is the well-known most commonly used method of controlling weeds. Chemical control includes pre-emergence (applied before emergence) and post emergence (applied after planting) (N2Africa, 2014). Commercially available weedicides include; Dual Magnum (S-metolachlor) for broad leaved and grass weeds, Poast for annual and perennial grass weeds (Sethoxydim: 2-[1-(ethoxyimino) butyl]-5-[2-ethylthio) propyl]-3-hydroxy-2-cyclohexen-1) and BareSpot (Sodium Chlorate and Sodium Metaborate) for grass weeds, Pursuit (Imazethapyr) for Broadleaved weed and Fulisade (aryloxyphenoxypropionate) for grasses and volunteer weeds (N2Africa, 2014).

vii Insect pests

Major insects' pests that affect cowpea include; cowpea aphid, foliage beetles, flower bud thrips, legume pod borer, sucking bug, leaf eating beetles and storage pest (Bruchid). The most prevalent insect pests in Limpopo province are aphids and pod sucking buds.

- Cowpea aphids

Cowpea aphid (*Aphis craccivora* Koch.) is from class Insecta, order Homoptera, family Aphididae and genus Aphis (Dixon, 1985). They are one of the world's most injurious insect pests affecting various plants worldwide (Ali *et al.*, 2013). It is rated as an important pest affecting cowpea production in Africa, Asia and Latin America (Souleymane *et al.*, 2013; Jackai and Singh, 1988). Its origin is not documented. Cowpea aphid is described as a 1.5-2 mm long, shiny black, pear shaped pest divided into a head, thorax and abdomen (Jackai *et al.*, 2001; Potarot, 2012), while its nymph is smoky grey and waxy (Sariah, 2012). Under favourable conditions,

aphid lifecycle can be completed in 13 days. However, during cold condition an adult aphid take about 22 days to fully develop (Amoah Amoah, 2010). The females can give birth to 100 nymphs in 30 days, hence the rapid population (William, 2014). Cowpea aphid causes damage by piercing and sucking terminal shoots, flowers and pods of a cowpea plant and extracting the phloem sap that is essential for plant growth and reducing photosynthesis by secreting honeydew that promotes fungal growth on the leaves (Soffan and Aldawood, 2014; Aliyu and Ishiyaka, 2013). Aphid infestation results in leaf distortion, stunting and reduction in pod set (Potarot, 2012). Cowpea aphid is a vector of viruses such as Broad bean yellow mosaic virus and Bean leaf roll virus (Aliyu and Ishiyaka, 2013).

- Foliage beetles

Foliage beetle (*Oothea mutabilis*) not only feeds on cowpea plant but it is also a vector of Cowpea mosaic virus. Its adult colour varies from shiny reddish brown, black to brown with an oval shaped body that is 6 mm long. The adult feeds on the cowpea leaves, punching holes into leaves while the larvae feed on roots (Singh and Allen, not dated).

- Flower thrips

Flower thrips (*Megalurothrips sjostedti*) appears as shiny black (Singh and Allen, not dated). Their life circle takes about 14-18 days, the pupae is produced in soil and the eggs are laid in the flower buds (Singh and Allen, not dated). They mainly attack first the flower buds and then the flowers resulting in flower abortion. Thrips distort malform and discolour floral parts of cowpea. When severe, flower thrips cause plants to stop producing flowers or flower buds (N2Africa, 2014).

- Legume pod borer

Legume pod borer (*Maruca vitrata*) belongs to the order Lepidoptera, family Crambidae and Genus *Maruca* (Taylor, 1967 in Murna, 2014). It is most prevalent in Sub-Saharan Africa where temperatures and relative humidity are high. They thrive well under tropical and subtropical conditions. Legume pod borer larvae are identified as two pairs body segmented whitish in colour with black spots while the adult is brown in colour with large eyes, long antenna with brown forewings and mixture of brown on the vertex (Murna, 2014). Both the larvae and adult attack

cowpea from flower initiation to pod maturation and they are nocturnal. Legume pod borer cause damage to cowpea plant by producing silk like material that forms a web and stick together flower pods, stems and leaves. After sticking them together the pod borer then feeds on them piercing holes into them.

- Pod sucking bugs

There are various species of pod sucking bugs namely; *Anoplocnemis curvipes*, *Riptortus dentipes*, *Acanthomia spp* and *Nezara viridula*. They are all dominant in the tropics and *Nezara viridula* is both dominant in tropics and subtropics (Singh and Allen, not dated). *Anoplocnemis curvipes* life cycle takes about 24-84 days and their eggs are grey in colour and the adult are black in colour and 3 cm long. *Riptortus dentipes* adult are white with yellow lines on their sides and are cylindrically shaped (Singh and Allen, not dated). *Acanthomia spp* are grey coloured with cylindrical shape and longer spines while *Nezara viridula* is shiny black with bright spots (Singh and Allen, not dated). They all cause damage by sucking sap from green pods resulting in dry shrivelled seeds and further loss of seeds.

- Flower beetles (Blister beetles)

Blister beetles (*Mylabris spp.*) are pre-dominant in Africa and Asia. *Mylabris spp.* Includes; *M. farquharsoni* and *M. bipartia* which are common in Africa and *M. pustulata* which is common in Asia (Singh and Allen, not dated). They appear as bright coloured with broad black yellow or red bands, elongated and narrow shaped. They feed on flowers resulting in total crop damage and yield loss (N2Africa, 2014).

- Storage pest

Storage pests are pests that affect cowpea in storage. The most common storage pest in cowpea is the weevil (Bruchid). Cowpea weevil, (*Callosobruchus maculatus*) belongs to family Coleoptera Bruchidae, genus *Callosobruchus*. The female weevil is large and dark coloured while the male is small and light coloured. Its life cycle takes up to 35 days and it consists of egg, larva, pupa and adult stages (Letsoalo, 2015). Cowpea weevil attacks stored seeds causing damage to endosperm, reducing dry weight, grain quality and seed viability. The larvae feed on the cotyledons and it emerges out of the seeds as adult piercing holes on the seed (N2Africa, 2014). Within a storage period of 100 days, the resistant varieties show about 25%

damaged seeds in contrast to 95% damage for the susceptible varieties (N2Africa, 2014).

2.1.6 Methods of insect control

i Physical and cultural control

Physical control involves altering the environment or controlling pest through the use of devices (traps, barriers, fences and nets), machines (plows, disks, mower and cultivator) and other physical methods to reduce the important sources that pests needs for survival (food, shelter and water) (Hartzfield and Sargent, 2017). Machines (plow, disks, mower and cultivator) remove weeds that offer shelter to pests and removing dead plant parts and crop residues. Traps physically catch pests by snaring or sticking them. One example is using traps such as *Crotalaria juncea* to divert insect attack away from cowpea crop and using flies and wasps to consume the legume pest for its own nutrition is also efficient way of reducing legume pod borer population on cowpea plants. Location is one of the most important things to look at. The location must provide optimum conditions that suit the crop and it must be thoroughly prepared or cleared to avoid pests and weeds from previous crop from surfacing (Croplife International, 2014). However, tillage promotes infiltration, thus minimum tillage together with resistant varieties can be used to reduce crop protection measures (N2Africa, 2014). Crop rotation is also effective in controlling pests as it discourages pest build up by breaking the pest life cycle and also improves soil fertility i.e. legume-cereal crop rotation.

ii Biological control

Biological control includes using pests' natural enemies (insects or pathogens) to control or suppress pest (insects, mites, fungus, animal and weed) populations (Hartzfield and Sargent (2017). It involves the release of large numbers of natural enemies into the field to control specific pests periodically because they do not give long term results. There are two major classes of natural enemies namely; specialist and generalist natural enemies (Snyder and Ives, 2009). Specialists have a narrow diet (feed on specific prey species) and require specific environmental conditions and if the environment does not suit it, it will die and can ultimately become extinct (Hanski *et al.*, 2001; Snyder and Ives, 2009). Generalists survive on many different preys and give themselves a sort of environmental buffer when resources begin to

run out such that they can adapt to a changing environment (Hanski *et al.*, 2001; Snyder and Ives, 2009). Example of generalists includes; arthropods, herbivores, detritivores, predators and detritus. Arthropod serves as prey for bigger predators and one of their examples is the lady beetle (Oor, 2009). Another example of generalist is *Orias* spp. which is an enemy of aphids and white flies (Karuppaiah and Sujayanad, 2012). Specialist example includes *Aphidius ervi* which is natural enemy for pea aphid and it is effective because it has high reproduction rates than the prey (multiplies quickly) (Hanski *et al.*, 2001). To control weeds, nematodes and fungi are used as natural enemies, predatory mites control spider mites and lady beetles are used to control various insect pests (Croplife international, 2014). Biological control does not pollute the environment; it uses natural enemies of pests for their control (Wikipedia, 2015). It is free of side effects, safe to handle or use, occurs naturally, has high degree of host specificity, is cost effective, survives at low host density and is self-perpetuating (Khaliq *et al.*, 2014).

iii Chemical control

Chemical control methods are the most effective yet they pose serious negative impacts such as residual toxic effects to humans, target plants, foods and other living things, induction of insect/pathogen resistance resulting to ineffectiveness of pesticides; and have harmful effects to non-target beneficial organisms and unbalanced ecosystem due to pollution of soil, water and environment (Fatakun, 2009). Furthermore most farmers are not well knowledgeable to protect themselves against chemicals and chemicals are expensive. Thus chemicals are used mostly by commercial farmers than subsistent farmers. The following insecticides are commercially available for control of insects in cowpeas; Adios (Diphacinone and Imidacloprid), Azadirachtin (Azadirachtin), *Bacillus thuringiensis* (*Bacillus thuringiensis* subspecies), Capture (Bifenthrin), Di-Syston (Disulfoton), Gaucho (Imidacloprid), Insecticidal soap (Potassium laurate), Lorsban (Chlorpyrifos), Methaldehyde (Methaldehyde), Methomyl (Carbamate), Pyrethrin (Pyrethrin, Cinerin I & II and Jasmolin I & II), Sevin (Carbaryl), Success (Spinetoram), Telone (1,3 dichloropropene- Telone) and Trilogy (clarified hydrophobic extract of neem oil) (N2Africa, 2014).

iv Host plant resistance

Host plant resistance includes breeding plants and animals to resist specific pests (Hartzfield and Sargent (2017). Hartzfield and Sargent (2017) further explained that for plants to become resistant to certain insect pests, genetic material from certain insect destroying organisms are transferred into hybrid seeds and the host plant's health and nutrition must be maintained. However, certain plants are naturally resistant to pests and some are naturally repellent to certain insect pests.

Genetic engineers have designed *Bacillus thuringiensis*, a soil bacterium that produces a parasporal crystal made up of Cry- proteins that is toxic to specific group(s) of insects (Karuppaiah and Sujayanad, 2012). There are *B. thuringiensis* containing crops, trees, and microbes to control a range of insect pests like, maize borer, cotton bollworm, potato beetle and beet armyworm (Oor, 2009). However researches have pointed out non *B. thuringiensis* genes that have potential to interfere with the development and nutritional requirements of different insect pests namely; Chitinase, Proteinase Inhibitors, Plant Lectins, Bean α -Amylase Inhibitors and venoms. Chitinase enzymes target chitin in the mid gut causing severe and fatal abrasion of the insect's gut lining that results in disrupted feeding and insect mortality (Hanski *et al.*, 2001). Proteinase Inhibitors interfere with the activity of mid gut and help in drawing the insects' essential nutrients thus causing nutritional limitations (Hanski *et al.*, 2001). Plant lectins represent a class of a heterogeneous group of proteins that specifically bind with sugars to cause sugar limitations that result in no energy for insects. Venoms are poisonous insect species such as scorpions and spiders that form a specific class of heterologous proteins, which exert a neurotoxic effect in other insect species (Oor, 2009).

v Integrated pest management (IPM)

Croplife international (2014) and Kusi *et al.* (2016) define Integrated pest management (IPM) as incorporating all available pest control techniques (cultural, physical, biological and chemical methods) that maintain pest population below threshold level while conserving the environment and people's health at low cost. According to Hartzfield and Sargent (2017), IPM keeps a balanced ecosystem as it uses minimal chemical and more of both physical, cultural and bio-control methods to control pests. Minimal chemical use helps to avoid destroying certain species that

are beneficial (i.e. natural enemies). Chemical use leaves few natural mechanisms to consume pests. Besides these, chemical use is mostly non sustainable as it requires training, specific precautionary measures, proper application rate and proper timing (Croplife international, 2014). Moreover, pests have a habit of becoming resistant to pesticides and applied pesticides can be washed off by irrigation. IPM saves money as pesticide use will be minimised, that is; pesticides will only be used when infestation has reached economic threshold (Economic threshold is population level where control should be started to stop pest population from causing yield loss) (Herzfield and Sargent, 2017). IPM practices promote a healthy environment as IPM ensures low contamination of groundwater and low pesticide container disposal. The question is how does IPM use one or combinations of insect pest control methods to reach its goal? Kasi *et al* (2016) explained that IPM uses three components namely; prevention, monitoring and intervention. Prevention includes use of physical and cultural control methods such as crop rotation, tillage, selection of a conducive growth location and many more to prevent pest build up while monitoring includes scouting for pests and diseases and intervention is where control measures such as physical, biological and chemical methods that pose less harm to the environment and people are used depending on the severity of the pest (Kasi *et al.*, 2016). Barrier of IPM is that most farmers are reluctant to adopt IPM practices (Murna, 2014). Farmers are afraid of change and prefer doing things the old. Pretty (2001) reported that the high cost of purchasing improved technologies such as hybrid seeds also results in farmers being reluctant to adapt to IPM. Murna (2014) further explained that farmer participatory approach can be used to help farmers to adopt this practice.

2.2 Cowpea breeding

2.2.1 Hand crossing

Cowpea hand crossing is usually done under greenhouse conditions to reduce the effect of insect mediated outcrossing, high air temperatures and low humidity. However, the number of flowers per plant is low which limits the number of cross pollinations that can be made (Rachie *et al.*, 1975). Successful crosses can still be made under field conditions with many plants per plot that enable the possibility of having more crosses, in other words, the more crosses you have the higher the percentage of success (Zary and Miller, 1982). Crosses also depend on synchronisation of flowers. It is wise to plant late maturing varieties ahead of early

maturing ones, particularly when used as females to avoid delay in flowering (Rachie *et al.*, 1975).

Cowpea is self-pollinating by nature, so in hand crossing; emasculation must first be done (Gomez, 2004). Emasculation and pollination can be done any time of the day but pollination is preferable at the close of evening when temperatures are cool, as hot temperatures during the day dry up the pollen and make it non-viable (Rachie *et al.*, 1975) Flowers which ought to open the following morning are emasculated by cutting the keel gently with a forceps to expose the anther and the stigma, the anthers are then removed without injuring the stamen as it is fragile and without busting the anthers to avoid self-contamination, this will be used as the female (Vaz *et al.*, 1998). Flower chosen as male is slipped downwards and a mass of pollen on the style is used as brush to introduce pollen grains on stigma of an emasculated flower (Rachie *et al.*, 1975). Flowers drop within 24 hours if pollination is unsuccessful.

2.2.2 Outcrossing (Insect mediated crossing)

Outcrossing is a breeding practice where genetic variability is increased through mating or crossing different plants of the same species that are not closely related. A simple definition is that it is unintended pollination by an outside source of the same crop during hybrid seed production. Ants, thrips, (Rachie *et al.*, 1985) bumble-bees and honeybees (Asiwe, 2009a) are reported to be pollinating agents in outcrossing genotype fields. Outcrossing is not usually desired by breeders as it creates unwanted genotypes (offtypes) in the field and mixes up seed lots thereby reducing genetic purity of the seed lot (Asiwe, 2009a). Asiwe (2009a) further stated that they leads to a lot of costs as offtypes need to be rouged out and mixed seed lots have to be sorted, which is labour intensive. However, in cases where there are no favourable conditions for hand crossing, breeders resort to well-planned outcrossing for the development of promising varieties. Insects and bees responsible for outcrossing are destroyed by insecticides in the field which constitutes to low outcrossing rates (Fohouo *et al.*, 2009). It is advised not to spray any insecticides in outcrossing field and the fields have to be planted in isolation from other fields where insecticides are used for successful outbreeding. Few works have been documented on outcrossing. Asiwe (2009a) reported that in cowpea, outcrossing ranges from

0.50-0.85 % and it can be enhanced through alternate row arrangement of parents needed in the crossing. In rape and white mustard, outcrossing rate has been reported to be 20-35 % (Gosta, 1960).

Literature has shown that when you cross white cowpea with brown cowpea the offspring (F1) will be black seeded (Asiwe, 2009a). This concept is usually used to determine outcrossing success despite that there are genetic markers that can be used to test this, which are expensive (Fahouo *et al.*, 2009). However, this is disadvantageous, as breeders cannot tell through mere observation whether outcrossing was successful between brown and brown seed coated cowpea and between white and white seed coated cowpea. This also limits breeders to which traits to cross.

2.2.3 Isolation plot for open pollination

Isolation distance is the minimum separation required between two or more varieties of the same species for the purpose of keeping seed pure or to avoid contamination by foreign pollen (McCormack, 2004). The greater the degree of purity desired, the farther the isolation distance required. Isolation distance limits the number of plants per plot hence it is always wise to decrease the isolation distance using the following techniques (McCormack, 2004);

- i Collect seed from the centre block plantings as seeds produced at the centre of the block are more likely to produce pure seeds.
- ii Use barrier crops to distract pollinators by providing alternative pollen and nectar sources which may attract pollinators away from the main seed crop.
- iii Mechanical isolation can be used if isolation in time and space is not practical. This involves using physical barriers such as building pollination cages or bagging flowers to prevent unwanted pollination.

In cucumber, treatment combinations are planted in isolation blocks separated by a minimum of 2 km to prevent intercrossing of treatments (FAO, 2012). According to Setimela *et al.* (2006), in maize open pollinated varieties, isolation distanced of 250-350 m is required to ensure that pollen from neighbouring fields does not contaminate the field and breeders prefers to plant in small plots for better monitoring. Setimela *et al.* (2006) further explained that isolation in time is done by

delaying planting of other field by 4-6 weeks to delay tasseling and silking. Asiwe (2009a) recommended alternative row planting as a technique for enhancing cowpea outcrossing.

2.2.4 Heterosis (Hybrid vigor)

Heterosis and hybrid vigour are both used interchangeably as synonyms, however, according to Hagberg (not dated), heterosis is a mechanism which results in hybrid vigour, while hybrid vigour is the end result of heterosis. A better and more comprehensive definition was given by Lamke and Edwards (not dated) that heterosis is a phenomenon which results in improved performance and traits observed in F1 progeny when compared to their parents. Heterosis was first discovered during the early 1970s by Joseph Koelreuter when he realised that height of F1 progeny of tobacco plant was greater than that of its parents (Ryder *et al.*, 2014). Heterosis can be negative or positive as it can increase a trait or decrease a trait in comparison with its parents. Both negative and positive heterosis are desired as negative heterosis results in early maturity and positive heterosis results in increase in yield (Kyu, 2011).

In agriculture, heterosis is important as it alters traits such as flowering time, biomass tolerance to biotic and abiotic stress and maturity time (Ryder *et al.*, 2014). High values of heterosis indicate good genetic diversity among cowpea varieties. Large number of pollinations must be made to obtain enough F1 seeds to test as this will give a good range of choice for selection (Fatokun *et al.*, 2002). Choice of selection is made on desirable traits that are superior to their parents (Lamke and Edwards, not dated).

2.2.5 Mechanism of resistance in cowpea to insect pests

Insect pests are a major constraint in cowpea production (Fatokun *et al.*, 2002). Every stage of the cowpea life cycle has at least one major insect pest that could damage the crop or completely wipe it out resulting in huge yield losses (Asiwe, 2009), therefore the crop needs to be sprayed with insecticides very often. However, insecticides are expensive and certain insects tend to develop resistance towards insecticides, thus breeders are introgressing genes of resistance into cowpea varieties. Progress has been reported in improving cowpea resistance to aphid,

thrips and bruchid (Fatokun *et al.*, 2002). However, in South Africa some of the released varieties are susceptible to some major cowpea pests and yield of cowpea remains low (Ayo-Vaughan, 2013 and Asiwe, 2009a).

Mawandu (1985) reported that a cowpea plant is likely to be infested by cowpea aphid on the stem and leaves than on any other plant part. He further concluded that between the two varieties she studied, variety Kano white is more susceptible to cowpea aphid than Ife brown (Mawandu, 1985). However, IITA scientists have developed improved cowpea varieties that are high yielding to safeguard food security in Africa, but not specifically in South Africa (IITA, 2014). Some of the varieties that are cowpea aphid resistant include TVu 408, TVu 3417 and TVu 3509 (IITA, 1982).

Painter (1958) listed and described three mechanism for resistance namely; non preference, antibiosis and tolerance

i Antibiosis

Antibiosis is a situation when the insect feeds on the plant it changes its behaviour (loss of appetite), insect biology (mating) and physiology (loss of weight) (Painter, 1958).

ii Tolerance

Tolerance is a state where a plant, despite infestation by large population of insects that could possibly cause severe damage, recovers from damage, withstands damage or suffers little damage instead (Painter, 1958; Salikutty and Peter, 2007). He further reported that cowpea varieties with pigmented calyx, petioles and pod are less susceptible to pod borer (*Maruca vitrata*). Tolerant hosts expose the pests to their natural enemies and thus keeping pest population at their lowest.

iii Non-preference

Non preference is a situation where the plant is unattractive to the insect resulting in the reduction of the use of the plant by the insect for feeding, shelter and oviposition purposes or both (Painter, 1958). In non-preference Asare (2012) further explained that a plant may have physical features or exudate chemical properties that will pull away the insect. Non preferred crops escape infestation by pests as they host pests,

the pests lay few eggs and thus produce low populations as host does not provide optimum conditions for them to multiply rapidly.

2.3 Management of segregating populations

Segregating populations are defined as “genetically diverse progeny from crosses between parents that differ for one or more traits that are under genetic control” (Bailey, 2015). F1 Hybrid seeds are grown along with their parental plants for comparison. Phenotypic traits that are easily observable such as pod colour of the purported hybrid seed is used for comparison (Jayaprakash *et al.*, 2017). Segregating populations are managed using agronomic practices that raise a good crop. Usually, 4 metre rows at spacing of 60 x 20 cm are recommended for planting segregating cowpea along with their parents (Biradar *et al.*, 2007).

2.4 Varietal selection

Varietal selection depends on the mode of reproduction of your plant species. The mode of reproduction determines the genetic composition of the crop and thus the type of breeding program and selection methods suitable for that crop (Chahal and Gosal, 2002). The mode of reproduction includes; self-fertilisation, cross pollination, asexual propagation and apomixis. Different selection methods applied in crop breeding viz; mass selection, single-plant pedigree, bulk population, backcross, recurrent, F1 hybrid and single seed descend.

- Self-fertilising crops
 - Mass selection

Selection is made based on phenotypic performance and it is only effective for highly heritable traits. Selected plant seeds are bulked for next generation (Takebayashi and Morell, 2001).

- Single plant selection

Single plant is selected and their seeds are kept apart and used to perform offspring tests (studying breeding behaviour).

- Selection of cross pollinated crops
 - Mass selection

Individual plants are selected based on their performance and then bulked to produce next generation seeds.

- Recurrent selection

Plants with desired characteristics are selected and then self-pollinated for genetic recombination. The progenies are inter-crossed in all possible ways and their resulting offspring are selected to then constitute to the next generation (Frisch and Melchinger, 2005).

- Selection of segregating populations
 - Pedigree method

Individual plants are selected at F2 generation based on performance and selection is continued until the progeny shows segregation. A pedigree record of progeny and parent is kept (Frisch and Malchinger, 2005). F1 plants are grown to produce F2 seeds. At F2 selection of individual plants begins based on the plant behaviour. The selected plants are harvested and record is kept (Biradar *et al.*, 2007). At F3 selected plants are planted along with their parents for comparison. Promising individual plants are re-selected, individually harvested and record is kept (Biradar *et al.*, 2007). Progeny selected at F3 is grown to produce F4 families. One F3 plant constitutes a family. At F4 selected plants are grown in rows and variations within families decreases (Jayaprakash *et al.*, 2007). At F6-F8 preliminary yield trials of selected families are done. Multi-locations trials are conducted at F7-F12. After all selection steps are taken the selected families are then released as varieties.

- Single seed descent method

At F2, large numbers of plants are carried forward through single seed from each plant until F6 generation (Biradar *et al.*, 2007). The seeds of single plants are then planted in rows for multiplication trials.

- Bulk method

Seeds are harvested in bulk from F2-F6 generations and individual plants are selected afterwards (Biradar *et al.*, 2007). F1 plants are grown along parents and selfed progeny are removed. F2 are then harvested. From F2 to F4, F2 seeds are planted and harvested in bulk, and samples of harvested seeds are collected (Biradar *et al.*, 2007). This process is repeated in F3 and F4. F5 seeds are harvested. Bulked F5 seeds are planted and desirable plants are then selected and

harvested individually. At F6, single plant progeny rows are then planted, uniform, promising progenies are selected and selected progenies are bulk harvested separately (Jayaprakash *et al.*, 2007). Preliminary yield trials are conducted from F7-F9, tests are then repeated in multi-locations at F9 and the last progeny is selected and released as a new variety (Biradar *et al.*, 2007).

2.5 Work not done on the research problem

Cowpea breeding in South Africa is springing up. Cowpea varieties that are high yielding and insect pest resistant have been introduced and released in South Africa with single trait of either yield or insect resistance. Thus there is a need to introduce stacked genes of high yielding and insect pest resistance into a variety with good agronomic background to save costs of insecticides, to produce cowpea within a short period of time, evade drought and increase availability of varieties suited to smallholder farmers, who often do not have resources for insecticides.

CHAPTER 3

MATERIALS AND METHODS

3.1 Study Site Description

All experiments were performed at University of Limpopo Experimental Farm, Syferkuil. Syferkuil is about 40 km east of Polokwane in Limpopo Province South Africa. The site is located at 23°49'S and 29°49'E with average temperatures ranging from 28-30°C during summer and 400-600 mm of rainfall throughout the year. The soil at Syferkuil farm is sandy loam with a neutral and alkaline pH values. The first study was conducted during December 2015 while the second experiment was conducted during January 2017. Both experiments were not on irrigation. Supplemented irrigation was used for seedling establishment.

3.2 Procedures, Treatments and Research Design

i Methods for achieving objective 1 (Development of F1 population)

The first study was to develop F1 population. Parental lines that have good promising traits viz; TX08-30-1 (white), IT98K-205-8 (white), GEC (white), IT98K-589-2 (white), IT97K-499-35 (white), IT08K-827-11 (white), IT07K-243-1-2 (white) and IT10K-815-5 (white) were planted in alternate rows of 1m apart with GLENDA (brown) and TX08-30-5 (brown) for pollination by insects as described by Asiwe (2009a). F1 seeds that had black color were selected as progeny from outcrossing between female and any other participating member of the population. According to Asiwe (2009a), evidence has shown that when you cross brown cowpea seed with black eye and white cowpea seed with black eye, F1 seed coat will be black. Black seeds were harvested from the plots as F1 segregating population. It is on this basis that materials planted in isolation for outcrossing by insects were selected for advancement.

ii Methods for achieving objective 2 (Assessment of population)

The seeds derived from the crosses were planted in the field for multiplication and selection that constituted the F2 population. Materials selected at F2 were evaluated as F3 populations for development of high yielding combined resistant cowpea genotypes. Randomised complete block design (RCBD) was used in three

replications to assess and test the developed F3 genotypes as segregants along with their parental lines (Figure 1). Land was prepared using a disc plough and harrow. The field was laid out and demarcated using a measuring tape. Single row plots with 1 m inter row spacing and 35 cm intra row spacing was used. The rows were 4 m long. F3 seeds were planted at a recommended depth of 2-5 cm. Irrigation was scheduled weekly and manual weeding was carried out as when needed. Diseases were scouted weekly. Insect pest control was effected using Aphox (Carbamate) and Karate (Lambda-cyhalothrin) insecticides at the application rate of 1L per hectare as described by Asiwe *et al.* (2005) (Figure 2). At harvest, selection was made based on visual evaluation (desired traits such as long pod length, bigger seed size, erect pods and yield).



Figure 1: Field trial for the development of high yielding and pest resistant cowpea F3 genotypes.



Figure 2: Spraying Aphox insecticide to control Cowpea aphids.

3.3 General data collection

The following phenological and yield potential data were collected;

i Canopy height and width

Canopy height and width were measured using a ruler. They were randomly sampled from three plants of each genotype at maturity from every pedigree screened. Plants with broader canopy widths also are desired for fodder and vegetable.

ii Peduncle and pod length

Metre ruler was used to measure peduncle length. Three plants were randomly selected and three peduncles and three pods of each plant were measured per genotype at maturity.

iii Number of pods per plant

Three plants were randomly sampled from each genotype per pedigree to determine number of pods per plant by counting.

iv Pod weight

Pod weight was weighed at harvest. Overall weight of all genotypes per pedigree was done using electronic scale.

v 100 seed weight

100 seeds were sampled from three plants of each genotype per pedigree and weighed using electronic scale CBK Bench check weighing scale CBK 8H (max 8kg d=0,1 g) to determine 100 seed weight. Farmers desire Cowpea with large seeds as they tend to have a high germination percentage and appeal to the eye.

vi Grain and fodder yield

Grain was measured from each pedigree using an electronic scale CBK Bench check weighing scale CBK 8H (max 8kg d=0,1 g) and hanging scale was used for fodder yield. For grain yield, pods from all plants in each pedigree were harvested; sun dried, shelled and weighed while fodder yield was determined by uprooted all plants at physiological maturity, dried and measuring them per pedigree. Grain and fodder yields determine the type of cowpea. Varieties with more grain yield than fodder have the potential of being further evaluated for grain type traits while those that have both greater grain and fodder yield quantity can further be evaluated for traits of dual purpose cowpea type.

vii Aphid incidence

Before insecticide application, aphid incidence was taken by counting number of plants with or without aphids and expressed as a proportion (%) of total number of plants per plot. Aphid incidence expresses whether they were any occurrence of aphids or not.

viii Aphid severity

Aphid severity was determined using 9-point scale described by (Jackai and Singh 1988, Jackai *et al.*, 2000). Resistance was measured using 9-point scale where 1=3 resistance, 4=6 medium resistance and 7=9 susceptible.

ix Genotypes advanced

Genotypes were advanced through selection. Selection was made based on farmer's needs, phenotypic fitness and overall performance of the genotype as described by Sarutayophat (2008).

3.4 Data analysis

Data were analysed using non parametric data analysis. Means of genotypes were arranged into frequency tables to determine the number of genotypes that were better than the parent or less than the parent.

CHAPTER 4

RESULTS AND DISCUSSION

4.1 Assessment of population

Means of genotypes showed significant differences among themselves and their parents; where most parents were superior over their progeny. However, this conclusion was drawn based on averages. Among those genotypes, there were roughly 4-5 plants that showed superiority over their parents as means are affected by extreme values therefore frequency distribution tables were used. Results were taken from seven pedigrees; however, only five pedigrees had enough genotypes (Table 2). Therefore, pedigree with the large number of genotypes were presented and discussed (Table 2).

Table 1: Parental traits

Parents	Aphid resistance
GEC	Susceptible
GLENDA	Susceptible
TX08-30-5	Resistant
IT97K-499-35	Resistant
IT98K-205-8	Resistant
IT98K-589-2	Resistant
TX08-30-1	Resistant

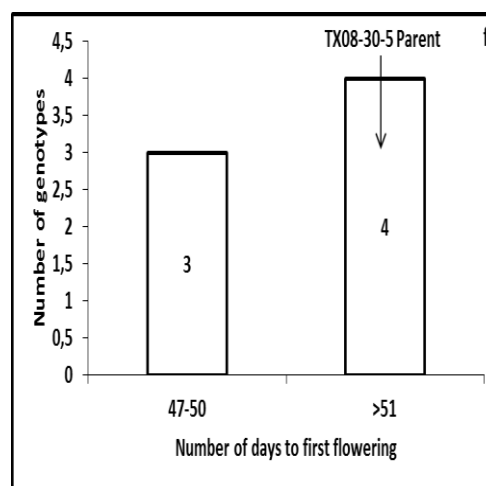
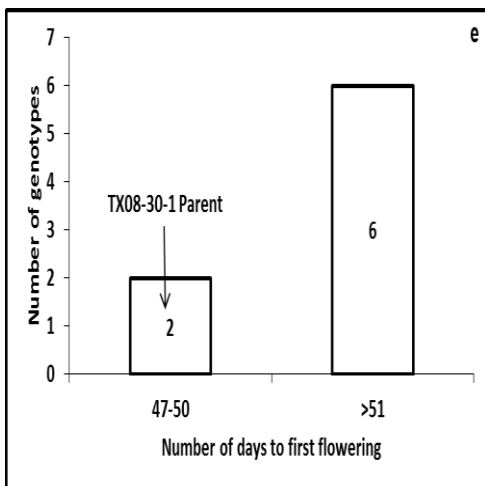
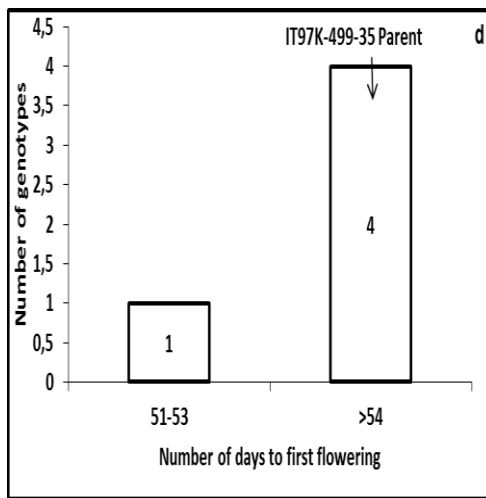
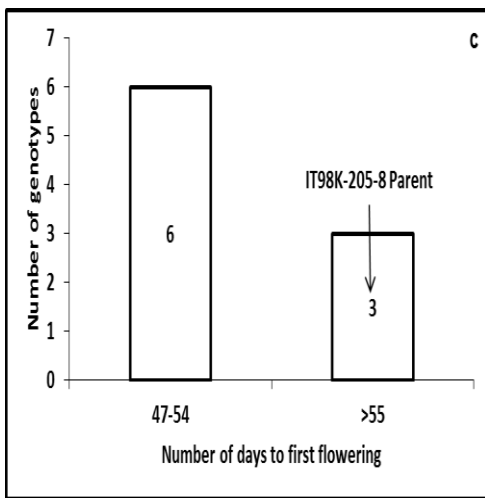
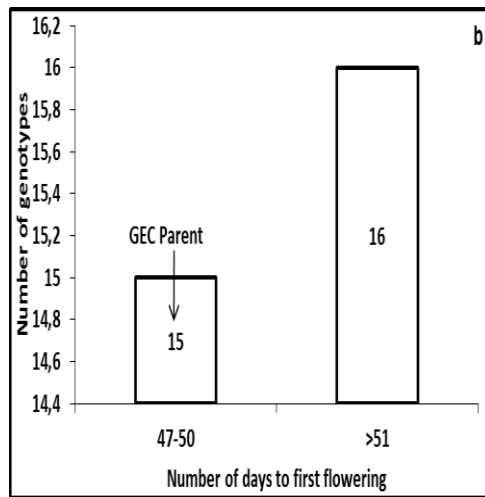
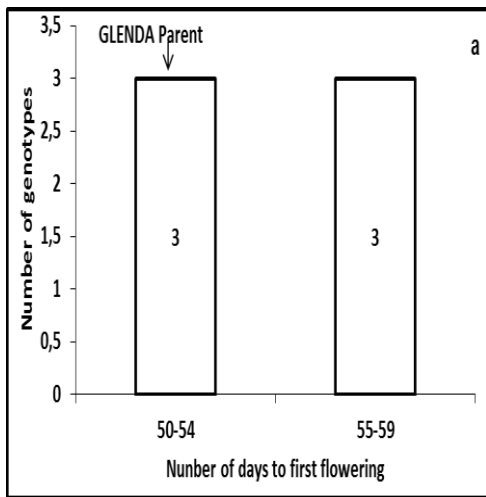
Table 2: Number of genotypes at F2 and F3 per pedigree

Parents	F2 genotypes	F3 genotypes
GEC	7	31
GLEND A	10	6
TX08-30-5	10	7
IT97K-499-35	1	5
IT98K-205-8	6	9
IT98K-589-2	1	3
TX08-30-1	5	8

4.1.1 Number of days to first flowering

The results as shown in Figures 3 (a, b, c, d, e and f), illustrate that genotypes obtained from pedigrees IT98K-205-8, TX08-30-5 and IT97K-499-35 were identified as promising pedigrees with potential to be further evaluated for early maturity. Six genotypes from pedigree IT98K-205-8 flowered (47-54 days) earlier than their parent while three genotypes from pedigree IT97K-499-35 flowered (47-50 days) earlier than their parent and one genotype from pedigree TX08-30-5 flowered (51-53 days) earlier than their parent. These materials will be further evaluated as promising early maturing segregating populations (Ayovaugan et al., 2013). Also, they have the potential to be selected for drought prone areas to help evade drought. Number of days to first flowering is important as it helps farmers to predict when the crop can be ready for harvest and in preparations of other agronomic activities.

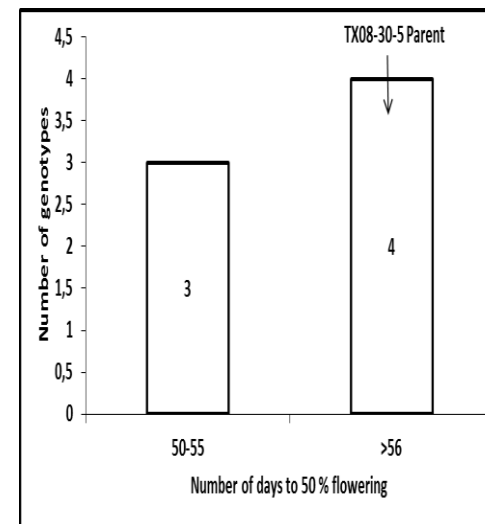
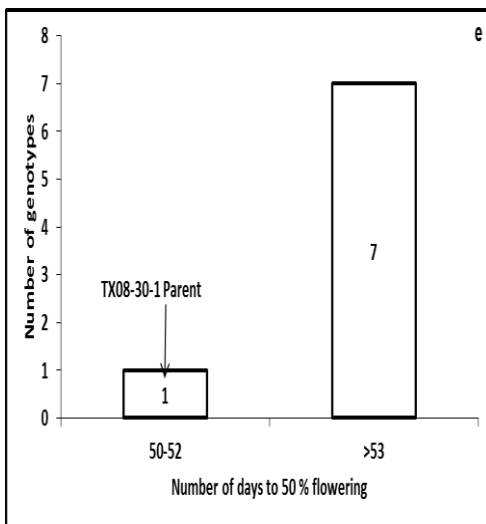
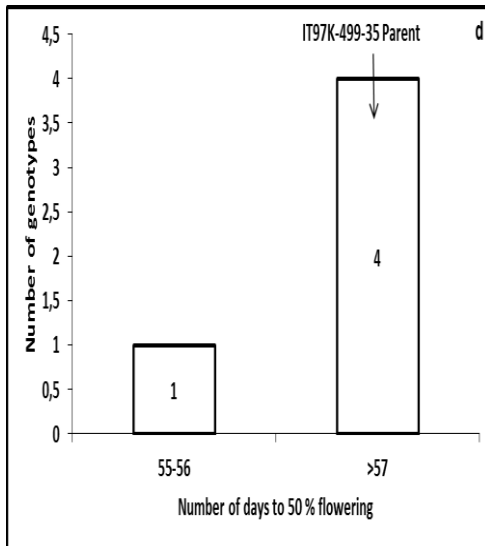
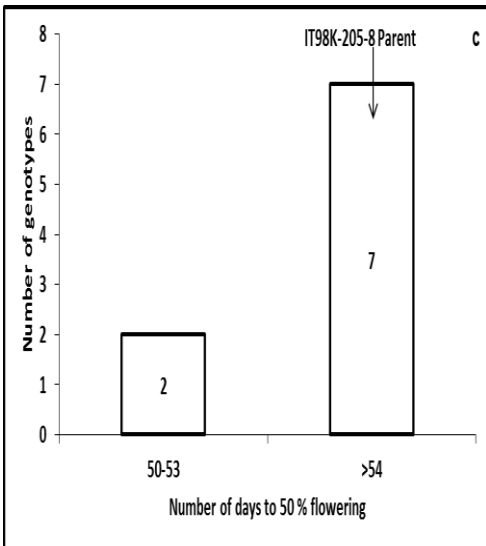
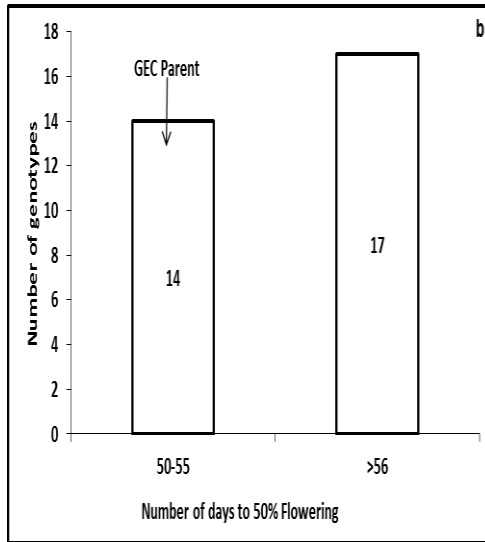
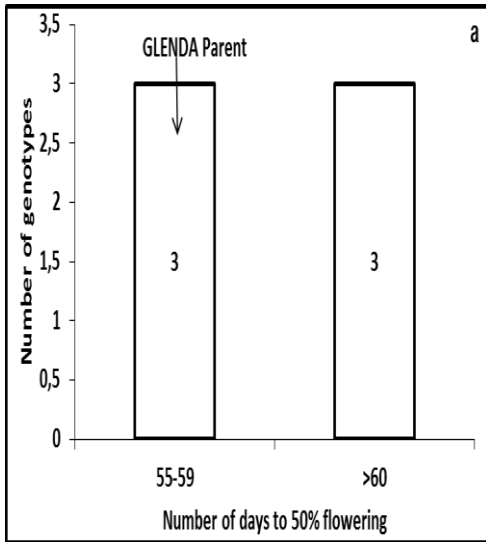
These were compared to genotypes obtained from pedigrees GLEND A, GEC and TX08-30-1 which flowered later than their parents. Genotypes derived from pedigree GLEND A had three genotypes that flowered at 55-59 days, while genotypes obtained from pedigree GEC had sixteen genotypes that flowered (>51 days) and genotypes obtained from pedigree TX08-30-1 had six genotypes that flowered (>51 days) later than their parents. Genotypes from pedigree GLEND A, GEC and TX08-30-1 did not inherit flowering earliness of their female parent but rather they might have inherited that of the unknown male parent. However, they still expressed medium to early maturity traits. Therefore, these materials have the potential to be selected for areas with abundant rain.



Figures 3 (a, b, c, d, e & f): Frequency distribution of number of days to first flowering of cowpea genotypes.

4.1.2 Number of days to 50 % flowering

Results for 50% flowering are shown in Figures 4 (a, b, c, d, e & f). Three genotypes derived from pedigree TX08-30-5 reached 50 % flowering at 50-55 days which was earlier than their parent compared to other genotypes followed by genotypes from pedigree IT98K-205-8 and IT97K-499-35 which had two and one genotypes that reached 50 % flowering (50-53 and 55-56 days) earlier than their parents, respectively. Early varieties can be grown in areas of low rainfall and can be used in intercropping. Whether a variety is short or long season can be determined by 50 % flowering. Short season varieties are desired for sole cropping in areas with erratic rainfall and multiple (double or triple) cropping in areas with adequate rainfall. Most of these pedigrees screened reached 50 % flowering later than their parents. Seventeen genotypes from pedigree GEC, three from GLENDA, and seven from TX08-30-1 reached 50 % flowering >56 days, >60 days and >55 days later than their parents, respectively. Heterosis can either be negative or positive, that is it can decrease or increase a trait in comparison with parent. In this case positive heterosis was observed as most pedigrees flowered later than their parents. Pedigrees that flowered later than their parents have potential of late maturity trait exhibition which may qualify them for semi erect indeterminate cowpea types (Moalafi *et al.*, 2010). Moalafi *et al.* (2010) further argued that late maturing cowpea types are usually produced for fodder use while early maturing type are grown for grain. Long season cowpea (late maturing), produce more branches which help sustain livestock farmers with winter feed. Medium maturing types are dual purpose. They serve both as grain and fodder type and they are suited in various cropping systems. Limpopo Province (Syferkuil) experiences high temperatures which might have affected flowering. Shiringani (2007) reported fewer numbers of 50 % flowering at Syferkuil as compared to other locations studied. Shiringani (2007) further argued that combination of high temperatures and long days (summer) can slow or inhibit floral bud development and thus few flowers being produced.



Figures 4 (a, b, c, d, e & f): Frequency distribution of number of days to 50 % flowering of Cowpea genotypes.

4.1.3 Number of days to 90 % maturity

The results obtained from the current study showed that three pedigree exhibited earlier maturity traits while three pedigree exhibited late maturity traits (Figure 5a, b, c, d, e & f). Eleven genotypes from pedigree GEC, four genotypes from pedigree TX08-30-1 and three genotypes from GLENDA reached 90 % maturity (>91 days) later than their parents. Besides these, five genotypes from pedigree TX08-30-5 and four genotypes from pedigree IT98K-205-8 and IT97K-499-35 reached 90 % maturity (80-90 days) earlier than their parents. According to Moalafi *et al.* (2010) variation in number of days to 90 % maturity gives meaning to different cowpea types which are; early maturity type (dual purpose) and late maturity type (fodder type). Those that reached 90 % maturity within 90 days are medium maturing and those that reached 90 % maturity within 109 days are late maturing. GEC, GLENDA and TX08-30-1 can be further selected and evaluated for fodder use while IT98K-205-8, IT97K-499-35 and TX08-30-5 can be further selected and evaluated for dual purpose.

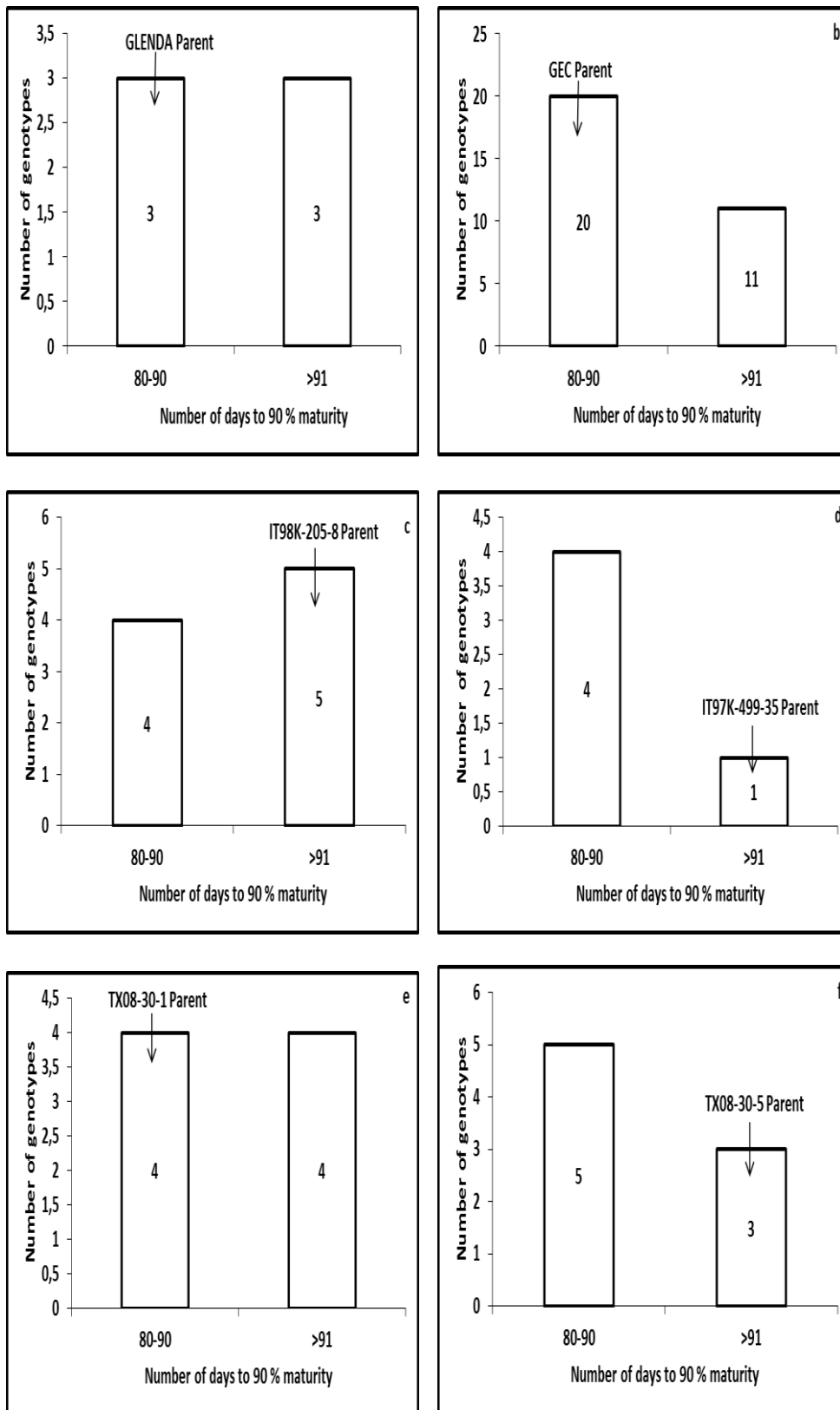
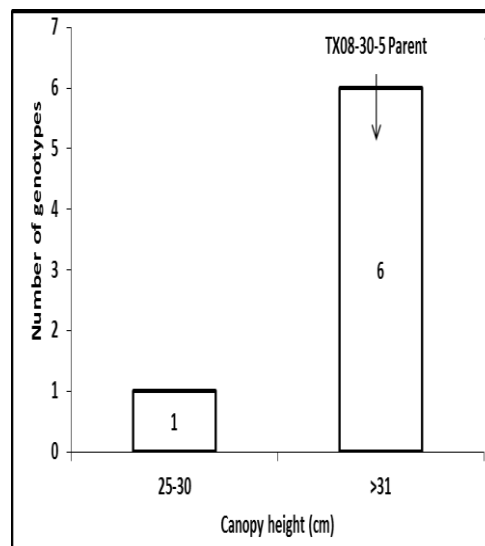
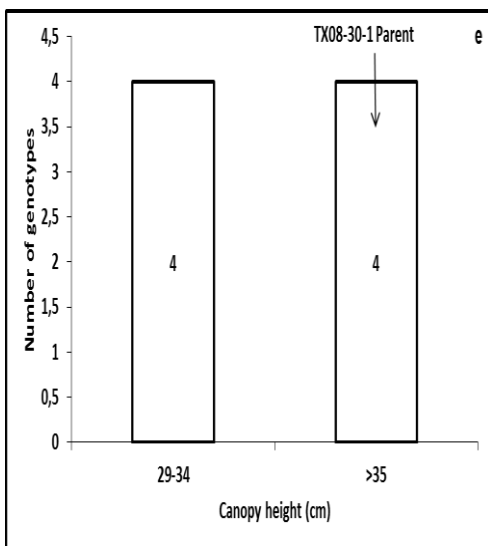
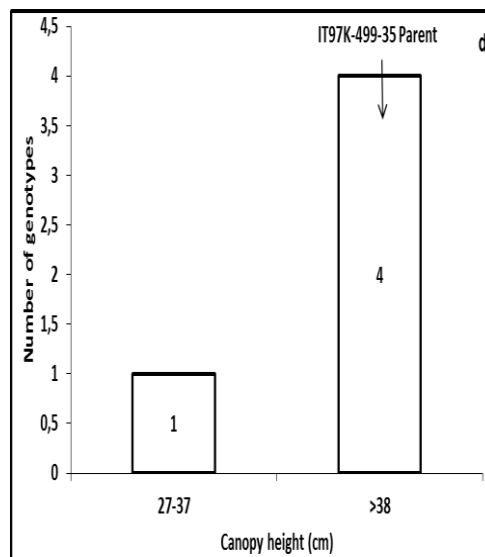
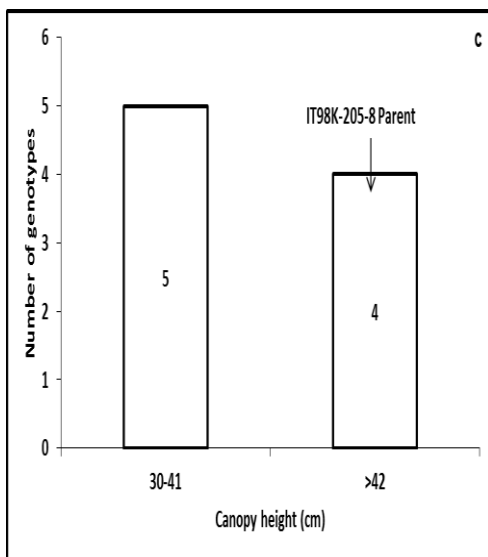
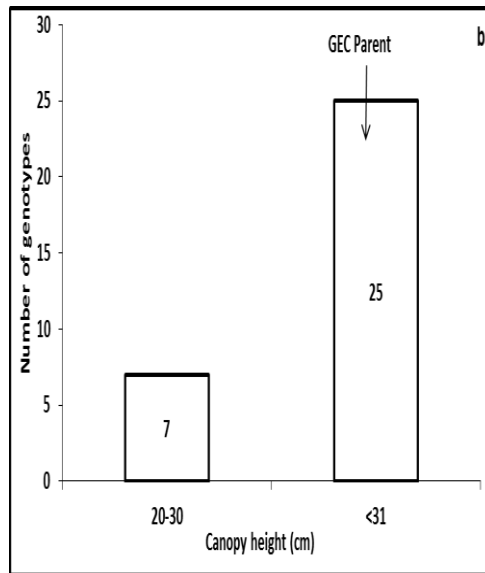
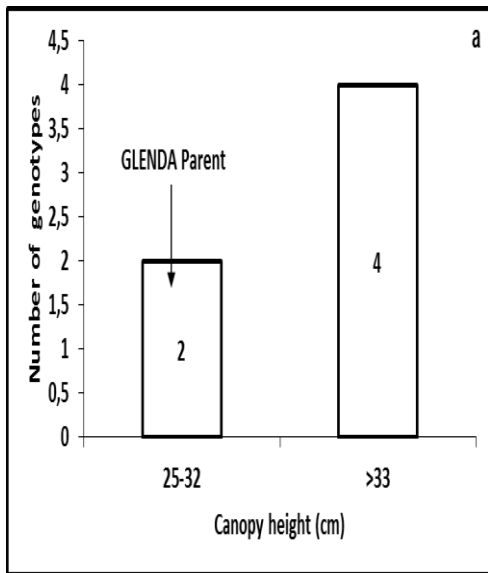


Figure 5 (a, b, c, d, e & f): Frequency distribution of number of days to 90 % maturity of Cowpea genotypes.

4.1.4 Canopy height

Significant variations were also observed among progeny as shown in Figures 6 (a, b, c, d, e & f). Most of the cowpea pedigrees studied had a shorter canopy height as compared to their parents. Genotypes from pedigrees GEC, IT98K-205-8, IT97K-499-35, TX08-30-1 and TX08-30-5 attained the lowest canopy height (20-30 cm, 30-41 cm, 27-37 cm, 29-34 cm and 25-30 cm) than their parents, respectively. These results were in accordance with that of Swarto and Nurchasanah, (2015) who obtained high mean canopy length at F3 generation (114.24 cm) than the parent for rice. Only genotypes from pedigree GLENDA exhibited four genotypes with long canopy height (>33 cm) than their parent. Longer canopy height is not always desirable as lots of energy and resources are directed into plants' tallness than into pod formation and pod filling. Dwarf or short canopy height helps in water conservation, as short plants do not require much water as tall ones. However, in dwarf plants pods crawls on the ground making them prone to diseases and rotting (Aremu, 2011). Thus, tall canopy height lifts and improves the pod clearance from the ground. Genotypes from pedigree GLENDA attained higher canopy than the parent. This shows transgressive segregation. These might have resulted from recombination of both parents' alleles (Rieseberg *et al.*, 1999). The results indicate the potential for selectable traits for plant architecture. Tall canopy results in more branches per plant, which allows or results in more peduncles per plant and the more the peduncles, the more likely the number of pods per plant and the more the grain yield depending on the seed size. However, plants with long canopy heights consume more water than shorter ones.

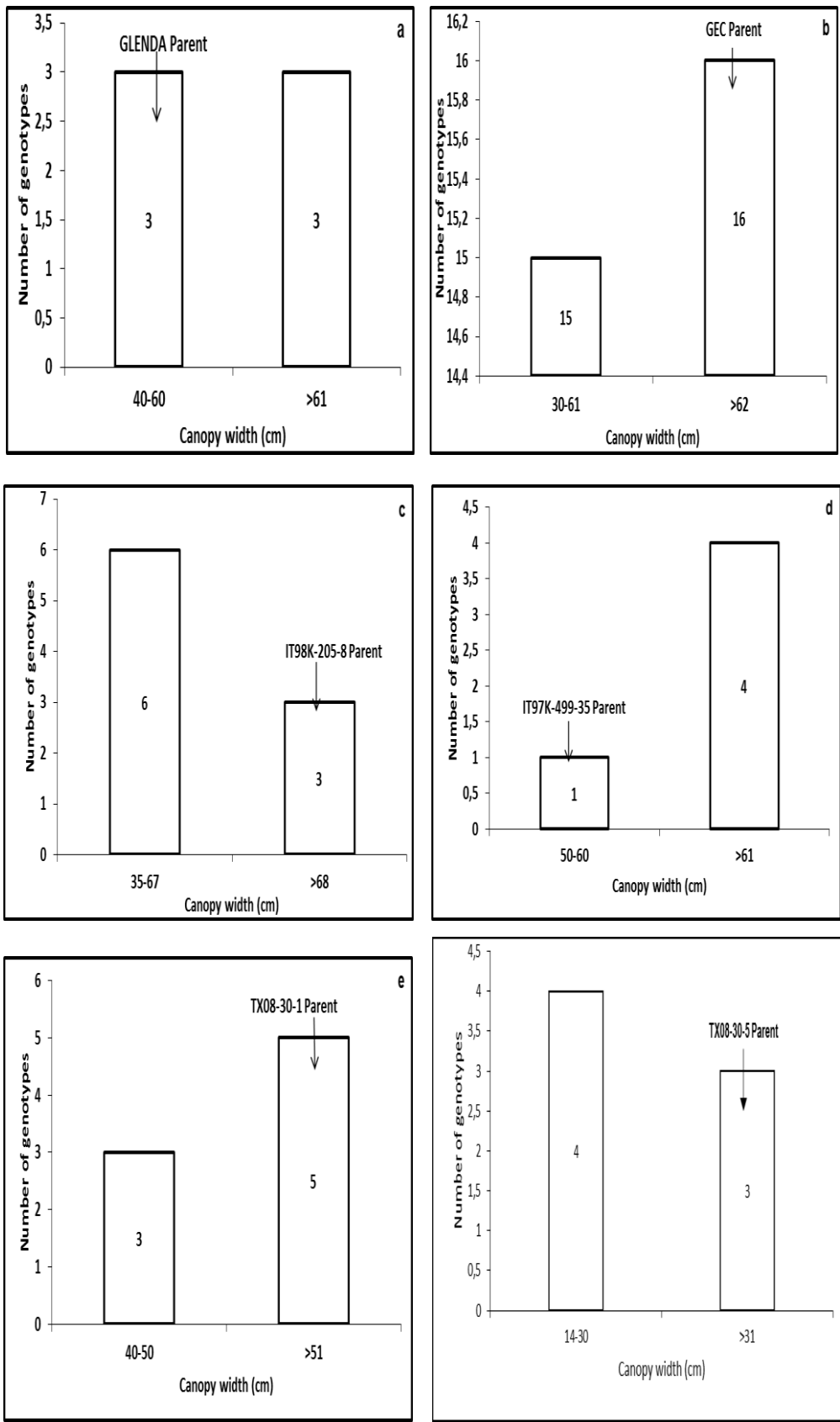


Figures 6 (a, b, c, d, e & f): Frequency distribution of canopy height of Cowpea genotypes

4.1.5 Canopy width

Transgressive segregations of progeny to parents on canopy widths were obtained from GLENDA and IT97K-499-35 pedigrees (Figures 7a & d). Three and four genotypes from pedigrees GLENDA and IT97K-499-35 both attained wider canopy width of >61 cm which was better than other offsprings and parents. This implies that these genotypes have potential of carrying wide canopy width trait, which is a yield component desired by livestock farmers. So when selecting for yield and fodder attributes they can be a good source of germplasm. In addition, exhibition of wider canopy by progeny over their parents shows that they are vigorous. Broader canopy widths are desired for fodder and vegetable use. They are also desired as they provide more ground cover which reduces risk of erosion, prevent evaporation and promotes surface water infiltration (Fatokun *et al.*, 2002). However, the broader the canopy width the more the intra row spacing needed which will use up more land but with less seeding rate per hectare. They also suppresses weeds therefore there would not be need for herbicides, and they help as good ground cover. However, they require more spacing which requires more land for planting and they make it difficult for machinery to pass through if not properly spaced.

Results obtained show that there was greater variability on canopy width because fifteen, six, four and three genotypes from pedigrees GEC, IT98K-205-8, TX08-30-5 and TX08-30-1, respectively showed the narrowest canopy width (30-61 cm, 35-67 cm, 40-50 cm and 15-30 cm) than their parent, respectively. Nwofia *et al.* (2015) explained that early planting increases canopy width through increasing number of branches per plant due to peak rains. F3 was planted in early January which might have contributed to narrow canopy widths as it was planted late. Exhibition of a wider canopy by progeny over their parents shows potential of the pedigrees to be vigorous.

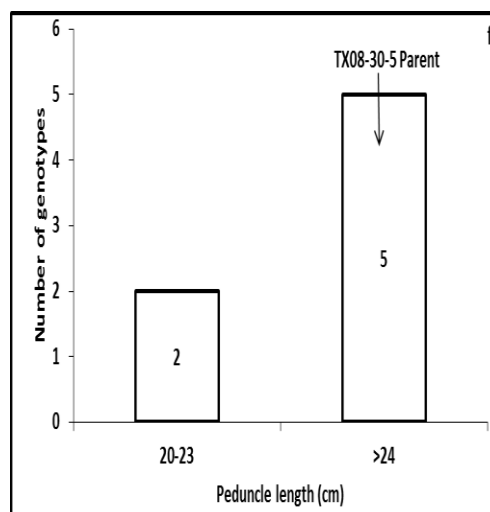
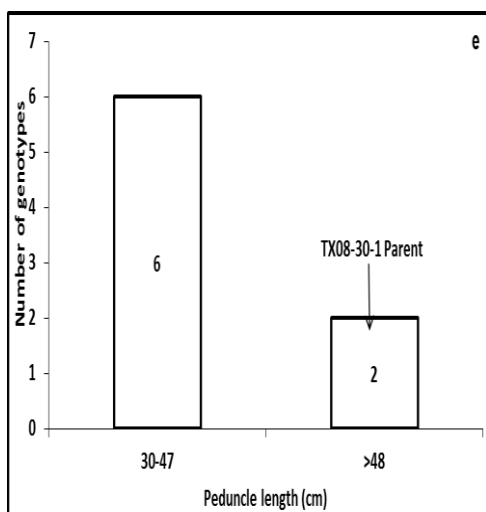
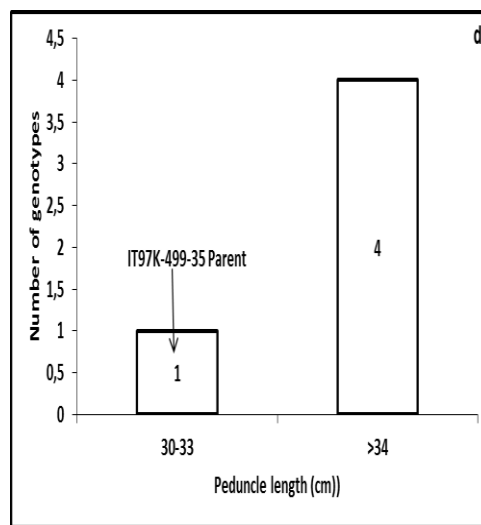
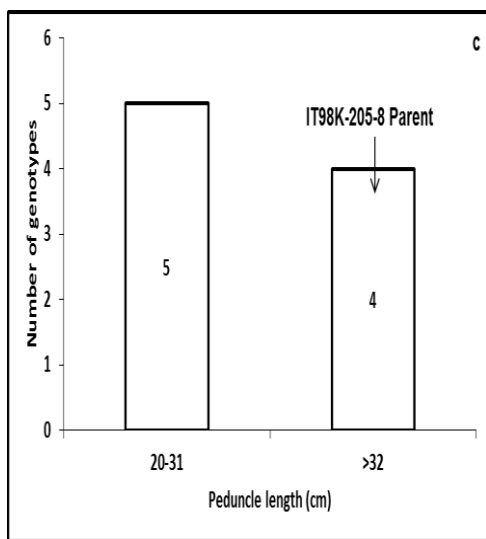
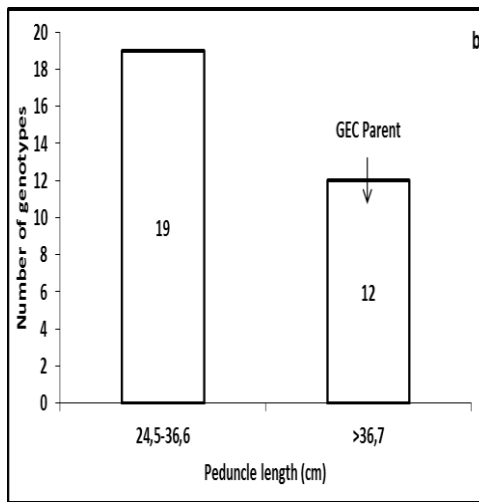
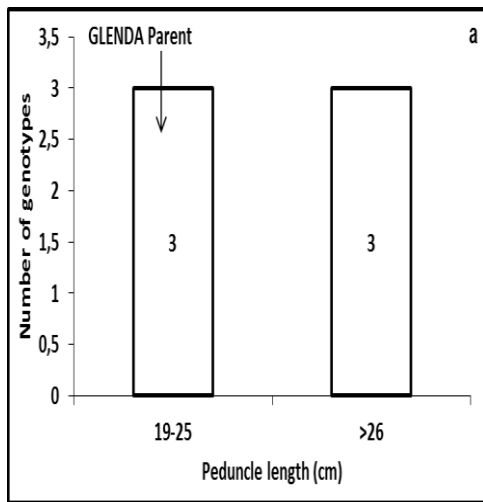


Figures 7 (a, b, c, d, e & f): Frequency distribution of canopy width of Cowpea genotypes.

4.1.6 Peduncle length

Peduncle length expresses the length in which the pod will hang above the soil surface. It is an important characteristic that farmers use in selection of varieties. Farmers prefer varieties with long peduncle lengths as they are compatible to use of combine harvesters and they are not vulnerable to rotting like those that touch the ground while those that have short length crawls on the ground making them vulnerable to pests (reducing seed quality) and making it difficult for use of combine harvester (Labour intensive).. Out of all pedigrees screened, seven genotypes (three from pedigree GLENDA and four from pedigree IT97K-499-35) exhibited long peduncle length of >26 cm and >34 cm which was longer than parent while the remaining had shorter peduncle length than their parents (Figure 8a and d). These results differ with that of Kumar *et al.*, (2009) who evaluated Lentil progeny and parents with long and short peduncles where F2 peduncle lengths segregated into 3 long: 1 short peduncle length. The position of pod on these progeny will be high above plant canopy than that of their parents, these will in-turn avoid rotting of pods and diseases (Aremu, 2011) and have ease when harvesting both by machinery and manually.

Peduncle length of nineteen genotypes from pedigree GEC, six from pedigree TX08-30-1, five from pedigree IT98K-205-8 and two from pedigree TX08-30-5 were lower (24.4-36.6 cm, 30-47 cm, 20-31 cm and 20-23 cm) than their parents, respectively (Figure 8b, c, e and f). These results differ with that of Nsabiyela *et al.* (2013) where they observed that pedicel length at F3 was better than their parents. This means that peduncle length did not express transgressive segregation in this study.



Figures 8 (a, b, c, d, e and f): Frequency distribution of peduncle length of Cowpea genotypes.

4.1.7 Number of pods per plant

Results in Figures 9 (a, b, c, d, e & f) show that out of six pedigrees screened, four performed better than their parents. Twenty-four genotypes from pedigree GEC, four from pedigree GLENDA, five from pedigree TX08-30-5 and six from pedigree IT97K-499-35 acquired higher of pods per plant (>41, >29, >26 and >29) in comparison with their parents, respectively. The remaining two pedigrees (TX08-30-1 and IT98K-205-8) had six and one genotypes respectively, which acquired less number of pods per plant (40-51 and 20-31) than their parents. Genetic variability observed amongst number of pods per plant was due to genetic factors. Ali et al. (2009) supported this statement with the findings of their research where they found that it was due to varietal differences. Erect and semi-erect cowpea plant types tend to produce more number of pods per plant while prostrate cowpea produce less number of pods per plant (Moalafi et al., 2010) probably because erect and semi cowpea types have more number of branches than the prostrate types.

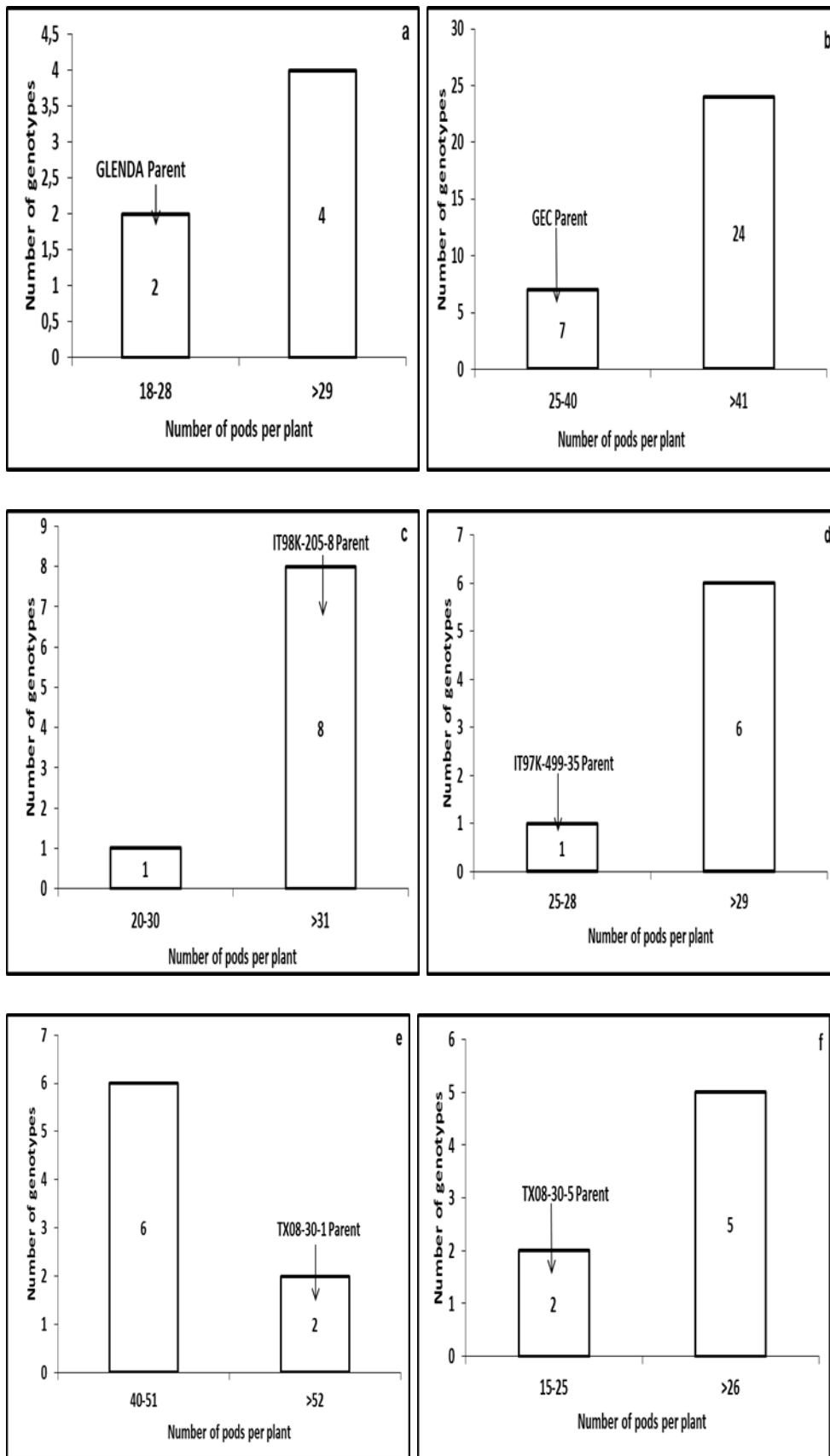
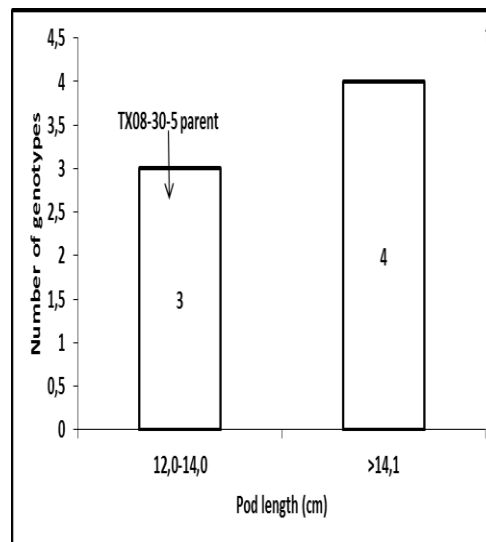
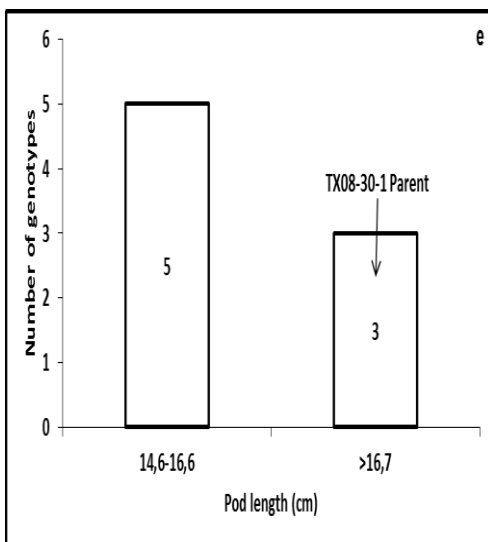
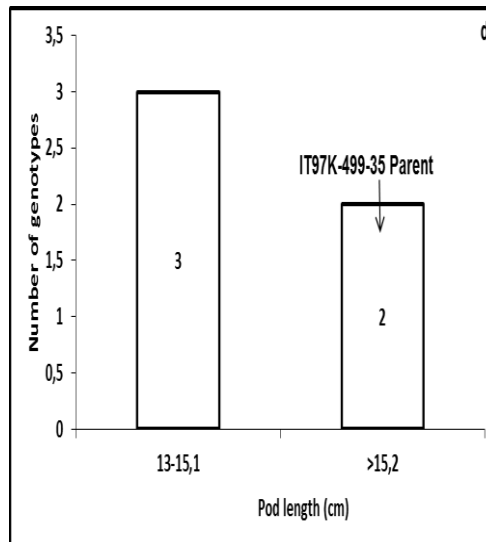
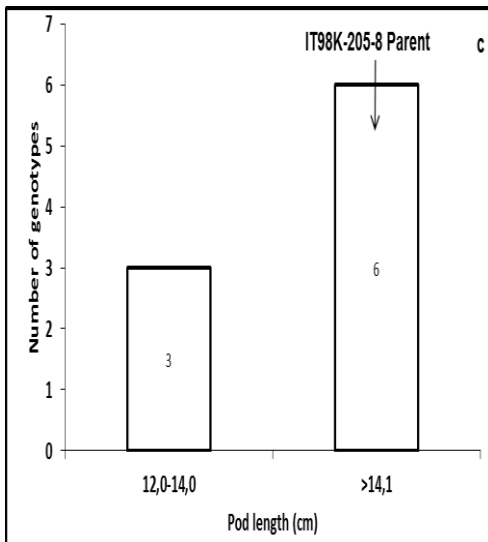
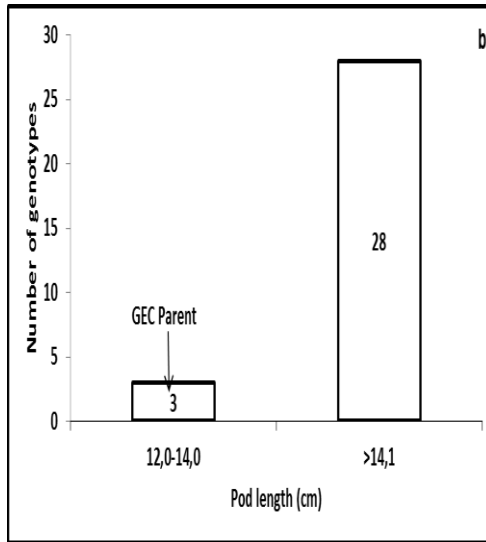
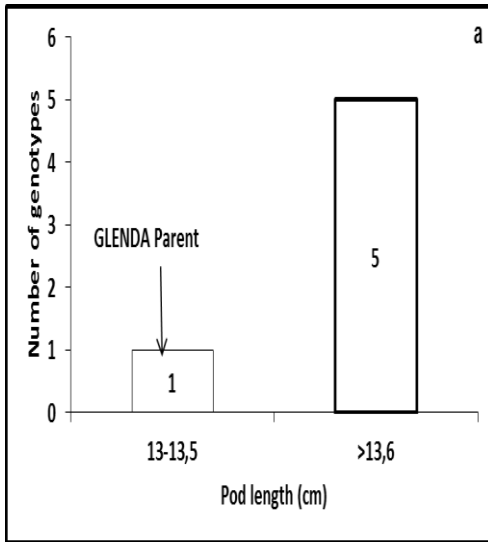


Figure 9 (a, b, c, d, e & f): Frequency distribution for number of pods per plant of Cowpea genotypes

4.1.8 Pod length

Long pod length has the potential of having more seeds per pod depending on seed size. Thirty-seven genotypes with promising traits of long pod length were observed for pedigrees GEC, TX08-30-5 and GLENDA. GEC pedigree had twenty-eight genotypes, TX08-30-5 pedigrees had four genotypes and GLENDA had five genotype with long pod length than its parent (Figure 10a, b and f). These results are in agreement with that of Aliyu and Makinde (2016); they reported a significant difference among progeny and their parent on pod length (longest pod length being 21.4 cm). These indicate that the progeny had more seeds per pod as compared to their parents because the longer the pod the more the seeds it will contain provided that the pods were filled with grains. Fruit length was improved at F3 for pepper (Nsabiyera *et al.*, 2013). This suggests that these hybrids continue segregating into better traits despite the environmental factors endured during 2017 and 2016 production years. These results are similar to that of Moalafi *et al.* (2010), who reported that at F2 mean pod length of six best progenies were higher than that of their parents.

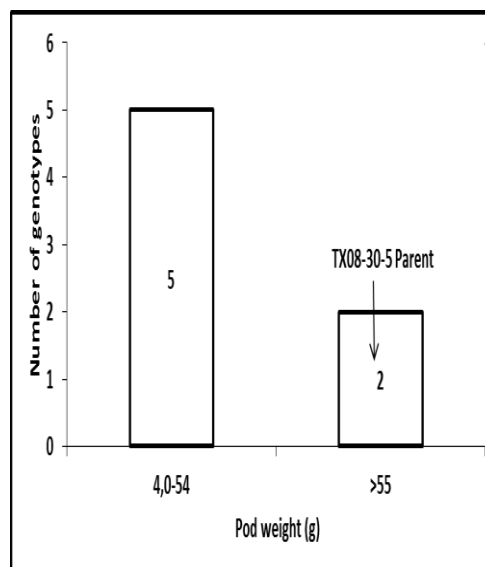
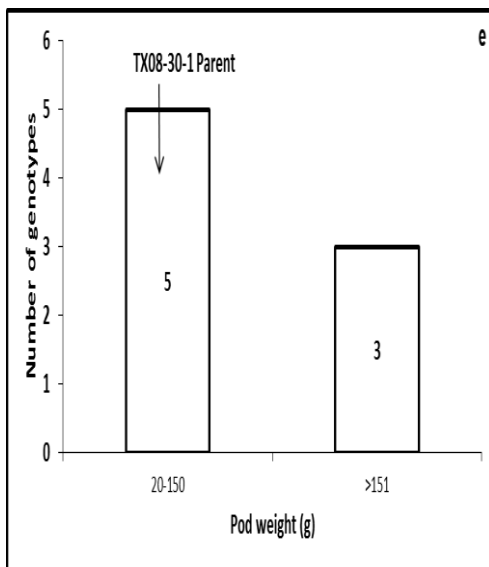
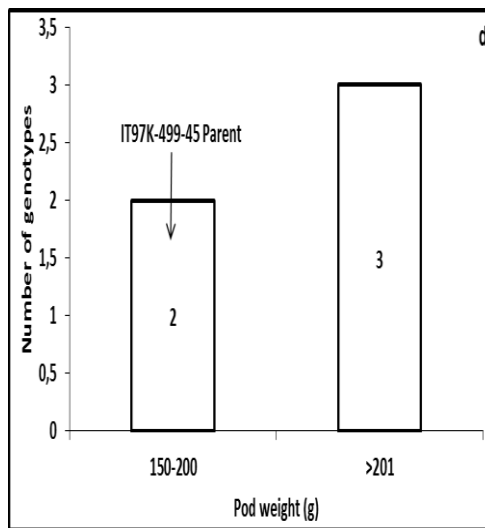
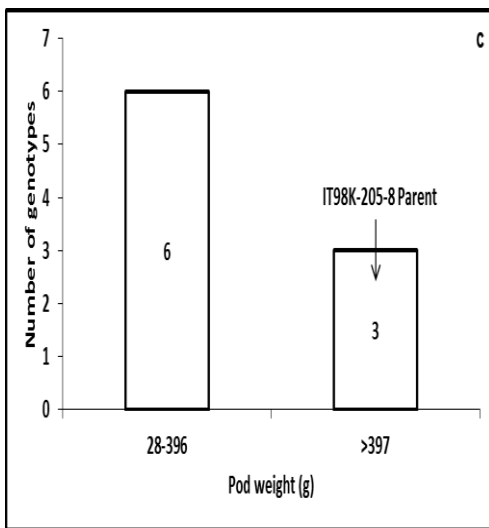
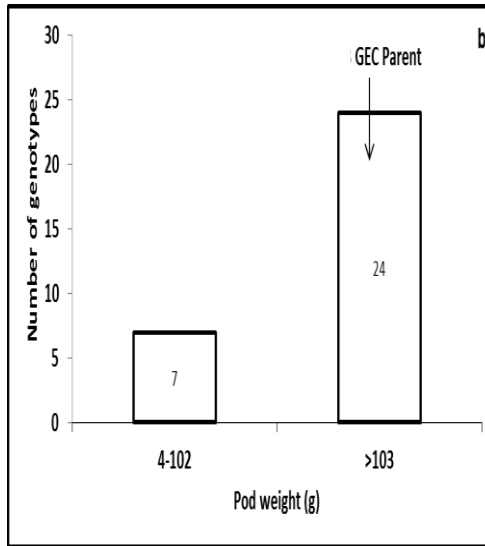
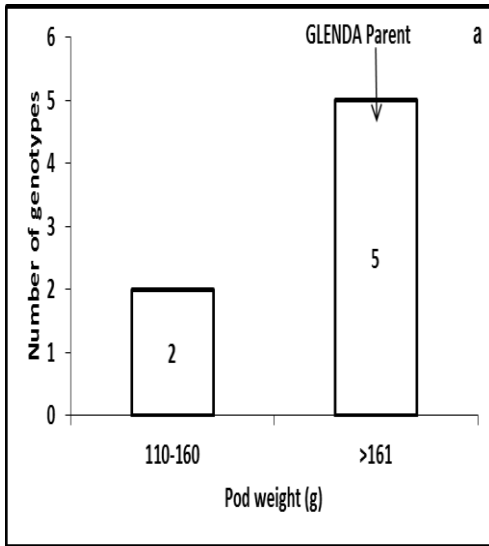
Besides this, three pedigrees had eleven genotypes that expressed shorter range of pod length than the parent. Pedigrees from TX08-30-1, IT98K-205-8 and IT97K-499-35 had five, three and three genotypes respectively that expressed short (14.6-16.6 cm, 12.0-14.0 cm and 13-15 cm) pod length compared to parent (Figure 10c, d and e). Optimum temperature for pod formation is 27 °C. In cowpea, heat induces suppression of floral buds development which can result in two weeks delay in flowering when plants are found in very hot field environment under long days which can result in poor pod formation (Ekpo *et al.*, 2012). This according to Ekpo *et al.* (2012) might have been the reason for some genotypes from pedigrees IT98K-205-8, TX08-30-1 and IT97K-499-35 exhibiting the shortest pod length than their parents. However, GLENDA, GEC and TX08-30-5 still expressed heterosis.



Figures 10 (a, b, c, d, e & f): Frequency distribution for pod length of Cowpea genotypes.

4.1.9 Pod weight

Figures 11 d & e, shows that genotypes from pedigrees IT97K-499-35 and TX08-30-1 had high pod weight (>201 g and >151 g) than their parents. Each of the pedigrees had three genotypes that attained better pod weight than parents. Shreya *et al.* (2017) reported transgressive segregation on pod weight among F2 and F3 genotypes of groundnut. These results are in agreement with those of Shreya *et al.*, (2017), where groundnut pod weight attained was superior to the better parent. These means that pod weight, number of pods per plant and grain yield might be interrelated. This implies that pedigrees with more number of pods per plant or pod weight have potential to produce high yield. Pod weight also determines the grain yield through seed size that means the more the weight the higher the yield. However, most of the pedigrees had low pod weight than their parents. Genotype derivatives from pedigree GEC (seven genotypes), from pedigree IT98K-205-8 (six genotypes), from pedigree TX08-30-5 (five genotypes) and from pedigree GLENDIA (two genotypes) obtained 4-102 g, 28-396 g, 4-54 g and 110-160 g pod weight less than the parent did (Figures 11a, b, c and f). However, the few genotypes (IT98K-205-8, IT97K-499-35 and TX08-30-1) that exhibited higher pod weights than their parents form the selectable promising lines.

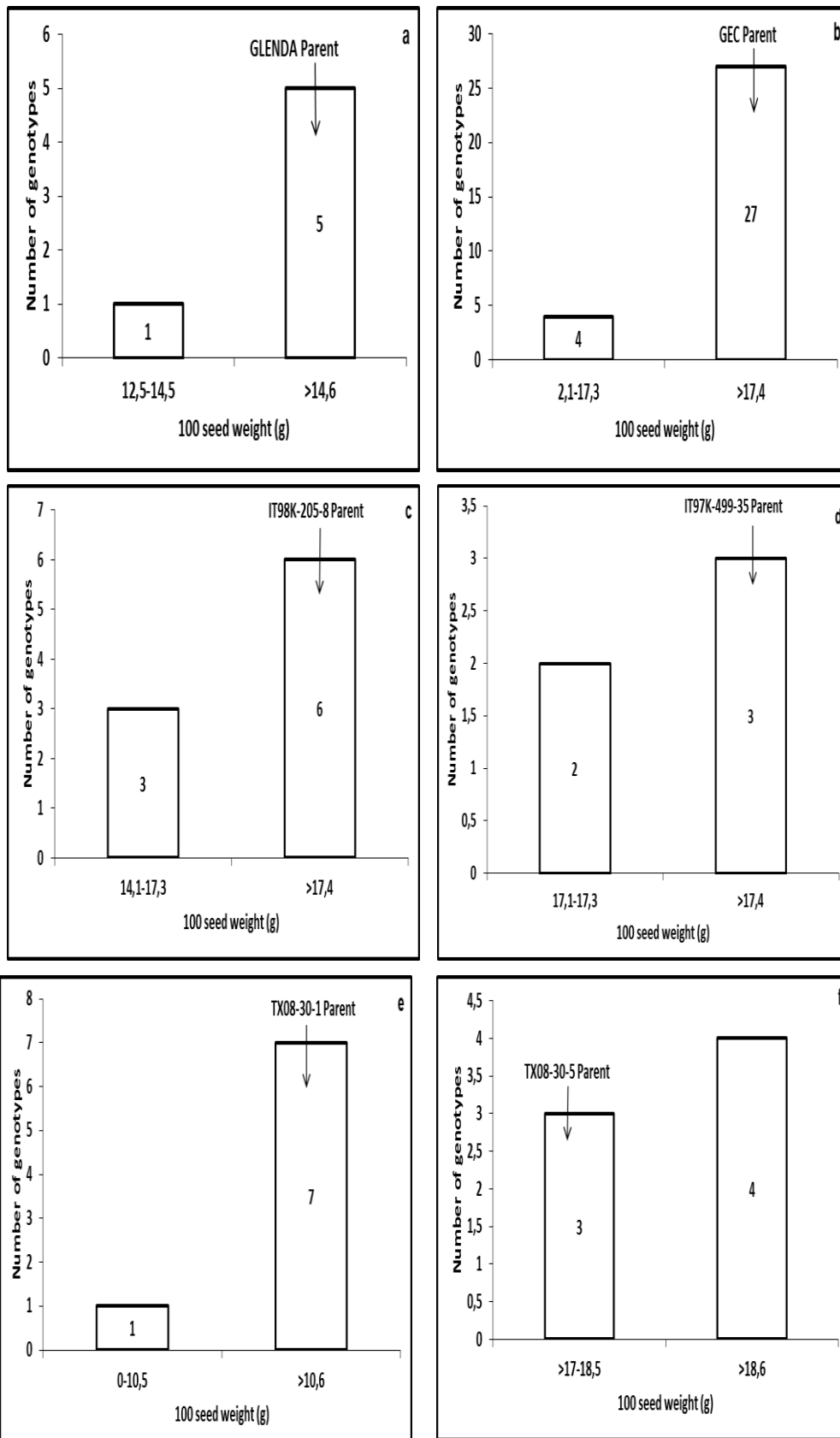


Figures 11 (a, b, c, d, e & f): Frequency distribution for pod weight of Cowpea genotypes.

4.1.10 Hundred seed weight

Figures 12 a, b, c, d, e & f show frequency distribution of six pedigrees for 100 seed weight. Results indicated that four genotypes derived from pedigree TX08-30-5 attained the highest hundred seed weight (>18.6 g) than its parent. Furthermore; TX08-30-5 was the only pedigree that performed better than its parent. Hundred seed weight measures the seed size. Larger seeds are desired as most of them have a high germination percentage, they are easy to handle, easy to sort and appeal to the eye. Transgressive segregation of both F2 (23.9 g) and F3 (22.9 g) generations was observed on buckwheat over their parent (20.5 g) (Li *et al.*, 2012).

GLENDIA and TX08-30-1 pedigrees had one genotype that obtained less hundred seed weight (12.5-14.5 g and 0-10.5 g) than the parent, while four genotypes from pedigree GEC, three genotypes from IT98K-205-8 and two genotypes from IT97K-499-35 exhibited hundred seed weight range of 2.1-17.3 g, 14.1-17.3 g and 17.1-17.3 g, respectively, that was lower than their parents. In Pigeon pea, high genetic variability of medium to low 100 seed weight was observed among three crosses where means of the crosses ranged from 4.5-16.9 g and 3.1-15.4 g at F2 and F3, respectively (Ajay *et al.*, 2014). Variability reduction with each generation is in agreement with the study of Ajay *et al.* (2014) on pigeon pea where they reported that it is due to selfing.



Figures 12 (a, b, c, d, e & f): Frequency distribution for 100 seed weight of Cowpea genotypes.

4.1.11 Grain yield

Figures 13 (a, b, c, d, e & f) illustrates results obtained from six pedigrees on grain yield. Two cowpea pedigrees had 10 genotypes, which exhibited good performance in terms of grain yield. Pedigree GEC had eight genotypes and pedigree IT97K-499-35 had two genotypes that obtained high grain yield (50-100 g/plot and 110-160 g/plot) as compared their parents, respectively. These pedigrees have potential to be selected for further evaluation. They were spotted as promising high yielders during this experiment as they expressed superiority over their parents; which means transgressive segregation was observed. Similarly, average grain yield of yardlong bean expressed significant differences between F₂, F₃ and F₄ (320.9, 330.0 and 367.3 g/plant) respectively, in comparison with their parents (260.0 g/pant) (Sarutayophat, 2008). Sarutayophat (2008) further reported that most traits studied constantly increase in succeeding generations with selection. Heavy yielders were likely to be the ones that had more pods per plant. This means that grain yield and number of pods per plant are interrelated. More yield returns help sustain the farmers for longer periods even on winter months. They are able to get more profits than from normal yielders.

On the other hand, four pedigrees had twenty-two genotypes with low grain yield as compared to their parents; of which eight genotypes were from pedigree IT98K-205-8, six from TX08-30-5, five from TX08-30-1 and two from GLENDA with grain yield of 20-321 g/plot, 2,0-22.0 g/plot, 15-40 g/plot and 110-160 g/plot respectively.

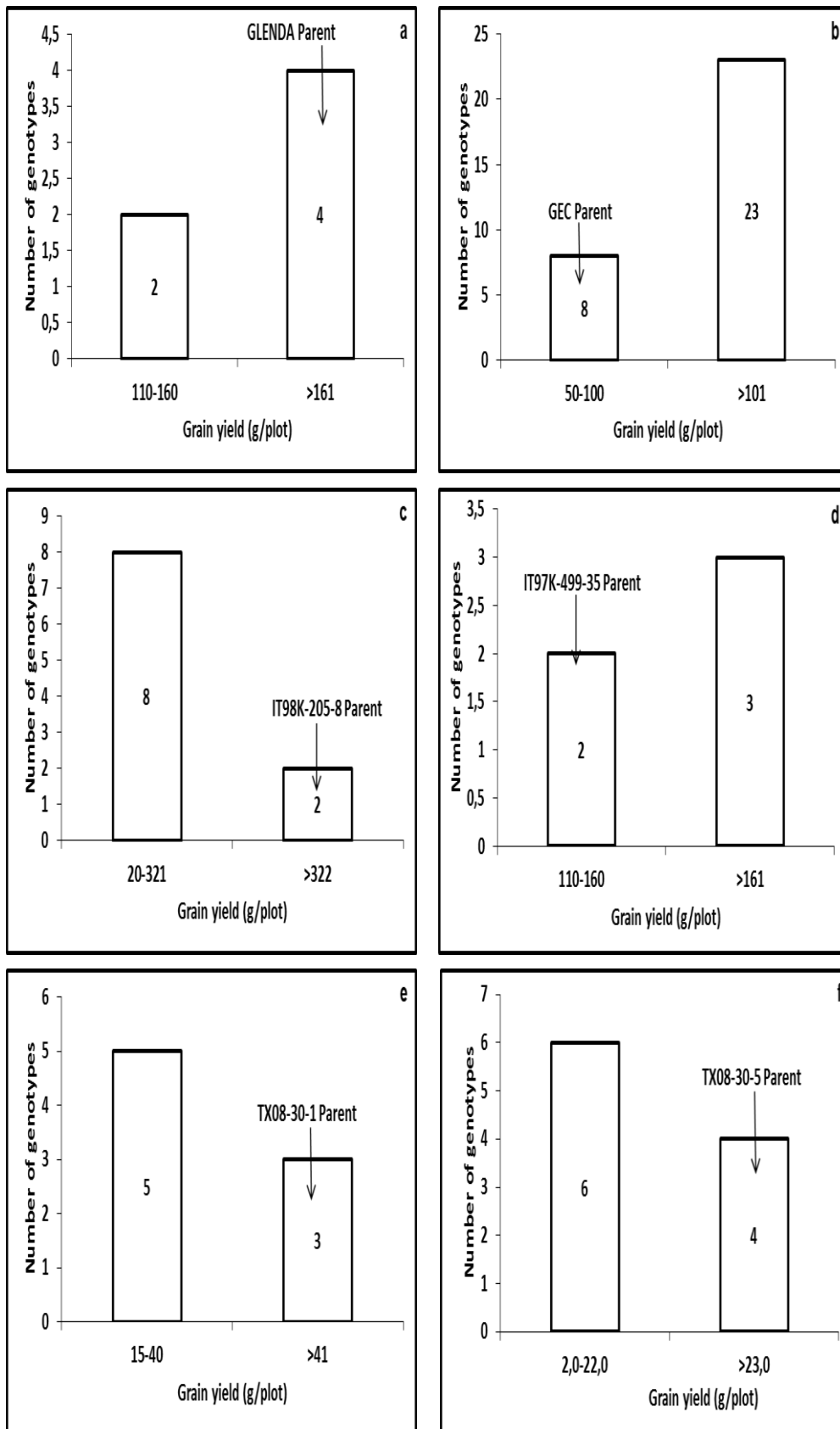


Figure 13 (a, b, c, d, e & f): Frequency distribution for grain yield of Cowpea genotypes.

4.1.12 Fodder yield

Figure 14 (a, b, c, d, e & f) illustrates performance of six cowpea pedigree on fodder yield. The best overall pedigrees that showed good performance in terms of fodder yield in increasing order were TX08-30-1 and IT97K-499-35. These pedigrees showed heterosis as most of their genotypes were superior to their parents. They were the only pedigree that produced fodder yield better than their parents. Genotype from pedigree IT97K-499-35 had six genotypes that yielded (>0.8 g/plot) better than the parent. Pedigree from TX08-30-1 had four genotypes that yielded (>0.8 g/plot) better than their parent. The remaining pedigrees had fodder yield that were less than their parents. Pedigrees from IT98K-205-8 and TX08-30-5 each had three genotypes that obtained lower fodder yield (0.2-0.7 g/plot) than their parents while pedigree from GEC had eleven genotypes and pedigree from GLENDA had two genotypes that attained lower fodder yield (0.2-0.7 g/plot) than their parents.

Overall performance of the genotypes was more than their parent. This suggests that fodder yield may continue to segregate into better traits during succeeding generations (Sarutayophat, 2008). Genotypes from TX08-30-1, IT98K-205-8 and IT97K-499-35 expressed heterosis. Genotypes that expressed heterosis have the potential to be further evaluated for fodder use. Samireddypaile *et al.* (2017) reported similar results; he observed high significant ($P \leq 0.0001$) variation on fodder yield of cowpea which ranged from 590-690 g/plant.

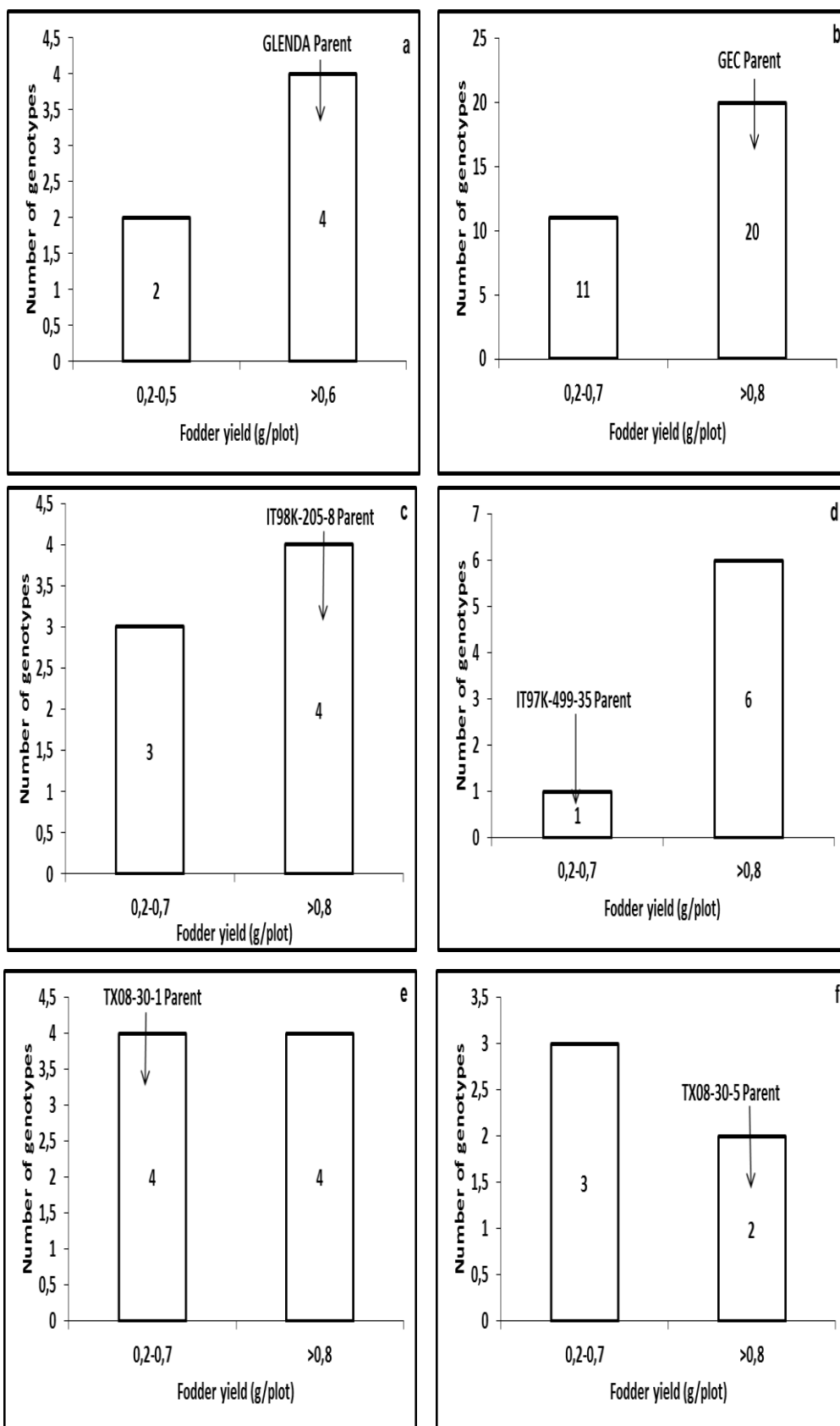


Figure 14 (a, b, c, d, e & f): Frequency distribution for fodder yield of Cowpea genotypes.

4.1.13 Aphid incidence

Only three out of six pedigrees recorded 0 aphid incidence as compared to parents as illustrated in Figures 15 (a, b, c, d, e & f). Three genotypes from pedigree GLENDA and IT98K-205-8 and four genotypes from pedigree TX08-30-5 had an aphid occurrence of 0, which means there was no aphid infestation in these pedigrees. However, twenty-seven genotypes from pedigree GEC, eight from pedigree TX08-30-1 and four from pedigree IT97K-499-35 had an aphid incidence of >1 than their parents. Variation in aphid incidence might be due to preference of the crop or unattractiveness of the genotypes to the insect resulting in reduction of use of the plant by the insect for feeding, shelter and oviposition purposes (Painter, 1958).

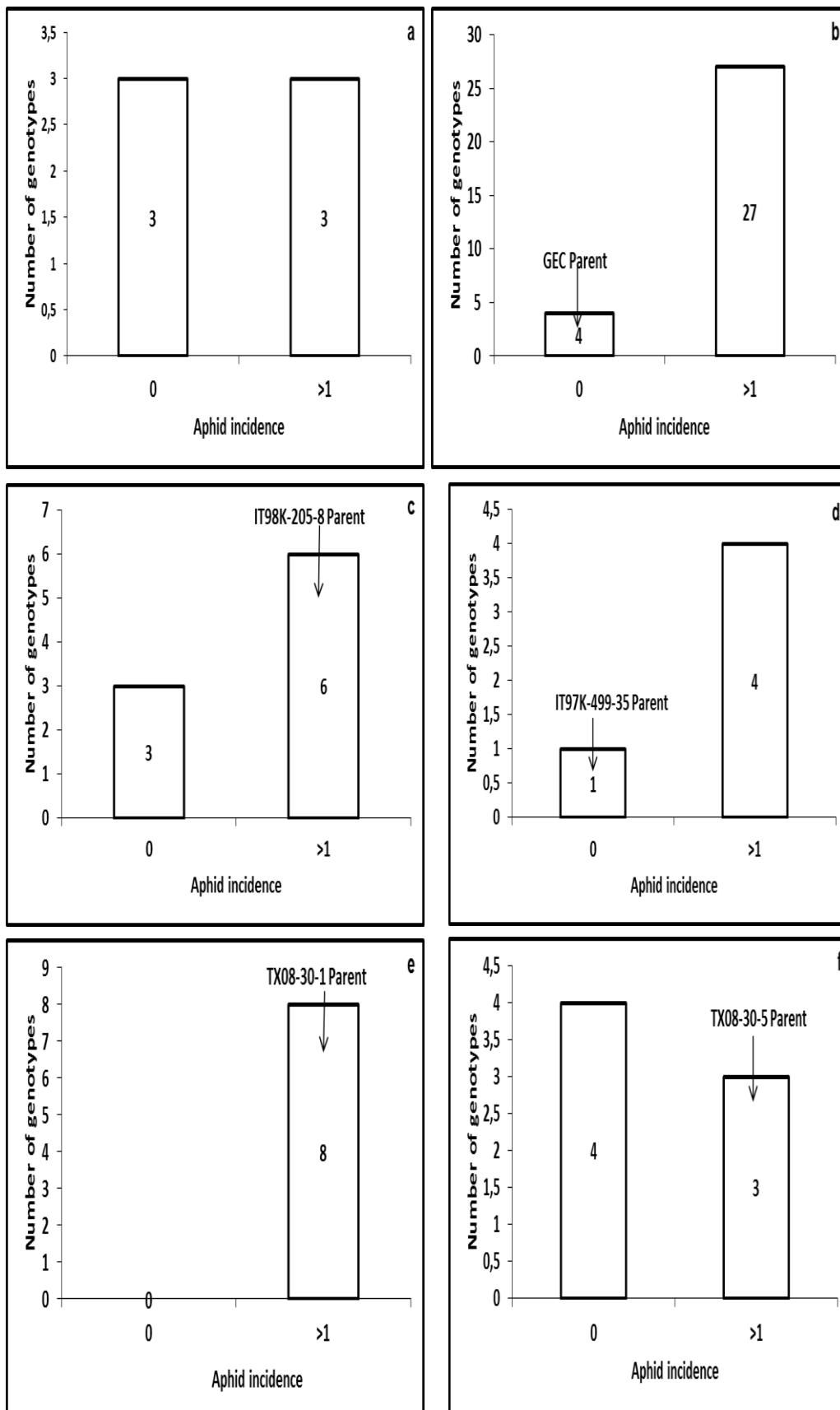


Figure 15 (a, b, c, d, e & f): Frequency distribution for aphid incidence

4.1.14 Aphid severity

Figures 16 (a, b, c, d, e, & f) shows result for aphid severity of six pedigrees. Results show that only one cowpea pedigree had an aphid rating that was less than its parent. The remaining five pedigrees had more aphids rating score than their parents. According to Jackai and Singh (1988) resistance is measured using the 9-point scale where 1=3 resistance, 4=6 medium, resistance and 7=9 susceptible. One genotype from pedigree TX08-30-1 scored 1-2 aphid rating score which was better than its parent. Sixteen genotypes from pedigree GEC, four from IT97K-499-35, three from GLENDA, two from TX08-30-5 and one from IT98K-205-8 obtained range from >0, 4-3 more than their parents. However, all pedigrees expressed resistance towards aphid attack. This might be due to tolerance state, where a plant, despite infestation by large population of insects that could possibly cause severe damage, suffers little damage instead. Therefore all these pedigrees have potential to be screened further for aphid severity for the development of insect resistant cowpea lines that will save farmers from the cost of insecticide application.

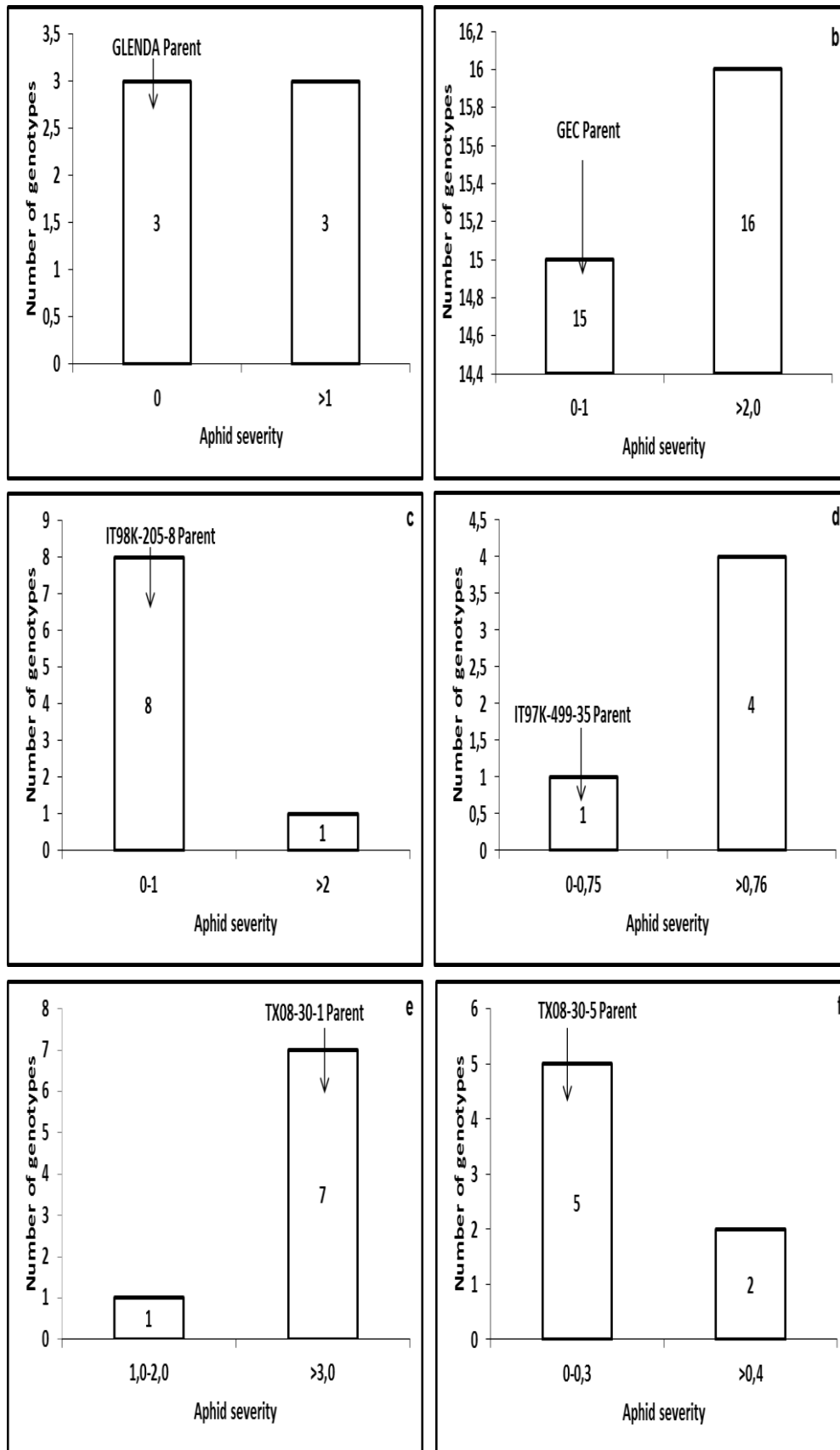


Figure 16 (a, b, c, d, e & f): Frequency distribution for aphid severity of Cowpea genotypes.

4.1.15 Genotypes advanced

Forty selections were recorded for GEC genotypes, while seven genotypes selected from IT97K-499-35 and no genotypes were selected from GLENDA genotypes for advancement as shown in Figure 17. Genotype selection depends on breeders and farmer's needs. GEC genotype was mostly selected which means that it has the potential to be developed into promising variety than others. Breeders mostly select based on farmer's needs, genotype quality traits and overall performance of the genotype (Sarutayophat, 2008). In this study, selection was based on grain type, seed size, grain yield, aphid resistance and plant type.

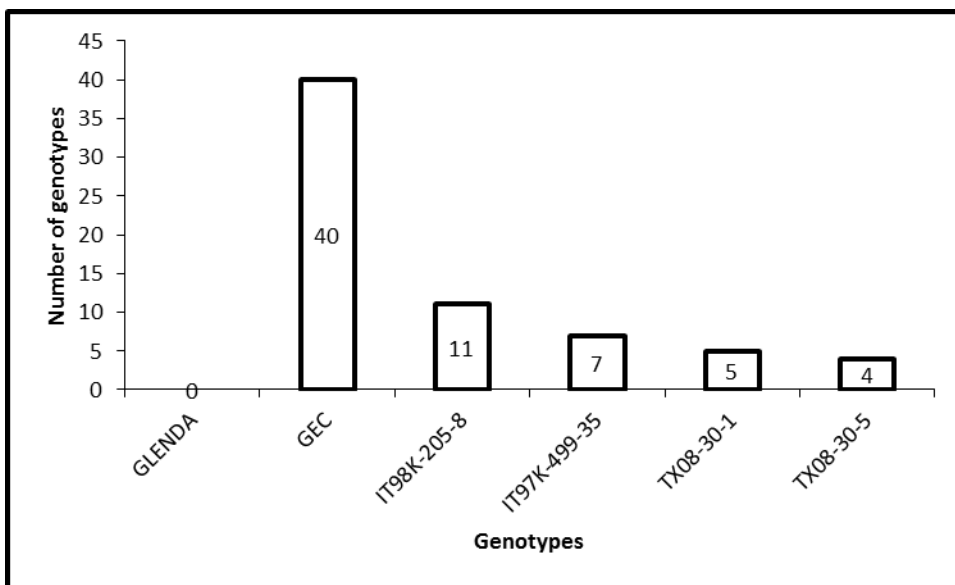


Figure 17: Frequency distribution for number of genotypes selected

CHAPTER 6

SUMMARY AND CONCLUSION

There are several promising genotypes which can be further evaluated and developed into adapted varieties. It was evident that the cowpea germplasm has a great potential for genetic improvement for most plant characteristics studied. High genetic variability was observed for aphid resistance, aphid severity, number of days to first flowering, 50 % flowering, canopy height, canopy width, pod length, peduncle length, number of pods per plant, grain yield, pod weight and 100 seed weight. The null hypothesis of this study stated that F3 hybrid populations of high yielding and insect pest resistant genotypes did not differ with their parents. However, the results of this study were evident enough to reject the null hypothesis. That is; the evaluated genotypes differed from their parents at F3. These might have resulted from genetic variability. Genotypes from pedigrees TX08-30-5, IT98K-205-8 and IT97K-499-35 expressed early maturity trait by reaching 50 % flowering earlier than other genotypes. Early maturity is desired for relay cropping and to evade drought in erratic rainfall environments. Results showed that genotypes from pedigrees TX08-30-5, TX08-30-1, GEC, IT97K-499-35 and GLENDA exhibited yield components (canopy height, canopy width, pod length, peduncle length, number of pods per plant, grain yield, pod weight, 100 seed weight) values that were greater than their parents. Genotypes that showed better trait performance over generations can be further screened for development of promising varieties. They will also help in improving the farmer's productivity. For aphid resistance the following varieties TX08-30-5, IT97K-499-35, TX08-30-1, GEC, GLENDA and IT98K-205-8 were found to have medium to high resistance which means they can be further evaluated for aphid resistance. Aphid resistance will help farmers reduce crop loss through aphid damage and save them on cost of pesticides. Top 5 pedigrees that were selected include; genotypes from pedigrees GEC, IT98K-205-8, IT97K-499-35, TX08-30-1 and TX08-30-5. These genotypes can be further evaluated at different locations for adaptation.

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