

**INHERITANCE OF RESISTANCE TO *ALECTRA VOGELII* IN COWPEAS
(*Vigna unguiculata* [L] Walp.)**

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**A dissertation submitted to the University of Zambia in partial fulfilment of the
requirements for the Degree of the Master of Science in Plant Breeding and
Seed Systems**

**SCHOOL OF AGRICULTURAL SCIENCES, DEPARTMENT OF PLANT
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DECLARATION

I, Mbwando Ally hereby declare that the work presented in this dissertation was my own and has never been submitted for a degree at this or any other university.

Signature.....

Date.....

APPROVAL

This dissertation of Mr Mbwando Ally was approved as fulfilling part of the requirements of the award of Master of Science in Plant Breeding and Seed Systems by the University of Zambia

Examiner's Name and signature

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ABSTRACT

Alectra vogelii (benth) is a parasitic weed which causes significant yield reduction in cowpea (*Vigna unguiculata* Walp) in Tanzania. The objective of this study was to (i) identify the type of gene action controlling the trait for resistance to *Alectra vogelii* in cowpeas and its heritability (ii) determine the effect of *Alectra vogelii* infestation to yield, yield components and seed protein content. Seven genotypes of cowpea were mated in half diallel and their F₂ progeny including parents were evaluated for reaction to *Alectra vogelii* infection in the field in two locations using Randomized Complete Block Design (RCBD) with three replications. High significant ($P < 0.001$) differences were found for *Alectra* emergency and infestation at Ilonga. General combining ability (GCA) effects and specific combining ability (SCA) effects for both *Alectra* emergency and infestation were significant ($P < 0.001$) and $P < 0.05$ respectively). The estimate of Baker's ratio for *Alectra* emergency and infestation were 0.62 and 0.66 respectively. Thus indicating that both additive and non-additive gene actions influenced the trait for resistance to *Alectra* emergency and infestation with additivity being predominant. Narrow sense heritability estimates were found to be 0.41 and 0.44 respectively. Correlation between *Alectra* infestation and emergency with yield components revealed that the number of pods were the most negatively affected ($P < 0.001$). This implies that indirect breeding for resistance or tolerance to *Alectra* infestation can endeavor to screen or breed for cultivars with high number of pod. The finding on Baker's ratio means crossing carefully selected genotypes with resistance gene followed by selection at early segregating generation is the best method for improving this trait for resistance to *Alectra vogelii* in cowpea.

DEDICATION

To the Almighty God, for making everything possible for me to accomplish my studies

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CHAPTER ONE

1.0 INTRODUCTION

Cowpeas (*Vigna unguiculata* (L) Walp) is one of the most important grain legume pulse crops which is grown in the tropics with varying environmental conditions ranging from arid to humid. It does not require highly fertile soils and it is tolerant to high temperatures and drought. Although it is tolerant to drought and well adapted to sandy and poor soils, best yields are however obtained in well-drained sandy loam to clay loam soils with the pH between 6 and 7 (Dugje *et al.*, 2009).

Cowpeas are important in human dietary needs, especially for resource poor families and it is the source of quality protein for human and animal nutrition. On dry weight basis, cowpea grains contain 23.4 % protein, 1.8 % fat and 60.3 % carbohydrates and it is a rich source of calcium and iron (Gupta 1998, Tarawali *et al.*, 2002). It provides quality vegetable protein and can be used to replace or supplements the meat protein in human diet. This makes it easier and possible for resource poor families to have access to protein. In Southern Africa, cowpea has been used as a cheap source of protein almost in all rural population at an affordable cost (Mbwaga *et al.*, 2010). It contributes more on alleviating the problem of protein-energy malnutrition to children of under 5 years old in the predominantly carbohydrates based diet societies (Singh *et al.*, 2006).

Its tendency to produce a heavy vegetative growth that provides full ground cover enables it to be used in controlling soil erosion. As a leguminous crop its root system has high ability of associating with the different species of *Rhizobia* bacteria in the soil to fix atmospheric nitrogen. It fixes about 70–240 N kg ha⁻¹ of atmospheric

nitrogen per year (Berner and Williams, 1998) and residue of fixed N deposit of 60-70 N Kg ha⁻¹ can be left to the soils for the successive crop. As a result of this, cowpeas are grown in rotation or mixed with many cereals and tuber food crops.

In Tanzania, cowpea is third most important legume crop, it is grown on an average of 158000 ha of land with a total yield of 70000 MT per year behind the yields and land space occupied by beans and ground nuts (ICRISAT 2011, FAOSTAT 2012). It can grow in areas where beans does not perform well and forms a large portion of protein content in the diets of the resource poor families which cannot manage to access meat protein. Apart from its dietary importance, cowpeas is also a source of income for many rural household families in Tanzania who are dependent on agricultural employment by acquiring income through selling some of their produce though in relatively small quantities (Stahley *et al.*, 2012).

1.2 Research Problem and Justification

Yield of cowpea by small scale farmers are generally low compared to potential yield in all cowpea growing areas in Tanzania due to a number of constraints. Reliance on late maturing local landrace cultivars, low plant densities in intercropping with cereals, poor access to quality seeds for planting and pest attack contributes much to the reduction in cowpea yields. An even greater problem is a parasitic weed *Alectra vogelii* (Benth) which attaches to the roots of cowpea plants and divert the assimilate from roots and hence cause the reduction of the total biomass and yield (Singh and Emechebe 1997, Mbwaga *et al.*, 2010). The problem of *Alectra vogelii* appears to be widely spread in Southern and Eastern Africa (Kabambe *et al.*, 2005; Mbwaga *et al.*, 2010 and Karanja *et al.*, 2013) and high cowpea yield losse has been reported in

areas infested by the parasitic weed (Bagnall-Oakley *et al.*, 1991; Karanja *et al.*, 2013 and Mbwaga *et al.*, 2007 and 2010).

Different control measures have been widely proposed for controlling *A. vogelii* and this includes hand weeding, chemical control, crop rotation and use of trap crop but with little success (Bouker *et al.*, 2004). However, use of host plant resistance is an alternative approach that is most effective economically and environmentally friendly method in controlling *A. vogelii* (Mainjeni 1999, Rubiales *et al.*, 2006).

High yielding cowpea germplasm with resistance to emergence of *A. vogelii* and those that show resistance by delaying the emergence of *A. vogelii* exist (Mbwaga *et al.*, 2007, Mbwaga *et al.*, 2010). These can be employed in cowpea breeding programmes as parents as donors of resistant genes. However, in order to effectively use the germplasm, the nature of gene action conditioning resistance to *A. vogelii* needs to be fully understood. This is important as it forms the basis for any crop improvement methods designed for improving resistance to *A. vogelii* and improve yield in cowpeas. Plant breeders pay attention to the nature of gene action and their heritability to formulate the strategic method to use in incorporating the gene for resistance to *A. vogelii* in susceptible or elite genotypes (Acquaah, 2007).

Breeding for resistance to *A. vogelii* requires the breeders to understand the ideotype form of plants to select in a population of interest in order to come up with genotypes with better quality and high yielding grains. Grain quality in cowpea takes into considerations of various factors like grain size, grain colour and grain weight (Mbwaga *et al.*, 2010) but often nutritional contents especially protein content of the grain determine the grain quality in cowpea (Afiukwa *et al.*, 2013). Based on this fact, special attention with regards to seed protein content needs consideration during cowpea selection in the cowpea breeding program since cowpea is the cheap source

of quality protein for resource poor families (Vadivel and Pugalenti, 2010). Therefore, interaction between yield, yield components, seed protein content and *A. vogelii* infestation need to be investigated in order to determine which yield component can be used as indirect selection criteria for resistance or tolerance to *A. vogelii* (Geleta, 2010) as well as high in protein content.

1.3 OBJECTIVES

1.3.1. General objectives

The general objective of this study was to determine the genetic resistance of cowpea to *Alectra vogelii* (Benth) and its effects to yield.

1.3.2. Specific objectives

1. To determine the nature of gene action conferring resistance to *A. vogelii* in cowpeas and its heritability.
2. To determine the effect of *Alectra vogelii* infestation on yield, yield component and seed protein content.

1.4 Research Hypothesis

This study was conducted on the premise that, there exists considerable genetic variation for *A. vogelii* resistance in cowpeas which can be used in breeding for host plant resistance against this pest in cowpeas.

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1. Origin, Domestication and Geographic Distribution of Cowpeas

The specific origin of cowpea (*Vigna unguiculata*) has remained a controversial study to many plant botanist especially the specific primary centre of cultivation in Africa, since it is the only place around the world where the diversity of wild forms of cowpeas are found (Steele, 1979). Cowpea is considered to have been domesticated in Africa from its wild ancestral form, *V Unguiculata subsp dekindtiana* (Harms) Verdc (Ng and Marechal, 1985). However, the precise location of origin where cowpea was first domesticated is still under speculation.

According to Ba *et al.*, (2004), the crop was probably domesticated in West Africa. Flight, (1976), reported that the centre of diversity of cowpea is West Africa since carbon dating of cowpea (or Wild cowpea remaining from the Kintampo rock shelter in Central Ghana) has been carried out and was found to be the oldest archaeological evidence of cowpea found in Africa. Some evidence show that the North-eastern part of Africa is the primary place of cowpea domestication based on molecular studies (Caulibaly *et al.*, 2002). South Africa is also regarded as the centre of origin but it is not yet clear where the crop was first planted in the region (Timko and Singh, 2008).

Cowpea was probably introduced from Africa to the Indian subcontinent approximately 2000 to 3500 years ago (Allen, 1983). Cowpeas reached Europe from Asia (Tosti and Negri, 2002). From the West Indies, cowpea was taken to the USA in about 1700 BC (Purseglove, 1984). The slave trade from West Africa resulted in the crop reaching the southern United States of America early 18th century, However,

many US cultivars appear more closely related to germplasm from Asia or southern Europe than West Africa (Fang *et al.*, 2007). Presently, cowpea is grown throughout the tropics and subtropics regions around the world.

2.2. Taxonomy and Botany of Cowpea

Cowpea [*Vigna unguiculata* (L) Walp.] is a dicotyledonous crop in the order *Fabaceae*, subfamily *Faboideae* (Syn. *Papillionoideae*), tribe *Phaseoleae*, sub tribe *Phaseolinae*, genus *Vigna*, and section *Catiang* (Verdcourt 1970). It is a diploid plant having 22 chromosomes (Timko and Singh, 2008) and its nuclear genome size is estimated to cover 620 million base pairs (Mbp) (Timko *et al.* 2008). The genus *Vigna* is pan tropical and highly variable. *Vigna unguiculata* subspecies *unguiculata* includes four cultigroups; *unguiculata*, *biflora* (or cylindrical), *sesquipedalis*, and *textilis* (Ng and Marechal, 1985).

Vigna unguiculata is an herbaceous, prostrate, climbing or sub erect annual plant, growing 15-80 cm high. Leaves are alternate trifoliolate with petiole 5-25 cm long. The lateral leaflet is opposite and asymmetrical, while the central leaflet is symmetrical and ovate. The inflorescence are racemose, flowers are white, cream, yellow or purple. Growth habit is either determinate or indeterminate. Seeds are variable in size and shape (kidney, ovoid, crowder, globose or rhomboid) (IBPGR 1983). Seeds are of various colours: white, brown, black, cream or grey, dotted (black, brown), purple or red. Pods length ranges from 8-22 cm with 10-20 seeds per pod (Timko and Singh, 2008)

2.3. Importance of Cowpea

Cowpea occupies a unique position in world agriculture by value of their high protein and starch contents and capacity to fix atmospheric nitrogen (Tarawali *et al.*,

2002). In many of the developing countries, cowpea is the major source of dietary protein. Their amino acids pattern is close to the perfect amino gram which is rich in lysine content. In fact lysine is the most limiting essential amino acid in cereals, which is very well supplemented by the pulses (Steele, 1985). Food legumes also serve as a feed crop in many farming systems and fetch higher prices compared to cereals and are increasingly grown to supplement farmers' incomes (Gowda *et al.*, 1996).

Cowpea is among the major legume food in sub-Saharan Africa, especially in Southern Africa where the grain and leaves are major sources of food and family income, particularly for the resource-poor households. The crop has a high protein content of around 25% in the grain (dry weight basis) (Bressani, 1985; Singh *et al.*, 2003), and also serves as an inexpensive source of vitamins and minerals. The crop enhances the quality of the cereal based diets when its high lysine content is combined with the high content of methionine and cysteine of cereals (Lambot, 2002). In addition, the crop improves the cropping systems and soil fertility by reducing soil erosion, suppressing the weeds and fixing atmospheric nitrogen which contributes to increased yields of non-nitrogen fixing crops grown with or after it (Tarawali *et al.*, 2002).

In Tanzania, cowpea has remained as one of the most important legume and supplements a large part of protein to resource poor farmers who live in drought prone areas where production of beans (*Phaseolus vulgaris*) is not suitable. It provides a less expensive source of protein and leaf vegetable for diets that tend to be heavily dependent on starchy foods based on millets, sorghum, maize and cassava. Women value cowpeas highly as early harvest of green pods and leaf provide a

source of vegetable in the “hunger months” prior to the main cereal harvest and cash income from sales of both grain and dried processed leaves (Mbwaga *et al.*, 2007; Dugje *et al.*, 2009). In terms of utilization, cowpeas can be used to prepare various dishes, which are traditionally acceptable and valued. The young tender leaves can be cooked and eaten as vegetable and the green pods can be cooked and eaten just like green beans. In addition, the seeds can be cooked when fresh (semi-ripe) or eaten as pulses when fully matured and dried. Like in other African countries, in Tanzania cowpea is used for preparing the stew that is either used together with cereal dishes or directly mixed with the cereals such as maize, wheat, sorghum and rice (Saria, 2010).

2.4. Production Constrains to Cowpeas Production

Cowpea is grown on 10 to 12.5 million ha representing 85% of the world production of which 3 million tons of grains are produced (FAOSTAT, 2012). Africa is the leading producer, with West and Central Africa accounting for about 64% of world production (Singh *et al.*, 2002). Nigeria is the world’s leading cowpea producing country which account for more than 50% of the total world cowpea grain production, followed by Brazil (Singh *et al.*, 2002; FAOSTAT, 2008). Other countries from West Africa which are important producers of cowpeas are; Niger, Burkina Faso, Senegal, Ghana, Cameroon and Mali (Fery, 2002; FAOSTAT, 2008). Considerable production also takes place in Asia and Oceania, the Middle East, Southern Europe, Southern USA, Central and South America (Singh *et al.*, 2002). The other important producers in eastern and southern Africa are Ethiopia, Kenya, Tanzania, Malawi, Botswana, Zimbabwe and Mozambique (Ehlers and Hall, 1997; NGICA, 2006).

Despite its widespread cultivation, the average yields on farmers' fields are low, averaging less than 300 Kg ha⁻¹ (Takim and Uddin, 2010). The low yields have been attributed to a number of biotic stresses such as insect pests, nematodes, diseases and parasitic weeds and abiotic stresses such as drought, high temperature, low soil fertility, low pH and aluminium toxicity (Ehlers and Hall, 1997; Hall, 2004)

In southern and eastern Africa, Tanzania is the largest producer producing about 1.5 million ha and 1 million (MT) metric tons respectively according to a report of the Ministry of Agriculture, Food Security and Cooperatives (MAFSC) of the government of Tanzania.

In Tanzania, cowpea is ranked fourth among the grain legumes after common beans, groundnuts and pigeon pea (Abate, 2011). The estimated area planted to cowpea is 12, 8000 ha, which is about 8.3% of the annual total area planted with grain legume and gives 578 Kg ha⁻¹ with a total annual yield of 74000 metric tons. (Abate, 2011; Stahley *et al.*, 2012). Mbwaga *et al.*, (2010), reported the yield of 319 Kg ha⁻¹ which was a lower than the potential yield. Many factors have been reported to contribute to the lower yield potential of cowpea (section 2.4). Other factor include economic, wrong intercropping practices, occasional drought interval encountered during the growing season, use of unimproved varieties, low plant density, use of late maturity varieties as well as inaccessibility to extension services by the farmers (Mbwaga *et al.*, 2007 Coulibaly *et al.*, 2009; Stahley *et al.*, 2012). On the other hand, a parasitic weed of *Alectra vogelii* was reported to be one of the noxious weed that contributes to yield losses in cowpea that need to be managed in order to ensure the high yield in production areas where this parasitic weed has found to be a problem (Mbwaga *et al.*, 2010).

2.5. *Alectra. Vogelii* as a Production Constraint in Cowpea

2.5.1. Origin and Spread of *Alectra vogelii*

Alectra vogelii (Benth) (Fig. 1) is a parasitic weed which attacks most of the legume crops in sub-Saharan Africa. Cowpea (*Vigna unguiculata*) is major host for this weed but other legume like groundnuts (*Arachis hypogaea* L.), soybean (*Glycine max.* (L) merril), common bean (*Phaseolus vulgaris*) and other tropical legume have been found to be infested by this parasitic weed of *A. vogelii* (Kureh *et al.*, 2005). It is annual weed which has spread in most cowpea growing areas in eastern Africa and yield losses of 50% to 100% has been observed in heavily infested fields by this parasitic weed (Kureh *et al.*, 1999; Mbwaga *et al.*, 2010; Karanja *et al.*, 2013).

Little is known of the origin of *A. vogelii*. It is presumably to have moved with the cowpea crop during human migrations from West and Central Africa (CABI, 2012). Although *A. vogelii* is already widespread in semi-arid areas of Africa, further spread might have occurred through contaminated seed shipments to markets or in grain samples distributed throughout sub-Saharan Africa for trials by research organizations. The accidental introduction of the related *Striga asiatica*, a noxious parasitic weed of maize and other cereals, into the USA in the 1950s (Parker and Riches, 1993) demonstrates that long-distance spread of the tiny seeds of these root parasites is possible.

In eastern and southern Africa cowpeas are heavily affected by *Alectra vogelii* which reduces yield and sometimes total yield losses may be observed in the field which are heavily infested by this parasitic weed (Atokple *et al.*, 1995, Singh and Amechebe 1997, Singh *et al.*, 1997). *A. vogelii* has spread in various parts of South and East Africa. Most parts in Malawi including Lilongwe, Kasungu plains, Dowa and

Blantyre Shile Highlands are known to be infested with *A. vogelii* (Mainjeni 1999, Kabambe *et al.*, 2005, Mbwaga *et al.*, 2007). Yield losses ranging from 20% to 100% has been observed in *Alectra vogelii* infected areas (Bagnall-Oakley *et al.*, 1991; Mbwaga *et al.*, 2007, Karanja *et al.*, 2010, Mbwaga *et al.*, 2010). *A. Vogelii* infestations have been observed in other legume crops grown in Eastern and Southern Africa in various regions. Groundnuts (*Arachid hypogeal* (L)), bambara (*Vigna subteraneane*), soybeans (*glysine max.*(L) Merr) and common beans (*Phaseolus vulgaris* (L)), have also observed to be infected by *A.vogelii* (Riches 1989, Riches *et al.*, 1992, Lagoke *et al.*, 1993).

2.5.2. Taxonomy and Botany of *Alectra vogelii*

Engler (1922) split the species into *A. angustifolia*, *A. merkeri* and *A. scharensis*. Although, in his taxonomic revision of the genus, Melchior (1941) considered these all to be characteristic of *A. vogelii* on the basis of the specimen collected in Guinea. All previous and subsequent major floras for West Africa (Hutchinson and Dalziel, 1963) and south-eastern Africa (Philcox, 1998) have maintained the name as *A. vogelii*. Although these accounts include the genus in the family Scrophulariaceae, a sequence analysis of three plastid genes suggested that it should be placed in the family Orobanchaceae along with other closely related parasitic genera (Olmstead *et al.*, 2001). No morphological or anatomical evidence for this reclassification has however been advanced. It was concluded that it placed in the family Scrophulariaceae (Timko and Singh, 2008)

Alectra vogelii Benth (Fig. 1) belongs to a family Scrophulariaceae, it is an annual herb, 20–50 cm high; stems erect, simple or with several branches arising from the base or above, base of stem and roots are orange-yellow. Leaves are opposite, ovate

7–25 mm long and 3–8 mm wide. Inflorescence of terminal racemes, generally compact; flowers solitary in the axils of leaf-like bracts; bracts linear to linear-lanceolate, 9–13 mm long, entire or with 1–3 blunt teeth, with 1–3 prominent veins; bracteoles linear, 5–6 mm, acute, hispid; pedicels 1–2 mm. Calyx campanulate, 5–6 mm long, 5-veined, veins not prominent, hispid; lobes ovate, 2–3 mm, acute, ciliate. Corollas are yellow with purple veins, 10–12 mm long with rounded lobes, filaments glabrous with a few hairs present right below the anther. Capsule ovoid, 5–6 mm long, \pm 5 mm in diameter, glabrous (Ghazanfar *et al*, 2008).



Figure 1. *Alectra vogelii* (Benth) plant

Source: *Sune Holt* 2014

2.5.3. Host Range of *Alectra vogelii*

Cowpea is the major crop host of *A. vogelii* (Parker and Riches, 1993). Alternative host include bambara, groundnuts, common bean, soyabean, mung-bean, and tepary beans and it has also reported to infest chickpea and runner bean (Parker and Riches, 1993). There is a clear geographic variation in the host range in different regions of Africa infested by *A. vogelii*. Although different population of *A. vogelii* infest the plant legume most are host specific infesting groundnuts, cowpea, bambara nuts or mung-bean (Riches *et al.*, 1992). Apart from legume crops, it has also been reported as a parasite on non-legume weeds including *Acanthospermum hispidum*, *Vernonia poskeana* (Compositae), *Euphorbia* (Euphorbiaceae) and *Hibiscus* (Malvaceae) species in addition to common legume weeds including *Indigofera* and *Tephrosia* species.

2.6. Nature of Parasitism of *Alectra vogelii* to Cowpea

2.6.1. Attachment of *Alectra vogelii* to Cowpea

The biology of the cowpea parasite, *Alectra vogelii* shares many biological characteristics with another cowpea parasite of *Striga gesnerioides*, and the histology of infected cowpea plant (Igbinnosa and Okonkwo, 1991; Samb and Chamel, 1992). *Alectra vogelii* seeds germinate when exposed to root exudates from cowpea, other host, and a few non-hosts. Radical elongates, showing a chemotropic response to a concentration gradient of roots exudates. Radicular apex develops numerous hair, which attach to host roots, once it comes into contact with the host roots. *Alectra* radical penetrates and stimulate cell division in the host root. The new host cells, together with growing parasite tissue, form a large haustorium, uniting the parasite with tissue in the host's stele which eventually permits transfer of water and nutrients

from host to parasite. *Alectra* tends to form a large haustorium than *Striga*, however, both *Striga* and *Alectra* shoots emerge from the haustorium about 2 weeks after infection and grow into plant which may be 15-25 cm tall (Agrios, 2005)

The haustorium is the nodal point in the identification of the parasitic habit in Angiosperms. It is a vital conducting organ for parasitism and forms an anatomical bridge through which physiological continuity is maintained between the parasite and host.

2.6.2. Symptoms of Infection by *Alectra vogelii* to Cowpea

The parasitism of *Alectra* begins about 2-3 weeks before emergence of parasitic weed above the soil, but symptoms of infection by *Alectra* can be noticed on cowpea plants much earlier before emergence of the parasite. Symptoms associated with *Alectra* Infected plant include general stunting and wilting with reduced numbers of flowers and pods, and plant may be completely wilted if there is acute moisture deficit (Emechebe *et al.*, 1991), however, time and level of infection by *Alectra* to cowpea determine the extent of yield reduction (Singh and Emechebe 1991; Parker and Riches 1993; Lagoke *et al.*, 1993).

2.7. Reproduction and Control of *Alectra vogelii*

With reference to non-parasitic weeds, the control of parasitic weeds has proved to be exceptionally difficult. The parasitic weeds are propagated by seed. The ability of the parasite to produce a tremendously high number of seeds, which can remain viable in the soil for more than ten years, and their intimate physiological interaction with their host plants, are the main difficulties that limit the development of successful control measures that can be accepted and used by subsistence farmers (Rugare *et al.*, 2013). The weed can be transmitted through contaminated seeds

shipments to markets or in grain samples distributed throughout sub-Saharan Africa for trials by research organizations as earlier eluded in section 2.5.1. Several control methods have been tried for the control of parasitic weeds (Parker and Riches, 1993; Kroschel, 2001 and Omany, 2001), details are explained in sub section 2.7.1 to 2.7.3.

2.7.1. Cultural Control

There are two options of using catch and trap-cropping, are available for reducing the size of the *A. vogelii* seed bank in the soil. Catch crops are susceptible species which are ploughed in and harvested after parasite attachment but before emergence and seed production. In a season of good rainfall, a quick-maturing crop of sunflower could then be grown with cowpea planted again in the following season. Trap-crops produce the *Alectra* germination stimulant in their root exudates but are not susceptible to attack by the parasite seedlings. Some grain or fodder cultivars of pearl millet and bambara, are not attacked by the local biotype of the parasite, but are potent stimulators of *A. vogelii* germination (Parker and Riches, 1993). These can be used in a rotation to cause suicidal germination of the parasite and hence reduce the number of seed in the soil. Improved cowpea cultivars which combine resistance to *A. vogelii*, the related parasitic weed *Striga gesnerioides*, and several insect pests and fungal diseases have been developed in West Africa. These include the cultivars IT90K-76-6 and IT90K-82-2 which have been released for commercial production in Nigeria (Singh *et al.*, 2006). These are not, however, resistant to biotypes of *A. vogelii* from southern Africa (Riches, 2001). The Botswana landrace accession B359 has been shown to be resistant to samples of the parasite from Botswana, Malawi and Kenya, so could be used as a parent for breeding improved cultivars for East and southern Africa (Riches *et al.*, 1992; Riches, 2001). Potentially useful levels of

resistance to *Alectra* have also been demonstrated in germplasm of bambara (Riches *et al.*, 1992) and cultivars of soyabean (Kureh and Alabi, 2003) but multi-location testing is needed to confirm the value of these lines in the field. In small scale farmers' fields, common traditional methods of weed control like hand weeding by hoe and crop rotation have been used since long time and they are still under practice to date since they are simple and affordable to anyone but not effective as much as parasitic weed control strategies are concerned (Riches, 1993; Bouker *et al.*, 2004).

2.7.2. Chemical Control

A. vogelii is predominantly a pest of crops grown by resource-poor small-holder farmers who rarely have the finance to access herbicides. Little attention has therefore been given to the development of chemical control. The potential for controlling the weed by treating cowpea seed with the herbicide imazaquin before planting has been demonstrated (Berner *et al.*, 1994). Farmers can reduce cowpea infection by *A. vogelii* when pre-emergence herbicide mixtures containing pre (metazachlor + antidote) are applied, followed by post-emergence application of imazaquin at 0.18 kg active ingredient/ha (Magani and Lagoke, 2009).

2.7.3. Integrated Control

Integrated control was found to be built around the use of resistant crop cultivars if possible, or choice of the least susceptible cultivar that is currently available. Timely destruction of legume crop residues is important to prevent parasite seed production after harvest and trap-crops should be included in the rotation to reduce the soil seed bank. Hand-pulling by uprooting the *A. vogelii* shoots carried out on lightly infested areas, particularly in fields which have not previously had a history of infestation has been also included as part of integrated control method (CABI, 2012). Moderate

tolerance varieties which help to reduce the amount of seed of *Alectra* soil seed bank when they are used in combination with other control measures was also found as good part of integrate control method of *A. vogelii* (Adetimirin *et al* 2000; Kim 2000).

2.8. Host Plant Resistance and Breeding for *Alectra vogelii* Resistance in

Cowpea

More progress has been made in Africa in controlling *Striga* and *Alectra* on cowpea by host plant resistance through a series of screening and breeding programmes. This has involved collaborative work among a number of national, regional and international programs over 20 years. Initial work was conducted by IITA scientists in Burkina Faso working in a joint project with International Development Research Centre (IDRC), Canada and the Semi- Arid Food Grain and Development (SAFGRAD) project of the organization of Africa Unity (Timko and Singh, 2008). Resistant varieties identified under field screening trials using different cowpea accessions at Kamboinse in 1981 (IITA 1982;1983), followed by evaluation by the IITA/SAFGRAD project at many location in Burkina Faso, Cameroon, Mali, Republic of Niger and Nigeria from 1983 to 1986 to ascertain the stability of *Striga* resistance across the west Africa Savanna. Gorom local and 58-57 had shown a high level of resistance to *Striga* in Burkina Faso, but their susceptibility in other countries had indicated the presence of different strains (Aggarwal 1991). Parker and Polniaszek (1990) and Emechebe *et al.*, (1991), reported the identification of two new source of resistant to *Striga* in B301 (a landrace from Botswana) and IT82D-849 (an improved breeding line from IITA). This new source showed stable resistance to *Striga* across Burkina Faso, Mali, Republic of Niger, and Nigeria. In addition, a number of other lines were identified which are less susceptible to *Striga*, as shown

by a lower number of *Striga* plants as well as delayed emergence of *Striga* (Singh and Emechebe 1991). However, IT82D-849 found to be resistant to *Striga* but susceptible to *Alectra*, whereas IT86D-534, IT86D-371 and IT84D-666 are moderate resistant to *Striga* and highly resistant to *Alectra*. B301 is completely resistant to both. Suvita-2 is highly resistant to the *Striga* strain from Burkina Faso, moderate resistant to *Striga* from Niger but highly susceptible to *Alectra* (Singh and Emechebe, 1990a, 1990b).

In Tanzania various series of pot and field screening trials have been conducted to screen for several lines of cowpea for resistance to *A.vogelii* at several Agricultural research institutions and Sokoine University of Agriculture under a McKnight Foundation Collaborative Crops Research Project commencing from 2006 to 2008, has revealed the existence of cowpea accessions which are resistant to *A.vogelii* (Mbwaga *et al.*, 2007; 2010). Various cowpea accessions from the research institutions in Tanzania, National Plant Genetic Research Centre (NPGRC), International Institute of Tropical Agriculture (IITA) and other seeds from farmers were assembled and screened for *Alectra* resistance in 2006/2007 season in various locations with high infestation of *Alectra*, and some few lines showed resistance to the growth of *A.vogelii*. In 2007/2008 season, advance screening under field condition in Tanzania and in the glass house in UK identified two promising lines of B301 and IT81D-994 from IITA which continued to show full resistivity to the growth of *A.vogelii* in all locations under field trials, supported by a glass house experiment in UK (Mbwaga *et al.*, 2010). B301 is a land race from Botswana and IT81D-994 is a line from the breeding Programme at IITA. There have been confirmed as sources of resistance in cowpea to *A.vogelii* in Tanzania as they have

been found to be resistant in West Africa (Singh *et al.*, 2006). However, for effective breeding for resistance to *A. vogelii* it is important that a breeder understand the genetics and mechanism for resistance to *A. vogelii* in cowpea.

2.8.1. Genetics of Cowpea Resistance to *Alectra vogelii*

Little has been done in establishing the nature of gene action conditioning the resistance to *A.vogelii* species. Previous work done, suggested that, the nature of gene action is conditioned by non-additive gene with dominant genes at play (Singh *et al.*, 1993; Atokple *et al.*, 1995). More work needs to be done to confirm this aspect since the selection for this resistance is dependant to the environmental and sensitive to *Alectra vogelii* biotype (Mbwaga *et al.*, 2010; Mainjeni, 1993).

2.8.2. Heritability of *Alectra vogelii* Resistance to Cowpea

Heritability studies of a trait is relative important because it determines phenotypic variances due to genetic causes that will be going to be passed or inherited on from parents to offspring. Heritability is often interpreted as the extent to which the phenotype is determined by genes from parent (Wray and Visscher, 2008). Heritability often tells the breeder on the progress they can make following a breeding method taken to improve a given trait (Morakinyo, 1996). The breeder can make rapid progress where heritability is high by using selection methods that are dependant solely on phenotype (e.g., Mass selection). However, where heritability is low the method of selection based on families and progeny testing are more effective and efficient. When over dominance predominate, the breeder can exploit short- term genetic gain very quickly by developing hybrid cultivar for the crop (Hamdi, 1996). However, it must be noted and so it is important to stress that, there is no one heritability value for a given trait in one species because heritability can and often

does differ among population and among environments (Conner and Hartl, 2004). Park *et al.*, (1998) and Singh and Munoz (1999) reported that, resistance to common bean bacterial blight to be controlled either quantitatively or qualitatively depending on the source of germplasm due to different genes being involved in resistance for pod and leaf respectively. Also Miklas *et al.*, (2000) and Yu *et al.*, (2004) suggested molecular marker linked to genes to be used in order to assist in speeding up selection and offer opportunities to incorporate both qualitative and quantitative resistance. Heritability can be broad sense or narrow sense heritability. The proportion of phenotypic differences due to all source of genetic variances is termed as broad sense heritability (H^2) while that proportion of phenotypic variances due to only additive genetic variance is a narrow sense heritability (h^2) (Acquaah. 2007).

Singh *et al.*, 1993 and Atokple and Emechebe (1995) had been studied the genetic and inheritance of *A. vogelii* resistance in cowpea by identifying and determine the type and number of genes which encoded the trait for resistance to *Alectra vogelii* in cowpea. The observed segregation pattern of the population in a generation mean analysis test for resistance to susceptible population was very well fit to 3:1 Mendelian ratio which confirmed that the duplicate dominant gene are at play for the inheritance of resistance to *A. vogelii* in cowpea. However, it is important also to consider this trait as quantitative traits, that's their gene effect do not always fall into clear cut categories and govern by gene with small individual effects and they are often described by their gene action rather than by the number of genes by which they are encoded. This is because the gene action is conceptually the same for major and minor genes but the essential difference being that, the gene action of a minor gene is small and significantly influenced by the environment (Conner and Hartl,

2001). Therefore, inheritance study for the resistance to *Alectra vogelii* by determining the gene action controlling the trait and their heritability is most important in order to plan for the strategic breeding program to be taken for crop improvement since breeders are often interested in more than one trait in breeding program which they seek to improve simultaneously.

2.8.3. Mechanism for Resistance to *Alectra vogelii* in Cowpea

Successfully infestation of cowpea by *A. vogelii* to occur requires good establishment of the parasite after germination, and development of a connection between the parasite and cowpea for supply of nutrients to the parasite. *A. vogelii* seeds germinate only when they sense the presence of chemical compound produced by the roots of cowpea. The germination of *A. vogelii* seed is stimulated by the root exudates called strigolactones (Lopez-Raez *et al.*, 2009). This also stimulates the germination of radical elongation toward the host plant roots, and parasitism established soon when the radical of *A. vogelii* come into contact with the host root. It develops numerous hairs which penetrate the host root and stimulates cell division in the host root from which new host cells, together with growing parasite tissue, form a large haustorium, uniting the parasite with tissue in the host's stele which permits transfer of water and nutrients from host to parasite (Lane *et al.*, 1991; Singh 1994). However, cowpea lines and varieties which have been screened, developed and released as resistant to infection by *A. Vogelii* has showed different way of expressing their mechanism of resistance to *A. vogelii* (Lane *et al.*, 1991; Samb and Channel 1992).

Low germination stimulation production which has been exploited as a successfully mechanism of resistance in breeding sorghum cultivars that are resistant to *Striga asiatica* (Ejeta *et al.*, 2000), was considered to play a very little role in resistance to

Scrophulariaceae in legume (terBorg, 1999), this trait has recently been found in accession of a range of legumes (Rubiales et al., 2006).

Mechanism of resistance to *A. vogelii* has been studied and in all cases results show that, there are at least two mechanisms of resistance but neither of them reduces parasite germination nor fails haustorial formation at the potential host. Parasite seeds germinate as usual and the radicle attaches to the roots, but the resistant roots do not permit haustorium development. Rapid necrosis of the host cells around the point of infection, leading to the death of the parasite in 3 to 4 days is the first mechanism of resistance which is described as analogous to the hypersensitive response shown in plant-pathogen interaction. The death of cowpea tissue localized to the sites of parasite invasion is what is explained as a specific response in the host is the first mechanism of resistance of cowpea to *A. vogelii*. The second type of resistance mechanism to parasitism is not as dramatic. In these interactions the majority of parasite seedlings penetrate the cortex and reach the host stele. Although tubercles start to develop on the host root surface but do not enlarge, remaining less than 0.5 mm in diameter or fail to expand their cotyledons and eventually the parasite fails to establish neither vascular bundle nor develop internal organization (Botanga and Timko, 2005, 2006; Timko et al., 2007; Timko et al., 2008). These two mechanisms were found to have similar negative impact on plant growth and performance (Singh, 2002).

2.9. Effect of *Alectra vogelii* Infestation on Yield Components

Cowpea crop is among of the legume crop which also suffers a serious damage from the infestation by *A. vogelii* in various parts of the Sub-Saharan African (Mbwaga et al., 2010; Kabambe et al., 2008). Different control strategies as mentioned in section

2.7 have been developed and used by farmers over the areas where *Alectra* appears to be a problem in order to alleviate the damage caused by this parasitic weed. However these proposed control methods have achieved little success and breeding for resistance has been advocated as the most sustainable and feasible approach for small scale farmers. Understanding the effect of *Alectra* infestation on yield components may help a breeder know which component (trait) many need considerable attention if indirect breeding for resistance is considered (Rubiales *et al.*, 2006). Other workers have found out that *A. vogelii* infestation causes a reduction in number of pods per plant, weight of pod, number of seeds per pod, weight of seeds and chlorophyll contents of the leaves among susceptible genotypes (Kutama *et al.*, 2013). Effect of *A. vogelii* infestation in cowpea was also observed in roots biomass, the thickness of the stem, root nodulation as well as reduction on plant biomass (Geleta, 2010; Omoigui *et al.*, 2012; Kutama *et al.*, 2013; Karanja *et al.*, 2013).

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1. Experimental sites

Field and screen house pot experiments were conducted at Ilonga research station (ARI-Ilonga), in Morogoro (06° S, 37° E, Altitude 506 M) and Hombolo research station (ARI-Hombolo), in Dodoma (5° 52' S, 35° E, Altitude 1100 M) Tanzania in 2014/ 2015. Rainfall at Ilonga station is monomodal with a tendency to bimodalism with a mean annual rainfall of 1000- 1064 mm and soil type of sandy clay loams, well drained, friable and dark coloured soil. Rainfall at Hombolo is monomodal with mean annual rainfall of 655 mm and has reddish, loamy sands soils with good and high drainage. The average annual temperature at Ilonga is 24.64 °C while that of Hombolo is 27.13 °C.

3.2. Experimental materials

A total of seven cowpea genotypes (Table 2) with varying reaction to *A. vogelii* infestation were collected from International Institute of Tropical Agriculture (IITA) and Ilonga Agriculture Research Institute (ARI- Ilonga).

3.2.1. *Alectra vogelii* seed collection and preparation

Alectra seeds were collected from natural infested field planted to cowpea at Ilonga and Hombolo research station and farmer's fields during the 2014/2015 cropping season. *Alectra* plants were air- dried and well dried *Alectra* floral heads was tapped gently with piece of wood to release the seed. The seed were then sieved and stored at room temperature till use.

Table 1. Soil analysis characterizing the soils for the two locations of Ilonga and Hombolo where the field evaluation for the reaction of cowpea genotypes to *Alectra vogelii* infestation conducted in 2015

Parameters												
Location	Particle size distribution			Textural class	T.N (%)	O.C (%)	Extractable P mg/Kg	CEC Cmol ⁽⁺⁾ /Kg	pH (-log)	Exchangeble Bases (Cmol ⁽⁺⁾ Kg)		
	Silt (%)	Clay (%)	Sand (%)							Ca ²⁺	Mg ²⁺	K ⁺
Ilonga	5.64	22.12	72.24	SCL	0.34	2.67	10	18.8	6.73	15.5	2.52	1.03
Hombolo	5.85	20.13	74.02	LS	0.48	2.4	1.7	10	7.1	1.8	0.8	0.7

SCL, sand clay loam; LS, loam sand TN, total nitrogen; OC, organic carbon; Ext. P, Bray 1 extractable P; CEC, cation exchange capacity

Table 2. Description of experimental materials used in 7x7 half diallel experiment to determine inheritance for resistance of *Alectra vogelii* in 2014/ 2015 cropping season at Ilonga and Hombolo Agriculture stations

Parental identity	Genotype	Reaction to <i>A. vogelii</i>	Source
P ₁	B301	Resistant	IITA
P ₂	IT99K-7-21-2-2-1	Resistant	ARI-Ilonga
P ₃	IT99K-573-1	Resistant	ARI-Ilonga
P ₄	IT99K-1122	Tolerant	ARI-Ilonga
P ₅	VULI-1	Susceptible	ARI-Ilonga
P ₆	VULI-2	Susceptible	ARI-Ilonga
P ₇	TUMAINI	Susceptible	ARI-Ilonga

IITA, International Institute for Tropical Agriculture; ARI, Agricultural Research Institute

3.3. Hybridisation and Evaluation Trial

Seven inbred lines were mated in a 7 x 7 half diallel (Method II and Model I by Griffing, 1956) under pots experiment in 2014 cropping season in the screen house at Ilonga station. Crossing procedures used were according to Rachie *et al.*, (1975). The 21 progenies produced were advanced to F₂ in the screen house. The F₂ families together with their parents were evaluated for their reaction to *A. vogelii* in the field in 2015 cropping season using a randomised complete block design with 3 replications at two locations (Ilonga and Hombolo). Fields used for evaluation were naturally infested by *A. vogelii* weeds though additional of artificial inoculation to increase seed population was done. *A. vogelii* was artificially infested in the soil by planting them with cowpea seeds at planting time. A full spoon of *Alectra* seeds calibrated to deliver about 1000 seeds of per hill was used.

An evaluation experiment was conducted at ARI-Ilonga, Morogoro and ARI-Hombolo, Dodoma between February and April 2015. Seeds were sown in a single row plot of 5.0 m length and the spacing between and within row were maintained at 0.75 and 0.3 m respectively. All trials were kept free of weeds by hand-hoe weeding for the first five weeks and then by hand weeding six weeks after planting. No fertilizer was applied to both trials.

3.4. Data collection

Data were collected on each plant on each row genotype in each replication and mean value of each measured parameter collected was calculated. The following parameters were assessed at both sites; number of cowpea plants infested per plot and number of *A.vogelii* shoots emerged per plot were collected at 10th weeks after planting (Geleta, 2010) as well as number of pods per plot, total grain yield per plot and 100 seed weight at 12th weeks after planting per each row. Data on number of *A.vogelii* shoot emerged per plot and number of cowpeas plants infested by *A. vogelii* per plot were transformed using square root transformation $\sqrt{(X+1)}$ where “X” was the number of emerged *Alectra* shoots per plot or number of cowpea plants infested by *A.vogelii* to stabilize the variance and ensure a normal distribution (Rugare *et al.*, 2013).

3.5. Seed protein content determination

Seed protein content determination was determined by Kjeldahl method (AOAC, 1984), on seed samples taken from seeds harvested and bulked from each row on each replication of the two locations.

The Kjeldahl procedure was used to determine the protein content using block digestion and steam distillation (Kjeltec™ 8200 Auto distillation unit 2012). 0.25g

of pre-dried sample was weighed and transferred into a digestion flask to which 2g of catalyst mixture (CuSO₄, K₂SO₄) were added followed by 6 ml of concentrated sulphuric acid. The contents of the flask were digested by heating in a fume chamber for about 1hour to allow the nitrogen held in the heterocyclic ring to be released. The content was connected to the nitrogen distillation unit containing 80 ml distilled water and 50 ml of 40% v/w NaOH, which convert ammonium (NH₄⁺) into ammonia (NH₃) thereafter steam distilled into a flask containing 30 ml boric acid solution with mixed indicators (bromocresol green and methyl red). Distillation was allowed to proceed until 100-150 mls were collected. The distillate was titrated with 0.1N HCl until colour change from blue to dirty green or orange endpoint, the volume of acid used for neutralization was noted. The percentage of crude protein was calculated as follows:

$$\% \text{ Nitrogen} = \frac{(1.401 \times (\text{titre} - \text{blank}) \text{ mls} \times \text{Concentration of acids in molarity})}{\text{sample weight (g)}}$$

$$\% \text{ crude protein} = \% \text{ N} \times \text{conversion factor}$$

A conversion factor of 6.25 was used for conversion of nitrogen into protein (Ezeagu *et al.*, 2002).

3.6. Data analysis

Genotypic responses on *Alectra* shoots count and number of cowpea plant infested by *Alectra* were analysed using Analysis of variances (ANOVA) and Regression analysis approaches in Gen Stat Discovery Edition 15th (Payne *et al.*, 2012). Diallel analysis was performed using Griffings (1956) method II, fixed model I. The relative contribution of GCA to SCA were analysed using bakers ratio (Baker 1978), computed as $2V_{\text{gca}} / (2V_{\text{gca}} + V_{\text{sca}})$. Narrow sense and broad sense heritability for

resistance to *Alectra* was determined by appropriate formulars (Acquaah, 2007).

Narrow-sense heritability (h^2) which measures the proportion of additive variance in the overall variance was estimated as follows:

$$h^2 = V_A / (V_A + V_D + V_E)$$

While, broad-sense heritability (H^2) which measures the proportion of both additive and dominance variances in the overall variance, was estimated as follows:

$$H^2 = V_A + V_D / (V_A + V_D + V_E)$$

Where: V_A = Variance component due to GCA;

V_D = Variance component due to SCA;

V_E , environmental or error variance;

Correlation between characters measured was performed using Gen stat discovery 15th edition (Pyne *et al.*, 2012).

Data for differences in seed protein content among the cowpea genotypes were equally performed by Genstat 15th (Pyne *et al.*, 2012) using ANOVA procedure, while least significance difference procedure was used to compute genotypes mean differences. Differences among genotypes were accepted when the p- value for both ANOVA and LSD was less than 0.05.

CHAPTER FOUR

4.0. RESULTS

4.1. Results

High significant differences ($P < 0.001$) were found in all measured parameters except seed protein content among genotypes at Ilonga site (Table 3). Significant differences ($P < 0.001$) were observed in number of pods per plot, 100 seed weight and total grain yield among the genotypes at Hombolo location (Table 4). The genetic component for resistance to *Alectra* was only computed from the results of Ilonga site where it was found to be significant. Both general combining ability (GCA) effects and specific combining ability (SCA) effects for both *Alectra* emergency and its infestation were significant ($P < 0.001$ and $P < 0.05$ respectively)

Table 3.

Table 3. Mean squares for 7x7 half diallel for cowpea genotypes and their parents evaluated for their reaction to *Alectra vogelii* at Ilonga site in 2015

Source of variation	d.f	No. <i>Alectra</i> Shoots	No. Plant infested	Total Yield	No. of Pods	100 seed weight	Seed Protein
Replication	2	2.13	0.56	74575	51700	2.654	25.345
Genotypes	27	1.07***	0.25***	77110***	52640***	13.84***	3.87
GCA	6	2.48***	0.61***				
SCA	21	0.66*	0.15*				
Error	54	0.38	0.09	23411	18654	2.971	3.658

* and *** significantly different at 0.5 and 0.001 probability levels respectively.

Table 4. Mean squares for 7x7 half diallel for cowpea genotypes and their parents evaluated for their reaction to *Alectra vogelii* at Hombolo site in 2015.

Source of variation	d.f	No. <i>Alectra</i> Shoots	No. Plant infested	Total Yield	No. of Pods	100 seed weight	Seed Protein
Replication	2	0.66	0.05	14993	11747	0.8517	0.421
Genotypes	27	1.12	0.16	25799***	27800***	2.90***	3.67
Error	54	1.29	0.15	5282	6446	0.5942	2.549

***; significantly different at 0.001 probability levels

Further analysis of genetic component of variation (Table 5) for number of *Alectra* shoots emerged and number of cowpea plants infested shows that broad sense heritability (H^2) for both parameters were greater than 0.5 while narrow sense heritability (h^2) was close to 0.5. Broad sense heritability of 0.66 and 0.67 were estimated for number of *Alectra* shoots emerged and number of cowpea plant infested respectively while narrow sense heritability of 0.41 and 0.44 were observed for number of *Alectra* shoots emerged and number of cowpea plant infested respectively. Likewise, mean square ratio of variances due to GCA to variances due to SCA ($2V_{gca}/(2V_{gca}+V_{sca})$) “Baker’s ratio”, for both number of *Alectra* shoots emerged and number of cowpea plant infested were observed to be 0.62 and 0.66 respectively.

The GCA effect (Table 6) estimates showed that, parent IT99K-7-21-2-2-1 and IT99K-573-1 exhibited significance ($P < 0.05$) negative GCA effects to both number of *Alectra* shoots emerged and number of cowpea plant infested. Likewise, parent VULI-2 showed a significant positive GCA effects ($P < 0.001$) in both number of

Table 5. Estimates of genetic parameters and their ratios in the genotypes of 7x7 half diallel and their parent observed for trait of resistance to *Alectra vogelii* in cowpea evaluated at Ilonga site in 2015.

Parameter	No of <i>Alectra</i> shoot emerge per plot	No of plant infested per plot
h^2_{ns} (%)	0.41	0.44
H^2_{bs} (%)	0.66	0.67
Ratio $2V_{gca}/(2V_{gca}+V_{sca})$	0.62	0.66

h^2 , Narrow sense heritability; H^2 , Broad sense heritability; ratio $2V_{gca}/(2V_{gca}+V_{sca})$, Baker's ratio; V_{gca} , variances for general combining ability; V_{sca} , variance for specific combining ability.

Table 6. Estimate of general combining ability (GCA) for number of *Alectra* shoots emerged and number of cowpea plant infested by *Alectra* among seven parents used for evaluation for reaction of cowpea to *Alectra vogelii* at Ilonga site in 2015.

Parent/Crosses	No of <i>Alectra</i> emerge per plot	No of plant infested per plot
B301 (P ₁)	-0.25	-0.08
IT99K-7-21-2-2-1(P ₂)	-0.56*	-0.27*
IT99K-573-1(P ₃)	-0.49*	-0.30*
1T99K-1122 (P ₄)	-0.19	-0.09
VULI-1(P ₅)	0.47	0.28*
VULI-2 (P ₆)	0.88***	0.39***
TUMAINI (P ₇)	0.14	0.07
Standard error	0.23	0.11

* and *** significantly different at $P < 0.5$ and $P < 0.001$ probability levels respectively.

Table 7. Estimates of specific combining ability (SCA) effects for number of *Alectra* shoots emerged and number of cowpea plant infested by *Alectra* among 21 F₂ genotypes of 7x7 half diallel evaluated for their reaction to *Alectra vogelii* in cowpea at Ilonga site in 2015

Parent/Crosses	No of <i>Alectra</i> emerge per plot	No of plant infested per plot
P ₁ xP ₂	0.46	0.34
P ₁ xP ₃	0.75	0.33
P ₁ xP ₄	-0.13	0.08
P ₁ xP ₅	-0.37	0.04
P ₁ xP ₆	-0.41	-0.23
P ₁ xP ₇	-0.01	-0.10
P ₂ xP ₃	0.78	0.62*
P ₂ xP ₄	0.29	0.07
P ₂ xP ₅	-0.44	-0.26
P ₂ xP ₆	-0.64	-0.30
P ₂ xP ₇	-0.39	-0.22
P ₃ xP ₄	-0.47	-0.28
P ₃ xP ₅	-0.38	-0.17
P ₃ xP ₆	-0.42	-0.34
P ₃ xP ₇	0.07	-0.02
P ₄ xP ₅	-0.60	-0.09
P ₄ xP ₆	0.70**	0.72**
P ₄ xP ₇	0.47	0.21
P ₅ xP ₆	1.86**	0.75**
P ₅ xP ₇	-1.20*	-0.54*
P ₆ xP ₇	0.33	0.28
Standard error	0.55	0.26

*, ** and *** significantly different at P < 0.05, P < 0.01 and P < 0.001 probability levels respectively

Alectra shoots emerged and number of cowpea plants infested while VULI-1 showed a significant ($P < 0.05$) positive GCA effects in number of cowpea infested.

Positive and negative SCA effects among the crosses (Table 7) were observed in number of *Alectra* shoots emerged and number of cowpea plants infested although only cross between VULI-1xTUMAINI showed a significant negative SCA effects ($P < 0.05$) in both number of *Alectra* shoots emerged and number of cowpea plant infested. IT99K-1122xVULI-2 and VULI-1xVULI-2 were among the crosses that showed a significant positive SCA effect ($P < 0.01$) in number of *Alectra* shoots emerged and number of cowpea plant infested while IT99K-7-21-2-2-1xIT99K-573-1 cross exhibited a positive SCA effect ($P < 0.05$) in number of cowpea plant infested only.

Table 8. Mean squares for 7x7 half diallel for cowpea genotypes and their parents evaluated for their resistance to *Alectra vogelii* across sites in 2015.

Source of variation	d.f	No. <i>Alectra</i> shoots	No. Plant infested	Total Yield	No. of Pods	100 seed weight	Seed Protein
Replication	2	2.51	0.38	3094	2415	2.5	14.5
Genotypes	27	2.56**	0.54***	7941***	5960***	13.9***	5.8
Location	1	3.59	3.57***	136452***	74307***	264.8***	61.4***
G x Location	27	1.77	0.37*	23498*	20832*	2.836*	1.68
Error	54	1.20	0.15	1515	1303	1.76	3.2

*** and ** significantly different at $P < 0.5$, $P < 0.01$ and $P < 0.001$ probability levels respectively.

The genotype by location interaction significance ($P < 0.05$) differences were observed in all parameters measured except for *Alectra* shoots emerged and seed

protein content (Table 8). Likewise, significance ($P < 0.001$) differences were observed for all parameters except for *Alectra* shoots emergency accounted among genotypes across location (Table 9).

A significant positive and negative correlation was found between *Alectra* emergency and infestation to yield, yield component and seed protein content (Table 10).

Table 9. Grand means of measured parameters for 7x7 half diallel for cowpea genotypes and their parents evaluated for their resistance to *Alectra vogelii* across sites in 2015

Location	No. <i>Alectra</i> Shoots	No. Plant infested	No. of Pods	Total Yield	100 seed weight	Protein content
Ilonga	5.5	2.19	379	454	12.17	21.58
Hombolo	1.85	1.37	245.9	273.7	9.66	19.37
LSD ($P < 0.05$)	7.46	0.63	130.6	140.8	1.5	1.34

Total grain yield had highly significant ($P < 0.001$) negative correlation coefficients with both number of *Alectra* shoots emerged and number of cowpea plant infested ($r = -0.392$ and $r = -0.460$ respectively). Likewise number of pods per plot also had a highly significant ($P < 0.001$) negative correlation coefficient with both number of *Alectra* shoots emerged and number of cowpea plant infested ($r = -0.371$ and $r = -0.442$ respectively). On the other hand, hundred seed weight also was found to have a significant ($P < 0.01$) negative correlation coefficient with both number of *Alectra* shoots emerged and number of cowpea plant infested ($r = -0.255$ and $r = -0.326$ respectively).

Total grain yield had recorded a positive significant ($P < 0.001$ and $P < 0.05$) correlation with number of pods per plot and seed protein content ($r = 0.94$ and $r = 0.21$ respectively), while negative significant ($P < 0.01$) correlation was observed with 100 seed weight ($r = -0.29$).

Significant seed protein content differences were observed between locations with a grand mean of 21.58 at Ilonga and 19.37 at Hombolo site (Table 11).

Table 10. Correlation coefficient among yield, yield components, seed protein content, number of *Alectra* shoots emerged and number of cowpea plants infested by *Alectra* for 7x7 half diallel population of cowpea and their parents evaluated for their reaction to *Alectra vogelii* in 2015

	No of Cowpea Plant infested	No of <i>Alectra</i> shoot emerged	100 seed Weight	No of Pods	Total Grain Yield	Seed Protein Content
No of Cowpea Plant infested						
No of <i>Alectra</i> shoot emerged	0.8773***					
100 Seed Weight	-0.3256**	-0.2545**				
No of Pods	-0.4415***	-0.3705***	-0.3314**			
Total Grain Yield	-0.4595***	-0.3920***	-0.2947**	0.9442***		
Seed Protein Content	0.0334	0.0109	-0.1093	0.2187*	0.2086*	-

***** significantly different at 0.05, 0.01, and 0.001 probability levels respectively

Table 11. Seed protein content grand means for 7x7 half diallel for cowpea genotypes and their parents evaluated for their resistance to *Alectra vogelii* across sites in 2015

Location	Seed Protein Content
Ilonga	21.58
Hombolo	19.37
LSD (P<0.05)	1.34

4.2. Discussion

Breeding for resistance to *Alectra vogelii* in cowpea will help to improve yield especially for small scale farmers in *A. vogelii* infested areas. In this study significance differences among genotypes in their reaction to *Alectra vogelii* was found at Ilonga site. No significant differences in their reaction to *A. vogelii* at Hombolo site in Dodoma were found. This was probably due to prolonged drought that affected both crop stand and *Alectra vogelii* degree of virulence (Mbwaga *et al.*, 2010). Significance differences detected among the genotypes in their reaction to *A. vogelii* indicate that genetic variability for this trait existed among the genotypes used in the study. These results are in agreement with Mbwaga *et al.*, (2010) who found similar genotypic response to *Alectra vogelii* in cowpea. The Baker's ratio for number of *Alectra* shoot emerged and number of cowpea plant infested were 0.62 and 0.66 respectively. This implies that this trait is conditioned by both additive and non- additive gene action effects with additivity being more predominant. This is desirable and necessary phenomenon for crop improvement since additivity effect is the heritable portion from parents to offspring. It will be possible to predict progeny response to *Alectra vogelii* infestation based on general combining ability of the parents. In addition, this trait can easily be selected for, it in the progeny in early segregating generation (Facloner, 1989 and Acquaah, 2007).

Parents IT99K-27-2-2-1 and IT99K-573-1 showed a negative GCA effects to both number of *Alectra* shoots emerged and number of cowpea plant infested. The negative estimate is desirable and important because it indicates the presence of a high gene frequency for *Alectra* resistance in the materials. Therefore, parents IT99K-27-2-2-1 and IT99K-573-1 were the best general combiners in hybridization for genetic improvement for this trait. Mbwaga *et al.*, 2010 recommended the same

parents as the best source of *Alectra* resistance in cowpea. VULI-2 had significant positive GCA effect for *Alectra* emergency and infestation implying that they were transmitting susceptibility genes to progenies. This is undesirable parents in hybridization, however, VULI-2 can be used in molecular mapping to identify QTL linked to the resistant trait to *Alecta vogelii* by crossing it with IT99K-7-21-2-2-1 or IT99K-573-1 (with negative significant GCA effects) and advancing up to filiar generation 8 mapping population (Acquaah, 2007).

Negative SCA effects in number of *Alectra* shoots emerged and number of cowpea plant infested by *Alectra* was observed in VULI-1xVULI-2. This suggests that, this cross VULI-1 x VULI-2 was significantly more resistance when compared to other crosses with one parent in common. The cross between VULI-1 and VULI-2 which was identified as best specific combiner derived from parents that proved to be poor general combiners for resistance trait, signifies the presence of non- additivity influence for this trait. This non additive gene action can be best exploited by multiple crosses followed by inter-mating among desirable segregates and selection (Singh *et al.*, 2006).

High narrow sense heritability was observed to both number of *Alectra* shoots emerged and number of cowpea plant infested suggest that there will be a gain from selection for this trait of resistance to *Alectra* in cowpea. Narrow sense heritability often shows the amount of genetic components that can be transferred or inherited from parents to offspring. Therefore, from this study it was established that there will be genetic gain from selection for the trait of resistance to *A. vogelii* in cowpea when visual phenotypic selection done from the segregating generation at early generation testing (Acquaah, 2007; Karademir *et al.*, 2007). Generally, the best breeding method to be undertaken to improve this trait should be based on hybridization and or

followed by selection in order to exploit both additivity and non- additivity effect of the trait and early generation testing can be done for the advanced generation with maximum success. On the other hand, high heritability observed for this trait in this study implies that resistance for *A. vogelii* in cowpea is conditioned by few major genes with minor effect of partial dominance (Falconer and Mackay, 1996; Faurie *et al.*, 2011). This finding is in agreement with Singh *et al.*, (1993) and Atokple and Emechebe (1995) who found that dominant genes condition the resistance to *Alectra vogelii* in cowpea.

Highly significant ($P < 0.001$) and strong positive correlation coefficient observed between number of *Alectra* shoots emerged and number of cowpea plant infested with *Alectra*, suggests either aspect (number of *Alectra* shoot emerged or number of cowpea plant infested) can be used as a selection criteria in breeding for resistance to *Alectra vogelii* in cowpea (Wallece and El- Zik, 1989).

Further correlation analysis revealed that, both total grain yield, number of pod and 100 seed weight had a significance negative correlation with number of *Alectra* shoots emerged and number of cowpea plants infested. The results suggested that, there were a decrease in number of pods formed, seed weight (100 seeds weight) and total grain yield in the cowpea with increasing in number of *A. vogelii* shoots. This could indicate that there could have been accumulation of dry matter in the cowpea roots at the expenses of the pods as a result of *A. vogelii* infestation as observed by Karanja *et al.*, (2013) so as to feeds the parasitic weeds of *Alectra vogelii* which leads to the reduction of the total grain yields.

The strong and positive correlation between yield and number of pods per plot suggested that yield improvement would be possibly achieved by selecting for the

number of pods per plant since the study revealed that, number of pods per plot is a strong yield determinant. It is negatively correlated ($r = -0.44$ and $r = -0.37$ for number *Alectra* shoots emerged and number of cowpea plant infested by *Alectra* respectively at ($P < 0.001$). Thus indirect breeding for improved resistance or tolerance under *Alectra vogelii* infestation would be achieved by screening or breeding for cowpea genotypes with high number of pods per plant.

The significance negative correlation between seed weight and both number of *Alectra* shoots emerged and number of cowpea plants infested by *Alectra* suggested that, *Alectra* reduces the quality of seed by affecting seed size (Mbwaga *et al.*, 2007 and 2010; Karanja *et al.*, 2013).

The study revealed that, there were no significant differences observed for seed protein content among genotypes in each location, but significant ($P < 0.001$) differences were observed across the locations, with grand mean at Ilonga and Hombolo of 21.58 and 19.37 respectively. This implies that seed protein content is influenced by the environment. On the other hand, specific genotypes in each location (Appendix 1) accounted for seed protein content of less than 30%. Seed protein content of 30% and above are regarded as high seed protein content (Afiukwe *et al.*, 2013) is suggesting that, the genotypes under study were of low seed protein content genetically. Most analysis done to determine the seed protein content in various genotypes of cowpea in different regions revealed that, seed protein content often range from 21% to 30% (Nielsen *et al.*, 1997; Chan and Phillips, 1994; Aluko and Yada, 1995; Mwasaru *et al.*, 1999). This suggest that, breeding program to improve *Alectra vogelii* resistance to cowpea should be also endeavour to improve seed protein content.

CHAPTER 5

5.1 Conclusion

The study revealed that, both additive and non-additive gene action contributed significantly to the inheritance of resistance to *Alectra vogelii* in cowpea with additivity being more important.

Parents IT99K-27-2-21 and IT99K-573-1 were the best parents with desirable GCA effect and could be used as parental materials in a breeding program for genetic improvement for resistance to *Alectra vogelii* in cowpea.

High heritability for *Alectra* emergence and infestation respectively suggests there were few genes among others numbers with major effect for resistance to *Alectra vogelii*.

The study also has shown that, indirect breeding for improved resistance or tolerance under *Alectra vogelii* infestation could be achieved by screening or breeding for cowpea genotypes with high number of pods per plant.

Lastly the study showed that, all genotypes under the study have low seed protein content therefore, any breeding program formulated to improve them for their reaction to *Alectra vogelii* should also intend to improve them for their seed protein content.

5.2. Recommendation

It is therefore recommended that marker assisted selection should be supplemented in selection process in order to increase the precision for selection due to effect of the environment as observed from *Alectra* infestation performance between two

locations. This will increase the effectiveness and efficiency of selection for the trait under the study.

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7.0 Appendices

Appendix 1. Means for seed protein content of cowpea parental lines and their 7x7 half diallel genotypes evaluated at Ilonga and Hombolo sites in 2015

Genotypes	Location	
	Ilonga	Hombolo
B 301(P ₁)	21.67	19.18
IT99K-27-2-21(P ₂)	21.25	21.57
IT99K-573-1(P ₃)	19.38	17.92
IT99K-1122 (P ₄)	20.32	20.76
VULI-1 (P ₅)	20.75	19.25
VULI-2 (P ₆)	22.5	22.23
TUMAINI (P ₇)	21.67	19.44
P ₁ xP ₂	21.4	19.73
P ₁ xP ₃	22.44	20.62
P ₁ xP ₄	20	19.83
P ₁ xP ₅	20.06	19.13
P ₁ xP ₆	22.46	20.91
P ₁ xP ₇	21.67	19.43
P ₂ xP ₃	22.57	21.19
P ₂ xP ₄	20.81	19.77
P ₂ xP ₅	23.34	22.17
P ₂ xP ₆	20	19.65
P ₂ xP ₇	21.03	20.24
P ₃ xP ₄	22.04	20.35
P ₃ xP ₅	21.15	20.23
P ₃ xP ₆	22.35	21.16
P ₃ xP ₇	23.2	21.22
P ₄ xP ₅	21.67	18.21
P ₄ xP ₆	20.5	21.13
P ₄ xP ₇	21.19	21.41
P ₅ xP ₆	21.69	21.71

P ₅ xP ₇	23.15	21.11
P ₆ xP ₇	23.15	20.74

Seed Protein Content > 30%= High, Seed Protein Content 30% - 20%= Medium, Seed Protein Content < 20%= low, seed protein content (Afiukwe *et al.*, 2013)