

Morphology and mineral content of cowpea lines in response to

planting date and zinc application rate.

by

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Thesis submitted in fulfilment of the requirements for the degree

Master of Technology: Agriculture

In the Faculty of Applied Sciences

At the Cape Peninsula University of Technology

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> Wellington December 2017

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ABSTRACT

Cowpea (*Vigna unguiculata* (L.) Walp) is an important grain and fodder legume grown around the world. It is a dual purpose grain legume crop, providing food for man and livestock. Cowpea is identified as a potential crop to diversify food production, minimize production input by improving soil fertility and improve micronutrients of seed, therefore, improving human nutrition. There is limited information available on cowpea production and suitable agronomic practices including planting date to best suit different environmental conditions in South Africa. The objective of this study was therefore to i) evaluate two soil types (sandy and clay soil) and its effect on cowpea, yield components and mineral composition, ii) the effect of different planting date and iii) assess the effect of zinc fertilizer application rate on vegetative, reproductive parameters and mineral content of cowpea seed.

A field trial was conducted in Agricultural Research Council (ARC), in two locations Nietvoorbij (clay loam soil) and Bien Donne' (sandy soil) during the 2015 summer planting season. The trial layout was conducted in a randomised complete block design (RCBD) with five replicates. The factors of the study include three cowpea lines: Cowpea Veg1, M217 and Qukawa with zinc application rate of (0, 15 and 30 kg/ha) through soil application and two planting date (2 October and 2 November 2015). The following agronomic variables were collected, in both locations: germination rate, number of leaves, number of branches, plant height, number of seed per pod, number of pods per plant, pod length, pods per treatment, pod weight, 100 seed weight, morphological traits, moisture content and seed mineral content. Vegetative data was collected on a fourth-night basis on six middle plants per treatment and reproductive parameters were taken after harvest. The variables were subjected to ANOVA using software SAS (2012). Treatments were tested at 5% level

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of significance and differences between treatments were separated using LSD and DMRT of the SAS 2012 test.

The results indicated that vegetative and reproductive parameters measured varied significantly among cowpea lines in each location and across locations due to different cowpea lines and soil type. Line Cowpea Veg1 and Qukawa were the best performing line in both vegetative and yield parameters across the two planting dates in 2015. These lines significantly obtained higher plant height than line M217. Yield and yield parameters were significantly affected by cowpea line. Qukawa obtained the highest seed yield at Bien Donne' with a mean of 1184.2 kg/ha and seed yield of 686.25 kg/ha for Cowpea Veg1 at Nietvoorbij. The second planting date (2 November 2015) improved germination of plants across the two locations, therefore improving vegetative growth. Zinc (Zn) fertilizer significantly improved plant height across all treatments. An inconsistent response to yield parameters due to Zn application rate was observed. However, though not significant, Zn application of 15 kg/ha increased most of the measured parameters. It was concluded that line Cowpea Veg1 and Qukawa were the best performing lines. The second planting date (2 November) increase germination rate for both locations. It is therefore, recommended that future research should evaluate Zn fertilizer time of application.

Keywords: cowpea lines, planting date, Zn application, location, soil type, mineral content

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ACKNOWLEDGEMENTS

I wish to thank:

- God almighty for grace, strength, wisdom and opportunity to attain this level of my study.
- My sincere appreciation goes to my supervisors Prof F.B Lewu and Dr A.R Mulidzi for their guidance and technical support in the course of the study.
- Agricultural Research Council (ARC) research farm managers and staff for field support which made agronomic practices possible in the duration of the study. Your support was immensely appreciated.
- My family for their love and understanding.
- Department of Agriculture post-graduate students who contributed physically and emotional to the study, your support was appreciated.
- CPUT Research Fund and Faculty of Applied Science for financial support.

If you have received funding from the NRF, include the following:

The financial assistance of the National Research Foundation towards this research is acknowledged. Opinions expressed in this thesis and the conclusions arrived at, are those of the author, and are not necessarily to be attributed to the National Research Foundation.

DEDICATION

I dedicate this thesis to my family especially my mother Mrs Jabu Eunice Mfeka for believing in me and the dream that I had. Her prayers that gave me strength and courage.

CONTRIBUTION TO THE THESIS

Conference oral presentation

Mfeka, N., Lewu, F.B., Mulidzi, A.R. Morphology and mineral content of cowpea lines in response to planting date and zinc application rate. 2nd Society of Medicinal Plants and Economic Development (SOMPED), International Conference. Birchwood Hotel & OR Tambo Conference Centre, Johannesburg, South Africa from 27th to 30th August, 2017.

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GLOSSARY

Abbreviation Description			
	Description		
pH v	potential of hydrogen		
%	percentage		
ANOVA	analysis of variance		
ARC	Agricultural Research Council		
BD	Bien Donne'		
BNF	biological nitrogen fixation		
cm	centimeters		
DAFF	Department of Agriculture, Forestry and Fisheries		
DFH	days to first harvest		
EU	experimental unit		
Fe	iron		
g	grams		
L1	Cowpea Veg1		
L2	M217		
L3	Qukawa		
GR	germination rate		
К	potassium		
kg	kilograms		
Kg/ha	kilograms per hectare		
Mg/kg	milligram per kilogram		
N	nitrogen		
N ₂	nitrogen gas (di-nitrogen)		
NH ₃	ammonia		
NVB	Nietvoorbij		
°C	degrees Celsius		
Р	phosphorus		
RCBD	randomize complete block design		
scl	sandy clay loam		
sl	sandy loam		
WAP	weeks after planting		
Zn	zinc		
ZnSO ₄	zinc sulphate		
KCI	Potassium chloride		
DMRT	Duncan's Multiple Range Test		

CHAPTER 1

GENERAL INTRODUCTION

1.1 Introduction

Cowpea is a significant grain and fodder pulse grown around the world (Sebetha *et al.*, 2010, Iqbal *et al.*, 2006). It serves as a dual purpose grain legume crop, providing food for human consumption and fodder for livestock (Singh *et al.*, 2003). Cowpea is a basis of protein and less expensive than meat (Singh *et al.*, 2011; Mamiro *et al.*, 2011). The crop provides food during food scarcity in most African countries (Kebe and Sembene, 2011). According to Ojo *et al.*, (2014), cowpea contains some phytochemicals which are lacking in meat. Cowpea is a staple crop in most African countries (Ndema *et al.* 2010). In eastern Africa for instance, most of the farmers prefer to cultivate a dual purpose cowpea genotype of which both leaves and grains are important dietary products for humans and livestock (Ehlers and Hall, 1997). Cowpea is able to cover the soil surface through its production of a dense green canopy and thus protect the soil against unfavorable weather conditions such as excessive sunshine, high rain-drop impact leading to splash, soil wash and erosion; and hence conserve physical properties and soil moisture (Barrett *et al.*, 1997).

According to Gomez (2004), Africa is the leading continent in cowpea production by 68% followed by Brazil 17%, Asia 3%, United State 2% and 10% is produced by the rest of the world. Africa alone accounts for 10 million hectares under cowpea production (Mamiro *et al.*, 2011) and the crop is indigenous to Africa (Khalid *et al.*, 2012; Singh *et al.*, 2003). Cultivation of cowpea had spread to other continents such as North America, South America and Asia (Davis *et al.*, 1991). The production of cowpea in South Africa (SA) is mainly in Limpopo, Mpumalanga, North-West and KwaZulu-Natal respectively, (DAFF, 2011). Langyintuo *et al.*, (2003), stated that information on cowpea marketing and trade is lacking in Africa and data on

production is scattered. Cowpea is identified as an indigenous crop that could assist in solving Africa's food crisis and food security challenges (Momberg, 2006).

Economic and environmental costs of excessive use of nitrogen (N) fertilizers in agriculture are a global concern (Bohlool *et al.*, 1992; Pla and Cobos-Porras, 2015). According to FAO (2006), N deficiency is widespread, research showed out of 142 million tons of plant nutrients applied worldwide through mineral fertilizer, 58 million tons is N. The ability of leguminous plants to obtain their N needs from the atmosphere is a well-established phenomenon (Balasubramanian and Nnadi, 1978). The use of cowpea is to maintain the N status of soil thereby reducing the costs of commercial N fertilizers (Frankenberger and Abdelmagid, 1985). According to Bloem *et al.* (2009), biological nitrogen fixation (BNF) of leguminous crops has been used by South African farmers to increase crop yield through affordable and sustainable biological technologies that enhance soil fertility. According to Ofori and Stern (1986), this is due to crop residue and N fixation to the soil. Cowpea does not require a high rate of N, the nodules soil bacteria *Rhizobia* fix N from the atmosphere (Nkaa *et al.*, 2014). According to Davis *et al.* (1991) cowpea can be used as green manure crop.

The Agricultural Research Council (ARC) identified cowpea as a potential crop to diversify the food production base and reduce food and nutritional insecurity, particularly for resource-poor house-hold in marginal cropping areas (Shargie, 2016).

1.2 Problem statement

Cowpea is identified as a neglected and underutilized crops species which still need further research in Africa and as a result cultivation of cowpea lacks good agronomic practices, seeds (Chivenge *et al.*, 2015; Degri *et al.*, 2013) and less commercial interest (Shiringani and Shimelis, 2011). Research and production of cowpea have been neglected in SA over the years due to lack of funding and research interest, as a result, the cultivation and the agronomic information is lacking (Asiwe, 2009). Recent results have shown that cowpea it still an underutilized crop (Mabhudhi and Modi, 2016). According to Quass, (1995) no coordinating body exists for cowpea production in SA and therefore no available data on production. Kiari *et al.*, (2011) also stated that in Niger no seed companies exist to supply good quality cowpea seed. Which result in poor cultivating practices, production and a lack of useful agronomic information such as proper planting date (Shiringani, 2007; Degri *et al.*, 2013; Ewansiha *et al.*, 2014; Mojaddam and Nouri, 2014).

Cultivated soils in SA are generally very low in organic matter and are susceptible to acidification through cultivation and N fertilization (FAO, 2005b). Bloem *et al.*, (2009) reported that arable soils of South African are depleting in nutrients due to continuous mono-cropping, fertilization and leaching due to rainfall. Nitrogen fertilizer usage in SA has increased drastically over the years, which improved optimum yield but had a negative effect on soil acidity and organic matter (Barnard and Du Preez, 2004).

The use of chemical fertilizers has, therefore, become an essential practice to optimize crop production. The excessive use of N fertilizer is a great concern in agriculture globally (Bohlool *et al.*, 1992). The amount of N required by a crop is high compared with the natural N reserve in most soils. A lack of nutrients availability in cultivated soils of SA has been reported on a study done by Adeyemi *et al.*, (2012) in

Eastern Cape Province. The use of chemical fertilizers has, therefore, become an essential practice to optimize crop production.

Therefore this study documented cultivation of cowpea in the Western Cape, Boland region and necessary conclusion and recommendation were made which indicated uses of the crop and if it can be used to minimize the use of nitrogen fertilizer (cover crop or intercrop) and as crop (cash crop). To achieve this, the study was conducted as a field experiment on three cowpea lines on two different soil types evaluating their morphological and mineral content.

1.3 Objectives of the study

1.3.1 Overall objective

To determine which planting date and a zinc application rate that will best suit the production of cowpea and its effect on morphology and mineral content.

Specific objectives are:

- To evaluate the two different soil types (sandy and clay) and their effects on cowpea morphology, yield and mineral composition.
- Evaluate the performance of the different cowpea lines as affected by planting date.
- Assess the effect of Zn fertilizer application rate on the vegetative, reproductive growth and cowpea mineral content.

1.4 The research questions

- On which soil type will cowpea production be favorable?
- Will the different planting dates have influence on cowpea yield and yield components?
- What is the influence of zinc fertilizer on the mineral content of cowpea?
- Which cowpea lines will have high production in relation to different planting dates?
- Can cowpea be used to minimize nitrogen fertilizer?

CHAPTER 2

LITERATURE REVIEW

2.1 Classification and origin of cowpea

Cowpea (*Vigna unguiculata* (L.) Walp) belongs to the class Dicotyledonea, order Fabales, family Fabaceae, subfamily Faboideae, tribe Phaseoleae, subtribe Phaseolinae, and genus Vigna (Padulosi and Ng, 1997). All cultivated cowpeas are grouped under the species *Vigna unguiculata*, which is subdivided into four cultivar groups: unguiculata (the common cowpea) grown as pulse, biflora (the catjang) mainly used as forage, sesquipedalis (the yard-long bean) grown as vegetable and textiles (used for fibres) (Ng and Marachel, 1985).

According to DAFF (2013), the origin of cowpea is not known, but it is believed that it originated from countries in West Africa and SA. It is one of the oldest leguminous crops known to human with its centre of origin and domestication being more related to pearl millet and sorghum in Africa (Chivenge *et al.*, 2015). In SA the crop is known as "swartbekboon" (Afrikaans), "dinawe" (Ndebele), "dinaba" (Shangaan), "imbumba" (Zulu), "intlumayo" (Xhosa), "dinawa" (Tswana and Sotho), "monawa" (Pedi) (DAFF, 2013). Limpopo has been identified as the province with the most diverse genotypes of cowpea (Shimelis and Shiringani, 2010) and with the most primitive wild varieties (Ng and Marachel, 1985).

2.2 Growth habit

The growth habit of cowpea plants is an important morphological trait into choosing suitable planting system which affects plant spacing (Egbadzor *et al.*, 2014). Growth forms vary with different cowpea genotypes as follows: erect, trailing, climbing and bushy (Magloire, 2005). Egbadzor *et al.* (2014) stated that spreading cowpea type have a vigorous character. According to Kabululu (2008), there are two main groups

of growing-habit of cowpea indeterminate with early and late maturing habit while determinate with early maturing type. Singh *et al.* (2003) classified cowpea into three categories early maturing (60 - 75 days), medium duration (80 - 85 days) and long-duration genotypes (110 - 130 days). Early maturing with erect or semi-erect growth habit is most ideal (Singh *et al.*, 1997). Early maturing and medium duration genotype are grown mainly for grain and long-duration for fodder (Singh *et al.*, 2003).

Asiwe (2007) concluded that some farmers in SA prefer early-maturing cowpea genotype to avoid drought and frost damage while others prefer bushy type, late maturing because they are high in fodder for animal feed.

2.3 Morphological characters

Researchers around the world have studied the morphology of cowpea genotypes and its response to the environment (Pasquet, 1998; Malgoire, 2005; Adeyemi, 2012; Egbadzor *et al.*, 2014; Gerreno *et al.*, 2015).

Cowpea plant emergence is epigeal, as it does not regenerate buds below cotyledons buds which make the plant more susceptible to seedling injuries (Timko *et al.*, 2007). According to Ige *et al.* (2011) cowpea plants have a strong tap-root and many lateral roots in the soil surface.

For cowpea, leaf shape is important for taxonomic classification purposes, as well as distinguishing of genotypes (Pottorff *et al.*, 2012). Many cowpea relatives have narrow leaf shape whereas most cultivated genotypes have ovate leaf shape (Pottorff *et al.*, 2012). The first pair of leaves is simple and opposite, while the rest are arranged in an alternate pattern and are trifoliate (Timko and Singh, 2008; Olotauh and Fadare, 2012). The two lateral leaves are asymmetrical and the terminal leaf is symmetrical (Sheahan, 2012). The leaves are usually dark green in colour and show considerable variation in size (6-16 x 4-11cm) and shape (long, pointed to oval)

depending on the variety. The leaf petiole is 5 to 25cm long (DAFF, 2009; Ige *et al.*, 2011). The structure of a mature plant varies depending on genotype, growth form, temperature and the photoperiod in which the plant grows (Timko and Singh, 2008). Cowpea stems vary from fine to slightly hairy and some have purple shades (DAFF, 2009).

Flower buds of cowpea are born on peduncles emerging in the axils of the main stem or branches (Kumar and Narain, 2005). When the flower buds develop, elongation of the peduncle occurs rapidly, reaching the height of 5 to 10cm in length at anthesis (Ehlers and Hall, 1997). Flower buds complete their development in one to two weeks and start flowering at approximately in 30 days after sowing with temperatures above 30°C (Ntombela, 2012; Ehlers and Hall, 1997). Night temperatures above 20°C can damage reproductive processes of cowpea (Ehlers and Hall, 1997). Typically each flower raceme produces 2 to 4 flowers sequentially over several days (Ehlers and Hall, 1997). Flowers are borne in axillary racemes on peduncles (stalks) of 15-30cm long. Ribeiro et al., (2013) identifies cowpea flowers as papilionaceous which means the upper petal called standard, two sides petal called wings and two lower, inner petals called keel. Cowpea is a day-neutral self-pollinating plant (Shiringani, 2007; Ehlers and Hall, 1997), but only 2% of out-crossing can be expected (Ngalamu et al., 2015). According to Ige et al. (2011) cowpea flowers open in the early day, close at midday and after booming they wilt and collapse. Cowpea plants attract ants which may play a role in fertilization (Xaba, 2007).

Pods are pendulous smooth 10-23 cm with a thickly curved beak and contain 10-15 seeds. According to Flynn and Idowu (2015), at pod fill, cowpea vegetative production ceases and the plant supplies nutrients to the seeds. Pod length is an essential morphological trait in cowpea for plant breeders in developing new species directed on seed set (Ekpo *et al.*, 2012). Seeds are 4-8 mm long, 3-4 mm broad,

variable in size, colour and texture (Magloire, 2005). The seeds range in shape from round to kidney-shaped (Timko and Singh, 2008; Nassar *et al.*, 2010), and the shape of the seed will depend on the restrictiveness of the pod until maturity is reached (Gomez, 2004). According to Ehlers and Hall (1997), cowpea seed coat varies from smooth to wrinkle.

Delay planting in the season reduces the number of days to flowering, also reduce the number of days to maturity and decrease the length of regulative and reproductive periods of development (Yogoub and Hamed, 2013).

Shimelis and Shiringani (2010), stated that cultivation practices such as proper planting date, insect pest infestation and photoperiod sensitivity contribute to the low productivity of cowpea in SA. According to Dube and Fanadzo (2013), planting date is an important factor in the production of cowpea in relation to drought stress at the vegetative stage of the crop.

2.4 Suitable environmental conditions for cowpea production

According DAFF (2009 and 2011), cowpea is planted on different soil types but it prefers sandy soil which is less restrictive to root growth, with a pH range of 5.5 to 6.5 (Davis *et al.*, 1991; Ndakidemi and Semoka, 2006). This crop is most successful in most regions due to its survival ability in low fertile soils (Elowad and Hall, 1987), where it can withstand alkaline soils (West and Francois, 1982). The most important soil nutrient supplied by cowpea residues is nitrogen and phosphorus (Kumar and Goh, 1999; Viktor *et al.*, 2003).

Cowpeas are sensitive to chilling temperature with rate of germination and emergence reduced at soil temperature below 19°C (Ntombela, 2012). Increased chilling tolerance at emergence would be valuable in subtropical zone where it allow earlier planting and provides longer growing season (Ehlers and Hall, 1997). Soil

moisture and environment temperature are the most important factors that affect germination (Peksen *et al.*, 2002). The growing period of cowpea ranges from 70-180 days with mean temperature of 20°C, with the day length of 12 hours per day during the growing season (Ewansiha *et al.*, 2014; Timko and Singh, 2008; Bastos *et al.*, 2002, Shiringani, 2007). Cowpea plants prefers cold, humid conditions while high, dry temperatures results in flowers closing early (Ige *et al.*, 2011). According to Ehlers and Hall (1997), cowpea is adapted to high temperatures, drought and low fertile soil and are warm-season crops (Peksen, 2004; Agyeman *et al.*, 2014; Shiringani, 2007). Early flowering cowpea genotypes can produce a crop of dry grain in 60 days, while longer season genotypes may require more than 150 days maturing depending on photoperiod (Timko *et al.*, 2007). According to Olubaya and Port (1997), correct harvesting time reduces infestation of cowpea weevil which affects postharvest harvest handling and storage.

Ofori and Stern (1986), stated that excessive nitrogen result in excessive growth, thus suppresses reproductive growth. On the study that was conducted by Peksen (2004) on different genotypes of cowpea, results showed that plants height, colour of pod, number of branches per plant, number of seeds per pod will differ accordingly. High density will decrease the number of pods per plant (Addo-Quaye *et al.*, 2011). DAFF (2009) stated fertilization on cowpea production depends on the anticipated yield and soil fertility. On a study done by Abayomi *et al.*, (2008) showed that application of fertilizer increase most yield parameter and grain but different genotype respond different to fertilizer application.

Cowpea is a warm-season legume that requires a minimum of 18°C temperatures for all growth stages (Timko and Singh, 2008). Dugje *et al.* (2009) stated that different genotype suit different areas according to weather. Planting date of the crop is determined by the onset and duration of rain, to avoid maturity on rainy days under

dry-land condition (Dugje *et al.*, 2009). Due to lack of research regarding cowpea, soybean planting guideline has been used over the years in South Africa for planting date as they are both classified under summer legume (Shiringani 2007).

Cowpea can be planted as a mono-crop but usually planted with other crops. It suppresses weeds and protects the soil against soil erosion (Singh *et al.*, 2003).

2.5 Economic and management importance

Many producers of cowpea in SA are small-scale farmers under dry-land farming conditions (DAFF, 2011). Intercropping cowpea with other crops is commonly used practice in SA and only about 6% of producer's cultivate cowpea as sole crop (Asiwe, 2009). Cowpea is economically important indigenous African legume crop (Langyintuo *et al.*, 2003) mainly in the dry region covering 12.5 million hectares (FAO, 2005a) with annual production of about 5.2 million tons of 5.4 million tons produced worldwide (IITA, 2003). Africa is leading country with 68% of the world total cowpea production (Figure 2.1), and Nigeria with the highest yield in Africa (Table 2.1).

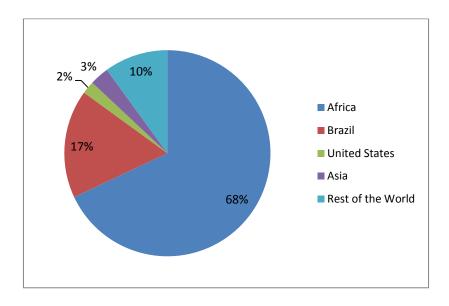


Figure 2.1: World cowpea production (Gomez, 2004)

Data from FAOSTAT (2015) indicate that total yield produced in South Africa has declined over the five years as indicated in (Figure 2.2).

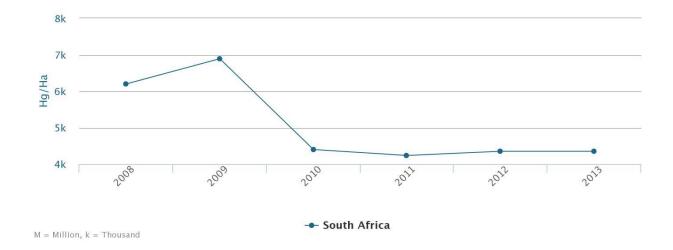


Figure 2.2: Cowpea production in SA (FAOSTAT, 2015)

A survey study done by Coker *et al.* (2014) in Niger State showed that production of cowpea in the area improved by 3% over a period of 12 years. Cowpea tends to give high yield when planted early in the growing season under wet season (Ehlers and Hall, 1997). According to Sangakkara (1998), during the dry season the total yield is not affected by time of planting (early or mid-season). Lawn (1982) reported that highest seed yield of cowpea can be obtained under irrigating with 2.31 t ha⁻¹ compared to 0.99 t ha⁻¹ under rain-fed conditions. Cowpea have high phosphorus (P) requirement, which simulate root growth and plant growth (DAIS, 2008). Legumes have a high P requirement, P is reported to stimulate root and plant growth, initiate nodule formation, as well as influence the efficiency of the rhizobium-legume symbiosis (Bationo *et al.*, 2002). According to Majaddam and Nouri (2014) one of the most important factors determining the yield of cowpea is appropriate sowing date. Delay in sowing date in the growing season will decrease yield affecting both vegetative and reproductive growth (Mojaddam and Nouri, 2014). Cowpea seeds differ in size, and a single pod can contain about 10 – 20 seeds (Ntombela, 2012).

Early application of nitrogen during the flowering stage on sole cowpea increases the yield by 600 kg ha⁻¹ (Elowad and Hall, 1987).

Research done by Ofori and Gamedoagbao (2005) and Ewansiha *et al.*, (2014), showed that when cowpea is intercropped with maize the yield of cowpea decrease due to shading and competition for nutrients. When cowpea is used as an intercrop, crop selection is of vital importance as far as planting date is concern. Ofori and Gamedoagbao (2005), suggested that cowpea is planted 10 day prior to planting maize. According to Nguluu *et al.*, (1996) and Muhammad *et al.*, (2010), cowpea is intercropped with maize mainly to sustain the nitrogen deficiency that could arise when mono-cropped. Mono-cropping of cowpea is becoming important as production is becoming commercialised to meet the demands of rapidly increasing urban population (Ehlers and Hall, 1997). Makoi *et al.*, (2009), shows that cowpea grown in monoculture gives better plant growth, higher nodule formation and increased tissue nitrogen concentration than those in mixed cropping system. Bloem and Barnard (2000), concluded that intercropping with cowpea in the same growing season does not benefit the other crop as mineralization of nitrogen occurs in the nodules once maturity is reached.

Research is underway in the Limpopo province on varieties that will improve yield with fewer days to maturity (60-65 days). These varieties have been tested in Taxes and were introduced in SA in 2012, and have yielded between 1.5 to 2.0 t/ha (Ledbetter, 2013).

Country	Area under cowpea (ha)	Production (tons)	Yield (kg/ha)
Nigeria	6770446	3560706	11212
Niger	6503081	862,455	2442
Burkina Faso	1133404	540571	8979
Mali	556945	161827	5842
Senegal	318604	70541	5685
Tanzania	277684	85526	5821
Kenya	204757	102295	8176
Malawi	148105	100,353	12939
Uganda	114210	104,526	17143
Mauritania	67330	24671	6544
Cameroon	54467	118600	40735
South Africa	25000	12440	9446
Myammar	158167	119062	12977
Haiti	91532	63319	13117
Sri Lanka	39617	27401	17244
USA	12065	16301	22669

Table 2.1: Major cowpea production countries in the world (1990-2007).

Coulibaly et al., 2009

According to Stoilova and Pereira (2013), increasing principal components of grain yield such as pod/plant, pod length, seed/pod and seed size will allow improving cowpea yield potential

2.6 Seed mineral composition

Cowpea seed as well as the vegetative parts makes major nutritional contribution to human's diet (Kebe and Sembene, 2011), as presented in Table 2.2. On a survey that was done by Mamiro *et al.*, 2011, results showed that cowpea varieties were high in protein, fibre, carbohydrates and calcium. Cowpea seed contains 25% protein and 64% carbohydrates (Magloire, 2005; Singh *et al.*, 2012) protein very within different genotypes (Afiukwa *et al.*, 2013) and 27-34% protein in the leaves (Moswatsi *et al.*, 2013; Belane and Dakora, 2012).

Cowpea line has different chemical composition and they vary with each line and its environment (Tshovhote *et al.*, 2003; Boukar *et al.*, 2011; Sallam and Ibrahim, 2016) and this can differ with edible leaves and grain (Belane and Dakora, 2012; De Moraes and Angelucci, 1971). Harvesting of leaves and grain (dual-purpose genotypes) from the same plant is a common practice in most African countries (Dube and Fanadzo, 2013). Results obtained from a study done by Belane and Dakora (2012) shows trace elements and macronutrients tend to be high on cowpea leaves than grain. Total nitrogen and protein of pod and seed increases rapidly during early stages of growth and the concentration decreases as maturity is reached (Peoples *et al.*, 1985). On the study done by Iqbal *et al.* (2006), where cowpea was compared to other legume crops on their nutritional quality, cowpea had the highest values of potassium, magnesium and phosphorus.

Components	Percentage (%)		
	Seed	Hay	Leaves
Carbohydrates	55-66	-	80
Protein	22-24	18.0	47
Water	11.0	9.6	85.0
Crude fibre	5.9-7.3	23.3	2.0
Ash	3.4-3.9	11.3	-
Fat	1.3-1.5	26	0.3
Phosphorus	0.146	-	0.063
Calcium	0.256	-	0.256
Iron	0.005	-	0.005

Table 2.2. Chemical composition (%) of different parts of cowpea

Source: Kumar and Narain (2005).

2.7 Cowpea pest and diseases

Insect pest have remained the most setback to cowpea production, because each phase attracts a number of different insect pest (DAFF, 2009; Ngalamu *et al.* 2015). Most damage usually occurs during the seedling stage (Sheahan, 2012). According to Dugje *et al.*, (2009) and Timko and Singh (2008) the main insect pest during the growing season are aphids, flowering thrips, blister beetle and pod sucking bugs. Generally, two to three sprays with insecticides are required for a good crop of cowpea, depending on the severity of insect attack and also on the cowpea variety (Ehlers and Hall, 1997). Late-maturing varieties require more sprays than early maturing varieties because of the staggered flowering period (DAFF, 2011; Dugje *et al.*, 2009).

In cowpea, diseases seldom occurs but several fungal, bacterial and viral disease attack different growth stages. Symptoms and control measure of these disease are well documented by Kumar and Narain, (2005); Singh and Allen, (1979). Plant breeders still lacks well adapted pest and disease resistant parental lines, so to breed cultivars that are resistant to pest and diseases (Ehlers and Hall, 1997).

2.8 Nitrogen in agriculture

Nitrogen (N) is a vital macronutrient required for plant growth and is an integral part of protein, amino acid and is also important to support optimal plant growth (Metuzals, 2014). Approximately 80% of the atmosphere is nitrogen gas (N₂). Unfortunately N₂ is unusable by most living organisms (Santi *et al.*, 2013; Pla and Cobos-Porras, 2015). Plants, animals and microorganisms can die of N deficiency; surrounded by N₂ they cannot use (Santi *et al.*, 2013). Rates of N₂ fixation tend to be highest when plant-available mineral N in the soil is limiting but water and other

nutrients are plentiful (ACIAR, 2008). Dube and Fanadzo (2013) stated no N fertilization is recommended post emergence but a basal dose of N at planting is ideal.

All organisms use the ammonia (NH₃) form of N to manufacture amino acid, protein, nucleic acid and other N-containing components necessary for life (Musyoka, 2014). N is the most limiting factor for crop production growth and N fertilizers represent one of the major costs in crop production (Hardarson, 1993; Hardarson and Danso, 1993). All harvested crops use up nutrients naturally available to the soil (FAO, 2006). As a result crops produced on nutrient-poor soils have limited micronutrients and pose major health issues (Belane, 2010). The N fixation efficiency of cowpeas indirectly affects its contribution to soil fertility in any farming system and the higher the efficiency of N fixation the higher will be the N concentration in most various plant parts including seed and the larger will be the amount of N added to soil through N rich cowpea residues (Balasubramanian and Nnadi, 1978). Flynn and Idowu (2015), stated that most of the nitrogen fixed during the growing season in removed from the field as grain. N is released into the soil in two ways: decomposition of plant remains and rhizo-deposition present in the soil called nodules (Metuzals, 2014). Decaying roots and nodules is the most important direct transfer of N (Peoples and Craswell, 1992).

Plants obtain N in two principal sources: soil, through commercial fertilization, manure or mineralization of organic matter and the atmosphere through symbiotic N_2 fixation (Vance, 2001; Musyoka, 2014). Balasubramanian and Nnadi (1978) reviewed the distribution of N in various parts of the plants and concluded that the seed production of cowpea tend to reduce N residue to the soil as N is taken up by the root during maturity to the seeds. Lindemann and Glover (1996) stated that legume nitrogen fixation starts with the formation of a nodule. Wagner (2011) states the

formation of nodules as bacteria colonize the host plant's root system and cause the roots to form nodules to accommodate the bacteria. The bacteria then start to fix the nitrogen required by the plant once the plant has reached physical maturity (Bloem and Barnard, 2000). Access to the fixed nitrogen allows the plant to produce leaves fortified with nitrogen that can be recycled throughout the plant. This allows the plant to increase photosynthetic capacity, which in turn yields nitrogen-rich seed. The consequences of legumes not being nodulated can be quite substantial, especially when the plants are grown in nitrogen-poor soil. The resulting plants are typically chlorotic, low in nitrogen content, and yield very little seed (Wagner, 2011).

A common soil bacterium, *Rhizobium*, invades the root and multiplies within the cortex cells and plant provides all the necessary nutrients and energy for the bacteria (Santi *et al.*, 2013). Within a week after infection, small nodules are visible with the naked eye and in the field, small nodules can be seen 2-3 weeks after planting, depending on legume species and germination conditions (Lindemann and Glover, 2003). On a study done by Kahn and Schoeder (1999), results showed a non-significant difference in nodulation of plants on seed that were inoculated with rhizobium and those that were naturally infected in the soil. Low pH of less than 5.5 negatively influences rhizobia affecting their growth and ability to function (Ferguson and Gresshof, 2015).

When nodules are young and not yet fixing nitrogen, they are usually white or grey inside (ACIAR, 2008). As nodules grow in size they gradually turn pink or reddish in colour, indicating nitrogen fixation has started (Figure 2.3). According to Hardarson and Danso (1993), nodule number and weight collate to the amount of nitrogen fixed. The pink or red colour is caused by leghemoglobin (similar to haemoglobin in blood) that controls oxygen flow to the bacteria (Dakora and Atkins, 1990; Flynn and Idowu 2015).



Figure 2.3 Mature cowpea nodules (Flynn and Idowu, 2015).

2.9 Biological nitrogen fixation

Biological nitrogen fixation (BNF) is an important feature of sustainable and environmental friendly food production and long-term crop productivity (Van Kessel and Hatley, 2000). Wagner (2011) stated that BNF was discovered by Beijerinck in 1901 by a specialised group of prokaryotes. According to Shantharam and Mattoo (1997) cowpea genotype fixes atmospheric nitrogen in a process called BNF due to specialized interaction between soil microorganisms for nitrogen and it offer ecological means of reducing external inputs and improving internal resources (Bohlool *et al.*, 1992). Proportion between total amounts of plant N, whether a plant is a contributor of N to the soil: plant species, the rhizobia present in the soil, environment and cultivation practices will determine N fixed (Giller and Cadisch, 1995). The BNF process that fixes nitrogen gas (N_2) which is changed to useful ammonia (NH_3) that will be available to plants (Lindemann and Glover, 2003). This process will be affected by the environmental factors such as soil moisture, soil acidity nutrition, disease and soil salinity (Pla and Cobos-Porras, 2015), cultivation practices performed (Ledgard and Steele, 1992; Warembourg and Roumet, 1989) and different cultivar (Danso *et al.*, 1993). For this process to occur the plant must contribute a significant amount of energy in a form of photosynthate and nutritional factors for the bacteria (Lindemann and Glover, 1996; Pla and Cobos-Porras, 2015) and conducive conditions (Peoples and Craswell, 1992). One of the methods identified by Danso *et al.* (1993) to measure BNF is the number of nodule and the weight that legume plants will give a positive correlation to the amount of N fixed. However, Appiah *et al.* (2015) reported differently stating that the quantity of nodules per plant will not determine the ability to fix nitrogen. According to Awonaike *et al.*, (1990), the effectiveness of BNF is also influenced by the cowpea inoculation used.

Thobatsi (2009) stated that BNF by legumes is a key process in Low External Input Agriculture (LEIA) technologies as it potentially results in a net addition of N to the system. According to Hardarson (1993) BNF is responsible for reducing ground water pollution, enhancing protein production, contribute N for succeeding crops and build up soil fertility. BNF is a viable alternative for providing the needs of plant species for N compounds (Rodrigues *et al.,* 2013).

2.10 Zinc deficiency in agricultural soils

Zinc (Zn) deficiency is one of the most widespread nutritional problems affecting almost one-third of the world population (WHO, 2002). Soil deficiency of Zn limits crop production (Johnson *et al.*, 2005; Khoshgoftarmanesh *et al.*, 2010; Alloway, 2008) by restricting plant growth and reducing crop yield (Nyoki and Ndakidemi, 2014; Das and Green, 2013). Zn deficiency in plant includes small leaves and short internode resulting in poor vegetative growth and fodder quality (Rathore *et al.*, 2015; Malakouti, 2007). Plant zinc deficiency is known as 'chlorotic dieback' (Tahir *et al.*, 2014). Tabassum *et al.*, (2014) stated that Zn is one of the eight trace elements which are essential in healthy growth and reproduction of crop plant and any shortage of each element restrict plant growth. Bioavailability of these minerals can be poor due to the presence of phytate which is insoluble complex that binds calcium, zinc and iron which inhibit absorption (Sandberg, 2002). According to Mamiro *et al.*, (2011) the soil mineral content influences the mineral uptake of the plant which also affects the mineral available in leaves and seeds.

Seeds from Zn-deficient soils tend to have low germination rate, reduce seedling vigour and poor competitiveness with weeds (Rengel *et al.*, 1999). Zn deficiency can be corrected by Zn fertilization which is soil applied, foliar application and seed treatment (Johnson *et al.*, 2005). On a study done by Rengel *et al.* (1999) proved that priming seeds with Zn has resulted in high crop yield in Zn deficient soils. Soil or foliar applications of Zn fertilizer increases Zn concentration in phloem-fed tissues, such as fruits, seeds, and tubers (Rengel *et al.*, 1999). Opposite results were obtained on a study done by Bobrenko *et al.* (2013), which concluded that Zn soil application was more effective than seed treatment.

According to Singh *et al.* (2012) Zn is one of the nutrients required in seed production of cowpea. On a study done by Aytac *et al.* (2014) shows that Zn fertilizer does increase both vegetative and reproductive growth especially for seed production.

Increasing Zn density in stable crops has been recommended to be a potential viable and cheaper method in sustainable agriculture rather than using food supplement (Moswatsi *et al.*, 2013; Rengel *et al.*, 1999), and this addition is referred to as fortification (FAO, 2001). Improving the micronutrient status of plants would

increase yield and increase micronutrient content of seeds therefore improving nutrition of crops and human nutrition (Johnson *et al.*, 2005; Rengel *et al.*, 1999).

2.11 Uses of cowpea at different growing stages

Cowpea can be utilized in a number of ways, cowpea leaves can be harvested as early as 21 days after planting and seed after 60 days (Gomez, 2004). Cowpea leaves and tender pods can be used as vegetables, grains can be cooked or made into bakery products (biscuits, moin-moin, akara) or used in feed mixtures and the whole plant as fodder (Mathews, 2010). It can be harvested at all growth stages, the young leaves for salad, and the crop as vegetable (Sebetha et al., 2010; Naim et al., 2010). Harvested fresh leaves of cowpea are often consumed by many South Africans rural people either merely as *imfino* or in combination with stiff porridge (Moswatsi, 2015; Dube and Fanadzo, 2013). Leaves are among the top leaf vegetable used in many parts of Africa and are sold in fresh and dry forms in many African countries (Coulibaly et al., 2009). Spreading, prostate varieties are mainly used for leafy vegetables (Van Rensburg et al., 2004). On a study done in Ghana by Egbadzor et al. (2013), shows that producers for grain production favour white/cream or cream seed as it is mostly preferred by consumer because of short period in cooking. The dry seed grain is commonly milled and consumed in numerous traditional main dishes of Africa as porridge (Kebe and Sembene, 2011; Kiari et al., 2011).

Improving the nutritional standard of cowpea fodder for use by livestock is of vital important to improving the productivity and profitability of the farming systems (Tarawali *et al.*, 1997). Livestock is an important component in the agricultural production system, and plays a crucial role in the economy (Hasan *et al.*, 2010).

Coker *et al.* (2014) emphasised the importance of cowpea forage as animal feed mainly during the dry season as it vital importance. Cowpea fodder is an important resource for livestock, the aboveground part of cowpea except pod which is grain (Inaizumi *et al.*, 1999). Heuze *et al.* (2015) stated that cowpea fodder includes vines and the leaves either fresh or as hay as feed for livestock and especially during dry seasons in West Africa (Tarawali *et al.*, 1997)

Peoples and Craswell (1992), has outlined the important role of legume plants in the farming system as follow:

- i. Cropping system where legume are grown in rotation or intercropped with cereals either as crops in their own right or as green manure.
- ii. Grazing system including extensive grazing of natural vegetation in semiarid regions, intensive pastoral type of agriculture and 'cut-and-carry' systems.
- iii. Planting systems where legume cover crops, food crops or shade trees are grown in the inter-row space of trees crops such as cocoa, coffee, tea, rubber and oil palm.
- iv. Agroforestry system in which multipurpose tree and shrubs legume are utilised in combination with crops and animal to increase the productivity and the sustainability of the farming system.

2.12 Conclusion

Previous research done in SA concluded that cowpea is an underutilised crop due to lack of proper cultivation practices, information and market. Cowpea plant have the ability to strive under unfavourable conditions. Cowpea plant (leaves and grain) contribute to animal and human diet due to nutritional content during different growing stages of the plant, especially in developing countries.

Production of cowpea to its optimum can be challenged by number of factors such as planting time, quality of seed, pest control and postharvest practices which need further research.

Cowpea can be used in an intercropping or crop rotation system this is beneficial as the plant fixes atmospheric nitrogen to usable nitrogen by the plant. Therefore minimum input is required in production of cowpea.

CHAPTER 3

MATERIALS AND METHODOLOGY

3.1 Seed Collection

The three cowpea lines used in the study were: Cowpea Veg1, M217 and Qukawa. These lines were obtained from Agricultural Research Council Vegetable and Ornamental Plant Institute (ARC-VOPI) gene bank Pretoria-South Africa. The seeds were counted and cleaned; all the damaged seeds were removed. Seeds were kept in a cold storage at a temperature of 4°C and were placed at room temperature prior to planting.

3.2 Experimental sites

The field experiment was conducted at ARC Neitvoorbij-Infruitec in Stellenbosch, Western Cape. The experiment was done on two research farms at Bien Donne and Nietvoorbij during the year 2015 growing season. The description of the experimental sites and soil texture is presented in Table 3.1. Western Cape Province has a Mediterranean climate. The area has an annual rainfall of 278mm of which approximately 178mm falls from March to August (Fourie, 2011). The average difference of the minimum temperature of the two sites is 1.02°C in 2 October 2015 which was the first planting date of the experiment. The average maximum temperature differs with 0.97°C on second planting date 2 November 2015 and temperature generally increase throughout the growing season. Winter rainfall of the Western Cape delayed planting from the intended September to October 2015 because of cold rainy weather. In October the average rainfall dropped to 0.18 (Nietvoorbij) and 0.24 (Bien Donne') mm for both sites as presented in Table 3.2.

Table 3.1	. Description	of the study	y locations.
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Location	Soil texture	Percentage (%)	Coordinate
Nietvoorbij (Stellenbosch) Bien Donne'	sandy clay Ioam	Sand (69.8%), silt (9.6%), clay (20.6)	33 [°] 55'S, 18 [°] 52'E
(Paarl)	sandy loam	Sand (77%), silt (8%), clay (14.2)	33 [°] 56'S, 18 [°] 36'E

Table 3.2. Average monthly minimum, maximum temperature and cumulative rainfall from Bien Donne' and Nietvoobij during2015 growing season.

Month/Year	Niety	voorbij (Stellenbos	sch)	Bien Donne'(Paarl)				
	Min. Temp [°] C	Max. Temp [°] C	Total rainfall (mm)	Min. Temp [°] C	Max. Temp [°] C	Total rainfall (mm)		
October 2015	10.62	27.37	0.18	11.64	26.4	0.24		
November 2015	10.1	28.05	0.7	12.66	27.18	0.55		
December 2015	13.45	30.86	0.97	15.62	30.83	0.89		
January 2016	18.3	34.03	0.29	20.27	33.4	0.61		
February 2016	14.35	31.67	0.41	16.55	31.04	0.23		
March 2016	11	29.16	0.33	14.6	27.25	1.21		

Source: ARC experimental station records.

Min temp= minimum temperature, Max temp= maximum temperature

3.3 Soil sampling and analysis

Prior to the establishment of the trial, soil samples at a depth of 0-30cm was collected using soil auger and put into 1kg nylon bag each (Adeoye and Agboola, 1984). Five samples for both experimental sites were taken randomly from each field and taken to commercial laboratory (BEMLAB, 2015) for physical and chemical properties of the soil. The soil samples were bulked for each site and analyses were done accordingly. The samples were analysed for soil available P, K, Cu, Zn, Mn, B, Fe, C%, N%, pH and exchangeable cations (Na, K, Ca and Mg). Procedure and methods used are out lined in Table 3.3. But for the purpose of this study only the following will be discussed in details pH, P, K, Zn, C% and N%. After harvest soil analysis was taken per treatment at a depth of 0-30cm.

Chemical properties	Standard specification, techniques/equipment used
В	ICP-OES Hot water extraction
Ca, K, Mg, Na	ICP-OES, Ammonium Acetate extraction
Р	ICP-OES Bray II
рН	KCI method
Organic carbon	Walkey- Black method
Ν	Leco total combustion method
<u>Cu, Zn, Mn</u> BEMLAB, 2015	EDTA extraction method

3.4 Experimental design and treatments

The trial layout was a randomised complete block design (RCBD) fitted into a factorial arrangement and replicated five times as illustrated in Appendix A for both sites. Each replicate consisted of eighteen experimental units (EU), each EU had twenty plant stands, and each EU was $2 \times 1 \text{ m}^2$ (Musyoka, 2014). Spaces between block was 2 m and 1 m between EU. Experimental area per site was $54m \times 13m$ (702 m²). The experiment had four factors: the two planting dates (2 October and 2 November 2015), soil types (sandy clay loam and sandy loam soil), three cowpea lines (Cowpea Veg1, M217 and Qukawa) and three levels of zinc (1= 0% control, 2 = 50% and 3 = 100%).

Treatment consisted of zinc fertilizer at three application rates. Zinc fertilizer was applied as ZnSO₄ (33% Zn). Fertilizer was applied directly to the soil by placement method (Moswatsi *et al.*, 2013). Application of fertilizer was made at eight weeks after planting (8WAP) for both planting dates. The recommended rate of zinc fertilizer was 30 kg ZnSO₄/ha (Alloway, 2008). The trial consisted of 9 treatments per location (Table 3.4).

Cowpea line	Name	Zinc level	Application rate
1	Cowpea Veg1	1	0% Control
1	Cowpea Veg1	2	50% Zn
1	Cowpea Veg1	3	100% Zn
2	M217	1	0% Control
2	M217	2	50% Zn
2	M217	3	100% Zn
3	Qukawa	1	0% Control
3	Qukawa	2	50% Zn
3	Qukawa	3	100% Zn

Table 3.4. Treatment details of Zn fertilizer

3.5 Cultivation practices and management

Fields were prepared using plough and then disc-harrowed (Olaoye, 2002) with a tractor, this was done in mid-September 2015. Plots were levelled and ridged to 2x1 m per experimental unit. Ridging promoted uniform slope between experimental plots. The layout was marked using tape measure, poles and ropes to demarcate experimental units. Beds (plots) were demarcated and manually prepared to obtain 2 x 1m area using spade. The same land preparation was done for both experimental sites. Ropes and meter rule were used to measure plant spacing.

Prior to planting, irrigation system were installed in both experimental sites. Micro sprinklers with 360° wetting pattern was used for the study (Geetha and Varughese, 2001). Immediately after planting, both fields were irrigated to stimulate germination. Plants were irrigated three times a week, one hour per day (12 mm/hour) and then selected irrigation was done according to matured EU. The cowpea seeds were hand planted at a spacing of 40cm x 20cm (Addo-Quaye *et al.*, 2011) and seeds were planted using dibbling method (Kumar and Narain, 2005). Two seeds were sown per hole at a depth of about 3cm (Sarkin *et al.*, 2015). Basal NPK 2-3-2 (14) fertilizer at a rate of 30 kg/ha was applied at planting (Atakora *et al.*, 2014). The fertilizer was incorporated to the soil using hand spades. The planting dates were 2nd of October and 2nd of November and planting dates were the same for the two experimental sites.

Germination was monitored and recorded two times a week till seedlings were well established. At second trifoliate stage 4WAP for first planting date and 2WAP for second planting date, the plants were thinned to one plant per stand (Legwaila *et al.*, 2012; Sallam and Ibrahim, 2016; Gerreno *et al.*, 2015). Thereafter plant gaps were refilled where needed. Recording number of germinated plants was done after

thinning to one plant per stand. Vegetative data of all treatments were taken from six middle plants (Aikins and Afuakwa, 2010; Nyoki and Ndakidemi, 2014; Gerreno *et al.*, 2015).

From the number of germinated plants, germination percentage was obtained by the following equation (Pahla *et al.*, 2014):

(GP)= <u>no. of g/T</u> x 100 =%

g is the number of germinated seeds per T (treatment) and 20 is for the total number of seeds. GP is the germination percentage.

Cutworms were observed for the first planting date in both fields and a pesticide (Cutworm Bait) was applied at 4WAP. This powder-form pesticide was applied once around each plant for the infested experimental units.

Weeding was done by hand within EU at four weeks after germination and subsequent weeding was done at three weeks interval and using spades between blocks. No weed control was necessary at highest plant population. General overview of practices performed during the experiments are shown on Appendix B.

Aphids were first observed at 8WAP in Bien Donne' for the first planting date and were regularly controlled with Kemprin 200 EC sprayed at the rate of 1.0 ml per 10 litre of water as per manufacturer' recommendation using backpack sprayer. Spraying started at initial flowering and repeated at 14 days interval where necessary. Zinc sulphate (ZnSO₄) was applied at initial flowering; this was indicated by the identification of flower bud on plants. The treatment application of the fertilizer was applied at the same date for both the areas, with different dates for the two planting dates.

3.6 Data collection

3.6.1 Vegetative parameters

Vegetative parameters measured were germination rate, plant height, number of primary branches per plant and number of leaves. Vegetative data were taken at 4, 6, 8, and 10WAP (second planting date) and 6, 8, 10, and 12WAP (first planting date), then continued at an interval of two weeks till reproductive growth commenced. Number of leaves borne on each plant at full leave maturity was counted and expressed as number of leaves per plant from data plants. Number of branches on the main stem of data plants on each experimental unit (EU) was counted from data plants. Plant height (cm) was measured from the main stem from ground level to the tip of the plant using a meter rule (Agbogidi and Egho, 2012; Ngalamu *et al.*, 2013).

3.6.2 Reproductive parameters

Reproductive parameters were taken at maturity; this was indicated by the change in colour and texture of pods (IBPGR, 1983). Matured pods from data plants were harvested separately from the border rows (Naim *et al.*, 2012; Ewansiha *et al.*, 2014; Ezeaku *et al.*, 2015). Harvesting of pods was done and ten pods were selected and threshed, seeds were counted and the average number of seeds per pod of each EU was calculated. Pod length (cm) of ten pods per treatment was randomly measured using a meter ruler. This was done at frequent interval as pods are ready for harvest. Number of pods per plant for data and yield plant were counted separately. Pods from data plants and boundary plants were counted per treatment and expressed as number of plants per treatment. Pods for each treatment were weighed using a digital weighing scale and an average pod weight was obtained. Hundred seeds weight was determined by randomly counting 100-seed (Naim and Jabereldar, 2010; Ngalamu *et*

al., 2013) from the threshed pods from each EU by using a digital weighing scale. Average seed weight was obtained. Moisture content of each treatment was measured using GAC 2100b Grain Analysis Computer. An average per treatment was obtained.

3.7 Chemical analysis of cowpea seed

At maturity two replicates of 100g samples per site, were sent to a private laboratory for mineral content analysis of the seed. Analysis was determined using fruit standard and trace element procedure (<u>www.bemlab.co.za</u>). Minerals analysed included percentage of N using the (Leco-combustion method) and P, K, Ca, Mg, Na, Mn, Fe, Cu, Zn, B using Hydro-chloric total acid digestion.

3.8 Qualitative and quantitative cowpea traits

Morphological characters from each line was observed and recorded. Assessment of sixteen traits were collected according to the International Board for Plant Genetic Resources (IBPGR, 1983) cowpea descriptors and scoring scale as follows:

Table 3.5. Qualitative and quantitative traits of the three cowpea lines.

									Tra	aits						
Lines	GP	GH	LC	FC	NMB	PPP	PA	PL	PS	NPPP	PC	SPP	SC	ST	SS	SW
Cowpea Veg1	2	3	7	4	4.3	3	3	15.8	3	40.7	1	15.1	3	1	5	11.7
M217	1	2	5	2	4.5	3	5	10.1	5	26.2	2	8.4	1	3	1	14.3
Qukawa	2	5	5	4	4.4	2	3	17.25	3	18.7	3	10.8	2	1	1	14.1

GP= Growth pattern: 1= Determinate, 2= Indeterminate; **GH= Growth habit**: 1= Acute erect (branches form acute angle with main stem), 2= Erect (branching angle less acute that above), 3= Semi-erect (branches perpendicular to main stem, but not touch ground), 4= Intermediate (most lower branches touch the ground), 5= Semi-prostrate (main stem reaches 20 cm or more), 6= Prostrate (plants flat on ground),7= Climbing; SC= Seed colour: 1= cream, 2= brown, 3= grey; **LC= Leaf colour**: 3= pale green, 5= intermediate green, 7= dark green; **NMB= Number of main branches**: average of 10 randomly selected plants; **FC= Flower colour**: 1= white, 2= yellow, 3= red, 4= purple; **ST= Seed texture**: 1= smooth, 3= smooth to rough, 5= rough, 7= rough to wrinkled, 9= wrinkled; **SS= Seed shape:** 1= kidney, 2=

ovoid, 3= crowder, 4= globose, 5= rhomboid; **PA= Pod attachment**: 3= pendant, 5= 30- 90° down from erect, 7= erect; **PS= Pod shape**: 0= straight, 3= slightly curved, 5= curved, 7= coiled; **PPP= Pods per peduncle**, average of mature pods from 10 randomly selected plants. 1= 1-2, 2= 2-3, 3=3-4; **PC= Pod colour**: 1= pale tan or straw, 2= dark tan, 3= dark brown, 4= black or dark purple, 5= other; **PL= Pod length (cm)**: average of 10 mature pods from 10 randomly selected plants; **SW= Seed weight (g)**: weight of 100 seeds per treatment; **NPPP= Number of pods per plant**: average number of pods from 10 selected plants; **SPP**= Seed per pod: average of 10 pods.

3.9 Statistical Analysis

Data on vegetative and reproductive parameters as well as mineral content of the seed were subjected to analysis of variance (ANOVA) procedure using SAS (SAS, 2012). Treatments were tested at 5% level of significance and differences between treatments were separated using LSD and DMRT of the SAS 2012.

CHAPTER 4

RESEARCH RESULTS

4.1 Bien Donne' (Paarl)

4.1.1 Vegetative parameters

4.1.1.1 Seed Germination

The different cowpea lines had a significant (P<0.05) effect on the germination percentage of plants at Bien Donne', cowpea line 1 had the highest germination percentage 89% followed by line 3 with a germination percentage of 85% and line 2 had the least germinated plants of 75% in the first planting date.

There was generally a slight improvement in seed germination in November compared with October for all cowpea lines (Table 4.1).

Cowpea line	02-Oct-15	02-Nov-15
1	89 ^a	94 ^a
2	75 ^b	93 ^a
3	85 ^{ab}	93 ^a
LSD _{0.05}	12.25	15.85

Table 4.1. Effect of planting date on germination at Bien Donne'.

LSD_{0.05}= Least significant difference; Cowpea line 1= Cowpea Veg1, 2= M217, 3= Qukawa: 02-Oct-15= 02 October 2015, 02-Nov-15= 02 November 2015.

Means with different letter within the column shows significant difference (P< 0.05).

4.1.1.2 Number of leaves per plant

There was generally no significant (P>0.05) difference in the number of leaves as affected by the different cowpea lines (Table 4.2). However, an increase in the number of leaves per plant where zinc fertilizer was applied at level 2 (50%) was observed compared to the control plants. An increment of zinc to level 3 (100%) resulted in plants with less number of leaves (Table 4.2) for cowpea lines 1 and 2. Planting date did not have a significant effect on the number of leaves per plant across all cowpea lines (Table 4.3).

4.1.1.3 Number of primary branches per plant

The number of branches per plant was significantly (P<0.05) affected by the three cowpea lines and Zn application levels. Line 1 at second Zn application level obtained the highest mean number of branches and was significantly different to line 2 at Zn level 3 which obtained the least number of branches per plant with a mean of 4.93 (Table 4.2). Planting date had a significant influence on the number of branches per plant. The highest number of branches per plant was obtained on the first planting date (2 October) and was significantly different from those of second planting date (2 November) with a decline of 55% across all cowpea lines (Table 4.3).

4.1.1.4 Plant height (cm)

The results of the study indicate there was a significant difference (P<0.05) for the measured plant height in Bien Donne'. Cowpea line 1 and 3 recorded higher mean plant height across all treatments regardless of Zn level and were significantly different to line 2 with lower plant height (Table 4.2). Plant height was significantly

affected by planting date as taller cowpea plants were obtained when planted in November (Table 4.3).

Table 4.2. Effect of cowpea lines and Zn application rate on vegetative parameters.

Cowpea line	Zn level	No. of leaves	No. of branches	Plant height (cm)
1	1	65.26 ^a	7.06 ^a	15.52 ^a
1	2	67.37 ^a	7.21 ^a	14.05 ^a
1	3	50.06 ^a	6.36 ^{ab}	12.63 ^a
2	1	47.19 ^a	5.66 ^{ab}	5.10 ^b
2	2	54.59 ^a	6.20 ^{ab}	5.14 ^b
2	3	46.91 ^a	4.93 ^b	5.47 ^b
3	1	56.77 ^a	6.90 ^{ab}	15.17 ^a
3	2	52.18 ^a	6.48 ^{ab}	12.74 ^a
3	3	52.36 ^a	5.61 ^{ab}	14.17 ^a
LSD _{0.05}		25.16	2.09	4

LSD_{0.05}= Least significant difference; No = number, Cowpea line: 1= Cowpea Veg1, 2= M217, 3= Qukawa; level 1= Control, 2= Zn at 50% and 3= Zn at 100%. Means with different letter within the column shows significant difference (P< 0.05).

Planting date	No. of leaves	No. of branches	Plant height (cm)
2-Oct-15	55.13 ^a	8.17 ^a	10.09 ^b
2-Nov-15	54.36 ^a	4.52 ^b	12.23 ^a
LSD _{0.05}	7.27	0.87	1.68

 $LSD_{0.05}$ = Least significant difference; No. = number; 02-Oct-15= 02 October 2015, 02-Nov-15= 02 November 2015. Means with different letter within the column shows significant difference (P< 0.05).

In general, the second planting date at Bien Donne' improved germination capacity for all cowpea lines. The highest number of germinated plants was recorded from cowpea line 1. Cowpea line 1 had more number of branches than other lines. Line 1 and 3 had taller plants and was significantly different to line 2 with shorter plants. The highest number of branch was obtained from the first planting date across all cowpea lines. The second planting date significantly increased plant height of all cowpea lines.

4.1.2 Reproductive parameters

4.1.2.1 Number of pods per plant

There was no significant (P>0.05) effect on the number of pods per plant across all treatments. Regardless of the insignificance between means, a trend was observed across all treatments with an increase in the number of pods per plant at Zn level 2. A further increment of Zn to level 3 for line 1 and 3 resulted in less number of pods obtained. Planting date did not have a significant effect on the number of pods per plant (Table 4.5).

4.1.2.2 Total harvested pods

There was a significant (P<0.05) difference of the number of harvested pods per treatment. Cowpea line 1 at Zn level 1 obtained the highest number of harvested pods with a mean of 1046.1 but was not significantly different from Zn level 2 and 3 (Table 4.4). Similar trend of results were obtained for cowpea line 2 and 3 in response to Zn level. Planting date had a significant (P<0.05) effect on the number of total harvested pods. Planting cowpea early in the growing season (2 October) increased the total number of harvested pods per treatment (Table 4.5).

4.1.2.3 Pod weight (g)

Treatment had a significant (P<0.05) effect on pod weight. Cowpea line 1 and 3 had highest pod weight regardless of Zn level. Line 2 had significantly less pod weight and there was no significant difference between the Zn levels. In Bien Donne' pod weight from the first planting date (2 October) were heavier than those of the second planting date (2 November).

4.1.2.4 Number of seed per pod

The number of seed per pod was significantly different. Cowpea line 1 recorded the highest number of seed per pod and was significantly different to line 2 and 3 across all Zn application rates. Line 2 obtained the least number of seed per pod with the means ranging from 9.26 – 9.65 (Table 4.4). There was no significant difference in the number of seed per pod in cowpea line 3. Planting date did not have a significant effect on the number of seed per pod.

4.1.2.5 Pod length (cm)

A distinct significance between the three cowpea lines was observed, as cowpea line 3 recorded longer pods with a mean of 17.81cm followed by line 1 (16.42cm) and shorter pods were obtained in line 2 (11.97cm) (Table 4.4). No significant (P>0.05) difference was observed within the three cowpea lines as affected by Zn level. Pod length in Bien Donne' was not significantly affected by planting date.

4.1.2.6 One-hundred seed weight (g)

Cowpea line 2 and 3 obtained significantly higher one-hundred seed weight and were not significantly different from each other across all treatments. Line 1 obtained less one-hundred seed weight and was statistically different from the two cowpea lines. Seeds harvested from the second planting date (2 November) obtained heavier onehundred seed weight than those from first planting date (2 October).

4.1.2.7 Moisture content

Line 3 and 1 obtained higher moisture content and was not significantly different from each other. Line 2 at Zn level 3 obtained the least moisture content with a mean of 10.76% (Table 4.4). Seeds harvested late in the growing season (2 November) obtained higher moisture content than when planted early (2 October).

Cowpea line	Zn level	No. of pods/plant	Total harvested pods	Pod weight (g)	No. of seed/pod	Pod length (cm)	100 seed weight (g)	Moisture content (%)
1	1	30.02 ^a	1046.1 ^a	2086.3 ^a	16.53 ^{ab}	16.35 ^b	11.4 ^b	12.17 ^a
1	2	34.62 ^a	1027.0 ^{ab}	2221.5 ^a	16.71 ^a	16.42 ^b	11.5 ^b	12.16 ^a
1	3	31.61 ^a	995.5 ^{ab}	2019.3 ^a	15.75 ^{abc}	16.18 ^b	12.2 ^b	12.34 ^a
2	1	25.99 ^a	831.9 ^{abc}	1021.9 ^b	9.26 ^d	11.97 ^c	14 ^a	11.02 ^c
2	2	30.02 ^a	780.9 ^{bc}	1104.7 ^b	9.65 ^d	11.78 ^c	13.8 ^a	11.36 ^{bc}
2	3	32.72 ^a	598.0 ^c	973.8 ^b	9.37 ^d	11.52 ^c	13.4 ^a	10.76 ^c
3	1	19.64 ^a	854.3 ^{ab}	2157.5 ^a	15.49 ^{bc}	17.81 ^a	13.7 ^a	12.08 ^{ab}
3	2	21.61 ^a	869.6 ^{ab}	2229.7 ^a	14.92 ^c	17.56 ^a	14 ^a	12 ^{ab}
3	3	20.17 ^a	926.6 ^{ab}	2368.4 ^a	15.54 ^{bc}	17.57 ^a	13.4 ^a	12.47 ^a
LSD _{0.05}		15.26	246.59	545.43	51.08	0.57	1.08	0.78

Table 4.4. Influence of Zn application rate and cowpea lines on reproductive parameters at Bien Donne'.

LSD_{0.05}= Least significant difference; L1= Cowpea Veg1; L2= M217; L3= Qukawa; T1= Zn at 0%; T2= Zn at 50% and T3= Zn at 100%; No. = number. Means with different letter within the column shows significant difference (P< 0.05)

Planting date	No. of pods/plant	Total harvested pods	Pod weight (g)	No. of seed/pod	Pod length (cm)	100 seed weight (g)	Moisture content (%)
2-Oct-15	28.86 ^a	936.32 ^a	1926.8 ^a	13.89 ^a	15.28 ^a	12.6 ^b	10.56 ^b
2-Nov-15	25.82 ^a	835.10 ^b	1673.4 ^b	13.50 ^a	15.27 ^a	13.49 ^a	13.08 ^a
LSD _{0.05}	4.63	73.82	209.74	0.6	0.33	0.52	0.36

 $LSD_{0.05}$ = Least Significant Different; No. = Number; 02-Oct-15= 02 October 2015, 02-Nov-15= 02 November 2015. Means with different letter within the column shows significant difference (P < 0.05)

In summary, cowpea line 1 had the highest total harvested pods. Line 3 at Zn level 3 attained heavier total pod weight. Cowpea line 2 and 3 obtained higher 100-seed weight and was significantly different to line 1. The first planting date generally improved the total number of harvested pods and therefore improved pod weight for all lines. At Bien Donne' 100-seed weight was obtained from the second planting date.

4.1.2.8 Mineral composition of cowpea seed

The results of the analysed seed chemical composition of the three cowpea lines are presented in Table 4.6. The three cowpea lines responded differently in the uptake of Zn application rate. There was no significant (P>0.05) difference in potassium content of the dried seed across all Zn levels. Nitrogen, phosphorus, sodium, iron, zinc, calcium and magnesium were significantly (P<0.05) affected by the three cowpea lines. A similar nitrogen percentage of 4.08 was obtained for line 1 and 3 at Zn level 2 which was the highest percentage. However, a non-significant difference (P>0.05) was observed within each cowpea line for all Zn levels. Line 1 and 3 at Zn level 2 obtained a significantly higher nitrogen percentage than line 2 at Zn level 2.

Cowpea line 3 at Zn level 2 obtained the highest phosphorus percentage and was significantly different to line 2 at Zn level 2 and line 1 at Zn level 1 and 2.

Sodium concentration was the highest in cowpea line 1 at Zn level 3 and was significantly different to line 2 and 3 both at Zn level 3, the percentage decrease of 65 and 59 respectively.

Cowpea line 1 had the highest concentration of iron at Zn level 1 with a mean of 88.75 and was significantly different to Zn level 3 with a mean of 77.50, this element was the lowest in cowpea line 2 at Zn level 1 with a mean of 67.00. Application of Zn fertilizer in Bien Donne' for cowpea line 1 suppressed available iron content in dried seed by 15% as shown on Table 4.6. Opposite results were observed with line M217, application of zinc increased iron content in the seed with the mean of 86.5 and 82.25 respectively.

Zinc concentration in cowpea seed was the highest for line 3 Zn level 2 and as significantly different to line 1 at Zn level 1 and 3, also line 2 at Zn level 2. Calcium content differed significantly in each cowpea line.

The highest concentration of calcium was obtained in line 2 at Zn level 1 and 3 and was significantly different to line 1 and 3 across all Zn levels with lower calcium concentration.

Magnesium concentration was the highest in cowpea line 1 at Zn level 1 and was significantly different to Zn level 3 with lower concentration. Line 2 obtained it highest magnesium concentration at Zn level 2 but was not significantly different to Zn level 1 with 11%. Magnesium concentration was the same across all Zn levels for line 3.

Minerals									
Cowpea	7	Percentage (%)			mg/kg				
line	Zn level	N	Р	K	Na	Fe	Zn	Ca	Mg
1	1	3.96 ^{ab}	0.5 ^{cd}	1.57 ^a	119.25 ^{ab}	88.75 ^a	35.00 ^b	0.12 ^{bc}	0.21 ^a
1	2	4.08 ^a	0.49 ^d	1.53 ^a	126.00 ^a	84.25 ^{ab}	39.00 ^{ab}	0.11 ^{cd}	-
1	3	3.96 ^{ab}	-	1.46 ^a	128.50 ^a	77.50 ^{bc}	37.25 ^b	0.10 ^{cd}	0.20 ^b
2	1	3.86 ^{ab}	0.52 ^{bcd}	1.47 ^a	107.25 ^{ab}	67.00 ^c	38.25 ^{ab}	0.15 ^a	0.18 ^c
2	2	3.78 ^b	0.53 ^{abc}	1.48 ^a	106.25 ^{ab}	86.50 ^{ab}	36.25 ^b	0.14 ^{ab}	0.20 ^b
2	3	3.88 ^{ab}	0.53 ^{ab}	1.49 ^a	77.75 ^b	82.25 ^{ab}	28.75 ^{ab}	0.15 ^a	0.19 ^{bc}
3	1	3.94 ^{ab}	0.54 ^{ab}	1.52 ^a	111.25 ^{ab}	79.25 ^{ab}	39.00 ^{ab}	0.10 ^d	0.20 ^b
3	2	4.08 ^a	0.56 ^a	1.57 ^a	107.25 ^{ab}	85.00 ^{ab}	42.00 ^a	0.11 ^{cd}	0.20 ^b
3	3	3.98 ^{ab}	0.53 ^{abc}	1.53 ^a	80.75 ^b	80.50 ^{ab}	38.25 ^{ab}	0.11 ^{cd}	0.20 ^b
LSD _{0.05}	-	0.26	0.03	0.13	37.56	10.85	4.12	0.02	0.01

Table 4.6. The response of cowpea seed mineral content to Zn application rate at Bien Donne'.

LSD_{0.05}= Least significant difference; L1= Cowpea Veg1; L2= M217; L3= Qukawa; T1= Zn at 0%; T2= Zn at 50% and T3= Zn at 100% Means with different letter within the column shows significant difference (P< 0.05).

The results of seed mineral analysis showed a non-significant (P>0.05) difference in the mean content of nitrogen, potassium, sodium and magnesium (Table 4.7). However, a significant difference in phosphorus, sodium, iron, zinc and calcium as affected by planting date was observed. Seeds from the first planting date (2 October 2015) was high in the mineral content, this content decreased on seed from the first planting date.

In summary, the highest nitrogen percentage was obtained in cowpea line 1 and 3 at Zn level 2. Cowpea line 1 seed mineral content was high in sodium, iron, magnesium and calcium. Generally, zinc content was high in cowpea line 3. Seeds from the first planting date revealed a high mineral composition.

	Minerals								
Planting date	Percentage (%)			mg/kg					
	Ν	Ρ	К	Na	Fe	Zn	Са	Mg	
2-Oct-15	3.92 ^a	0.53 ^a	1.52 ^a	118.11 ^a	88.06 ^a	39.56 ^a	0.13 ^a	0.19 ^a	
2-Nov-15	3.98 ^a	0.51 ^b	1.50 ^a	96.17 ^a	74.39 ^b	36.83 ^b	0.12 ^b	0.19 ^a	
LSD _{0.05}	0.15	0.02	0.04	28.19	7.35	2.28	0.01	0.01	

LSD_{0.05}= Least significant difference; 02-Oct-15= 02 October 2015, 02-Nov-15= 02 November 2015. Means with different letter within the column shows significant difference (P < 0.05).

4.2 Nietvoorbij

4.2.1 Vegetative parameters

4.2.1.1 Seed Germination

Germination at Nietvoorbij differed significantly (P<0.05). Cowpea line 1 and line 3 obtained the highest germinated plants with means of 81% and 79% respectively. Line 2 with a mean of 53% obtained lowest germination rate and was significantly different from the two above lines. The second planting date improved all cowpea lines germination capacity and a significant increase in germination by 75% was observed in line 2 when planted in November (Table 4.8).

Cowpea line	02-Oct-15	02-Nov-15
1	81 ^a	97 ^a
2	53 ^b	93 ^a
3	79 ^a	95ª
LSD _{0.05}	14.6	15.85

LSD_{0.05}= Least significant difference; cowpea line 1= Cowpea Veg1, 2= M217, 3= Qukawa; 02-Oct-15= 02 October 2015, 02-Nov-15= 02 November 2015. Means with different letter within the column shows significant difference (P< 0.05).

4.2.1.2 Number of leaves per plant

There was no significant difference in the number of leaves per plant (P>0.05). But a similar trend to that of Bien Donne' was observed with cowpea line 1 and 2, where Zn level 2 increased the mean number of leaves per plant (Table 4.9). Planting date had a significant difference in the number of leaves per plant. Generally planting cowpea

in November in this location increased the number of leaves for all treatments by 201% (Table 4.10).

4.2.1.3 Number primary of branches per plant

At Nietvoorbij the number of branches per plant did not differ significantly. Planting date had a significant effect on the number of branches per plant. The highest number of branches were obtained on the second planting date as shown in Table 4.10.

4.2.1.4 Plant height (cm)

The results of the study indicate there was a significant difference in the measured plant height in Nietvoorbij. Cowpea line 1 and 3 obtained higher plant height with no significant difference between the two cowpea lines and line 2 was significantly different with shorter plants regardless of Zn level. Planting cowpea in November significantly increase plant height with a mean on 17 cm (Table 4.10).

Cowpea line	Zn level	No. of leaves	No. of branches	Plant height (cm)
1	1	59.75 ^a	5.17 ^a	15.60 ^a
1	2	63.26 ^a	4.93 ^a	14.60 ^a
1	3	48.49 ^a	5.06 ^a	14.24 ^a
2	1	45.39 ^a	3.08 ^a	5.26 ^b
2	2	58.38 ^a	5.36 ^a	5.49 ^b
2	3	60.50 ^a	4.53 ^a	6.25 ^b
3	1	56.09 ^a	5.03 ^a	15.75 ^a
3	2	56.40 ^a	5.30 ^a	14.37 ^a
3	3	48.32 ^a	3.87 ^a	15.16 ^a
LSD _{0.05}		25.39	1.64	5.3

Table 4.9. Effect of cowpea lines and Zn application rate on vegetative parameters.

 $LSD_{0.05}$ = Least significant difference; L1= Cowpea Veg1; L2= M217; L3= Qukawa; T1= Zn at 0%; T2= Zn at 50% and T3= Zn at 100%. Means with different letter within the column shows significant difference (P<0.05).

Table 4.10. The effect of	planting date on vegetative	parameters at Nietvoorbij.

Planting date	No. of leaves	No. of branches	Plant height (cm)
2-Oct-15	27.12 ^b	3.58 ^b	6.82 ^b
2-Nov-15	81.71 ^a	5.94 ^a	17 ^a
LSD _{0.05}	13.37	1.27	2.28

LSD_{0.05}= Least significant difference; No. = number; 02-Oct-15= 02 October 2015, 02-Nov-15= 02 November 2015. Means with different letter within the column shows significant difference (P<0.05).

In summary, a similar trend to the effect of planting date and cowpea line of germination to that of Bien Donne' was observed in Nietvoorbij. Cowpea line 1 and 3 obtained the highest plant height. The second planting date generally increased all the measured vegetative parameters at Nietvoorbij across all cowpea lines.

4.2.2 Reproductive parameters

4.2.2.1 Number of pods per plant

The number of pods per plant was significantly influenced by the different cowpea lines. Cowpea line 1 at Zn level 1 obtained the highest mean number of pods per plant and was significantly different to cowpea line 2 and 3. Cowpea line 2 obtained the highest mean number of pods per plant at Zn level 2 but was not significantly different to Zn level 1 and 3. Similar results were obtained for cowpea line 3. Planting date had a significant effect on the number of leaves per plant. The first planting date in Nietvoorbij improved the number of pods (Table 4.12).

4.2.2. Total harvested pods

Cowpea line 1 at Zn level 3 obtained the highest number of harvested pods and was significantly different to cowpea line 2 and 3 across all treatments. Planting date did not significantly affect total harvested pods per treatment.

4.2.2.3 Pod weight (g)

Total pod weight differed significantly across treatments. Highest pod weight was obtained in cowpea line 1 Zn level 2 with a mean of ranging from 1074.2- 1372.5 g and line 2 had lighter pod weight with a mean ranging from 121.4 – 180.2 g (Table 4.10). Even a non-significant different was observed, a trend with line 1 and 3 was observed on the response on Zn application rate. Zn level of 2 increased (28% and 2%) pod weight for the two above cowpea lines and a further increment of Zn level 3 reduced pod weight (5% and 22%) respectively. Planting date did not have a significant effect on pod weight.

4.2.2.4 Number of seeds per pod

Cowpea line 1 and 3 obtained the highest number of seed per pod across all Zn levels and was significantly different to cowpea line 2. Cowpea line 2 at Zn level 2 obtained the highest number seed per pod and was significantly different to Zn level 1 with the lowest number of seed per pod. Pods for the second planting date (2 November) had the highest number of seed per pods than pods from the first planting date (2 October) as shown in Table 4.12.

4.2.2.5 Pod length (cm)

Longer pods were obtained in cowpea line 1 and 3 however, a non-significant difference in Zn level was observed across these two lines. Cowpea line 2 had shorter pods at Zn level 1 and 3 and was significantly different to Zn level 2 with longer pods. Pod length in Nietvoorbij was affected by planting date, pods from second planting date (2 November) obtained longer pods than those from the first planting date (2 October).

4.2.2.6 100-seed weight (g)

Treatment had a significant difference in 100-seed weight (Table 4.11). Line 1 and 3 generally obtained heavier seed weight, but was not significantly different from the two line, while cowpea line 2 obtained less 100-seed weight. Cowpea seeds harvested from the second planting date (2 November) was significantly different with heavier seeds from those harvested from first planting date (2 October).

4.2.2.7 Moisture content (%)

Treatment had a significant effect on seed moisture content, cowpea line 1 had highest seed moisture and line 2 obtained lowest seed moisture (Table 4.11). Seeds from the first planting date (2 October) in Nietvoorbij had highest moisture content compared to those from the second planting date (2 November).

Cowpea line	Zn level	No. of pods/plant	Total harvested pods	Pod weight (g)	No. of seed/pod	Pod length (cm)	100 seed weight (g)	Moisture content (%)
1	1	38.31 ^a	542.20 ^{ab}	1074.2 ^{ab}	13.78 ^a	16.14 ^a	12.22 ^{abcd}	11.51 ^{abc}
1	2	25.47 ^{abc}	640.57 ^a	1372.5 ^a	13.27 ^a	15.78 ^a	12.11 ^{abcd}	12.22 ^a
1	3	33.15 ^{ab}	647 ^a	1296.7 ^a	13.95 ^a	16.02 ^a	12.5 ^{abc}	11.97 ^{ab}
2	1	12.01 ^d	121.50 ^d	121.4 ^c	5.23 ^c	8.24 ^c	9.1 ^d	10.31 ^{de}
2	2	17.27 ^{cd}	121.67 ^d	127.3 ^c	7.87 ^b	11.58 ^b	10.6 ^{bdc}	10.31 ^{de}
2	3	12.29 ^d	180.38 ^{cd}	180.2 ^c	6.25 ^{bc}	9.26 ^c	9.4 ^{cd}	10 ^e
3	1	17.25 ^{cd}	417.50 ^b	1006.2 ^{ab}	12.74 ^a	17.23 ^a	14.1 ^a	11.16 ^{bcd}
3	2	21.13 ^{bcd}	431.80 ^b	1029.5 ^{ab}	13.65 ^a	17.79 ^a	14.22 ^a	11.63 ^{abc}
3	3	14.39 ^{cd}	356.20 ^{bc}	838.1 ^b	12.24 ^a	16.87 ^a	12.9 ^{ab}	11 ^{cd}
LSD _{0.05}		12.85	195.61	421.79	1.75	2.07	3.3	0.96

Table 4.11. Influence of Zn application rate and cowpea lines on reproductive parameters at Nietvoorbij.

LSD_{0.05}= Least significant difference, L1= Cowpea Veg1; L2= M217; L3= Qukawa; Zn level 1= Zn at 0%; 2= Zn at 50% and 3= Zn at 100%

Means with different letter within the column shows significant difference (P< 0.05).

Planting date	No. of pods/plant	Total harvested pods	Pod weight (g)	No. of seed/pod	Pod length (cm)	100 seed weight (g)	Moisture content (%)
2-Oct-15	27.61 ^a	405.64 ^a	854.90 ^a	10.56 ^b	13.71 ^b	10.74 ^b	11.54 ^a
2-Nov-15	15.52 ^b	374.91 ^a	763.16 ^a	11.75 ^a	15.09 ^a	12.98 ^a	10.85 ^b
LSD _{0.05}	5.55	63.67	179.37	0.83	0.9	1.37	0.52

Table 4.12. The effect of planting date on reproductive parameters at Nietvoorbij.

 $LSD_{0.05}$ = Least Significant Different, No. = Number; 02-Oct-15= 02 October 2015, 02-Nov-15= 02 November 2015. Means with different letter within the column shows significant difference (P< 0.05).

In general, at Nietvoorbij cowpea line 1 was the best performing line with yield parameters such as number of pods per plant, total harvested pods and pod weight being the highest from the other two lines. Line 3 obtained longer pods and was significantly different to line 2 with shorter pods. The first planting date at Nietvorbij significantly increased the number of pods per plant for all treatments. The second planting date increased the number of seed per pod, pod weight and 100-seed weight for all lines.

4.2.2.8 Seed mineral composition

There was no significant (P>0.05) difference in the nitrogen content of the dried seed across all the different cowpea lines and Zn levels. Phosphorus, potassium, sodium, iron, zinc, calcium and magnesium was significantly (P<0.05) affected by the three cowpea lines. Phosphorus content differed significantly in the studied cowpea lines. A similar trend in response to Zn level was observed in line 1 and 3 for phosphorus content. Line 2 contained the highest percentage of phosphorus at Zn level 3 with a mean of 0.55 % (Table 4.13).

Potassium percentage was higher in line 1 at Zn level 2 and 3, this percentage was significantly different to line 3 across all Zn levels with the lowest percentage.

The highest sodium content of 356.75 mg/kg was obtained in line 1 at Zn level 2 and was significantly (P>0.05) different to line 3 with lower content across all Zn levels.

Seed mineral content analysis results (Table 4.13) clearly show that iron content differed in the three cowpea lines. The highest iron content was obtained in cowpea line 2, Zn level 3 and was significantly different Zn level 2 with lower iron content. There was no significant difference in iron content for line 1 and 3 obtaining the lowest iron content.

Cowpea line 1 obtained the highest Zn content at Zn level1 and was significantly different to all treatments. Application of Zn fertilizer for line cowpea line 1 suppressed available Zn content in dried seed (Table 4.13) as application-level increases Zn content in the seed decreased. Line 2 obtained the lowest Zn mineral content at Zn level 2 but was not significant from the other two Zn levels.

Line 2 at Zn level 2 obtained the highest calcium content but was not significantly different to Zn level 1 and 3. A significant lower calcium content was observed for line 1 and 3 across all Zn levels.

Cowpea line 1 had a significant higher magnesium content of 0.20 mg/kg and it significantly differed from line 2 and 3 with lower magnesium content.

					Minerals				
Cowpea line	Zn level	Percentage (%)							
		Ν	Р	K	Na	Fe	Zn	Ca	Mg
1	1	3.77 ^a	0.47 ^b	1.55 ^{ab}	321.5 ^{abc}	75.50 ^c	62.00 ^a	0.14 ^c	-
1	2	3.77 ^a	0.51 ^{ab}	1.59 ^a	356.75 ^a	75.50 ^c	57.50 ^b	-	0.20 ^a
1	3	3.88 ^a	0.49 ^{ab}	1.59 ^a	346.75 ^{ab}	73.00 ^c	54.50 ^{bc}	0.13 ^c	0.20 ^a
2	1	3.81 ^a	0.51 ^{ab}	1.53 ^{abc}	277.50 ^{abcd}	92.00 ^{ab}	50.00 ^{de}	0.17 ^{ab}	0.18 ^b
2	2	3.86 ^a	0.52 ^a	1.55 ^{ab}	287.50 ^{abc}	83.50 ^{bc}	49.25 ^e	0.19 ^a	0.19 ^b
2	3	3.79 ^a	0.55 ^a	1.54 ^{ab}	285.25 ^{abc}	97.75 ^a	51.25 ^{cde}	0.18 ^a	0.19 ^b
3	1	3.94 ^a	0.48 ^b	1.45 ^c	210.25 ^{cd}	77.00 ^c	53.25 ^{cd}	0.14 ^c	0.19 ^b
3	2	3.97 ^a	0.49 ^{ab}	1.45 ^c	159.00 ^d	76.00 ^c	54.00 ^{bc}	0.13 ^c	0.18 ^b
3	3	3.82 ^a	0.51 ^{ab}	1.48 ^{bc}	229.00 ^{bcd}	77.50 ^c	54.25 ^{bc}	0.15 ^{bc}	0.19 ^b
LSD _{0.05}		0.39	0.07	0.09	120.77	13.45	3.76	0.03	0.01

Table 4.13. The response of cowpea seed mineral content to Zn application rate at Nie	tvoorbij.
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LSD_{0.05}= Least significant difference, L1= Cowpea Veg1; L2= M217; L3= Qukawa; T1= 0% Zn; T2= Zn at 50% and T3= Zn at 100% Means with different letter within the column shows significant difference (P< 0.05).

At Nietvoorbij, planting date had a significant (P<0.05) effect on phosphorus (%) and sodium (mg/kg). The first planting date improved the seed mineral content of the above minerals across all treatments. No significant difference was observed for the following minerals: nitrogen, potassium, iron, zinc, calcium and magnesium (Table 4.14).

Diamitima	Minerals										
Planting date	Per	centage	(%)	mg/kg							
	Ν	Р	к	Na	Fe	Zn	Ca	Mg			
2-Oct-15	3.85 ^a	0.52 ^a	1.54 ^a	286.56 ^a	80.67 ^a	53 ^a	0.15 ^a	0.19 ^a			
2-Nov-15	3.84 ^a	0.50 ^b	1.51 ^a	263.11 ^b	81.06 ^a	53.53 ^a	0.15 ^a	0.19 ^a			
LSD _{0.05}	0.2	0.01	0.05	13.89	7.49	2.8	0.01	0.003			

Table 4.14. The effect of planting date on seed mineral content at Nietvoorbij.

 $LSD_{0.05}$ = Least significant difference; 02-Oct-15= 02 October 2015, 02-Nov-15= 02 November 2015. Means with different letter within the column shows significant difference (P<0.05).

4.3 Interactions

4.3.1 Vegetative parameters

The highest number of leaves per plant was obtained in 2 November 2015 for cowpea line 1 at Zn level 2 and was significantly different to Zn level 3 with the lower number of leaves per plant (Table 4.15). Application of Zn at level 3 significantly increased the number of leaves per plant or cowpea line 2 planted in 2 November 2015. A non-significant difference response to application of Zn was observed in line 3. The number of leaves in the first planting date for line 1 was significantly lower at Zn level 1 and 2. Cowpea line 2 at Zn level 3 significantly had the lowest number of leaves for the first planting date. Line 3 planting in 2 November 2015 significantly increased the number of across all treatments.

The highest number of branches per plant was obtained in 2 October 2015 in line 2 Zn level 2 and was significantly different to Zn level 3. Line 2 at Zn level 1 planted in 2 November 2015 had the lowest number of branches.

Line 1 and 3 planted in November significantly obtained higher plant height across all treatment compare to cowpea plants height of 2 October 2015. Cowpea line 2 pant height was not affected by planting date (Table 4.15).

Planting Date	Cowpea line	Zn level	No. of leaves	No. of branches	Plant height (cm)
2-Oct-15	1	1	44.19 ^{def}	6.67 ^{ab}	10.99 ^b
2-Oct-15	1	2	43.62 ^{def}	6.27 ^{abc}	9.95 ^{bc}
2-Oct-15	1	3	40.78 ^{def}	5.92 ^{abcd}	9.74 ^{bcd}
2-Oct-15	2	1	43.93 ^{def}	6.03 ^{abcd}	4.22 ^e
2-Oct-15	2	2	52.04 ^{cdef}	7.35 ^a	4.88 ^e
2-Oct-15	2	3	35.77 ^f	4.60 ^{bcd}	3.90 ^e
2-Oct-15	3	1	39.87 ^{ef}	5.98 ^{abcd}	11.09 ^b
2-Oct-15	3	2	37.15 ^f	5.41 ^{abcd}	9.98 ^{bc}
2-Oct-15	3	3	38.35 ^{ef}	5.43 ^{abcd}	10.11 ^b
2-Nov-15	1	1	80.83 ^{ab}	6.10 ^{abcd}	20.13 ^a
2-Nov-15	1	2	87 ^a	5.86 ^{abcd}	18.69 ^a
2-Nov-15	1	3	57.77 ^{cdef}	5.50 ^{abcd}	17.13 ^a
2-Nov-15	2	1	48.36 ^{def}	3.87 ^d	5.93 ^{cde}
2-Nov-15	2	2	60.05 ^{bcde}	4.52 ^{bcd}	5.66 ^{de}
2-Nov-15	2	3	71.65 ^{abc}	4.86 ^{bcd}	7.80 ^{bcde}
2-Nov-15	3	1	72.99 ^{abc}	5.95 ^{abcd}	19.83 ^a
2-Nov-15	3	2	71.43 ^{abc}	6.36 ^{ab}	17.13 ^a
2-Nov-15	3	3	62.33 ^{bcd}	4.04 ^{cd}	19.21 ^a
LSD _{0.05}			22.21	2.25	4.15

Table 4.15. Influence of planting date on vegetative parameters across both locations.

LSD_{0.05}= Least significant difference; Oct= October; Nov= November; L1= Cowpea Veg1; L2= M217; L3= Qukawa; T1= 0% Zn; T2= Zn at 50% and T3= Zn at 100%; 02-Oct-15= 02 October 2015, 02-Nov-15= 02 November 2015. Means with different letter within the column shows significant difference (P< 0.05).

4.3.2 Reproductive parameters

Line 1 at Zn level 1 obtained the highest number of pods per plant for the first planting date and was significantly high to those planted on the second planting date across all treatments. Cowpea line 2 obtained its highest number of pods per plant in 2 October 2015 at Zn level 2 with a mean of 32.43 (Table 4.16). A decline in the number of pods per plant was observed for this line when planted in 2 November 2015 with a mean of 16.37. There was no significant difference in the number of pods per plant for line 3 for the two planting dates across all Zn levels.

Line 1 Zn level 1 and 2 from the first planting date had higher total harvested pods per treatment compared to those planted in 2 November 2015 and the non-significant difference was observed with Zn level 3. Planting date did not have a significant effect on the total number of harvested pods for cowpea line 2 and 3.

Pod weight for line 1 Zn level 1 and 2 decreased on the second planting date, a nonsignificant difference was observed at Zn level 3. Planting date did not significantly affect pod weight of cowpea line 2 and 3 across all treatments.

The number of seed per pod was not significantly affected by planting date for cowpea line 1 and 3 across all treatments. Line 2 at Zn level 1 obtained the lowest number of sees when planted in 2 October 2015.

Planting date did not have a significant effect on pod length for line 1 and 3 across all treatments. Cowpea line 2 Zn level 1 and 3 from the first planting date had shorter pods.

A non-significant difference was observed for line 1 and 3 across all treatments in 100-seed weight. Second planting date significantly increased 100-seed weight for line 2 across all treatments.

Cowpea seed from the first planting date had less moisture content compared to the seeds from second planting date (Table 4.16).

Planting Date	Cowpea line	Zn level	No. of pod/plant	Total harvested pods	Pod weight (g)	No. of seed/pod	Pod length (cm)	100 seed weight (g)	Moisture content (%)
2-Oct-15	1	1	41.75 ^a	915.30 ^{ab}	1816.6 ^{ab}	15.66 ^a	16.40 ^{bcd}	11.70 ^{defg}	11.38 ^{def}
2-Oct-15	1	2	33.29 ^{abc}	975 ^a	2038.3 ^a	15.22 ^{abc}	16.16 ^{bcd}	12 ^{cdef}	11.70 ^{bcdef}
2-Oct-15	1	3	37.79 ^{ab}	870.20 ^{abc}	1776.6 ^{ab}	15.55 ^{ab}	16.44 ^{bcd}	12.80 ^{bcdef}	11.75 ^{abcde}
2-Oct-15	2	1	23.90 ^{cdefghi}	539.38 ^{fgh}	719.3 ^d	6.43 ^f	8.65 ^f	9.30 ^h	9.97 ^h
2-Oct-15	2	2	32.43 ^{abcd}	514 ^{gh}	688.3 ^d	8.48 ^e	11.61 ^e	10.90 ^{fgh}	10.18 ^{gh}
2-Oct-15	2	3	16.14 ^{hi}	341.57 ⁱ	610.7 ^d	6.71 ^e	9.14 ^f	9.70 ^{gh}	9.95 ^h
2-Oct-15	3	1	22.06 ^{defghi}	613.90 ^{efg}	1497.5 ^{bc}	14.14 ^{bcd}	17.53 ^{ab}	13.60 ^{abcde}	10.92 ^{efg}
2-Oct-15	3	2	25.98 ^{cdefgh}	649.70 ^{defg}	1626 ^{bc}	14.72 ^{abcd}	17.92 ^a	13.11 ^{abcde}	11.56 ^{cdef}
2-Oct-15	3	3	20.12 ^{efghi}	617.89 ^{efg}	1794.5 ^{ab}	13.43 ^d	16.96 ^{abcd}	12.30 ^{bcdef}	11.37 ^{def}
2-Nov-15	1	1	26.57 ^{cdefgh}	673 ^{def}	1344 ^c	14.64 ^{abcd}	16.08 ^{cd}	11.88 ^{cdef}	12.30 ^{abc}
2-Nov-15	1	2	27.57 ^{bcde}	734.38 ^{cde}	1622.3 ^{bc}	14.95 ^{abc}	16.10 ^{cd}	11.6 ^{efg}	12.63 ^a
2-Nov-15	1	3	26.96 ^{cdefg}	772.30 ^{bcd}	1539.4 ^{bc}	14.15 ^{bcd}	15.75 ^d	11.9 ^{cdef}	12.56 ^{ab}
2-Nov-15	2	1	15.29 ⁱ	442.44 ^{hi}	543.6 ^d	8.91 ^e	11.91 ^e	13.8 ^{abcd}	11.29 ^{def}
2-Nov-15	2	2	16.37 ^{ghi}	427.80 ^{hi}	599.8 ^d	9.04 ^e	11.75 ^e	13.5 ^{abcde}	11.58 ^{cdef}
2-Nov-15	2	3	27.20 ^{bcdef}	404.75 ^{hi}	586.3 ^d	8.91 ^e	11.63 ^e	13.1 ^{abcde}	10.80 ^{fgh}

 Table 4.16. Influence of planting date on reproductive parameters at Nietvoorbj and Bien Donne'.

2-Nov-15	3	1	14.82 ⁱ	657.90 ^{defg}	1666.2 ^{abc}	14.09 ^{bcd}	17.50 ^{ab}	14.2 ^{ab}	12.34 ^{abc}
2-Nov-15	3	2	16.75 ^{fghi}	651.70 ^{defg}	1633.1 ^{abc}	13.85 ^{cd}	17.42 ^{abc}	15 ^a	12.07 ^{abcd}
2-Nov-15	3	3	14.43 ⁱ	604.22 ^{efg}	1430 ^{bc}	14.47 ^{abcd}	17.48 ^{ab}	14 ^{abc}	11.98 ^{abcd}
LSD _{0.05}			10.61	144.1	410.74	1.48	1.37	2.11	0.91

LSD_{0.05} = Least significant difference; Oct= October; Nov= November; L1= Cowpea Veg1; L2= M217; L3= Qukawa; T1= 0% Zn; T2= Zn at 50% and T3= Zn at 100%; 02-Oct-15= 02 October 2015, 02-Nov-15= 02 November 2015. Means with different letter within the column shows significant difference (P< 0.05).

CHAPTER 5

DISCUSSION

5.1 Effect of cowpea lines and zinc application levels at the two locations.

The germination percentage of cowpea was affected by the different cowpea lines. The different levels of zinc fertilizer did not have effect on germination as it was applied at initial flowering. Cowpea Veg1 and Qukawa had the highest mean germination rate, while line M217 had the lowest number of germinated plants per experimental unit (EU). The obtained results were similar to those reported by Wada and Abubakar (2013), who did a germination test on different cowpea lines and concluded that seed size and viability of seed are the factors that affect germination on different cowpea lines. The authors further identify imbibition by seed as a factor that can possibly delay emergence which is affected by the permeability of the seed coat. Germination rate was significantly affected by soil type; in sandy loam soil germination was more efficient than in sandy clay loam soils due to the different soil textural percentages (Table 3.1) of the two locations. Similar results were obtained from a study done by Pahla *et al.*, (2014), which showed a higher percentage of germination and emergence on sandy loam soils.

In this study, the three cowpea lines varied in their morphological characters as affected by the two planting date and the two locations. The qualitative and quantitative trait of the cowpea line studied varied (Table 3.5), the traits outlines the morphology of the three cowpea lines studied. Morphological traits of cowpea lines and the importance of these traits are well documented by Egbadzor *et al.*, (2014); Pasquet (1998). Significant difference at 5% probability level was observed in the number of branches (Bien Donne') and plant height for both locations. A non-significant difference in the number of leaves per plants for all lines was observed

across the two locations. Comparable findings were documented by Olatunji *et al.*, (2016). Contrary to this study, Ekpo *et al.*, (2012), reported a significant difference in number of leaves on eight cowpea lines in a period of six weeks.

There was a significant difference in the number of branches per plant in Bien Donne'. Cowpea Veg1 at Zn application level of 15 kg/ha obtained the highest mean number of branches. Variation in the number of branches was only observed between the lines. This is in agreement with findings from Ayan et al., (2012); Gerreno et al., (2015); Agbogidi and Egho (2012), these authors state that vegetative growth of cowpea is affected by its genotypic makeup, season and location. According to Egbadzor et al., (2014), plants that are classified as indeterminate are most vigorous. Cowpea Veg1 under study had indeterminate or spreading type plant pattern which concurs with above statement. In both locations Cowpea Veg1 and Qukawa obtained highest plant height and line M217 obtained shorter plants. This could be associated with the genetic makeup of each lines, as the morphological trait of the two lines showed a similar growth pattern. On a study by Rathore et al., (2015), results showed that application rate 10 kg/ha of Zn fertilizer significantly increased plant height on all cowpea lines under study. Line M217 morphologically has a determinate growth pattern. These results concur with the study reported by Shiringani (2007) on the same line.

The two different farms had different soil texture, analysis by farm showed that Nietvoorbij which have clay loam soils had the highest number of branches and plants obtained higher plant height compared to those of Bien Donne'. The results regarding the number of branches are in agreement with the result obtained by Shiringani (2007).

Results showed that application of fertilizer did not have an effect on the growth rate of the number of leaves and number of branches after the application of Zn fertilizer (Appendix G), but a significant increase in plant height was observed for all treatments. The time of application could be the reason for no growth as it was applied 8 weeks after germination, which meant that the plant had already completed its vegetative growth and only reproductive growth was active. These findings are contrary to the results by Elowad and Hall, (1987), who concluded that early flowering soil application of fertilizer increases the number of branches and pods of cowpea lines. Lewu *et al.*, (2017), stated that vegetative growth parameters increase to its maximum in early stages and it decrease or ceases at maturity. At Nietvoorbij the growth rate of the number of branches and plant height increased after Zn was applied. Application of Zn fertilizer increased plant height growth rate for all treatment, plant height growth increased from a rate of 1.95 to 2.62. In Bien Donne' the application of zinc did not affect plant height growth rate (Appendix G).



Figure 5.1 Vegetative growth of the three cowpea lines in Bien Donne' at 8 weeks after planting for the first planting date.



Figure 5.2 Vegetative growth of the three cowpea lines in Nietvoorbj at 8 weeks after planting for the first planting date.

5.2 Effect of planting date on cowpea lines

The results of the study indicate that line Cowpea Veg1 and Qukawa can successfully be sown early (2 October) or late (2 November) in the Western Cape region. Furthermore, line M217 will successfully germinate to its highest capacity if sown later (2 November) in the growing season when temperatures are between 10 and 30°C (Table 3.2). A progressive increase in maximum temperatures during the growing season from November 2015 till March 2016 was observed. Planting date had a significant effect on germination rate and it affected days to physiological maturity, as plants sown in October took more days to mature. Observation from the study generally indicates that at Bien Donne' plants sown in October took 116 days to first harvest (DFH) and 93 DFH for plants sown in November while at Nietvoorbij plants sown in October took 131 DFH and 121 DFH for plants sown in November. These results are in agreement with a study done by Akande et al., (2012), who found that planting date affects cowpea period to attain maturity. Dugje et al., (2009), classified cowpea into three categories that matures in 60 days as extra early, 61-80 as early and above 80 days as late. Aligned with these classifications all the lines from the study were late maturing lines. This could have been influenced by the environmental conditions of the two locations. Inconsistent on germination was observed on first planting date which could have been due to low temperatures especially on sandy clay loam at Nietvoorbij. The findings from this study are in agreement with the results reported by Ngalamu et al., (2013), on the importance of sowing date. The interaction between planting date and cowpea lines was significant. The second planting date improved germination for all cowpea lines (Appendix C).

Cowpea Veg1 was identified as the best performing line across the two locations and planting dates. Different planting date showed variation in vegetative growth, plants sown on the second planting date obtained a higher number of leaves, higher plant height but obtained less number of primary branches. The variation in different cowpea lines in response to planting date are well documented by Akande *et al.*, (2012). Vegetative growth of the three cowpea lines is presented in Figure 5.1 and 5.2, where it can be clearly seen that plants at Nietvoorbij struggled when planted in October because of delayed or no emergence compared to plants at Bien Donne'. The pictures illustrated were taken eight weeks after planting for both locations. At Bien Donne' the first planting date produced plants with the highest average number of branches per plant and opposite results were obtained at Nietvoorbij across all cowpea lines. Environmental conditions including the soil texture could have possibly have been the cause of retarded growth. The first planting date showed positive results on the overall number of branches. A strong interaction (P<0.001) between planting date x treatment and planting date x farm (Appendix F).

The results of the study showed that planting date greatly influenced plant height of all cowpea lines. The second planting date enhanced plant height across the two locations. This can be associated with the environmental conditions during the growing season which enhances plant vigour. These results are in agreement with findings in a study done by Ngalamu *et al.*, (2013), on soybeans. Plants from the first planting date showed an increase in plant height growth rate but decreased for the second planting date. This can be due to the increase in temperature as the season progresses (Table 3.2). Analysis by planting date showed an increase in the number of leaves growth rate on first planting date and there was no significant effect on the other vegetative parameters. The overall influence of planting date on vegetative parameters showed a significant (P<0.05) difference (Table 4.13). The second planting date increased the number of leaves per plant for Cowpea Veg1 and Qukawa across both locations. Similar trend in response to that of number of leaves per plant was observed for plant height. The similar response of these lines in their

vegetative growth can be related to the same growth pattern and the environmental condition which enhanced maximum growth. Line M217 was the least performing line across all planting date. This is due to the growth habit that the line has a dwarf, erect growth habit. The findings regarding line M217 were in agreement to those reported by Shiringani (2007).

In this study, cowpea lines showed remarkable variation across the two planting dates and locations in seed yield and yield parameters. The first planting date significantly increased the total number of harvested pods and pod weight at Bien Donne' across all lines. Whereas, at Nietvoorbij the second planting date favoured yield parameters such as the number of seed/pod, pod length and 100-seed weight across all lines. Planting cowpea in 2 October at Bien Donne' increased the total number of pods harvested per treatment and therefore increasing the weight for all cowpea lines, those parameters greatly influence yield and yield components This variation in yield parameters could be environmental conditions such as temperature and photoperiod which enhances photosynthesis. These findings coincide with the results obtained by Shiringani (2007).

Planting date greatly influenced the reproductive parameters across the two locations. Generally, the first planting date increased the number of pods per plant, number of seeds per pod, total harvested pods and seed yield. A significant increase in seed yield was observed line Cowpea Veg1 at Zn application rate of 15 kg/ha. Seed yield was not affected by planting date for line M217 and Qukawa across locations (Appendix E). Generally, yield and yield components were higher on the first planting date for all treatments.

Seeds from the first planting date in Bien Donne' had highest content of phosphorus, sodium, iron, zinc and calcium. The variation due to planting date can be associated

genotypic makeup, climatic and environmental conditions that enhances effective absorption and translocation of nutrients to the sink. The soil available minerals content could have been another factor contributing to seed mineral content in both locations.

5.3 The effect of cowpea lines and zinc application rate on yield parameters

The results of the study showed that reproductive parameters were significantly affected by the three cowpea lines. This was due to the genetic variation between the different lines, soil texture and climatic conditions that the plants were subjected to due to the different locations. Line Cowpea Veg1 and Qukawa were recognised as the best performing lines which can be cultivated on the two locations. M217 was observed as a least performing line but the observation from the study indicated that it was the first line to flower and bear pods. These observations were supported by Khan et al., (2010), they concluded that dwarf cowpea genotype tends to mature early than taller plants. The study further states that dwarf plant tends to produce a low yield. These findings concur with the performance of line M217 which consistently gave lower yield across all locations in the current study. Parameters such as the number of pods per plant and number of seed per pod contributed to the total yield harvested of the two lines. The mean number of pods in this study ranged from 14.39 -32.72/plant. Similar results on 12 genotypes were reported by Peksen and Peksen, (2012). Generally, Cowpea Veg1 was the best performing line across all cowpea lines and M217 was the least. Shiringani (2007), obtained contrary results on the performance of cowpea line M217 which obtained the higher number of pods per plant in two locations studied. At Nietvoorbij the highest number of pods per plant was obtained in Cowpea Veg1 and a non-significant different was observed in Zn application levels. This response to Zn application rate was observed in most of the parameters measured. The inconsistency response of all lines to Zn application can be associated with the time of application and genetic makeup of each line and its ability to absorb and translocate the nutrients to the sink (Naim et al., 2012). Rathore et al., (2015), stated that the ability of a plant to absorb available zinc in the soil is

also influenced by other plant nutrients. These nutrients can promote or hinder the plant ability absorb and translocate Zn to all parts of the plants.

Pod quality for each location is shown in Figure 5.3, different lines differed in pod size, weight and size of the seed. Generally, Qukawa (at Zn 0 kg/ha at Bien Donne' and 30 kg/ha Zn at Nietvoorbij) had longer pods with small seed size and opposite results were obtained for line M217. Cowpea line M217 had the shortest pods across both locations. On a survey done in Ghana consumers prefer white/cream seed coloured varieties as they are identified as sweet and the period of cooking is less (Egbadzor *et al.*, 2013). In the current study line M217 has cream seed colour (Figure 5.4).

The variation in pod length was observed in the three cowpea lines studied. Similar results were reported by Bhattarai *et al.*, (2017), the authors studied six lines under rain-fed conditions and reported significant variation in pod length between the lines. Pod length in the current study was significantly (P<0.05) affected by the cowpea lines. Line Qukawa obtained the highest pod length with a mean ranging from 16.87 - 17.81 cm across both locations. Line M217 had shorter pods. Pod length is an important morphological traits for breeders and farmers as it indicate the potential seed set (Ekpo *et al.*, 2012). According to Naim *et al.*, (2012), different cowpea line genetic make-up has an influence on the number of seeds per pod. These contradicting results can be due to climatic conditions and seed quality. As the study was done under different climatic conditions Western Cape Province which has winter rainfall and the Limpopo Province with the summer rainfall.

Seed yield was significantly affected by the different cowpea lines across the two locations. Yield at Bien Donne' ranged from 486.9-1184.2 kg/ha and at Nietvoorbij it ranged from 60.7-686.25 kg/ha (Appendix D). Line Qukawa obtained the highest

seed yield (1184.2 kg/ha) and was not significantly different to Cowpea Veg1 at Bien Donne' across all Zn application levels. At Nietvoorbij the highest seed yield was obtained from Cowpea Veg1 at an application of 15 kg/ha Zn (686.25 kg/ha) but these results were not significantly different to the control and Zn application rate of 30 kg/ha (537.1 and 648.35 kg/ha). An increase, though not significant in seed yield was observed with Zn fertilizer application to 15 kg/ha for all lines. A further increment of Zn application rate to 30 kg/ha decreased the yield with an exception for line Qukawa. Zinc application rate and its effect on yield and yield components are well documented by Moswatsi, (2015) and Malakouti, (2007). The author further concluded that zinc application rate of 5 kg/ha increased yield and an increment of the application had a negative effect on seed yield. Low seed yield was recorded for line M217 in both locations.

The 100-seed weight was significantly (P<0.05) affected by the cowpea lines. According to Aliyu and Makinde, (2016); Addo-Quaye *et al.*, (2011), the 100-seed weight is used as a seed size indicator. Fascinating findings from the study showed that line M217 in Bien Donne' obtained the highest 100-seed weight (14 g/100 seed). These results were significantly higher than line Cowpea Veg1 and this line was regarded as the best performing line. The findings clearly indicated that line M217 have bigger seed size even though it lacks most of the parameters measured. Bigger seed size are mostly prefers for home consumption (Doumbia *et al.*, 2013). Aliyu and Makinde, (2016), concluded that seed size related to the number of days to flowering and pod formation period. These findings agree with the observation of the current study, as line M217 was the first line to flower and bear pods. Khan *et al.*, (2010), found highly significant difference 100-seed weight of 24 cowpea genotypes.

Seed moisture percentage determines storage duration of the grain which also serve as an indication in minimizing pest infestation. According to DAFF, (2013), moisture

percentage of 12% is best suited for short-term storage and 8-9% long-term storage. Results of the study indicate that line M217 can be possibly stored for a longer period as the moisture content of this line ranged within a range of long-term storage. The other two lines high moisture content could have been attributed to an early change of pod colour which does not necessary mean seeds were dry. This can be associated with the environmental condition and storage at harvest.

In this study, cowpea Veg1 best performed where no zinc fertilizer was applied on most of the reproductive parameters. According to Ehlers (1994), cowpea line that performs best under limited inputs can be recommended into an intercropping system since it performs well as a sole crop due to its performance. This line has intermediate growth habit, dark green leaf colour (enhancing photosynthesis), a high number of pods and number of seed per pod, therefore, contributing to high yields (Table 3.5).

Analysis by planting date shows that reproductive parameters from the first planting date (2 October) had the highest mean number of the following parameters: number of pods per plant, the total number of harvested pods and pod weight. At Bien Donne' heavier pods were obtained than pods at Nietvoorbij, which is the result of the total number of pods harvested and the first planting date performed better. Climatic conditions could be the possible reason for the variation in reproductive parameters. The effect of planting date and location in cowpea lines is well documented by Shiringani, (2007). Planting date had a huge effect on most of the parameters measured for each location (Appendix H). This could be due to the differences in soil chemical properties as and climatic conditions.

In this study inconsistence response across all Zn levels were observed in seed mineral composition, due to genetic variation in different cowpea lines. The nutritional

content of the seed is greatly affected by cowpea line, availability of nutrients in the soil and uptake of those nutrients. According to Cakmak et al., (1999), the capacity of genotype to take up larger amounts of Zn under Zn deficiency is a main plant trait responsibility and it depends on root uptake and translocation. Davis et al., (1991), illustrated nutritional concentration of cowpea at different growing stages to be different. Tshovhote et al., (2003) showed variation in different cowpea lines chemical composition. Application of Zn fertilizer significantly increased iron content in cowpea line M217 in both locations. Harvested seed from Nietvoorbij had higher Zn content; this could be due to the soil available chemical content of the area prior to planting. The addition of Zn responded positively to the low phosphorus content and enhanced the uptake of Zn to the seed at Nietvoorbij. Malakouti (2007) stated that high level of phosphorus disturbs the uptake and translocation of Zn by the plants. Different cowpea lines vary in their mineral content as observed by Ayan et al., (2012). Rengel et al., (1999), stated that concentration of nutrients in seed depends on soil type, nutrient availability and crop species. The author further stated that increasing Zn content in the soil by fertilization increases Zn concentration in the seed.

Appendix I shows the response of seed mineral content as affected by planting date. Treatment varied in their response to planting date but was in a range of all chemical content observed. The location had an effect on chemical content, as seeds from Nietvoorbij obtained higher chemical content for most of the elements measured than those from Bien Donne'.

The overall performance of all reproductive parameters was high at Bien Donne' than Nietvoorbij. This was indicted by the mean number of pods harvest per location.



Figure 5.3 Illustration of pod quality per treatment for the two locations.

G1= Cowpea Veg1, G2= M217, G3=, Qukawa, C1= 0% zinc, C2= 50% zinc, C3= 100% zinc, BD= Bien Donne', NVB= Nietvoorbij



Figure 5.4 Illustration showing the different cowpea seed in the current study.

Cowpea Veg1= grey; M217= cream; Qukawa= brown seed colour.

CHAPTER 6

CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusions

The objective of the study was to determine which planting date will best suit cowpea production and its effect on morphological traits and seed chemical composition. Furthermore, the second objective was to document the effect of different zinc application rate on production at different locations. Results from the study showed that the three cowpea lines varied in their morphological trait. This variation greatly affected the performance of the lines under the two different soil conditions. Cowpea Veg1 and Qukawa were overall the best performing lines for both planting date with regards to vegetative and yield components. The results show that cowpea lines evaluated in Bien Donne' performed better than Nietvoorbij. Cowpea Veg1 obtained highest seed yield of 1110.75 kg/ha at Zn application rate of 15 kg/ha and 686.25 kg/ha at 15 kg/ha Zn at Nietvoorbij. Qukawa obtained its highest seed yield of 1184.2 kg/ha at Zn application rate of 30 kg/ha at Bien Donne' and 514.75 kg/ha at Zn application rate of 15 kg/ha. The environmental and soil conditions in Bien Donne' are conducive for the production of cowpea. Planting of cowpea in November improved germination, thereby, improving the overall vegetative performance of the crop. Reproductive parameters measured were significantly high from plants that were planted in October. The first planting date also improved the seed mineral contents of the cowpea lines and was a significant finding of this study across the two locations. Application of Zn fertilizer at flowering gave a substantial yield resulting in yield component increase. Zinc fertilizer significantly improved plant height and did not have an effect on the number of leaves and number of branches. An inconsistent response to zinc fertilizer for all measured parameters was observed. This inconsistency was affected by many factors such as the genotypic makeup of the

lines, environmental conditions and time of application. The application rate of 15 kg/ha significantly increases iron content in Bien Donne'. Analysis of the comparative mineral content of the three cowpea lines tested in the study suggests that Cowpea Veg1 generally recorded the highest level of nutrient concentration.

6.2 Recommendations

The results of the study showed variation in cowpea morphological traits, which differs from other areas. It is therefore recommended that more research on different cowpea lines be conducted in the area to validate the suitability and response of different lines to the area. Cowpea Veg1 and Qukawa are best lines which can be suitable for dual-purpose farming, planted as a sole crop and for intercropping. These cowpea lines are tolerant to low temperatures but planting cowpea in November was ideal for all lines. Planting cowpea in October best suit seed/grain production and planting in November increases the vegetative parameters therefore suitable for fodder production. Environmental conditions are a great influence on the performance of different cowpea lines, further in details regarding this factor need to be looked at. Zinc fertilizer time of application still needs further study in this province, as the inconsistent response to all treatments, was observed as this can be due to the soil properties and environmental conditions.

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APPENDICES

Appendix A: Design and layout of experiment for both locations.

Block 1		Block 2	Block	(3	Block 4	Block 5
2ndNov_G1_C1		2ndNov_G2_C1	2ndNov_(G1_C1	2ndNov_G2_C1	2ndNov_G1_C2
2ndNov_G2_C3		2ndNov_G3_C3	2ndOct_0	G3_C3	2ndOct_G1_C1	2ndOct_G3_C3
2ndNov_G2_C2		2ndNov_G2_C2	2ndOct_0	G1_C2	2ndNov_G1_C2	2ndNov_G1_C1
2ndOct_G2_C3	In	2ndOct_G2_C3	2ndOct_0	G1_C3	2ndNov_G3_C2	2ndOct_G2_C2
2ndNov_G1_C2	te	2ndOct_G3_C2	2ndOct_0	G1_C1	2ndNov_G3_C1	2ndNov_G2_C2
2ndNov_G2_C1	Interrow	2ndNov_G2_C3	2ndOct_0	62_C2	2ndOct_G3_C3	2ndNov_G3_C2
2ndOct_G1_C2	ž	2ndNov_G3_C1	2ndNov_(G3_C2	2ndNov_G1_C3	2ndNov_G2_C1
2ndOct_G1_C1	Sp	2ndNov_G1_C2	2ndOct_0	G2_C3	2ndOct_G2_C2	2ndNov_G1_C3
2ndNov_G3_C3	Spacin	2ndOct_G1_C2	2ndNov_(G2_C3	2ndOct_G1_C2	2ndOct_G3_C1
2ndNov_G1_C3	cin	2ndOct_G3_C1	2ndNov_(G1_C3	2ndOct_G1_C3	2ndNov_G3_C1
2ndNov_G3_C1	9 0	2ndNov_G3_C2	2ndOct_0	G3_C2	2ndOct_G3_C1	2ndOct_G1_C1
2ndOct_L2_C1		2ndOct_G1_C1	2ndOct_0	G3_C1	2ndNov_G3_C3	2ndOct_G1_C2
2ndNov_G3_C2	1m	2ndNov_G1_C3	2ndNov_(G1_C2	2ndOct_G2_C1	2ndOct_G1_C3
2ndOct_G2_C2		2ndNov_G1_C1	2ndOct_0	62_C1	2ndNov_G2_C3	2ndOct_G2_C1
2ndOct_G1_C3		2ndOct_G2_C2	2ndNov_(G3_C3	2ndOct_G2_C3	2ndOct_G2_C3
2ndOct_G3_C2		2ndOct_G3_C3	2ndNov_(G2_C1	2ndOct_G3_C2	2ndNov_G3_C3
2ndOct_G3_C1		2ndOct_G2_C1	2ndNov_(G3_C1	2ndNov_G1_C1	2ndNov_G2_C3
2ndOct_G3_C3		2ndOct_G1_C3	2ndNov_(G2_C2	2ndNov_G2_C2	2ndOct_G3_C2

2nd Nov= 2 November, 2nd Oct= 2 October, G1= Cowpea Veg1, G2= M217, G3= Qukawa, C1= Zn at 0%, C2= Zn at 50%, C3= Zn at 100%

Appendix B: General over view of the field trial and different cowpea growing stages.



Cowpea line	Planting Date	Mean
1	02-Nov-15	17.89 ^{ab}
1	02-Nov-15	17.70 ^{ab}
1	02-Nov-15	19.00 ^a
2	02-Nov-15	17.89 ^{ab}
2	02-Nov-15	18.60 ^a
2	02-Nov-15	18.30 ^{ab}
3	02-Nov-15	16.20 ^{bc}
3	02-Nov-15	17.70 ^{ab}
3	02-Nov-15	18.80 ^a
1	02-Oct-15	15.20 ^{cd}
1	02-Oct-15	14.78 ^{cd}
1	02-Oct-15	14.60 ^{cd}
2	02-Oct-15	7.40 ^e
2	02-Oct-15	6.90 ^e
2	02-Oct-15	7.40 ^e
3	02-Oct-15	14.30 ^{cd}
3	02-Oct-15	13.60 ^d
3	02-Oct-15	13.90 ^d
		$LSD_{0.05} = 2.2366$

Appendix C: Interaction between planting date and cowpea lines on germination.

LSD= Least significant difference; Cowpea line 1= Cowpea Veg1, 2= M217, 3= Qukawa; 2-Oct-15= 2 October 2015, 2-Nov-15= 2-November-15. Means with different letter within the column shows significant difference (P< 0.05).

В	ien Donne'		Nietvoorbij						
Cowpea line	Zn level	Yield (kg/ha)	Cowpea line	Zn level	Yield (kg/ha)				
1	1	1043.15	1	1	537.1				
1	2	1110.75	1	2	686.25				
1	3	1009.65	1	3	648.35				
2	1	510.95	2	1	60.7				
2	2	552.35	2	2	63.65				
2	3	486.9	2	3	90.1				
3	1	1078.75	3	1	503.1				
3	2	1114.85	3	2	514.75				
3	3	1184.2	3	3	419.05				

Appendix D: Average seed yield (kg/ha)

Cowpea line1= Cowpea Veg1; 2= M217; 3= Qukawa; Zn level 1= Zn at 0%; 2= Zn at 50% and 3= Zn at 100%.

Planting date	Cowpea line	Zn levels	Yield (kg/ha)
Oct	1	1	908.3
Oct	1	2	1019.15
Oct	1	3	888.3
Oct	2	1	359.65
Oct	2	2	344.15
Oct	2	3	305.35
Oct	3	1	748.75
Oct	3	2	813
Oct	3	3	897.25
Nov	1	1	672
Nov	1	2	811.15
Nov	1	3	769.7
Nov	2	1	271.8
Nov	2	2	299.9
Nov	2	3	293.15
Nov	3	1	833.1
Nov	3	2	816.55
Nov	3	3	715

Appendix E: Average seed yield as affected by planting date across both locations.

Cowpea line1= Cowpea Veg1; 2= M217; 3= Qukawa; Zn level 1= Zn at 0%; 2= Zn at 50% and 3= Zn at 100%; Oct= October; Nov=November.

Appendices F: P-values of ANOVA to determine interactions of all measured parameters as analysed by treatment, location and planting date the full model.

				No. of	Pod	No. of		Pod	100 seed	Moisture
GR	No. of leaves	No. of branches	PH (cm)	seed/p od	length (cm)	pods/ plant	No. of pods	weight (g)	weight (g)	content (%)
0.0455	0.9820	0.8550	0.9997	0.2031	0.0051	0.1605	0.2781	0.4617	0.0020	0.4168
<.0001	0.1742	0.4922	0.0275	0.0075	0.0014	0.0222	0.0265	0.3119	0.0089	0.3087
<.0001	<.0001	<.0001	<.0001	0.0075	0.0166	0.0045	0.2308	0.1437	0.0761	<.0001
0.0364	0.9549	0.8985	0.3833	0.0736	0.1640	0.2243	0.1060	0.1185	0.0908	0.6684
	0.0455 <.0001 <.0001 0.0364	0.04550.9820<.0001	GRleavesbranches0.04550.98200.8550<.0001	GR leaves branches PH (cm) 0.0455 0.9820 0.8550 0.9997 <.0001	No. of leaves No. of branches PH (cm) seed/p od 0.0455 0.9820 0.8550 0.9997 0.2031 <.0001	GR No. of leaves No. of branches PH (cm) seed/p od length (cm) 0.0455 0.9820 0.8550 0.9997 0.2031 0.0051 <.0001	GR No. of leaves No. of branches PH (cm) seed/p od length (cm) pods/ plant 0.0455 0.9820 0.8550 0.9997 0.2031 0.0051 0.1605 <.0001	GRNo. of leavesNo. of branchesPH (cm)seed/p odlength (cm)pods/ plantNo. of pods0.04550.98200.85500.99970.20310.00510.16050.2781<.0001	GR No. of leaves No. of branches PH (cm) seed/p od length (cm) pods/ plant No. of pods weight (g) 0.0455 0.9820 0.8550 0.9997 0.2031 0.0051 0.1605 0.2781 0.4617 <.0001	GR No. of leaves No. of branches PH (cm) seed/p od length (cm) pods/ plant No. of pods weight (g) 0.0455 0.9820 0.8550 0.9997 0.2031 0.0051 0.1605 0.2781 0.4617 0.0020 <.0001

	B	efore zinc was	applied	After application of zinc						
Treatment	No. of leaves	No. of branches	Plant height (cm)	No. of leaves	No. of branches	Plant height (cm)				
	12.66 ^a	1.43 ^a	1.95 ^b	12.88 ^a	0.10 ^b	2.62 ^a				
				$LSD_{0.05} = 0.73$	$LSD_{0.05} = 0.12$	$LSD_{0.05} = 0.16$				
Planting Date										
November	16.47 ^a	1.00 ^b	3.03 ^a	12.32 ^c	0.08 ^c	2.67 ^b				
October	8.16 ^d	1.95 ^a	0.68 ^c	13.62 ^b	0.12 ^c	2.55 ^b				
				$LSD_{0.05} = 1.04$	$LSD_{0.05} = 0.17$	$LSD_{0.05} = 0.22$				
Location										
Bien Donne'	16.32 ^a	1.85 ^a	2.32 ^b	12.17 ^c	0.17 ^d	2.45 ^b				
Nietvoorbij	8.72 ^d	0.99 ^b	1.55 ^c	13.63 ^b	0.39 ^c	2.80 ^a				
				$LSD_{0.05} = 1.03$	$LSD_{0.05} = 0.17$	$LSD_{0.05} = 0.22$				

Appendix G: Vegetative growth rate before and after application of zinc fertilizer.

LSD_{0.05}= Least Significant Difference; No= Number Means with different letter within the column shows significant difference (P< 0.05).

		Reproductive parameters and planting date													
	No of pods/plant		o of pods/plant Total yield		Pod w	Pod weight No		No. of seed/pod		Pod length		100-seed weight		e content	
Trt	2 Oct	2 Nov	2 Oct	2 Nov	2 Oct	2 Nov	2 Oct	2 Nov	2 Oct	2 Nov	2 Oct	2 Nov	2 Oct	2 Nov	
L1T1	41.75 ^a	26.58 ^{ab}	915.30 ^a	673.00 ^a	1816.6 ^{ab}	1344.0 ^a	15.66 ^a	14.65 ^a	16.40 ^a	16.08 ^b	11.70 ^{abc}	11.89 ^{cd}	11.38 ^{ab}	12.30 ^{ab}	
L1T2	33.30 ^{abc}	27.57 ^a	975.00 ^a	734.38 ^a	2038.3 ^a	1622.3 ^a	15.22 ^a	14.95 ^a	16.17 ^a	16.11 ^b	12.00 ^{abc}	11.60 ^d	11.70 ^a	12.63 ^a	
L1T3	37.79 ^{ab}	26.96 ^a	870.20 ^a	772.30 ^a	1776.6 ^a	1539.4 ^a	15.55 ^a	14.15 ^a	16.45 ^a	15.75 ^b	12.80 ^a	11.90 ^{cd}	11.75 ^a	12.56 ^a	
L2T1	23.91 ^{cd}	15.29 ^{bc}	539.38 ^b	442.44 ^{bc}	719.3 [°]	543.6 ^b	6.43 ^d	8.91 ^b	8.65 [°]	11.92 ^c	9.30 ^c	13.80 ^{ab}	9.98 ^c	11.29 ^{cd}	
L2T2	32.43 ^{abc}	16.37 ^{abc}	514.00 ^{bc}	427.80 ^{bc}	688.3 ^c	599.8 ^b	8.48 ^c	9.04 ^b	11.62 ^b	11.76 [°]	10.90 ^{abc}	13.50 ^b	10.19 ^c	11.59 ^{bcd}	
L2T3	16.15 ^d	27.21 ^a	341.57 [°]	404.75 [°]	610.7 ^c	586.3 ^b	6.71 ^d	8.91 ^b	9.14 ^c	11.63 ^c	9.70 ^{bc}	13.10 ^{bc}	9.96 ^c	10.80 ^d	
L3T1	22.06 ^{cd}	14.83 ^c	613.90 ^b	657.90 ^a	1497.5 ^b	1666.2 ^a	14.14 ^{ab}	14.09 ^a	17.53 ^a	17.50 ^a	13.60 ^a	14.20 ^{ab}	10.92 ^b	12.34 ^{ab}	
L3T2	25.99 ^{bcd}	16.76 ^{abc}	649.70 ^b	651.70 ^a	1626.0 ^{ab}	1633.1 ^a	14.72 ^{ab}	13.85 ^a	17.93 ^a	17.42 ^a	13.11 ^a	15.00 ^a	11.56 ^{ab}	12.08 ^{abc}	
L3T3	20.13 ^{cd}	14.43 ^c	617.89 ^a	604.22 ^{ab}	1794.5 ^{ab}	1430.0 ^a	13.43 ^b	14.47 ^a	16.96 ^a	17.49 ^a	12.30 ^{ab}	14.00 ^{ab}	11.38 ^{ab}	11.98 ^{abc}	
LSD _{0.05}	13.69	11.34	188.87	183.86	471.28	466.56	1.58	1.42	1.78	0.74	2.87	1.47	0.73	0.89	
Farm															
BD	28.86 ^a	25.82 ^a	936.32 ^a	835.10 ^ª	1926.8 ^a	1673.4 ^a	13.89 ^a	13.50 ^ª	15.28 ^ª	15.27 ^a	12.60 ^ª	13.49 ^a	10.56 ^b	13.07 ^b	
NVB	27.61 ^a	15.52 ^b	405.64 ^b	374.91 ^b	854.9 ^b	763.2 ^b	10.56 ^a	11.75 ^b	13.71 ^b	15.07 ^a	10.74 ^b	12.98 ^ª	11.54 ^a	10.85 ^b	
LSD _{0.05}	6.45	5.35	88.18	86.34	221.92	219.94	0.74	0.67	0.84	0.35	1.35	0.69	0.34	0.42	

Appendix H: Comparison of reproductive parameters as affected by planting date.

Trt= Treatment; LSD_{0.05}= Least significant different; L1= Cowpea Veg1, L2= M217, L3= Qukawa; T1= Zn at 0%, T2= Zn at 50% and T3= Zn at 100%; BD= Bien Donne', NVB= Nietvoorbij, 2 Oct= 2 October, 2 Nov= 2 November

Means with different letter within the column shows significant difference (P< 0.05).

	%							mg/kg									
Trt	N		Р		K		N	Na		Fe		'n	Ca		Mg		
	Oct	Nov	Oct	Nov	Oct	Nov	Oct	Nov	Oct	Nov	Oct	Nov	Oct	Nov	Oct	Nov	
LIT1	3.88 ^a	3.85 ^{ab}	0.49 ^c	0.50 ^{ab}	1.58 ^a	1.53 ^a	237.25 ^{ab}	203.50 ^{ab}	95.75 ^a	68.50 ^c	39.00 ^a	42.67 ^{ab}	0.15 ^{ab}	0.12 ^b	0.21 ^a	0.22 ^a	
L1T2	4.05 ^a	3.81 ^{ab}	0.50 ^c	0.51 ^{ab}	1.52 ^{ab}	1.60 ^a	238.25 ^{ab}	244.50 ^a	85.50 ^{abc}	74.25 ^{abc}	47.75 ^a	48.75 ^{ab}	0.12 ^b	0.12 ^b	0.20 ^{ab}	0.21 ^{ab}	
L1T3	3.90 ^a	3.94 ^{ab}	0.52 ^{bc}	0.47 ^b	1.57 ^a	1.48 ^{aa}	266.75 ^a	208.50 ^{ab}	78.00 ^c	72.50 ^{bc}	48.50 ^a	43.25 ^{ab}	0.13 ^b	0.12 ^b	0.21 ^{ab}	0.19 ^{bc}	
L2T1	3.92 ^a	3.75 ^b	0.53 ^{bc}	0.51 ^{ab}	1.51 ^{ab}	1.50 ^a	194.50 ^{abc}	190.25 ^{abc}	81.25 ^{bc}	77.75 ^{abc}	44.50 ^a	43.75 ^{ab}	0.17 ^a	0.16 ^a	0.18 ^d	0.19 ^{bc}	
L2T2	3.77 ^a	3.87 ^{ab}	0.56 ^{ab}	0.49 ^{ab}	1.56 ^{ab}	1.48 ^a	200.50 ^{abc}	193.25 ^{abc}	83.75 ^{bc}	86.25 ^{ab}	45.50 ^a	40.00 ^b	0.18 ^a	0.16 ^a	0.20 ^{bc}	0.19 ^{bc}	
L2T3	3.67 ^a	4.00 ^{ab}	0.58 ^a	0.50 ^{ab}	1.56 ^{ab}	1.49 ^a	194.75 ^{abc}	168.25 ^{bc}	91.50 ^{ab}	88.50 ^a	47.75 ^a	42.25 ^{ab}	0.17 ^a	0.16 ^a	0.20 ^{bc}	0.18 ^c	
L3T1	3.89 ^a	3.99 ^{ab}	0.53 ^{bc}	0.49 ^b	1.49 ^b	1.48 ^a	189.50 ^{abc}	132.80 ^c	81.75 ^{bc}	74.50 ^{abc}	45.50 ^a	46.75 ^{ab}	0.12 ^b	0.12 ^b	0.19 ^{cd}	0.19 ^{bc}	
L3T2	3.97 ^a	4.09 ^a	0.50 ^c	0.54 ^a	1.49 ^b	1.53 ^a	135.75 [°]	130.50 ^c	80.25 ^c	80.75 ^{abc}	46.50 ^a	49.50 ^a	0.13 ^b	0.12 ^b	0.18 ^d	0.19 ^{bc}	
L3T3	3.92 ^a	3.88 ^{ab}	0.52 ^{bc}	0.51 ^{ab}	1.52 ^{ab}	1.50 ^a	163.75 ^{bc}	146.00 ^{bc}	81.50 ^{bc}	76.50 ^{abc}	45.50 ^a	47.00 ^{ab}	0.14 ^{ab}	0.12 ^b	0.19 ^{cd}	0.19 ^{bc}	
$LSD_{0.05}$	0.43	0.31	0.04	0.05	0.08	0.13	78.55	66.48	10.67	15.08	5.35	9.11	0.04	0.03	0.01	0.02	
Farm																	
BD	3.92 ^a	3.98 ^a	0.53 ^a	0.51 ^a	1.52 ^a	1.50 ^a	118.11 ^b	96.17 ^b	88.06 ^a	74.39 ^a	39.56 ^b	36.83 ^b	0.13 ^b	0.12 ^b	0.19 ^a	0.19 ^a	
NVB	3.85 ^a	3.84 ^a	0.52 ^a	0.50 ^a	1.54 ^a	1.51 ^a	286.56 ^a	263.11 ^a	80.67 ^b	81.06 ^a	53.00 ^a	53.53 ^a	0.16 ^a	0.15 ^a	0.19 ^a	0.19 ^a	
LSD _{0.05}	0.16	0.17	0.03	0.03	0.04	0.67	30.73	32.2	5.83	7.35	2.28	2.23	0.01	0.01	0.01	0.01	

Appendix I: Effect of planting date on cowpea mineral content

Trt= Treatment; LSD_{0.05}= Least significant difference; Cowpea line 1= Cowpea Veg1, 2= M217, 3= Qukawa; T1= Zn at 0%; T2= Zn at 50% and T3= Zn at 100%, Oct= October, Nov= November; BD= Bien Donne', NVB= Nietvoorbij Means with different letter within the column shows significant difference (P< 0.05).