

**Effect of agronomic management on growth and yield of
selected leafy vegetables**

by

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UNIVERSITY OF SOUTH AFRICA

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STUDENT DECLARATION

Effect of agronomic management on growth and yield of selected leafy vegetables

I, Innocent Maseko, student number: 4968-618-6 declare that:

- (i) The research reported in this dissertation, except where otherwise indicated, is the result of my own endeavours in the College of Agriculture and Environmental Sciences, Department of Agriculture and Animal Health, UNIVERSITY OF SOUTH AFRICA;
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Regular consultation took place between the student and us throughout the investigation. We advised the student to the best of our ability and approved the final document for submission to the College of Agriculture and Environmental Sciences, Department of Agriculture and Animal Health, for examination by the university appointed examiners.

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I further declare that I have not previously submitted this work, or part of it, for examination at Unisa for another qualification or at any other higher education institution.

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DETAILS OF CONTRIBUTION TO PUBLICATIONS that form part and/or include research presented in this thesis (include publications in preparation, submitted, *in press*, published and details of the contributions to the experimental work and writing of each publication).

Publication 1

I. Maseko^{1*}, Y.G. Beletse², N. Nogemane¹, C.P. Du Plooy² and T. Mabhaudhi³, 2014. Growth, physiology and yield responses of *Amaranthus cruentus*, *Corchorus olitorius* and *Vigna unguiculata* to plant density under drip irrigated commercial production (*in press*).

Contributions: field trials, data collection, analysis and manuscript preparation were performed by the first author under the supervision of the other authors.

Publication 2

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Contributions: field trials, data collection analysis and manuscript preparation were performed by the first author under the supervision of the other authors.

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Contributions: field trials, data collection analysis and manuscript preparation were performed by the first author under the supervision of the other authors.

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CONFERENCE CONTRIBUTIONS

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I.Maseko^{1*}, Y.G. Beletse², N. Nogemane¹, C.P. Du Plooy² and T. Mabhaudhi³, 2013

Growth, yield and physiological responses of *Amaranthus cruentus* to plant density. The 4th Prof Humphrey Memorial Postgraduate Student Symposium, University of South Africa, Florida Campus - 29 July 2013.

Maseko I^{1*}, Beletse Y.G², Nogemane N¹, C.P. Du Plooy² and Mabhaudhi T³., 2013

Physiological, growth and yield responses of Chinese cabbage (*Brassica juncea*) to different agronomic management factors. Symposium on the Water Use and Nutritional Value of Indigenous and Traditional South African Underutilised Food Crops for Improved Livelihoods, Pretoria. 17 - 24 February 2014.

SUMMARY

Leafy vegetables are plant species of which the leafy parts, which may include young, succulent stems, flowers and very young fruit, are used as a vegetable. *Amaranthus cruentus*, *Corchorus olitorius*, *Vigna unguiculata* and *Brassica juncea* are African/traditional leafy vegetables with potential to improve nutritional values. Their promotion is partly hindered by the lack of agronomic information. The objective of the current study was to investigate the effect of nitrogen application rates and varying plant densities on growth, physiology and yield responses of *Amaranthus cruentus*, *Corchorus olitorius* and *Vigna unguiculata* during 2011/2012 and 2012/2013 summer season. The study further investigated the combined effect of nitrogen, planting date, irrigation frequency and plant density on growth, physiology and yield responses of *Brassica juncea* in 2011/2012 and 2012/2013 winter seasons. Marketable yield of *Amaranthus cruentus* and *Corchorus olitorius*, was attained at the lowest density of 50 000 plants ha⁻¹ while that of *Vigna unguiculata* was attained at the highest density of 100 000 plants ha⁻¹. Nitrogen rates used in the current study reduced marketable yield of *Vigna unguiculata*. In *Amaranthus cruentus* and *Corchorus olitorius* marketable yield was attained at 44 kg N ha⁻¹ and 100 kg N ha⁻¹ in 2011/12 and 2012/13 summer season respectively which were lower rates than recommended for *Amaranthaceae* species, Swiss chard (*Beta vulgaris L. var cicla*). Marketable yield of *Brassica juncea* planted during winter season was improved by planting early in June, irrigating thrice weekly, using lower plant densities of 50 000 plants ha⁻¹ and applying nitrogen at 50 kg ha⁻¹. Growth and yield parameters in the current study indicate that traditional leaf vegetables can be optimised through improved agronomic practise.

Key words: *Brassica juncea*, *Amaranthus cruentus*, *Corchorus olitorius*, *Vigna unguiculata*, irrigation, nitrogen, planting density, planting date.

ABSTRACT

African leafy vegetables have been shown and suggested to have potential to contribute to human diets and alleviate malnutrition; however, their levels of utilisation are currently low especially in South Africa. This is because there is limited access to these crops due to low availability in the market. Limited access is attributed, in part, to the lack of commercialisation as a result of limited agronomic information describing optimum management options for these leafy vegetables. Availability of such information would contribute to successful commercialisation of these crops. The primary objective of this study was to establish optimum agronomic management factors for *Amaranthus cruentus*, *Corchorus olitorius*, *Vigna unguiculata* and *Brassica juncea* for irrigated commercial production in South Africa.

Seeds of *Amaranthus cruentus*, *Corchorus olitorius* were obtained from the Agricultural Research Council seed bank; *Vigna unguiculata* were obtained from Hydrotech and *Brassica juncea* seeds were obtained from Stark Ayres. The project consisted of three field studies whose overall objective was to evaluate growth and yield responses of the selected African leafy vegetables to agronomic factors under irrigated commercial production. These field studies comprised of two single factors; summer trials (planting density and nitrogen on three selected crops) and a combined winter trial (nitrogen, irrigation, plant density and planting date on a winter crop).

Chapter three (3) investigated the effect of plant density on growth, physiology and yield responses of *Amaranthus cruentus*, *Corchorus olitorius* and *Vigna unguiculata* to three plant densities under drip irrigated commercial production. The plant density levels of 100 000, 66 666 and 50 000 plants/ha were used in the 2011/12 and 2012/13 summer seasons. Parameters measured included chlorophyll content index (CCI), chlorophyll fluorescence (CF), stomatal conductance (SC), leaf number, leaf area index (LAI) and biomass. *Amaranthus cruentus* and *Corchorus olitorius* showed better leaf quality at lower plant density of 50 000 plants ha⁻¹ than at 66 666 plants ha⁻¹ and 100 000 plants ha⁻¹. These results are based on bigger leaves expressed as leaf area index (LAI), better colour expressed as chlorophyll (CCI) and higher biomass per plant observed in these crops at 50 000 plants ha⁻¹ in comparison to 66 666 plants ha⁻¹ and 100 000 plants ha⁻¹. In *Vigna unguiculata* there were no responses observed in LAI and CCI. In *Amaranthus cruentus*, *Corchorus olitorius* and *Vigna unguiculata* fresh and dry mass yield of leaves were higher at 100 000 plants ha⁻¹ compared to other treatments. In A.

cruentus and *C. olerius*, higher leaf quality parameters (CCI, plant height, leaf number, biomass per plant and LAI) indicated that these crops can perform better at lower densities of 50 000 than at 66 666 plants ha⁻¹ and 100 000 plants ha⁻¹. Therefore, using 50 000 plants ha⁻¹ is suitable for commercial production of *A. cruentus* and *C. olerius*. In *Vigna unguiculata*, a plant density of 100 000 plants ha⁻¹ produced the highest fresh and dry mass per unit area without compromising quality in terms of the leaf size (LAI) and colour (CCI). Therefore 100 000 plants ha⁻¹ is a density recommended for commercial production in *V. unguiculata*.

Chapter four (4) was conducted to investigate growth, physiology and yield responses of *A. cruentus*, *C. olerius* and *V. unguiculata* to nitrogen application under drip irrigated commercial production. Three nitrogen treatments levels were used viz. 0, 44 and 88 kg N ha⁻¹ in 2011/12 season and four nitrogen treatments levels viz. 0, 50, 100 and 125 kg N ha⁻¹ were used in 2012/13 summer season. The nitrogen levels selected for each season were based on recommendations for *Amaranthaceae* species, Swiss chard (*Beta vulgaris L. var cicla*) derived from soil analysis of the trial (field) site. Parameters measured included chlorophyll content index (CCI), chlorophyll fluorescence (CF), stomatal conductance (SC), leaf number, leaf area index (LAI) and biomass. Results showed that application of nitrogen at 44 kg N ha⁻¹ in 2011/12 summer season and 100 kg N ha⁻¹ in 2012/13 summer season improved LAI, CCI, biomass per plants and yield in *A. cruentus*. A similar trend was observed in *C. olerius* except that 44 kg N ha⁻¹ improved stem fresh yield. Further increase in nitrogen fertiliser above 44 kg N ha⁻¹ during the 2011/12 season and above 100 kg N ha⁻¹ in 2012/13 summer season reduced leaf quality and yield in both crops. In *V. unguiculata*, nitrogen application showed a slight increase in yield values from 0 to 44 kg N ha⁻¹ followed by decrease at 88 kg N ha⁻¹ in 2011/12 summer season; however, this increase in yield was not significant. During the 2012/13 summer season, yield in terms of fresh weight was significantly (P<.001) reduced by applying nitrogen at various levels. However, leaf dry matter content increased significantly (P<.001) with increase in nitrogen from 0 kg up to 100 kg N ha⁻¹, then remained unchanged at 125 kg N ha⁻¹. Therefore, the current study recommends that *C. olerius* and *A. cruentus* could be commercialised at 44 kg N ha⁻¹ and 100 kg N ha⁻¹ which were lower nitrogen application rates than those recommended for *Amaranthaceae* species. In *V. unguiculata*, 50 kg N ha⁻¹ improved leaf number; however, this did not translate to any fresh yield advantage, implying that the optimum rate for nitrogen application might be lower than

50 kg N ha⁻¹. Therefore, nitrogen rates less than the ones used in the current study are recommended for *V. unguiculata*.

Chapter five (5) was conducted in winter and it was necessitated by observations made primarily in the previous studies which focused on the effects of single factors such as plant density, planting date and nitrogen deficits. Therefore, there was a need to address interactions between irrigation, nitrogen, spacing and planting date. The objective of this study was to evaluate growth, physiology and yield responses of *Brassica juncea* to different agronomic and management factors in the 2012 and 2013 seasons. The treatments were as follows: two planting dates in main plot (1 June and 18 July, 2012); two irrigation frequency in sub main plot (once and three times a week); three nitrogen levels (0, 50, 100 kg N ha⁻¹) and three plant densities (133 333, 80 000, 50 000 plants ha⁻¹) as subplots. Parameters measured included chlorophyll content index (CCI), chlorophyll fluorescence (CF), stomatal conductance (SC), leaf number, leaf area index (LAI) and biomass. Results from this study showed a significant interaction effect on plant height, LAI, CCI and CF. Crops irrigated thrice or once a week with 50 kg N ha⁻¹ combined with 50 000 plants ha⁻¹ produced tall plants and bigger leaves (LAI) in the early planting date (1 June) compared to other combinations. Irrigating three times a week combined with nitrogen application at 100 or 50 kg N ha⁻¹ improved CF for late planting date (18 July) in comparison to other combinations. Irrigating once a week combined with nitrogen application at 100 kg N ha⁻¹ increased CCI. There was no significant interaction effect on yield. Application of nitrogen at 50 and 100 kg N ha⁻¹ significantly ($P>0.05$) increased yield in early and late planting dates compared to the control (0 kg N ha⁻¹), in 2012 and 2013 winter season. Irrigating three times a week led to a significant ($P<0.05$) increase in yield in the late planting date (18th July) and early planting date (1st June) in 2013 season. Higher plant density of 133 333 plants ha⁻¹ resulted in significantly ($P<0.05$) higher yield in terms of fresh mass and leaf number in the late planting date 18 July in 2012 and 2013 seasons. However, leaf quality parameters such as leaf size and colour was compromised at 133 333 plants ha⁻¹ relative to 50 000 plants ha⁻¹. Therefore, farmers are recommended to plant early, apply 50 kg N ha⁻¹, irrigate thrice a week and utilise a spacing of 50 000 plants ha⁻¹. The current study indicates that growth and yield of traditional leaf vegetables can be optimised through improved agronomic practise.

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DEDICATION

Being a product of many hands in all walks of life, I dedicate this thesis to my colleagues of goodwill, who have always desired the best from me.

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LIST OF ACRONYMS

ANOVA:	analysis of variance
ALVs:	African leafy vegetables
<i>A. cruentus:</i>	<i>Amaranthus cruentus</i>
ARC	Agricultural Research Council
AWS:	Automatic weather station
CCI:	Chlorophyll Content Index
CF:	Chlorophyll Fluorescence
<i>C. olerius:</i>	<i>Corchorus olerius</i>
DAFF:	Department of Agriculture Forestry and Fisheries
ETo:	reference evapotranspiration
FAO:	Food and Agriculture Organisation
Fv/Fm:	ratio of quantum yield potential of photosynthesis
Ha:	Hectare
ISCW:	Institute of Soil, Climate and Water
:	litre
LAN:	lime (stone) ammonium nitrate
LAI:	Leaf Area Index
N:	Nitrogen
RCBD:	Randomised complete block design
SC:	Stomatal Conductance
VOPI:	Vegetable and Ornamental Plant Institute
<i>V. unguiculata:</i>	<i>Vigna unguiculata</i>
WAT:	weeks after transplanting

CHAPTER 1

INTRODUCTION

1.1 BACKGROUND

African leafy vegetables (ALVs) are defined as the collective of leafy vegetable species that form part of the culinary repertoire of particular contemporary African communities (van Rensburg *et al.*, 2007). African leafy vegetables embrace indigenous and recently introduced plant species (van Rensburg *et al.*, 2007).

There are many names by which African leafy vegetables are known by different authors including traditional leafy vegetables (Vorster *et al.*, 2008; Odhav *et al.*, 2007) and wild vegetables (Nesamvuni *et al.*, 2001). Ethnic groups in South Africa have their own names which vary from place to place. Collectively they are called *imfino* in isiZulu and isiXhosa, *morogo* in Sesotho or *miroho* in tshiVhenda (Maunder and Meaker, 2007). The plant species that are referred to as *imfino* or *morogo* vary from place to place (van Rensburg *et al.*, 2007). According to Faber *et al.*, (2002) *imfino* “is a collection of various dark-green leaves that is eaten as a vegetable; the leaves either grow wild or come from vegetables such as pumpkin and beetroot”.

In South Africa, the use of leafy vegetables is as old as the history of modern man (van Rensburg *et al.*, 2007). In the past leafy vegetables were obtained mainly by collecting from the wild and not by means of cultivation. The collection of edible plants was particularly important during times of emergency, when crops had failed, livestock herds had been decimated, or when hunters ran out of food (Peires, 1981). The collection of leafy vegetables and the knowledge associated with this practice was a female domain among both the Koisans (Parsons, 1993) and the Bantu-speaking tribes (van Rensburg *et al.*, 2004). Even in the twenty-first (21st) century, the collection of these vegetables continues to be widespread among black South Africans (van Rensburg *et al.*, 2004; Modi *et al.*, 2006). In the recent years limited broadcasting of the seeds of selected species in the field is being practised (Voster *et al.*, 2002; Hart & Voster, 2006).

Traditional leafy vegetables are well documented for their nutritional value. They contain nutrients such as calcium, iron and vitamins A, C, fiber and proteins. Therefore; they can play

a significant role in addressing the problems of low income, malnutrition, and poor health among resource poor households in sub-Saharan Africa (Smith and Eyzaguirre, 2007). Leafy vegetables such as *Amaranthus* and *Corchorus* are rich in protein and fiber (van Renseburg *et al.*, 2004) while *Brassica rapa var. chinensis* contains various nutrients such as fiber, vitamin C, antioxidants and anticarcinogenics (Podsdek, 2007). *Vigna unguicalata* can be used as a feed (grazed or harvested for fodder), or its pods can be harvested before maturity stage and eaten as a vegetable. Some people eat both fresh pods and leaves and the dried seeds are popular ingredients in various dishes (Hector & Jody, 2002). *Vigna unguicalata* contains (20-24%) protein, 63.3% carbohydrates and 1.9% fat (Davis *et al.*, 1991). *Cleome* is a highly nutritious leafy vegetable rich in vitamin A and C, calcium, iron and proteins (Abe & Imbamba, 1997).

Traditional leafy vegetables are also considered to be low management crops because they can grow in poor soils and in areas where the climate is not conducive to the production of exotic vegetables (van Averbeke *et al.*, 2012). They have also been reported to require less plant nutrients than their exotic counterparts, such as Swiss chard, (van Rensburg *et al.*, 2004; Maunder & Meaker, 2007). Traditional leafy vegetables are further reported to be resistant to drought, pests and diseases (DAFF, 2008). Therefore, the basic assumption is that they can grow better and produce higher yields when proper agronomic management factors such as fertilized soils and irrigation are established.

Although indigenous plant species have been used for human consumption for centuries (Vorster *et al.*, 2008; Adebooye & Opabode, 2004) and are noted for their good nutritional value, these crops have not been widely domesticated and are not cultivated on a wide scale, especially in South Africa. Their utilization is highly variable (van Rensburg *et al.*, 2007) and they are mostly gathered from cultivated fields, fallowed land and the veldt (Venter *et al.*, 2007). Low levels of utilisation are attributed in part to lack of agronomic information describing their production systems, such as optimum planting times, plant density, irrigation and nitrogen application rates in South Africa. There is a limited research on the cultivation practices of African leafy vegetables in South Africa. Due to this limited information, producers of African vegetables have been forced to rely on their own knowledge and experience when making agronomic decisions. Agronomic studies conducted on African leafy vegetables in other ecological regions (outside South Africa) show a possibility of

improvement in production in response to cultivation practises. However, it is difficult to adopt recommendations from other regions since the influence of agro climate and genotypic differences have been reported in various crops. Therefore, there is a need to conduct studies to determine cultivation practices that have the potential of improving production of African leafy vegetables under South African climate. Data obtained from this study will contribute to the scientific agronomic knowledge and to the development of production guidelines for the plants under study.

1.2. AIM AND OBJECTIVES

The aim of the study was to establish optimum planting density and nitrogen fertilizer application rates for three (3) selected summer crops (*Amaranthus cruentus*, *Corchorus olitorius* and *Vigna unguiculata*) under irrigated commercial production in South Africa. The study also sought to determine growth, physiology and yield responses of a winter crop, *Brassica juncea* (Chinese cabbage) to various agronomic and management factors.

1.2.1 Specific objectives

- To determine the effect of planting density on growth, physiology and yield responses of *Amaranthus cruentus*, *Corchorus olitorius* and *Vigna unguiculata*.
- To determine the effect of nitrogen fertilizer application rates on growth, physiology and yield responses of *Amaranthus cruentus*, *Corchorus olitorius* and *Vigna unguiculata*.
- To determine the combined effect of nitrogen, planting date, spacing and irrigation on growth, physiology and yield responses of Chinese cabbage (*Brassica juncea*).

1.2.2 Hypotheses

- Planting density of 66 666 plants ha⁻¹ has no effect on growth, physiology and yield responses of *Amaranthus cruentus*, *Corchorus olitorius* and *Vigna unguiculata*
- Nitrogen fertilizer application rates recommended for *Amaranthaceae species*, Swiss chard (*Beta vulgaris L.var cicla*) would lead to optimum growth, physiology and yield responses of *Amaranthus cruentus*, *Corchorus olitorius* and *Vigna unguiculata*.
- Planting density of 80 000 plants ha⁻¹, planting early in June, irrigating thrice a week and application of 50 kg N ha⁻¹ will provide optimum growth and yield in *Brassica juncea*.

1.3 SCOPE OF STUDY

The thesis is written in publication format with each chapter having its own separate introduction, materials and methods, results and discussion and reference sections. However, this being a trans-disciplinary study there is a general introduction, a general literature review and general discussion and recommendation sections. These general sections are intended to illustrate the relationships among various studies and show that, although the chapters have been written separately, they are linked. The structure of the thesis also necessitates that some themes and sections be repeated in the different sections since each of the research chapters share the same introduction and literature review.

CHAPTER 2 reviews literature in terms of general description and distribution of the crops under study and how crop growth, physiology and yield respond to various agronomic management factors.

CHAPTER 3 outlines experimental procedures and results on growth, physiology and yield responses of *Amaranthus cruentus*, *Corchorus olitorius* and *Vigna unguiculata* to varying plant densities under commercial scale production. It has been written in the format that fits the South African Journal of Plant and Soil (SAJPS).

Chapter 4 outlines experimental procedures and results on growth, physiology and yield responses of *Amaranthus cruentus*, *Corchorus olitorius* and *Vigna unguiculata* to nitrogen application under drip irrigated commercial production. It has been written in the format that fits the Journal of Field Crops Research.

Chapter 5 outlines experimental procedures and results on growth, physiology and yield responses of Chinese cabbage (*Brassica juncea*) to nitrogen, planting date, spacing and irrigation management. It has been written in the format that fits the Journal of Water, South Africa (Water SA).

Chapter 6 presents the general conclusions and recommendations of the study.

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CHAPTER 2

LITERATURE REVIEW

2.1 DESCRIPTION AND DISTRIBUTION OF SELECTED TRADITIONAL LEAFY VEGETABLES USED IN THIS STUDY

Leafy vegetables are plant species of which the leafy parts, which may include young, succulent stems, flowers and very young fruit, are used as a vegetable (van Rensburg *et al.*, 2007). Wehmeyer & Rose (1983) identified more than 100 different species of plants that are used as leafy vegetables in South Africa. Out of these 100 species, seven major groups of leafy vegetables species are of particular importance in South Africa (van Rensburg *et al.*, 2007). These include, *C. olerius* (*jute mallow*), *Amaranthus cruentus* (pigweed), *Citrallus lanatus* (bitter melon), *Vigna unguiculata* (cowpea), *Cleome gynandra* (spider plant) and *Brassica rapa subsp. chinensis* (Chinese cabbage). The leafy vegetables used in this study are among the seven major leafy vegetable of importance in South Africa listed by van Rensburg and co-authors. These include three (3) summer crops: *Corchorus olerius*, *Amaranthus cruentus*, *Vigna unguiculata L.* and *Brassica juncea*, a winter crop (Table 2.1). The criteria for selection of these crops included but was not limited to the extent of consumption in South Africa, the extent of their cultivation, the availability of seeds, the primary growing season and the potential to improve the Vitamin A and iron status of people based on observations of Oelofse & van Averbek (2012). The extent of consumption and production are considered to be important indicators of the potential of the leafy vegetables to be commercialised whilst their primary growing season - winter versus summer - are considered important for year round drip irrigated commercial production (Oelofse & van Averbek, 2012). Scientific names, local names and common names of the selected leafy vegetables are presented in the table below (Table 2.1).

Table 2. 1. Scientific names of the selected Indigenous leafy vegetables. (Fox & Norwood Young, 1982; Bromilow, 1995; Van Wyk & Gericke, 2000, Vorster *et al.*, 2002, van Rensburg *et al.*, 2007).

Scientific name	Common name	Local name	Photographs
<i>Corchorus olitorius</i>	Jew's mallow	Wilde jute in Afrikaans; <i>thelele</i> and <i>ligusha</i> in Sepedi, Sesotho and Setswana; <i>delele</i> in Tshivenda; and <i>guxe</i> , <i>ligushe</i> in Xitsonga and Shangaan.	
<i>Amaranthus (cruentus)</i>	pigweed, cockscomb and hell's curse	<i>Unomdlomboyi</i> , <i>imbuya</i> , <i>umifinoumyuthu</i> in isiXhosa, <i>imbuya</i> , <i>isheke</i> , <i>indwabaza</i> in isiZulu, <i>thepe</i> , <i>theepe</i> in IsiPedi, Sesotho and Setswana, <i>umbuya</i> , <i>isheke</i> in siSwati, <i>vowa</i> , <i>theebe</i> in Tshivenda, <i>theyke</i> , <i>chekein</i> Xitsonga, and <i>imbuya</i> , <i>tyutu</i> in Pondo.	
<i>Brassica juncea</i>	Chinese cabbage, rape or Chinese mustard cabbage	<i>Sjinesekool</i> in Afrikaans and <i>mut-shainain</i> Tshivenda and other local African languages.	
<i>Vigna unguiculata</i> L.	Cowpeas	<i>Akkerboontjie</i> , <i>koertjie</i> in Afrikaans; <i>dinawa</i> in isiNdebele; <i>iimbotyi</i> in isiXhosa; <i>imbumba</i> , <i>indumba</i> , <i>isihlumanya</i> in isiZulu; <i>monawa</i> in Sepedi; <i>monawa</i> , <i>dinawa</i> , <i>nawa</i> in Sesotho; <i>dinawa</i> , <i>nawa-ea-setswana</i> in Setswana; <i>munawa</i> (plant), <i>nawa</i> (beans) in	

		Tshivenda; <i>dinaba</i> , <i>munaoa</i> , <i>tinyawa</i> in Xitsonga; and <i>murowi we nyemba</i> in Shona	
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2.1.1. Amaranth (*Amaranthus* spp.)

Amaranthus cruentus belongs to the Amaranthaceae family and is an extremely variable, erect to spreading herb (van Rensburg *et al.*, 2007). The young leaves, growth points and seedlings of *Amaranthus* are used as vegetables (van Rensburg *et al.*, 2007). The ability of *Amaranthus* to thrive in the wild leads to a general belief among people that there is no need to cultivate it because it grows naturally. However, women in areas like Limpopo and Mpumalanga provinces do harvest and store seed of *Amaranthus*, which they broadcast in their fields when they observe a decline in the population (van Rensburg *et al.*, 2007). Furthermore, women also practise selective weeding to replenish natural seed reserves (Vorster *et al.*, 2002; Hart & Vorster, 2006). When practising selective weeding, African leafy vegetable species, such as Amaranth, are allowed to grow without being disturbed whilst other weed species, which are not used as food, are controlled. There are different species of amaranthus which are utilized all over South Africa, except in the arid south western areas (Schippers, 2000; Van Wyk & Gericke, 2000; Vorster *et al.*, 2002; Hart & Vorster, 2006). These include *Amaranthus thunbergii* (L), *A. greazicans*, (L), *A. spinosus* (L), *A. deflexus* (L), *A. hypochondriacus* (L), *A. viridus* (L) and *A. hybridus* (L). (Fox & Norwood Young, 1982; Schippers, 2000; Vorster *et al.*, 2002; Hart & Vorster, 2006).

2.1.2 Jew's Mallow (*Corchorus olitorius*)

Corchorus olitorius is an erect annual herb that varies from 20 cm to approximately 1.5 m in height and belongs to the Tiliaceae family (van Rensburg *et al.*, 2007). The stems are angular in shape with simple oblong to lanceolate leaves that have serrated margins and distinct hair-like teeth at the base. The bright yellow flowers are usually very small and the fruit is a straight, angular capsule. *Corchorus* seed show a high degree of dormancy which can be

broken by means of hot water treatment (Schippers *et al.*, 2002). Different types of *Corchorus* species are reported to be found in South Africa (Schippers *et al.*, 2002; Van Wyk & Gericke, 2000). These include *Corchorus asplenifolius*, *C. trilocularis*, *C. tridens* and *C. olitorius* (Van Wyk and Gericke, 2000). *Corchorus* has been established to prefer warm, humid conditions and this is observed by its occurrence mainly in the northern and eastern regions of South Africa (van Rensburg *et al.*, 2007). People in the northern regions of South Africa appreciate its sliminess than the people in the south regions (van Rensburg *et al.*, 2007). As a result the people in the southern regions add bicarbonate of soda to the cooking water to reduce the sliminess of *Corchorus* (Fox & Norwood Young, 1982; Van Wyk & Gericke, 2000; Schippers *et al.*, 2002).

2.1.3. Cowpeas (*Vigna unguiculata* L.)

Vigna unguiculata is a leaf and pulse crop that belongs to the Leguminosae family (van Rensburg *et al.*, 2007). It is an annual or perennial herbaceous plant with tri-foliolate leaves. Different varieties exist, which can either be indeterminate or determinate, types (van Rensburg *et al.*, 2007). The varieties mainly used as a leafy vegetable are the indeterminate types. *Vigna unguiculata* is indigenous to Africa and has been cultivated for a long time on the continent for seed production and as a fodder crop (Fox and Norwood Young, 1982; Schippers, 2000; Vorster *et al.*, 2002; Hart & Vorster, 2006). Various subspecies of cowpeas are found in the wild in the eastern parts of the KwaZulu-Natal and Mphumalanga (van Rensburg *et al.*, 2007). Its ability to fix nitrogen plays a good role in a crop rotation system.

2.1.4. Chinese cabbage (*Brassica juncea* -mustard spinach)

Brassica juncea (mustard spinach) is a leafy vegetable grown in Southern Africa under the name leaf mustard (*B. juncea* ssp. 'Rugosa') or rape. The name 'rape' is confusing since it also refers to the leafy equivalent of the oilseed crops *B. rapa* and *B. napus* (Schippers, 2002). Chinese cabbage belongs to the family of Brassicaceae or Crucifereae (van Rensburg *et al.*, 2007). According to Schippers (2002), *B. juncea* is not generally considered indigenous to Africa and it is a more important crop in China and South-East Asia, where it is known in a variety of forms. *Brassica juncea* is therefore indigenised in South Africa and is being cultivated in some areas of the country.

2.2 POTENTIAL CONTRIBUTION OF *VIGNA UNGUICULATA*, *CORCHORUS OLITORUS*, *BRASSICA JUNCEA* AND *AMARANTHUS CRUENTUS* TO FOOD SECURITY

African leafy vegetables have potential to alleviate micronutrient malnutrition. According to the Food and Agriculture Organisation (FAO, 1992), *Amaranthus cruentus* L. ranks among the best leafy vegetables in terms of its chemical composition and nutritional status. *A. cruentus* contains appreciable amounts of crude protein, minerals (calcium and potassium) and vitamins A and C that can contribute substantially to our daily requirements when consumed in reasonable quantities (Saunders & Becker, 1983; Rubatzky & Yamaguchi, 1997). *Corchorus olitorus* leaves are a rich source of iron, protein, calcium, thiamin, riboflavin, niacin, folate, and dietary fibre (Leung *et al.*, 1968). *Vigna unguiculata* is a major source of protein, minerals and vitamins (Bressani, 1985). Young leaves, green pods and green seeds are used as vegetables whereas dry seeds are used in a variety of food preparations (Nout, 1996; Nielsel *et al.*, 1997). Chinese cabbage (mustard greens) are a good source of dietary fibre, provitamin A, vitamin C, vitamin K, thiamine, riboflavin, vitamin B6, folate and mineral nutrients (van Wyk, 2005).

Production of *Vigna unguiculata*, *Corchorus olitorus*, *Brassica juncea* and *Amaranthus cruentus* will provide food to the rural household and the surplus could be sold to the urban market, even with the prospect of exporting (DAFF, 2008). Studies have shown that African leafy vegetables like *Vigna unguiculata*, *Corchorus olitorus*, *Brassica juncea* and *Amaranthus cruentus* have potential for income generation to small holder farmers and people involved in the economic activities that are linked to the production of these crops (Whitbread, 1986; Weinberger & Pichop, 2009; Manyelo, 2011). However, income generation through marketing of African leafy vegetables in South Africa is still limited and mostly restricted to dried products (Voster *et al.*, 2002; Hart & Voster, 2006).

2.3. EFFECT OF CULTIVATION PRACTICES ON GROWTH, PHYSIOLOGY AND YIELD OF CROPS

2.3.1 Crop responses to water stress

Crop responses to water/drought stress vary due to the intensity and duration of the stress (Chaves *et al.*, 2002). The major crop responses to water stress are discussed below.

2.3.1.1 Stomatal conductance

Stomatal conductance is the measure of the rate of passage of carbon dioxide (CO₂) entering or water vapour exiting through through the stomata of a leaf. Plants grown under drought conditions have a lower stomatal conductance in order to conserve water (Mafakheri *et al.*, 2010). Drought stress is defined as the moderate loss of water which results in stomatal closure and limitation of gas exchange (Jaleel *et al.*, 2009). It has been established that the closure of stomata is the first response of almost all plants to water stress (Mansfield & Atkinson, 1990; Cornic & Massacci, 1996). Transpirational water losses are reduced by stomatal closure. When stomata close, it decreases the flow of CO₂ into the leaves, followed by a decline in net photosynthesis leading to reduced plant growth (Modi & Mbahudhi, 2013). It has been widely reported that stomatal closure is the main reason for decreased photosynthesis under mild to moderate water stress (Cornic & Massacci, 1996; Chaves *et al.*, 2002; Yokota *et al.*, 2002). Studies have reported reduction of transpiration of vegetable amaranth due to reduction of stomatal conductance under drought stress (Liu & Stützel, 2002).

2.3.1.2. Chlorophyll content

Severe drought stress inhibits photosynthesis by causing changes in chlorophyll content, chlorophyll components and by damaging the photosynthetic apparatus (IturbeOrmaetxe *et al.*, 1998). Ommen *et al.* (1999) reported that leaf chlorophyll content decreased as a result of drought stress. Drought stress causes a large decline in chlorophyll a content, chlorophyll b content, and total chlorophyll content in all sunflower varieties investigated (Manivannan *et al.*, 2007). Studies have established that chlorophyll decreases under drought stress due to the damage of chloroplasts caused by reactive oxygen species (Smirnoff, 1995). Decrease in chlorophyll content due to water stress has been reported in various crops such as sunflower

plants (Kiani *et al.*, 2008), okra (Estill *et al.*, 1991 Ashraf *et al.*, 1994) and sesame (Mensha *et al.*, 2006).

2.3.1.3 Chlorophyll Fluorescence (CF)

Studies by Maxwell and Johnson (2000) showed that light energy absorbed by chlorophyll molecules in a leaf can be used to drive photosynthesis (photo-chemistry); excess energy can be dissipated as heat or it can be re-emitted as chlorophyll fluorescence (light). Furthermore, information about changes in the efficiency of photochemistry and heat dissipation can be gained measuring the yield of chlorophyll fluorescence (Maxwell & Johnson, 2000).

Chlorophyll fluorescence (CF) analysis has become one of the widely used techniques to obtain various physiological responses of a crop (Maxwell & Johnson, 2000). It indirectly measures photosynthetic efficiency (Krause & Weis, 1991). Furthermore, CF gives an insight into the ability of a plant to tolerate water stress and the extent of damage on the photosynthetic apparatus (Maxwell & Johnson 2000). Use of CF parameters, such as F_o (initial), F_m (maximum), F_v (variable = $F_m - F_o$), F_v/F_m to evaluate intact leaves; make it possible to estimate photosynthetic efficiency of the leaf under various conditions (Durães *et al.*, 2001). The F_v/F_m ratio (the measurement of quantum yield potential of photosynthesis, or maximal photochemical efficiency of PSII) has been shown to be a reliable stress indicator (Krause and Weis, 1991; Schreiber *et al.*, 1994). Severe levels of drought may irreversibly damage the photosynthetic apparatus (Kawamitsu *et al.*, 2000; Zulini *et al.*, 2007). Reduction in chlorophyll fluorescence due to stress has been reported in other crops such as tomatoes (Boamah *et al.*, 2011).

2.3.1.4. Plant growth and development

Plant growth is the irreversible increase in the size of the plant. It includes stages from germination, emergence, vegetative growth up to and including reproductive growth (Modi & Mabhaudhi, 2013). Plant growth is achieved through cell division (mitosis), expansion and finally differentiation (Modi & Mabhaudhi, 2013). The processes of cell growth are sensitive to water stress due to reduction in turgor pressure (Taiz & Zeiger, 2006). Reduction in germination and emergence is one of the effects of water stress (Harris *et al.*, 2002). Reduction in germination and seedling stand establishment has been reported in various crops, in sunflowers (Kaya *et al.*, 2006) and in indigenous crops such as wild mustard (Mbatha & Modi, 2010) and wild water melon (Zulu & Modi, 2010).

Water stress impairs mitosis, elongation and expansion, resulting in reduced plant height, leaf number and leaf area and generally reduced crop growth (Nonami, 1998; Kaya *et al.*, 2006). Studies on soya beans (Specht *et al.*, 2001; Zhang *et al.*, 2004) and potatoes (Heuer & Nadler, 1995) showed a reduction in plant height in response to water stress while in cowpea (Manivannan *et al.*, 2007) a reduction in leaf number and leaf area was observed. Furthermore, water stress reduced plant height and leaf number in wild mustard (Mbatha & Modi, 2010) and wild water melon (Zulu & Modi, 2010). Bhatt & Rao (2005) associated the reduction in plant height with a reduction in cell expansion. Leaf development is crucial to photosynthesis and dry matter production.

The importance of the root system with regards to a plant's ability to tolerate stress has been studied (Jaleel *et al.*, 2009). Under water stress, plants tend to elongate roots in search of water; however, genetic variations may limit potential maximum rooting depth (Blum, 2005). Increased root growth under stress has been associated with an increased root: shoot ratio; under stress, plants will allocate more assimilate to root growth (sourcing more water) while limiting stem growth (loss of water) (Modi & Mabhaudhi, 2013). Growth of *Corchorus* has also been reported to slow down considerably when plants are subjected to prolonged periods of water deficit (van Rensburg *et al.*, 2007). Rapid leaf area development and high stomatal conductance, rapid root and shoot growth after germination are part of the features that ensure the crop uses available soil water efficiently (Liu & Stutzel, 2002).

2.3.1.5 Yield

The objective of every farmer is to obtain high yields (Jaleel *et al.*, 2009) under all conditions, more so under drought stress. Yields show considerable variation under drought stress conditions (Jaleel *et al.*, 2009). Various yield-determining plant processes are affected by water stress (Farooq *et al.*, 2009). Studies have shown yield reduction in response to water stress in legume crops such as soya beans (Frederick *et al.*, 2001) and black beans (Nielson & Nelson, 1998). Cowpea thrives in arid and semi-arid conditions and is produced in areas with optimum rainfall conditions of 400 to 700 mm per annum (van Rensburg *et al.*, 2007). Cowpea is reported to be a drought and heat tolerant crop due to its adaptation to semi-arid regions where other legume crops do not perform well (Singh *et al.*, 2003).

Amaranth species are tolerant to adverse climatic conditions (Grubben, 2004; Maundu & Grubben, 2004) and they are quite drought-tolerant, but prolonged dry spells induce

flowering and decrease leaf yield (Schippers, 2000; Palada & Chang, 2003). Genotypic differences in these traits and their relations to plant performance under drought stress have been studied. Neluheni *et al.* (2003) reported on amaranth that the level of drought tolerance varied according to the specie's genetic makeup; *A. graezizans* was adapted to dry lands, while *A. cruentus* was less tolerant to harsh conditions such as drought. Chinese cabbage has also been reported to require adequate availability of soil water for optimum growth (Tshikalange & van Averbek, 2006) and does not tolerate poorly drained conditions (van Rensburg *et al.*, 2007).

2.3.2. Crop response to nitrogen fertiliser

Nitrogen is more important than any other elements for plant growth and its uptake is influenced by temperature, soil water availability, microbial activities and soil reaction (Splittstoesser, 1990). The major crop responses to nitrogen fertilizer are discussed below.

2.3.2.1 Stomatal conductance

The control of leaf stomatal conductance is a crucial mechanism for plants, since it is essential for both CO₂ acquisition and desiccation prevention (Dodd, 2003). Studies with maize have shown that nitrogen deficit can decrease (Dodd, 2003), increase or have no effect (Cechin & Fumis, 2004) on stomatal conductance.

Studies conducted on *Cleome* showed that stomatal conductance was affected by nitrogen fertilizer application rates regardless of the type of fertiliser used. In *Cleome* fertilised with 0-200 kg N ha⁻¹ (calcium ammonium nitrate) leaf stomatal conductance significantly increased with the increase in nitrogen fertilizer up to 150 kg N ha⁻¹ and it decreased afterwards (Ng'etich *et al.*, 2012a). Similar observations were made in *Cleome* fertilised with 0-15 t/ha of farmyard manure, in which leaf stomatal conductance significantly increased with the increase in farmyard manure up to 11.5 t/ha of farmyard manure and it decreased afterwards (Ng'etich *et al.*, 2012b). Ng'etich *et al.* (2012b) concluded that in the case of *Cleome*, low stomatal conductance in the control treatment is attributed to nitrogen deficiency and the reduction in stomatal conductance at the higher application rate is attributed to excess nitrogen in the soil. Furthermore, he reported that low stomatal conductance could lead to reduced yield due to reduction in gaseous exchange.

2.3.2.2 Chlorophyll content and Chlorophyll Fluorescence (CF)

Chlorophyll, a green pigment present in plants, captures radiation that is used in photosynthesis (Swain & Jagtap Sandip, 2010). Nitrogen is a constituent of chlorophyll, proteins, and amino acids (Sumeet *et al.* (2009). Leaves with different nitrogen content would, therefore, differ greatly in chlorophyll content (Witt *et al.*, 2005). Nitrogen deficiency generally results in stunted growth and chlorotic leaves caused by poor assimilate formation that lead to premature flowering and shortening of the growth cycle. The presence of nitrogen in excess promotes development of the above ground organs with abundant dark green (high chlorophyll) tissues of soft consistency and relatively poor root growth (Wolf, 1999). In *Amaranthus cruentus*, chlorophyll content increased with increase in palm oil mill effluent (POME) and inorganic fertiliser (NPK) application compared to plants with no nitrogen (Law-Ogbomo *et al.*, 2011).

Nitrogen also plays a role in photosynthesis and as well as carbon dioxide assimilation (Jasso-Chaverria *et al.*, 2005). Nitrogen is an essential nutrient in creating the plant dry matter as well as many energy-rich compounds which regulates photosynthesis and plant production (Wu *et al.*, 1998). Nitrogen fertilisation in maize has been reported to promote the net photosynthetic rate in leaves (Ng'etich *et al.*, 2013).

2.3.2.3 Growth and development

Plants absorb nitrogen mainly in the nitrate (NO_3) and ammonium (NH_4) forms, both of which are metabolised by plants (Ng'etich *et al.*, 2013). It stimulates vegetative growth resulting in large stems and leaves. Abidin & Yasdar (1986) reported that nitrogen application leads to increased above ground vegetative growth rate, net assimilation rate and leaf area index. Nitrogen also mediates the utilization of potassium, phosphorus and other elements in plants and the optimum amounts of these elements in the soil cannot be utilized efficiently if nitrogen is deficient in plants (Brady, 1984). Onyango (2002) reported that nitrogen promotes growth of leafy vegetables through its effect on cell division, expansion, and elongation.

Plants under low levels of nitrogen develop an elevated root: shoot ratio with shortened lateral branches. Higher levels of nitrate (NO_3) inhibit root growth and leads to a decrease in

the root: shoot ratio (Ng'etich *et al.*, 2013). Many researchers have established that nitrogen deficiency resulted in poor growth rate, earlier maturity and shortened vegetative growth phase in various crops (Jasso-Chaverria *et al.*, 2005; Wolf, 1999; Cao & Tibbttts, 1998). High nitrate rates in soil causes osmotic stress, which causes oxidative damage to many important cellular components, such as lipids, protein, DNA and RNA (Wei *et al.*, 2009). Claussen *et al.* (2006) reported that when some plants are exposed to elevated ammonium concentrations they accumulate many low molecular mass osmolytes, such as sugars, proline, organic acids, polyamines and others to become more tolerant.

2.3.2.4 Yield

Nitrogen fertiliser is an essential component of any system in which the aim is to maintain good yield (Law & Egharevba, 2009; Modhej *et al.*, 2008). Ojo & Olufolaji (1997) observed increases in yield of *Corchorus* treated with 70 kg N ha⁻¹ compared to plants with no nitrogen. Various researchers have shown that yield increases in cowpea due to nitrogen fertilizer application (Oliveira *et al.*, 2001; Chowdhury *et al.*, 2000; Mazaheri & Hoseini 2003; Abayomi *et al.*, 2008; Gohari *et al.*, 2010). Cowpea has been reported to have a lower soil fertility requirement than many other crops (van Rensburg *et al.*, 2007) because, as a legume, it has ability to fix nitrogen from the atmosphere (Schippers, 2000). Non-heading Chinese cabbage requires adequate availability of nitrogen for optimum growth (Tshikalange & Van Averbek, 2006).

Van Averbek *et al.* (2007b) observed increases in yield up to 200 kg N ha⁻¹ followed by a decline in Chinese cabbage treated nitrogen concentration between 0 kg up to 300 kg N ha⁻¹. Similarly Olaniyi *et al.* (2008) observed that grain yield and quality of the amaranth increased from 0 up to 45 kg N ha⁻¹ and declined thereafter while dry matter yield of the leaves and stem increased from 0 up 60 kg N ha⁻¹. Fresh and dry leaf yield of vegetables tend to increase with increases in applied nitrogen fertilizer up to optimum level followed by decline in fresh and dry matter production (Greef, 1994).

Researchers have established variations in the effect of nitrogen fertilizer on the levels of mineral elements in vegetables (Chweya, 1993; Ojeniyi & Adeniyi, 1999; Tarfa *et al.*, 2001; Safaa & Abd El Fattah, 2007). The variations arise due to differences in cultivars and environmental factors, such as season, temperature, day length and light intensity as well as

soil chemical and physical properties Takebe *et al.*, 1995; Chweya & Nameus, 1997; Grazyna & Waldemar, 1999; Singh, 2005; Aliyu & Morufu, 2006; Rickman *et al.*, 2007).

2.3.3 Crop response to planting densities

Ideal plant populations can lead to optimum yields, whereas too high or too low plant populations can result in relatively lower yields and quality. The effect of plant densities on crops is discussed below.

2.3.3.1. Stomatal conductance

Stomata occupy a central position in the pathways for both water loss and CO₂ uptake of the plants (Jones, 1998). Higher plant densities have been reported to cause competition for nutrients, physical spaces and water (Law & Egharevba, 2009). If competition for water is high, this will induce stress. Under drought stress, plants will close stomata leading to a decrease in the flow of CO₂ into the leaves, followed by a decline in net photosynthesis, and hence reduced plant growth.

2.3.3.2 Chlorophyll content and Chlorophyll Fluorescence (CF)

Sumeet *et al.* (2009) reported that chlorophyll was strongly related to nitrogen concentration in the soil of spinach beet. Since higher densities have been reported to lead to higher competition for nutrients, physical space and water (Law & Egharevba, 2009). Increased competition for nitrogen at higher plant densities may lead to reduced chlorophyll content. Higher plant densities may result in plants with lower chlorophyll content caused by increased mutual shading at such high plant densities. Lower plant densities may result in higher chlorophyll content due to less competition for nutrients. Aminifard *et al.* (2012) reported no variations in chlorophyll content of sweet pepper (*Capsicum annum* L.) due to increased plant densities.

2.3.3.3. Growth

As plant population density increases, competition for available water, mineral nutrients and light increases (Samih, 2008). This competition for available nutrients leads to reduced growth (Yarnia, 2010). In amaranth, it was observed that increase in density from 10 up to 30 plants m⁻² increased plant height significantly while increasing plant density further up to 40 plants m⁻² decreased plant height. On the other hand, plants have been shown to increase height under higher densities as they try to intercept higher radiation (Yarnia, 2010).

2.3.3.4. Yield.

Two general concepts are frequently used to explain the relationship between plant densities and yield. Firstly, maximum yield can be attained only if the plant community produces enough leaf area to provide maximum radiation interception during reproductive growth. Secondly, equidistant spacing between plants will maximize yield because it minimizes inter plant competition (Egli, 1988). Plant density and planting pattern are major causes of inability to achieve yield potential in irrigated and dryland production (Bell *et al.*, 1991).

Yield per unit area tends to increase as plant density increases up to a point and then declines (Akintoye *et al.*, 2009). The increase can be due to the fact that competition is less in low planting density than in high planting density (Law & Egharevba, 2009). The decline is probably caused by increased competition. The competition might be high for nutrients, physical space and water as density increases (Law & Egharevba, 2009). Also Carmi *et al.* (2006) reported that increasing density, decreases water availability for plants and so plants encounter water stress and thus yield is decreased. Lower plant densities have also been reported to cause low yields. Law & Egharevba (2009) reported that lower planting densities per unit area produced more vigorous crops than at higher population densities, but this sometimes cannot compensate for a reduced number of plants per unit area. In such cases the total yield increases with higher planting densities. This was probably due to increase in the number of plants per unit area, which might contribute to the production of extra yield per unit area leading to high yield (Law & Egharevba, 2009).

Effect of spacing on seed yield of cowpea has been widely studied (Ezedinma, 1974; Subramanian *et al.*, 1977; Shivashankar *et al.*, 1979; Korode & Odulaja, 1985). There is limited information describing its response to plant density as a leafy vegetable. Coetzee (1995) concluded that a plant density of 111 111 (0.9 m x 0.1 m) plants ha⁻¹ for upright growth types and 66 000 (1.5 m x 0.10 m) plants ha⁻¹ for both semi-vining and vining growth types was optimum. According to Schippers (2002), mustard spinach can be planted at a spacing of 0.50 m between rows and 0.30-0.45 m in the row (44. 444 - 66.666 plants ha⁻¹). Grubben & Denton (2004), however, suggested 0.30 - 0.50 m between rows and 0.20-0.40 m in the row (50 000 -160 000 plants ha⁻¹). Peirce (1987) and Matsumura (1981) recommended a plant density of 44 000 plants ha⁻¹ to 55 000 plants ha⁻¹. The optimum plant density and planting pattern at one site may not apply at other locations because regional variations in weather and soil (Azam-Ali *et al.*, 1993).

2.3.4. Crop response to planting date selection

Choosing an appropriate sowing date for a crop is one of the most important factors in its production when it is cultivated. Appropriate sowing date of a crop is a date when the plants can be well established and their susceptible growth stages do not coincide with adverse environmental conditions (Seghatoleslami *et al.*, 2013). The major crop responses to planting date are discussed below.

2.3.4.1. Stomatal conductance

Knowing the best sowing date helps to maximise yield. Different sowing dates imply that growing crops will face different soil temperatures and moisture levels, have different chances of being affected by a late frost, and that their growth cycles will last different lengths of time (Troinani *et al.*, 2004). If a crop is sown or planted in such that its growth stages coincide with low moisture levels or drought stress, plants will close stomata to reduce water losses. This will lead to a decrease in the flow of CO₂ into the leaves; hence there will be a decline in net photosynthesis (Modi & Mbahudhi, 2013).

2.3.4.2 Chlorophyll content and Chlorophyll Fluorescence (CF)

Chlorophyll level in plants is an important factor in maintaining their photosynthesis capacity (Jiang & Huang, 2001). El-Khoby (2004) reported that a delayed sowing date decreased the chlorophyll content of rice. In a study on the effect of five sowing dates (April 19, April 30, May 10, May 20 and May 31) on rice, Basyouni Abou-Khalifa (2010) found that the highest leaf chlorophyll content was obtained at sowing date of May 10 at the stages of heading and milky as well as at sowing date of April 30 at the stages of dough and maturity. In a study on the effect of four sowing dates on beans, Biswas *et al.* (2002) stated that the increase in leaf photosynthesis rate was accompanied with the increase in stomatal conductance. This could be due to a planting date coinciding with high levels of moisture.

2.3.4.3 Growth

Early planting is crucial to achieve an early yield and consequently a high price. However, there is risk of environmental stress associated with exposing young plants to cool weather after field setting. The sooner the planting date, the stronger the stress (Palada *et al.*, 1987). Each plant species has an optimum temperature when growth is rapid, while lower non-freezing temperature allows plant growth, but at a considerably reduced rate. Moreover, the response of plants to unfavorable temperature results in modification of many physiological and biochemical process leading to changes in the chemical composition (Nam *et al.*, 1995). The degree of these changes is mainly dependant on the temperature level, the temperature exposure duration and the stage of plant development. Planting date influences temperature and photoperiod (Roberts & Summerfield, 1987; Squire, 1990). Low temperature has been reported to reduce the growth of Chinese cabbage seedlings (Wiebe, 1990) and to influence their chemical composition (Sasaki *et al.*, 1996).

Planting date should be selected in such a way as to avoid climatic factors that cause stress during the growth process in plants. Plant height reduction by delayed planting is related to changes in temperature and day length during the growth season. Chinese cabbage (non heading) seedlings grow optimally at a temperature of 22 °C (Ope a, Kuo and Yoon, 1988). Slow and incomplete emergence due to varying planting dates has been reported in cowpea (Ismail *et al.*, 1997). Delay in planting was reported to cause plant height reduction in *amaranthus* (Yarnia, 2010).

2.3.4.4 Yield.

Studies have shown that higher temperature affects productivity of many crops which is often influenced by planting date (Hiler *et al.*, 1972; Hall, 1992). To achieve an economic yield an optimum planting date has to be established. Planting seeds of *Amaranthus* on a suitable date maximizes the growth duration and complete seed maturation which leads to maximum yield and reduces the risk of unfavourable environmental conditions on grain and forage quality (Yarnia, 2010). When the proper planting date is selected, a cultivar with a suitable growth period can flower and produce seed in a proper time (Yarnia, 2010). If a plant starts to flower early, maximum growth and yield will not be achieved. Early sowing can also increase risk of late freezing. Barros *et al.* (2004) expressed that early planting date increased amaranth leaf area duration and water absorption during the critical period between flower bud appearances to flowering..

Amaranth being a C4 plant grows optimally under warm conditions (day temperatures above 25°C and night temperatures not lower than 15°C, bright light and adequate availability of plant nutrients (van den Heever & Coertze, 1996; Schippers, 2000). Amaranth is photoperiod sensitive and starts to flower as soon as the day length shortens. Yarnia (2010) reported that delay in amaranthus planting from 20 April to 5 May, 20 May, 3- June and 18 June increased harvest index as 0.52, 2.68 and 1.84 and 10 %, respectively. Studies on *Amaranthus cruentus* L., *A. hypochondriacus* L. and *A. Mantegazzianus* sown at 15 day intervals during November, December and January showed that last planting dates yield was low because the plant life cycle was limited with temperature and lighting conditions (De Troiani *et al.*, 2004). Planting date has been reported to be the main important yield factor in the fresh leaf yield of Chinese cabbage (Juma *et al.*, 2005). Studies on small holder farmers in Limpopo reported that non heading Chinese cabbage is mainly planted during April and May (Van Averbeke *et al.*, 2007a). This earlier planting has the advantage of superior market conditions, but the disadvantage that it increases the incidence of pests (van Averbeke *et al.*, 2007a). Delayed planting date, poor stand establishment and drought have been identified as factors that limit growth of legumes (Sullivan, 2003). *Corchorus* prefers warm, humid conditions and performs well in high temperatures, 30°C during the day and 25°C at night (van Rensburg *et al.*, 2007). Furthermore, growth of *Corchorus* slows down considerably when the temperature drops below 15°C. According to Green *et al.* (1985), on maize he

reported that results of planting dates may vary and can be inconsistent between seasons and sites and that it is not unusual for late planted crops to out-yield the optimum planting.

2.4. EFFECT OF CULTIVATION PRACTICES ON QUALITY ATTRIBUTES OF FRESH PRODUCE

Abbot (1999) defined the term 'quality' as the degree of excellence of a product or its suitability for a particular use. Quality of produce encompasses sensory properties (appearance, texture, taste and aroma), nutritive values, chemical constituents, mechanical properties, functional properties and defects (Hussin *et al.*, 2010). It is important to know the quality attributes of vegetables as they may lead to a competitive advantage for the farmers (Hussin *et al.*, 2010). Indigenous leafy vegetable have potential for commercialization. Thus, it is vital for production of these vegetables to be based upon objective quality criteria of fresh produce so as to improve on the yields for marketing.

Different quality attributes of perishable crops include freshness, healthiness, flavour, nutrition, safety, appearance, price, environmental effect, certification, freshness, taste/flavour, cleanliness, high quality and good value (Groff *et al.*, 1993; Govindasamy *et al.*, 1997; Wolf, 2002).

Based on the study conducted by Hussin *et al.* (2010) the following was established:

- quality attributes for fruits such as watermelon include: absence of defects, absence of blemishes, freshness, ripeness, sweetness, nutritional values, flavours, absence of pesticides, absence of preservative, cleanliness, naturally ripened, size, weight, shape, colour, succulence and juiciness;
- product attributes (quality attributes) for leafy vegetable such as mustard include: colour, absence of defect, absence of blemishes, freshness, ripeness, flavour, nutritional value, absence of pesticides, absence of preservatives, cleanliness and naturally ripened shape is also important; and
- product attributes (quality attributes) for non-leafy vegetables such as long bean include: colour, absence of defect, absence of blemishes, freshness, ripeness, flavour, nutritional

values, absence of pesticides, absence of preservative, cleanliness and naturally ripened, size and shape.

Farmers need to realise that quality of fresh produce is a very important attribute that is always required by customers. Cultivation practices such as irrigation, nitrogen, planting density and planting date should be known with considerable precision as they have some impact on the quality attributes. According to Beletse *et al.* (2012), yields obtained under water stressed conditions may lack the quality needed to market the produce. Sowing at optimal seed rates results in optimal plant population density, reduced seed costs and lodging as well as ameliorating disease problems (Hosseini *et al.*, 2001). Nitrogen influences crop quality (Ng'etich *et al.*, 2013). It stimulates vegetative growth resulting in large stems and leaves. Early planting is crucial to achieve an early yield and consequently a high price.

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CHAPTER 3

Growth, physiology and yield responses of *Amaranthus cruentus*, *Corchorus olitorius* and *Vigna unguiculata* to plant density under commercial scale production

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3.1 ABSTRACT

The aim of the study was to evaluate the effect of plant density on growth, physiology and yield responses of *Amaranthus cruentus*, *Corchorus olitorius* and *Vigna unguiculata* under drip irrigated commercial scale production. Field trials were conducted at the Agricultural Research Council -Vegetable and Ornamental Plant Institute, Gauteng Province, South Africa over two summer seasons, 2011/2012 and 2012/2013. The trials were laid out in a randomized complete block design (RCBD) with three replications in the 2011/2012 first season and nine replications in the 2012/2013 season. The planting materials were three (3) African leafy vegetables, *Amaranthus cruentus*, *Corchorus olitorius* and *Vigna unguiculata*. The spacing treatments were plant density of 100 000, 66 666 and 50 000 plants ha⁻¹. Parameters measured included chlorophyll content index (CCI), chlorophyll fluorescence (CF), stomatal conductance (SC), leaf number, leaf area index (LAI) and biomass. Plant density of 50 000 plants ha⁻¹ resulted in significantly (P<0.05) higher LAI, CCI and biomass per plant in *A. cruentus* and *C. olitorius*. A higher leaf number was recorded in *V. unguiculata* grown at 66 666 plants ha⁻¹ relative to 50 000 to 100 000 plants ha⁻¹. In *V. unguiculata* LAI and CCI were not significantly affected by varying plant densities. SC and CF of *A. cruentus*, *C. olitorius* and *V. unguiculata* were not significantly affected (P>0.05) by variations in plant density. Total yield in *A. cruentus*, *C. olitorius* and *V. unguiculata* was

significantly ($P < 0.05$) higher at 100 000 plants ha^{-1} relative to 50 000 and 66 666 plants ha^{-1} . In *A. cruentus* and *C. olitorius*, higher leaf quality parameters (CCI, plant height, leaf number, biomass per plant and LAI) indicate that these crops can perform better at lower densities of 50 000 plants ha^{-1} . Therefore, using 50 000 plants ha^{-1} is suitable for commercial production of *A. cruentus* and *C. olitorius*. Even though leaf number was low in *V. unguiculata* planted at 100 000 plants ha^{-1} compared to other densities in the current study, other quality parameters such leaf size measured in terms of LAI and leaf colour measured in terms of CCI were not compromised. Furthermore, higher yields of *V. unguiculata* were obtained at 100 000 plants ha^{-1} . Therefore 100 000 plants ha^{-1} is recommended for commercial production of *V. unguiculata*.

Keywords: *Amaranthus cruentus*, *Corchorus olitorius*, *Vigna unguiculata*, planting density

3.2. INTRODUCTION

Wehmeyer & Rose (1983) identified more than 100 different species of plants that are being used as leafy vegetables in South Africa. Out of these 100 species, seven major groups of leafy vegetables species are of particular importance in South Africa (van Rensburg *et al.*, 2007). *Amaranthus cruentus* (pig weed), *Corchorus olitorius* (Jews mallow) and *Vigna unguiculata* (cowpea) are among the major seven groups of traditional leafy vegetables. *A. cruentus*, *V. unguiculata* and *C. olitorius* have been discovered to be highly nutritious (Leung *et al.*, 1968; Makus, 1984; Sussan & Anne, 1988; Stallknecht & Schaeffer, 1993, Enwere *et al.*, 1998). Marketing of *A. cruentus* and *C. olitorius* as leafy vegetables has been observed in South African informal markets (Whitebread, 1986; Weinberger & Pichop, 2009). Street vendors obtain these species by collecting them as weeds from fields and vegetable gardens. The development of *V. unguiculata* as a leafy vegetable has not been a major research objective. Research efforts have continued to focus on improvements of the grain and/or the entire herbage for animal feed (Singh *et al.* 2003).

Planting density is one of the important yield determinants for successful commercial production (Aminifard *et al.*, 2012). At lower plant densities crops produce vigorous growth due to less competition for resources while at higher plant densities crop growth is limited

due to competition for resources (Law & Egharevba, 2009). Crop yield per unit area tends to increase as plant density increases up to a point and then declines (Akintoye *et al.*, 2009). Increase in yield as density increases could be due to the number of plants per unit area or ability of plants to maximize resources. However, further increase in density may increase competition for water and nutrients resulting in limited vegetative growth and low yields (Knave, 1988). In certain crops, cultivar choice affects plant population. In *V. unguiculata* cultivars with upright growth forms have a higher plant density than vining or semi-vining types because the upright forms perform much better in narrow rows (Coetzee, 1995; Weber *et al.*, 1996).

Previous studies showed variation in crop growth and yield of *C. olerius* and *A. cruentus* in response to plant densities. In *C. olerius* optimum growth, leaf and seed yields were observed at 40 000 plants ha⁻¹ (Schippers, 2000). On the contrary, Madakadze *et al.* (2007) observed highest leaf and seed yield of *C. olerius* at 200 000 plants ha⁻¹ compared to 40 000 and 66 600 plants ha⁻¹ under rain fed with supplementary irrigation. Studies in *A. cruentus* showed that a plant density of 66 666 plants ha⁻¹ resulted in higher leaf number, dry matter, biological and grain yield in rain fed trials (Olofintoye *et al.*, 2011). Lower yields in *A. cruentus* were recorded at 40 000 plants ha⁻¹ and 50 000 plants ha⁻¹. In contrast, Yarnia (2010) observed an increase in growth and yield with an increase in plant density from 100 000 plants ha⁻¹ up to 300 000 plants ha⁻¹ in *Amaranthus* plants. Further increases in plant density up to 400 000 plants ha⁻¹ reduced growth and yield. The differences in crop yield in the former and latter studies could be due to differences in climate, among other factors, as the studies were undertaken in different agro ecological regions. The hypothesis was that plant density of 66 666 plants ha⁻¹ has no effect on growth, physiology and yield of *A. cruentus*, *C. olerius* and *V. unguiculata*. The objective of this study was to investigate growth, physiology and yield responses of *A. cruentus*, *C. olerius* and *V. unguiculata* to varying levels of plant densities under drip irrigation in South Africa.

3.3 MATERIALS AND METHOD

3.3.1 Plant material

Seeds of *A. cruentus* and *C. olerius* were obtained from the Agricultural Research Council (ARC), and the Vegetable and Ornamental Plant Institute (VOPI) seed bank. *V. unguiculata*

(Bechuana white, a runner type) was obtained from Hygrotech Seed (Pty) Ltd. There was no treatment done to the seeds.

3.3.2. Description of trial site

Field trials were carried out during the 2011/12 (season 1) and 2012/13 (season 2) summer seasons at ARC-VOPI Roodeplaat (25°35' S; 28°21' E; 1164 m a.s.l), tropical climate Pretoria, South Africa. The average seasonal rainfall (November to April) at Roodeplaat is 500 mm and is highly variable with maximum precipitation in December and January. The total amount of rainfall received at Roodeplaat during 2011/12 and 2012/13 summer seasons (November-February) was approximately 400 mm and 350 mm respectively (ARC, Meteorological Station). Supplementary irrigation supplied was 548 mm during 2011/12 and 435 mm during 2012/13 summer season. The average daily maximum and minimum temperature at Roodeplaat are 8°C and 34°C in summer (November – April). This climate data was obtained from the Agricultural Research Council–Institute for Soil, Climate and Water [ARC–ISCW] automatic weather stations network. The soil type in the field is classified as Hutton clay loam (South African soil taxonomic system) and its composition is shown in Table 3.1.

Table 3.1 Physical and chemical characteristics of the soil in the experimental field at ARC, Roodeplaat.

Parameter	2011/12 summer season	2012/13 summer season
P (mg kg ⁻¹)	17.3	13.6
K (mg kg ⁻¹)	227	203
Ca (mg kg ⁻¹)	756	774
Mg (mg kg ⁻¹)	234	289
Na (mg kg ⁻¹)	15.5	35.3
Exchangeable cation Ca (%)	56.7	51.6
Exchangeable cation Mg (%)	34	28.9
Exchangeable cation K (%)	7.8	16.1
Exchangeable cation Na (%)	1.5	3.4
pH	6.6	6.6
Clay (%)	25	25
Silt (%)	6	6
Sand (%)	69	69

3.3.3. Experimental design and treatments

The trial was laid out in a randomized complete block design (RCBD) with three replications in 2011/2012 and nine replications in the 2012/2013 summer season. The treatments were three plant density levels at 100 000 (0.5 m inter row x 0.2 m intra row), 66 666 (0.5 m inter row x 0.3 m intra row) and 50 000 (0.5 m inter row x 0.4 m intra row) plants ha⁻¹. The three leafy vegetables used were *Amaranthus cruentus*, *Corchorus olitorius* and *Vigna unguiculata*. The plot size was 72 m² (12 m x 6 m) for 2011/12 and 36 m² (9 m x 4 m) for 2012/13 summer seasons. The plot size was reduced in the second season because the number of replications was increased to better estimate the true effects of treatments and further strengthen the experiment's reliability and validity. In both seasons each plot had three (3) crops; each crop occupied three (3) ridges per plot. The crops were established on double rows per ridge at an interrow spacing of 0.5 m for both seasons. The outer rows in the ridge were meant to reduce border effects while inner rows were used as experimental plants. The drip irrigation system comprised of an electric powered pump, control unit (solenoid valves and controller), filter, water meters and polyethylene drip tape. The drippers were laterals at a spacing of 1 metre and connected to a main line with a flow metre, a valve and a pressure gauge at the entrance of the plots to control the operating pressure and measure the irrigation volume. Thin-wall drip tape with emitters at 0.3 m intervals/spacing and a drip rate of approximately 2.0 hr⁻¹ at an operating pressure of 150 kPa was placed on the centre of the raised beds. Irrigation scheduling for 2011/12 and 2012/13 summer seasons was based on daily reference evapotranspiration (ET_o) which was obtained from an automatic weather station (AWS) at the experimental site. Reference evapotranspiration (ET_o) was then adjusted by a crop factor (K_c). Plants were irrigated at a frequency of thrice a week throughout the trial. The AWS calculates ET_o according to FAO Penman-Monteith method (Allen *et al.*, 1998).

3.3.4. Agronomic practices

Soil samples were collected from 0 - 0.3m and 0.3- 0.6m depths from the field prior to land preparation and submitted for soil fertility analyses at the Agricultural Research Council - Institute of Soil, Climate and Water (ARC-ISCW). Land preparation included ploughing, disking, rotovating and ridging using a tractor to achieve a fine seedbed. The nitrogen used was limestone ammonium nitrate (LAN) which consists of 28% nitrogen. Nitrogen was

applied based on results of soil fertility analysis at 88 kg N ha⁻¹ and 125 Kg N ha⁻¹ for 2011/12 and 2012/13 summer seasons, respectively for both crops. Nitrogen was applied by banding in three split applications. The first application was at transplanting/sowing (50%), the second at four weeks after transplanting/sowing (25%) and the last 25% at 8 weeks after transplanting/sowing. Double super phosphate was applied at 88 kg P ha⁻¹ (10.5 % P) and 97 P kg ha⁻¹ at planting for 2011/12 and 2012/13 summer season based on results of soil fertility analysis. The crop that was previously planted in the experimental field was non heading Chinese cabbage. Seedlings of *Amaranthus cruentus* and *Corchorus olitorius* were grown in 250 cavity polystyrene trays filled with a commercial growing medium, Hygromix® obtained from Hygrotech Seed Pty. Ltd., South Africa.

Seedlings were then covered with vermiculate to minimize water losses from above the surface. Seedlings were transplanted at four weeks after sowing. Transplanting was done early in the morning to prevent transplanting stress. *Vigna unguiculata* was sown directly using seed at a rate of one (1) seed per station based on a previous germination test carried out at the experimental site. Routine weeding and scouting for pests and diseases was done to ensure best management practices for the trials as shown in Figure 3.1.



Figure 3.1. Weed free *Amaranthus* field trial at Roodeplaat during 2012 growing season.

3.3.5 Data collection

Weather data (maximum and minimum air temperature, maximum and minimum relative humidity, rainfall, wind speed and reference evapotranspiration (ET_0)) for the duration of the trials were monitored from an automatic weather station (AWS) situated within a 100m radius from the field trials. Plant height, leaf number, stomatal conductance, chlorophyll content index (CCI), chlorophyll fluorescence (CF) and leaf area index (LAI) were measured starting from four weeks after transplanting (WAT) and weekly thereafter.

A total of three (3) plants per replication was tagged for data collection for growth and physiology parameters. The outer rows were excluded for data collection to prevent border effects and data was only collected from the inner rows. All measurements were done on leaves that had at least 50% green leaf area. Plant height was measured using a measuring tape from the ground level to the tip or apex of the tallest stem. Leaf number was obtained by visual counting of green fresh leaves. Stomatal conductance measurements were done on the abaxial leaf surface using a steady state leaf porometer (Model SC-1, Decagon Devices, Inc., USA). Chlorophyll content index was determined on the adaxial surface using the CCM-200 *Plus* chlorophyll content meter (Opti-Sciences, Inc., USA). Leaf area index was measured using the LAI 2200 Canopy Analyzer (Li-Cor, USA). Photosynthetic efficiency as indicated by chlorophyll fluorescence (CF) was measured using a Handy Plant Efficiency Analyzer (PEA) chlorophyll fluorometer (Hansatech Instruments, U.K.). The measurement of CF was only done during the second season because the equipment was not available during the first season. Leaves were initially dark adapted (30 min) before CF measurements were taken. Values of F_v/F_m (the measurement of quantum yield potential of photosynthesis, or maximal photochemical efficiency of PSII) were recorded from the PEA and used for analysis. All measurements were done in-between irrigation weekly throughout the study.

Harvesting commenced at six (6) weeks after transplanting (WAT) or sowing and every two weeks thereafter. The sample size per yield was 1 m² for each replicate for 2011/12 and 2012/2013 summer seasons. During each harvest, the yield of *C. olitorius* and *A. cruentus* was determined by cutting the mass of the above ground portion of the plant, leaving 0.2 m of plant height above ground level. In *V. unguiculata*, harvesting was done by picking three to

four fresh marketable leaves including their tender stems towards the growing tip of each runner, leaving the first and second growing leaves from the tip. Marketable leaves in *V. unguiculata* were defined as young or green tender leaves. The harvested portion was then partitioned into leaves and stems. In order to obtain accurate results, the plants were weighed immediately after harvest to avoid reduction in mass due to loss of water. Biomass per plant was obtained by randomly sampling three plants per replicate at each harvest. Dry matter content was obtained by oven drying the leaves or stems at 70°C for 48 hours. Yield per hectare was obtained by conversion from measurements taken at 1 m² per replicate.

3.3.6. Statistical Analyses

All data were subjected to statistical analysis using analysis of variance in GenStat® (Version 14, VSN International, UK) statistical package. Means were compared by Duncan's multiple range test (DMRT) at the 5% level of significance.

3.4 RESULTS AND DISCUSSION

3.4.1 Growth parameters

3.4.1.1 Plant height and leaf number

Plant height in *A. cruentus* was not significantly ($P>0.05$) affected by plant density for all seasons although during the 2012/2013 season plant height increased with a decrease in planting densities (Table 3.2). Although not significant for 2011/2012 season, leaf number per plant at lower plant densities of 50 000 plants ha⁻¹ was higher relative to densities of 66 666 and 100 000 plants ha⁻¹. In the 2012/2013 season leaf number increased with approximately from 50 000 to 66 666 and then declined at 100 000 plants ha⁻¹. The difference in trends observed between the two seasons could be attributed to variation in fertiliser recommendations between the between two seasons among other factors. In *C. olitorius* plant height increased with a decrease in plant density during the 2011/2012 season, while during the 2012/2013 season, plant height tended to increase from 50 000 to 66 666 plants ha⁻¹ and then decline at 100 000 plants ha⁻¹. However, the difference observed in plant height of *C. olitorius* was not significant ($P>0.05$) for all seasons (Table 3.2).

Table 3.2. Effect of plant density on growth of three selected African leafy vegetables for two seasons at ARC, Roodeplaat.

Crops	Parameters	Plant density (plants/ha)							
		2011/12 summer (Season 1)				2012/2013 summer (Season 2)			
		50 000	66 666	100 000	LSD _(0.05)	50 000	66 666	100 000	LSD _(0.05)
<i>A.cruentus</i>	Plant height (m)	0.71 ^a	0.71 ^a	0.73 ^a	Ns	1.06 ^a	1.00 ^a	0.87 ^a	Ns
	Leaf number/plant	153 ^a	99 ^a	111 ^a	Ns	202 ^a	207 ^a	184 ^a	Ns
	LAI	2.3 ^a	2.2 ^a	2.6 ^a	Ns	3.3 ^a	3.1 ^a	2.7 ^b	0.38
<i>C.olitorius</i>	Plant height (m)	1.03 ^a	1.00 ^a	0.97 ^a	Ns	0.97 ^a	1.08 ^a	0.98 ^a	Ns
	Leaf number/plant	55 ^b	154 ^a	161 ^a	27	141.3 ^a	166.3 ^a	160.3 ^a	Ns
	LAI	2.0 ^a	1.7 ^a	1.8 ^a	Ns	3.1 ^a	2.4 ^b	2.9 ^b	0.44
<i>V.unguiculata</i>	Leaf number	62 ^a	69 ^a	112 ^a	Ns	168.8 ^b	234.3 ^a	194.8 ^b	50.2
	LAI	1.7 ^a	1.6 ^a	2.1 ^a	Ns	2.9 ^a	3.2 ^a	3.1 ^a	Ns

*Means followed by the same letters within a row are not significantly different according to Duncan's multiple range tests at P 0.05.

Leaf number in *C. olitorius* increased significantly ($P < 0.05$) from 50 000 to 66 666 plants ha^{-1} and remained unchanged at 100 000 plants ha^{-1} during 2011/2012 season (Table 3.2). The same trend was observed during the 2012/2013 season although the difference were not statistically significant ($P > 0.05$) (Table 3.2). There was no significant ($P > 0.05$) difference in leaf number in *V. unguiculata* during the 2011/2012 season (Table 3.2). However, during the 2012/2013 season leaf number increased significantly from 50 000 to 66 666 plants ha^{-1} . A further increase in plant density to 100 000 plants ha^{-1} significantly ($P < 0.05$) reduced leaf number. The results of leaf number doubled during the second season possible due to the fact that some measurements did not match growing periods, matching growing periods was difficult under field conditions.

Similar to results of the current study in *A. cruentus* during 2012/2013, Yarnia (2010) observed increases in plant height with decreases in plant densities in *Amaranthus*. Yarnia (2010) attributed the reduction in plant height at higher densities to competition for nutrients. Taller plants observed at 100 000 plants ha^{-1} relative to 50 000 and 66 666 plants ha^{-1} during 2011/2012 season, could be attributed to competition for light among other factors. As plants compete for light they elongate (Yarnia, 2010). In contrast to results of 2012/2013 season, Olofintoye *et al.* (2011) observed an increase in plant height with an increase in density from 40 000, 50 000 up to 66 666 plants ha^{-1} in *A. cruentus* treated with 100kg ha^{-1} phosphate fertiliser. The variations between the current study and that of Olofintoye *et al.* (2011) could be attributed to plant density levels among other factors. There is a possibility that if plant density levels were increased above 66 666 plants ha^{-1} , the response in plant height could be similar to the current study. Although in *C. olitorius* statistically analysis showed no significant differences in plant height in response to plant density in the current study, the trend observed concurs with the findings of Okunsanya *et al.* (1990) where plant height of *C. olitorius* decreased with increases in plant density.

In contrast, Makinde *et al.* (2009) observed an increase in plant height of *C. olitorius* in response to increases in plant densities. Reduction in plant height with increases in density observed in the current study could be attributed to competition for water and nitrogen (Law & Egharevba, 2009) while increases in plant height with increases in plant density could be

due to competition for light. As plants compete for light they increase in height to intercept higher radiation (Yarnia, 2010).

The difference in leaf number in *C. olitorius* concurs with Makinde *et al.* (2009) who observed a decrease in leaf number with increases in plant densities. This may imply that at above 66 666 plants ha⁻¹ population density reaches some threshold at which resources become limited. Weber (1966), Mohammed (1984), Alege & Mustapha (2007) attributed the reduction in leaf number in response to increase in density to the reduction in the number of branches in *V. unguiculata*. Increase in leaf number is an important quality attribute. Often, in markets where leafy vegetables are sold, they are not sold on a mass basis but rather using a bunch (a certain number of leaves). In such instances, this suggests that at 100 000 plants ha⁻¹ yield (in this case leaf number) may be compromised in *V. unguiculata*.

3.4.1.2 Leaf Area Index (LAI)

Planting density did not significantly ($P>0.05$) affect LAI in *A. cruentus* during the 2011/2012 season (Table 3.2). However, there were significant ($P<0.05$) differences among planting density levels during the 2012/2013 season (Table 3.2). Plant density of 100 000 plants ha⁻¹ significantly reduced LAI with about 20% compared to 50 000 and 66 666 plants ha⁻¹. In *C. olitorius* lower densities (50 000 plants ha⁻¹) showed a relatively higher LAI during the 2011/2012 season; however, there were no significant ($P>0.05$) differences observed that were not significant ($P>0.05$) (Table 3.2). During the 2012/2013 season significant differences ($P<0.05$) were observed by varying plant density levels (Table 3.2). Lower plant densities of 50 000 plants ha⁻¹ had significantly ($P<0.05$) higher LAI approximately 30 % in comparison to higher densities of 66 66 and 100 000 plants ha⁻¹. Results of *V. unguiculata* showed that, for both planting seasons, LAI was not significantly affected ($P>0.05$) by planting densities (Table 3.2).

Results of LAI in *A. cruentus* and *C. olitorius* concurred with studies by Lazim (1973) who showed a decrease in LAI in response to increasing plant densities. Studies in *Amaranthus* have also shown this decrease in leaf area with an increase in plant densities (Yarnia, 2010). This could be due to competition for resources at high densities of 100 000 plants ha⁻¹. Water stress has been shown to reduce leaf area (Chen & Dai, 1996). Reduction of leaf area is a drought avoidance mechanism, which reduces plant water use rate and hence conserves water

during periods of drought (Ludlow & Muchow, 1990; Jones, 1992). In *A. cruentus* plants were generally shorter with less leaves but with higher LAI at 50 000 plants ha⁻¹. Reduction in leaf size in terms of LAI at 100 000 plants ha⁻¹ might have compromised the quality of leaf crops which is important in commercial production. Small leaves can reduce marketability of *A. cruentus*. Lack of variation of LAI in *V. unguiculata* could mean that competition for nutrients and water due to varying densities was less. Therefore, the sizes of the leaves were not compromised.

3.4.2. Physiological parameters

3.4.2.1. Stomatal conductance and chlorophyll fluorescence (CF)

There were no significant ($P>0.05$) differences in stomatal conductance in response to varying plant density in *A. cruentus*, *C. olitorius* and *V. unguiculata* for both seasons (Table 2). Chlorophyll fluorescence (CF) showed no significant ($P>0.05$) response to varying plant density in *A. cruentus*, *C. olitorius* and *V. unguiculata* (Table 3.3). Therefore, this could indicate that plants were not subjected to any stress. Stomatal conductance is mostly related to soil water availability (Mabhaudhi *et al.*, 2013a, 2013b) while CF is related to the intactness of the photosynthetic apparatus (Mabhaudhi, 2009). The lack of effect in SC and CF due to varying plant densities may be attributed to the fact that experiments were conducted under optimum irrigation and fertilisation. Stomatal conductance was higher during the 2011/2012 season compared to the second season in all crops. This could be attributed to environmental effect that may have had an effect on the study.

Table 3.3. Effect of plant density on physiological parameters of three selected African leafy vegetables for two seasons at ARC, Roodeplaat.

Crops	Parameters	Plant density (plants/ha)							
		2011/12 summer (Season 1)				2012/2013 summer (Season 2)			
		50 000	66 666	100 000	LSD _(0.05)	50 000	66 666	100 000	LSD _(0.05)
<i>A.cruentus</i>	CCI	36.8 ^a	38.4 ^a	40.0 ^a	Ns	46.2 ^a	35.0 ^c	37.9 ^b	2.9
	SC (mmol m ⁻² s ⁻¹)	324.4 ^a	222.9 ^a	213.1 ^a	Ns	112.1 ^a	117.1 ^a	119.9 ^a	Ns
	CF (F _v /F _m)	-	-	-		0.745 ^a	0.721 ^a	0.723 ^a	Ns
<i>C.olitorius</i>	CCI	40.1 ^a	35.6 ^a	35.8 ^a	Ns	44.8 ^a	39.8 ^a	42.7 ^a	Ns
	SC (mmol m ⁻² s ⁻¹)	408.5 ^a	395.4 ^a	322.5 ^a	Ns	117.5 ^a	120.0 ^a	117.2 ^a	Ns
	CF (F _v /F _m)	-	-	-		0.720 ^a	0.706 ^a	0.712 ^a	Ns
<i>V.unguiculata</i>	CCI	46.1 ^a	45.3 ^a	42.3 ^a	Ns	60.6 ^a	64.1 ^a	66.3 ^a	Ns
	SC (mmol m ⁻² s ⁻¹)	482.7 ^a	424.3 ^a	446.1 ^a	Ns	117.5 ^a	120.0 ^a	117.2 ^a	Ns
	CF(F _v /F _m)	-	-	-		0.732 ^a	0.736 ^a	0.737 ^a	Ns

*Means followed by the same letters within a row are not significantly different according to Duncan's multiple range tests at P 0.05.

3.4.2.2 Chlorophyll content index (CCI)

Chlorophyll content index in *A. cruentus* was significantly ($P < 0.001$) higher at densities of 50 000 plants ha^{-1} than at 66 666 plants ha^{-1} and 100 000 plants ha^{-1} during the 2012/2013 season. During 2011/2012 increase in plant density did not significantly affect CCI (Table 3.3). Results of current study in *A. cruentus* concurs with that of Sumeet *et al.* (2009) that Chlorophyll is strongly related to nitrogen concentration in the soil. At lower plant densities of 50 000 plants ha^{-1} there is more nitrogen available to plants due to less competition than at higher densities of 66 666 plants ha^{-1} and 100 000 plants ha^{-1} . Therefore, low CCI observed at higher densities of 100 000 plants ha^{-1} may be due to competition for nitrogen at higher plant densities. Law & Egharevba (2009) reported that at higher densities competition might be high for nutrients, radiation and water. Furthermore, increased mutual shading at plant densities of 100 000 plants ha^{-1} relative to 50 000 plants ha^{-1} may account for the observation made. In *C. olitorius* there was no significant difference in CCI in response to varying densities for both seasons. However, the trend in CCI similar to *A. cruentus* was observed in *C. olitorius* in both seasons and in *V. unguiculata* during 2011/2012 season. Lack of significant differences in *V. unguiculata* among treatments may be due to the ability of plants to maximize resources even at higher densities of 100 000 plants ha^{-1} . Therefore, varying densities in *V. unguiculata* did not compromise leaf colour or greenness of the leaf.

3.4.3 Yield

3.4.3.1 Biomass accumulation per plant

In *A. cruentus* there was significant ($P < 0.05$) differences in fresh mass of stems, fresh of leaves and dry mass of leaves due to varying plant densities for both 2011/2012 and 2012/2013 seasons (Table 3.4). Increasing plant density from 50 000 to 66 666 plants ha^{-1} up to 100 000 plants ha^{-1} significantly ($P < 0.05$) reduced biomass with approximately 40% (Table 3.4). In *C. olitorius* biomass per plant increased with approximately 10% from 50 000 to 66 666 plants ha^{-1} and then declined at 100 000 plants ha^{-1} . However, the differences observed in *C. olitorius* were not significant ($P > 0.05$) during the 2011/2012 season. During the 2012/2013 season increasing plant density from 50 000 to 66 666 and 100 000 plants ha^{-1} significantly ($P < 0.05$) reduced biomass per plant. Mean separation showed that plants had similar biomass at 66 666 and 100 000 plants ha^{-1} (Table 3.4).

Table 3.4. Effect of plant density on the biomass of three selected African leafy vegetables obtained from two seasons at ARC, Roodeplaat.

Crops	Plant parts kg/plant	Plant density (plants ha ⁻¹)					
		2011/12 summer (Season 1)			2012/13 summer (Season 2)		
		50 000	66 666	100 000	50 000	66 666	100 000
<i>A. cruentus</i>	FM above ground	0.426 ^a	0.331 ^b	0.217 ^c	0.170 ^a	0.182 ^a	0.150 ^a
	FM leaves	0.214 ^a	0.165 ^{ab}	0.118 ^b	0.051 ^a	0.049 ^a	0.043 ^a
	FM stem	0.193 ^a	0.152 ^a	0.088 ^b	0.070 ^b	0.099 ^a	0.058 ^b
	DM leaves	0.027 ^a	0.021 ^{ab}	0.014 ^b	0.011 ^a	0.010 ^{ab}	0.008 ^b
	DM stem	0.031 ^a	0.023 ^a	0.010 ^b	0.011 ^a	0.010 ^a	0.009 ^a
<i>C. olerius</i>	FM above ground	0.089 ^a	0.105 ^a	0.059 ^a	0.185 ^a	0.139 ^b	0.118 ^b
	FM leaves	0.037 ^a	0.041 ^a	0.031 ^a	0.049 ^a	0.036 ^b	0.030 ^b
	FM stem	0.047 ^a	0.049 ^a	0.030 ^a	0.091 ^a	0.085 ^a	0.064 ^b
	DM leaves	0.008 ^a	0.009 ^a	0.005 ^a	0.049 ^a	0.036 ^b	0.030 ^b
	DM stem	0.008 ^a	0.010 ^a	0.006 ^a	0.019 ^a	0.013 ^b	0.008 ^c
<i>V. unguiculata</i>	FM stem + leaves	0.102 ^a	0.082 ^a	0.054 ^a	0.070 ^a	0.062 ^a	0.060 ^a
	FM leaves	0.054 ^a	0.042 ^b	0.025 ^c	0.040 ^b	0.045 ^{ab}	0.053 ^a
	FM stem	0.012 ^a	0.012 ^a	0.009 ^a	0.015 ^a	0.017 ^a	0.016 ^a
	DM leaves	0.007 ^a	0.005 ^a	0.007 ^a	0.009 ^a	0.009 ^a	0.009 ^a
	DM stem	0.001 ^a	0.001 ^a	0.001 ^a	0.019 ^a	0.013 ^b	0.008 ^c

*Means followed by the same letters within a row are not significantly different according to Duncan's multiple range tests at P 0.05. FM=Fresh mass, DM =Dry mass

During the 2011/2012 season leaf fresh mass in *V. unguiculata* was significantly ($P<0.05$) reduced as plant density increased from 50 000 up to 100 000 plants ha^{-1} (Table 3.4). During the 2012/2013 season, leaf fresh mass increased significantly as plant density increased from 50 000 to 66 666 plants ha^{-1} and declined at 100 000 plants ha^{-1} (Table 3.4). Stem dry matter content was significantly ($P<0.05$) higher at 50 000 plants ha^{-1} than at 66 666 and 100 000 plants ha^{-1} (Table 3.4). Results of biomass in *A. cruentus* and *C. olitorius* in the current study had a similar trend with the results of plant height, leaf number and LAI which reduced at 100 000 plants ha^{-1} than at 50 000 plants ha^{-1} . This suggests strong competition for resources such as nutrients, physical spaces and water among plants at higher plant densities of 66 666 plants ha^{-1} and 100 000 plants ha^{-1} . In *V. unguiculata*, previous studies attributed reduction in plant biomass to fewer branches per plant at higher plant densities (Alege & Mustapha, 2007; Weber, 1966; Mohammed, 1984).

3.4.3.2 Total fresh and dry yield

Both dry and fresh mass yield per unit area in *A. cruentus* and *C. olitorius* was not significantly ($P>0.05$) affected by plant density during the 2011/2012 season. However, during the 2012/2013 season significant ($P<0.05$) differences were observed among plant densities with respect to both fresh and dry mass yield per unit area (Table 3.5). Plant densities of 50 000 and 66 666 plants ha^{-1} produced significantly ($P<0.05$) lower yield approximately 30% in terms of stem and leaf yield for both crops relative to 100 000 plants ha^{-1} . Fresh and dry yield result trend across years was inconsistent for *A. cruentus* and *C. olitorius*. This could have been due to variation in recommended fertiliser application rates besides other biophysical factors. Often plants compete for nutrients; during the 2011/2012 season the recommended fertiliser application rates were lower, hence nitrogen could have been a limiting factor especially for higher densities. Although, they were done in different climatic conditions the trend of the current study in *A. cruentus* are similar with the findings of Olofintoye *et al.* (2011) who also observed an increase in leaf yield increased with increase in density from 40 000 plants ha^{-1} up to 66 666 plants ha^{-1} in *A. cruentus*. In *C. olitorius*, although carried in different agro ecological regions, the trends of the current study are similar with the findings of Madakadze *et al.* (2007) who also observed an increase in leaf yield with increase in density above 100 000 plants ha^{-1} in *C. olitorius*.

Table 3.5. Effect of plant density on the yield of three selected African leafy vegetables obtained from two seasons at ARC, Roodeplaat.

Crops	Plant parts (t ha ⁻¹)	Plant density (plants ha ⁻¹)							
		2011/12 summer (Season 1)				2012/13 summer (Season 2)			
		50 000	66 666	100 000	LSD _(0.05)	50 000	66 666	100 000	LSD _(0.05)
<i>A.cruentus</i>	FM above ground	17.910 ^a	18.790 ^a	17.930 ^a	Ns	8.500 ^c	12.110 ^b	15.020 ^a	2.504
	FM leaves	9.160 ^a	9.615 ^a	10.023 ^a	Ns	2.541 ^b	3.255 ^b	4.296 ^a	0.817
	FM stem	7.769 ^a	8.219 ^a	7.169 ^a	Ns	3.513 ^b	6.625 ^a	5.769 ^a	1.430
	DM leaves	1.245 ^a	1.344 ^a	1.308 ^a	Ns	0.534 ^b	0.647 ^b	0.836 ^a	0.147
	DM stem	1.248 ^a	1.283 ^a	0.940 ^a	Ns	0.561 ^b	0.689 ^{ab}	0.896 ^a	0.220
<i>C.olitorius</i>	FM above ground	4.472 ^a	7.028 ^a	5.917 ^a	Ns	9.250 ^b	9.280 ^b	11.780 ^a	1.804
	FM leaves	1.860 ^a	2.722 ^a	3.104 ^a	Ns	2.431 ^b	2.459 ^b	3.088 ^a	0.508
	FM stem	0.389 ^a	0.672 ^a	0.605 ^a	Ns	4.543 ^b	5.671 ^{ab}	6.431 ^a	1.228
	DM leaves	0.398 ^a	0.603 ^a	0.544 ^a	Ns	0.673 ^{ab}	0.607 ^b	0.823 ^a	0.156
	DM stem	0.389 ^a	0.672 ^a	0.605 ^a	Ns	0.831 ^a	0.849 ^a	0.949 ^a	Ns
<i>V.unguiculata</i>	FM stem + leaves	5.106 ^a	5.447 ^a	5.397 ^a	Ns	3.015 ^c	4.148 ^b	6.972 ^a	1.054
	FM leaves	2.682 ^a	2.806 ^a	2.474 ^a	Ns	2.025 ^c	2.986 ^b	5.282 ^a	0.691
	FM stem	0.621 ^a	0.811 ^a	0.974 ^a	Ns	0.758 ^b	1.134 ^b	1.611 ^a	0.429
	DM leaves	0.339 ^b	0.318 ^b	0.518 ^a	0.111	0.440 ^c	0.600 ^b	0.900 ^a	0.113
	DM stem	0.043 ^b	0.043 ^b	0.105 ^a	0.033	0.156 ^b	0.191 ^b	0.343 ^a	0.099

*Means followed by the same letters within a row are not significantly different according to Duncan's multiple range tests at P 0.05. FM=Fresh mass, DM =Dry mass

Increase in yield in *A. cruentus* and *C. olitorius* as plant density increase may be due to increase in the number of plants per unit area, which might contribute to the production of extra yield per unit area leading to high yield (Law & Egharevba, 2009). Plant density significantly ($P<0.05$) improved dry mass yield for stem and leaf in *V. unguiculata* for both seasons (Table 3.5). During the 2011/2012 season, plant density of 100 000 plants ha^{-1} led to significantly ($P<0.05$) higher stem and leaf dry matter content relative to lower densities. During the 2012/2013 season both fresh and dry yield components were significantly higher at 100 000 plants ha^{-1} than at 50 000 and 66 666 plants ha^{-1} during the second season (Table 3.5). Total fresh and dry yield components in *V. unguiculata* significantly ($P<0.05$) increased with increase in plant densities from 50 000, 66 666 up to 100 000 plants ha^{-1} . The effect of higher density of 100 000 plants ha^{-1} on stem and leaf dry matter content was consistent for both seasons. Lower planting densities per unit area in *V. unguiculata* produced more vigorous crops than at higher population density, but this could not compensate for the reduced number of plants per unit area. The total yield increased with higher planting densities.

3.5 CONCLUSION

Results from 2011/12 growing season indicate that there is no significant differences in terms of plant density on growth of the three plants tested except for leaf number per plant of *C.olitorius*. Similar findings were also observed in the following season, except for *V.unguiculata* that a medium planting density gave the highest leaf number and for *A. cruentus* and *C.olitorius* lowest planting density gave highest LAI. The reason for variation between seasons could be the weather variations between the two seasons.

No significant differences of plant density effects on physiological parameters were observed during the 2011/12 growing season. Results were also similar in the following season, with the exception of *A. cruentus*, which had significantly higher CCI at higher planting densities.

Higher plant densities generally improved fresh and dry mass of the three crops as it is evident in the 2012/13 season. However, in the 2011/12 growing season this finding was not clearly evident especially for *C.olitorius* and *V.unguiculata*.

In general, crop yield of the three crops significantly increased with the highest plant density tested, as evident in the 2012/13 growing season. However, in the 2011/12 growing season there were no significant benefits of increasing planting densities.

In conclusion, the use of low plant densities will be suitable for mechanization as it will allow the use of implements such as pesticide application and mechanical harvesting. Furthermore, this could also give a provision of reduced seed cost, as seeds of these crops have low availability in the market. Therefore, the study recommends a plant density of 50 000 plants ha⁻¹ for *A. cruentus* and *C. olitorius* and 100 000 plants ha⁻¹ for *V. unguiculata*.

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CHAPTER 4

Growth, physiology and yield responses of *Amaranthus cruentus*, *Corchorus olitorius* and *Vigna unguiculata* to nitrogen application under drip irrigated commercial production.

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4.1 ABSTRACT

Field trials were conducted at the Agricultural Research Council - Vegetable and Ornamental Plant Institute, Gauteng Province, South Africa over two summer seasons, 2011/2012 and 2012/2013. The aim of the study was to evaluate growth, physiology and yield responses of *Amaranthus cruentus*, *Corchorus olitorius* and *Vigna unguiculata* to nitrogen fertilizer application rates. The trials were laid out in a randomized complete block design (RCBD) with three replications in 2011/2012 and nine replications in the 2012/2013 summer seasons. The treatments were three nitrogen levels, viz. 0, 44 and 88 kg N ha⁻¹ for the 2011/12 season and four levels viz. 0, 50, 100 and 125 kg N ha⁻¹ for the 2012/13 season. Nitrogen treatments for both seasons were based on recommendations of *Brassica* species derived from soil analysis. Parameters measured included chlorophyll content index (CCI), chlorophyll fluorescence (CF), stomatal conductance (SC), leaf number, leaf area index (LAI) and biomass. Results showed that the application of nitrogen at 44 kg N ha⁻¹ (2011/12) and 100 kg N ha⁻¹ (2012/13) resulted in significantly (P<0.05) higher growth in terms of leaf number, LAI increased CCI, increased biomass and yield of *A. cruentus*. A similar trend was observed in *C. olitorius* except that 44 kg N ha⁻¹ only improved stem fresh yield. A further increase of nitrogen above 44 kg N ha⁻¹ and 100 kg N ha⁻¹ had diminishing returns in *A. cruentus* and *C. olitorius*. In *V. unguiculata*, the application of 50 kg N ha⁻¹ significantly (P<0.05) increased

leaf number. Fresh mass yield (fresh yield) was significantly reduced by applying nitrogen. However, dry matter content was significantly higher at 50 kg N ha⁻¹ and 100 kg N ha⁻¹, further increase to 125 kg N ha⁻¹ reduced dry matter content. In *C. olerius* and *A. cruentus* higher leaf quality parameters (CCI, plant height, leaf number, biomass per plant, LAI and yield) following nitrogen application at 44 kg N ha⁻¹ (2011/12) and 100 kg N ha⁻¹ (2012/13) indicate that these crops can perform better under nitrogen application. Therefore using lower nitrogen rates than recommended for *Amaranthaceae* species could be suitable for commercial production of *A. cruentus* and *C. olerius*. In *V. unguiculata*, 50 kg N ha⁻¹ improved leaf number; however, this did not translate to any fresh yield advantage implying that the optimum rate for nitrogen application might be lower than 50 kg N ha⁻¹. Recommendation of nitrogen below 50 kg N ha⁻¹ could lead to reduced production costs.

4.2 INTRODUCTION

Amaranthus spp. (pig weed) *Corchorus olerius* (Jews mallow) and *Vigna unguiculata* (cowpeas) are amongst the major African leafy vegetables of importance in South Africa (van Rensburg *et al.*, 2007). African leafy vegetables are generally considered to be low management crops because they require less plant nutrients than their exotic counterparts, such as Swiss chard, for growth (van Rensburg *et al.*, 2004; Maunder & Meaker, 2007). This is a desirable trait to farmers in terms of reduced financial cost of fertiliser, water, etc (van Averbek *et al.*, 2012).

Nitrogen fertilization is an important management practice in crop production. This is because it is a constituent of many organic compounds, nucleic acids and protein compounds that promote growth and yield of leafy vegetables (Madan & Munjal, 2009; Onyango, 2002). Nitrogen deficiency results in a poor growth rate; early maturity and shortened vegetative growth phase (Jasso-Chaverria *et al.*, 2005). Although traditional leaf vegetables are considered to be low management crops that can grow in poor soils, agronomic studies conducted in other ecological regions have shown an improvement in production of *Amaranthus cruentus* (Denton & Olufolaji, 2000; Manga, 2001; Olaniyi *et al.*, 2008; Ainika *et al.*, 2011) and *Corchorus olerius* (Ojo & Olufolaji, 1997; Olaniyi & Ajibola, 2008) in response to nitrogen application. Although *Vigna unguiculata* can fix its nitrogen (Kolawale *et al.*, 2000; Sanginga *et al.*, 2000), increases in growth and yield due to nitrogen fertilization

have been achieved (Oliveira *et al.*, 2001; Chowdhury *et al.*, 2000). A South African study has shown an improvement in yield through use of fertilizer in *Vigna unguiculata* and *Amaranthus cruentus* (van Averbek *et al.*, 2012). However, the studies were done in potting plants in a glasshouse. Nitrogen application promotes above ground vegetative growth leading to higher yields (Abidin & Yasdar, 1986). Therefore, there is need to conduct agronomic studies on nitrogen to optimize yield of these leafy vegetables in South Africa. Therefore, it was hypothesized that Nitrogen fertilizer application rates recommended for *Brassica species*, Swiss chard (*Beta vulgaris L.var cicla*) would lead to optimum growth, physiology and yield responses of *Amaranthus cruentus*, *Corchorus olitorius* and *Vigna unguiculata*. The objective of this study was to measure growth, physiology and yield responses of *Amaranthus cruentus*, *Corchorus olitorius* and *Vigna unguiculata* in response to nitrogen fertilizer application rates under irrigated commercial production in South Africa.

4.3 MATERIALS AND METHOD

4.3.1 Plant material

Seeds (*Amaranthus cruentus* and *Corchorus olitorius*) were obtained from the Agricultural Research Council seed bank. *Vigna unguiculata* (Bechuana white, a runner type) was obtained from Hygrotech Seeds (Pty) Ltd. There was no treatment done to the seeds. Seedlings of *A. cruentus* and *C. olitorius* were grown in 250 cavity polystyrene trays filled with a commercial growing medium, Hygromix® (Hygrotech Seed Pty. Ltd., South Africa) and covered with vermiculate to reduce water loss from above the surface. Seedlings were raised in the nursery and irrigation was done three times a week from the date of sowing. Transplanting was done early in the morning to prevent transplanting stress. Seedlings of *A. cruentus* and *C. olitorius* were transplanted four weeks after sowing. *V. unguiculata* was sown directly to the field using seed at a rate of one (1) seed per station based on previous germination tests carried at the experimental site.

4.3.2 Description of trial site

Field trials were conducted at the Agricultural Research Council - Vegetable and Ornamental Plant Institute (ARC-VOPI), Roodeplaas, Pretoria, South Africa (25°35' S; 28°21' E; 1164 m above sea level). The average seasonal rainfall (November to April) of Roodeplaas is c. 500mm and is highly variable with maximum precipitation in December and January. The

total, in season, amount of rainfall received at the experimental site (November - February) was approximately 400mm and 300mm during the 2011/12 and 2012/13 planting seasons (ARC, Meteorological Station). Supplementary irrigation supplied amounted to 548mm during 2011/12 and 435 mm during 2012/13. The average daily maximum and minimum temperatures for Roodeplaat are 8°C and 34°C in summer (November – April). The climate data was obtained from the Agricultural Research Council – Institute for Soil, Climate and Water [ARC–ISCW] automatic weather stations network). The soil type in the field is classified as Hutton clay loam (South African soil taxonomic system) and its composition is given Table 4.1.

Table 4.1 Physical and chemical characteristics of the soil in the experimental field at ARC, Roodeplaat.

Parameter	2011/12 summer season	2012/13 summer season
P (mg kg ⁻¹)	17.3	13.6
K (mg kg ⁻¹)	227	203
Ca (mg kg ⁻¹)	756	774
Mg (mg kg ⁻¹)	234	289
Na (mg kg ⁻¹)	15.5	35.3
Exchangeable cation Ca (%)	56.7	51.6
Exchangeable cation Mg (%)	34	28.9
Exchangeable cation K (%)	7.8	16.1
Exchangeable cation Na (%)	1.5	3.4
pH	6.6	6.6
Clay (%)	25	25
Silt (%)	6	6
Sand (%)	69	69

4.3.3 Experimental design and treatments

The experimental design was laid out in a randomised complete block design (RCBD) with three replications during the 2011/2012 summer season (season 1) and nine replications in the 2012/13 summer season (season 2). The nitrogen treatments for both seasons were based on soil fertility analysis and recommendations (ARC-ISCW, 2011/12 and 2012/13 season). The recommendations used were of the *Amaranthaceae species* species, Swiss chard (*Beta vulgaris* L.var *cicla*). The reason why Swiss chard was used is related to the similarities in terms of morphological and physiological characteristics. Furthermore, indigenous leafy

vegetables are neglected crops hence information on nitrogen requirements is very limited to do baseline line studies.

During the 2011/12 season, the treatments were applied at 0%, 50% and 100% of the recommendations. The treatments were three levels of nitrogen 0, 44 and 88 kg N ha⁻¹ in 2011/12. During the 2012/13 season, the treatments were applied at 0%, 40%, 80%, and 100% of the recommendations. The treatments were four levels of nitrogen 0, 50, 100, and 125 kg N ha⁻¹ in the 2012/13 season. The plot size was 72 m² (12 m x 6 m) for 2011/12 season and 27 m² (9 m x 3 m) for 2012/13 season. The plot size changed in the 2012/13 season because the number of replications and nitrogen levels were increased. For both planting seasons, the source of nitrogen fertilizer used was limestone ammonium nitrate (LAN, 28% N) which was applied by banding in three split-applications. The first application was at planting (50%), second at four weeks (25%) and the last (25%) at eight weeks after transplanting for *C. olerarius* and *A. cruentus*. For *V. unguiculata*, the first application was at sowing (50%), the second at four weeks after sowing (25%) and the last (25%) at eight weeks after sowing. Double super phosphate (10.5% P) was applied at 80 kg and 97 kg P ha⁻¹ at planting during the 2011/12 and 2012/13 planting seasons, respectively, based on soil fertility results. Potassium was deemed sufficient from soil analysis (ARC-ISCW) results for both seasons.

During the 2011/12 and 2012/13 seasons, each plot had three (3) crops; each crop occupied three (3) ridges per plot. The crops were established on double rows per ridge for both seasons. The outer rows in the ridges were meant to reduce border effects while the inner rows were used as experimental plants. The drip irrigation system comprised of an electric powered pump, control unit (solenoid valves and controller), filter, water meters and polyethylene drip tape. The drippers were laterals connected to a main line with a flow meter, a valve and a pressure gauge at the entrance of the plots to control the operating pressure and measure the irrigation volume. Thin-wall drip tape with emitters at 0.3 m intervals/spacing and a drip rate of approximately 2.0 hr⁻¹ at an operating pressure of 150 kPa was placed on the centre of raised beds. Irrigation scheduling for both seasons was based on daily reference evapotranspiration (ET_o) which was obtained from an AWS at the experimental site. Reference evapotranspiration (ET_o) was then adjusted by a crop factor (K_c). The AWS calculates ET_o based on the FAO Penmann-Monteith method (Allen *et al.*, 1998).

4.3.4 Agronomic practices

Soil samples were taken at 0.30 m and 0.60 m depths prior to land preparation and submitted for soil fertility analyses at ARC-ISCW. Land preparation included ploughing, disking and rotovating to achieve a fine seedbed installation of drippers. *Vigna unguiculata* were sown directly using seed at a rate of one (1) seed per station based on previous germination tests carried out at the experimental site. During the two seasons, a plant density of 66 666 plants ha⁻¹ was used. Routine weeding and scouting for pests and diseases were done to ensure best management practices for the trials as shown in Figure 4.1.



Figure 4.1. *Vigna unguiculata* and *Corchorus olitorius* growing under commercial production in the trials at Roodeplaat in 2012 season.

4.3.5 Data collection

Weather data (maximum and minimum air temperature, maximum and minimum relative humidity, rainfall, wind speed and reference evapotranspiration (ET_o)) were monitored from an automatic weather station situated within a 100m radius from the field trials. Plant height, leaf number, stomatal conductance, chlorophyll content index (CCI), chlorophyll fluorescence (CF) and leaf area index (LAI) were measured starting from four weeks after transplanting (WAT) and weekly thereafter.

A total of three (3) plants per replication was tagged for data collection for growth and physiology parameters. The outer rows were excluded for data collection to prevent border effects and data was only collected from the inner rows. All measurements were done on leaves that had at least 50% green leaf area. Plant height was measured using a measuring

tape from the ground level to the tip or apex of the tallest stem. Leaf number was done by visual count of green fresh leaves. Stomatal conductance measurements were done on the abaxial leaf surface using a steady state leaf porometer (Model SC-1, Decagon Devices, Inc., USA). The chlorophyll content index was determined on the adaxial surface using the CCM-200 *Plus* chlorophyll content meter (Opti-Sciences, Inc., USA). The leaf area index was measured using the LAI 2200 Canopy Analyzer (Decagon Devices, Inc., USA). Photosynthetic efficiency as indicated by chlorophyll fluorescence (CF) was measured using a Handy Plant Efficiency Analyzer (PEA) chlorophyll fluorometer (Hansatech Instruments, U.K.). The measurement of CF was only done during the second season because the equipment was not available during the first season. Leaves were initially dark adapted (30 minutes) before measurements were taken. Values of F_v/F_m (the measurement of quantum yield potential of photosynthesis, or maximal photochemical efficiency of PSII) were recorded from the PEA and used for analysis. All measurements were done between irrigation and during mid-day weekly.

Harvesting commenced at six (6) weeks after transplanting (WAT) or sowing and every two weeks thereafter. The sample size for the yield was 1 m² for each replicate for both seasons. During each harvest, *C. olitorius* and *A. cruentus* yields were determined by cutting the mass of the above ground portion of the plant leaving 0.2 m of plant height above ground level. For *V. unguiculata*, harvesting was done by picking three to four fresh marketable leaves with their tender stem towards the growing tip of each runner, leaving the first and second growing leaves from the tip. Marketable leaves in *V. unguiculata* were defined as fresh or green tender leaves. The harvested portion was then partitioned into leaves and stems. In order to obtain accurate results, the plants were weighed immediately after harvest to avoid reduction in mass due to loss of water. Biomass per plant was obtained by randomly sampling three plants per replicate at each harvest. Dry matter content was obtained by oven drying at 70°C for 48 hours. Yield per hectare was obtained by conversion from measurements taken at 1 m² per replicate.

4.3.6 Statistical analyses

All data were subjected to statistical analyses using analysis of variance in GenStat® (Version 14, VSN International, UK) statistical package. Means were compared using Duncan's multiple range test (DMRT) at the 5% level of probability.

4.4 RESULTS AND DISCUSSION

4.4.1 Growth parameters

4.4.1.1 Plant height and leaf number

Plant height in *A. cruentus* and *C. olitorius* was not significantly ($P>0.05$) affected by nitrogen rates during the 2011/12 season although in *A. cruentus* there was a tendency of plant height to increase from 0 kg to 44 kg N ha⁻¹ and decline thereafter (Table 4.2). During 2012/13 summer season, plant height in *C. olitorius* and in *A. cruentus* increased approximately 30% with increase in Nitrogen (N) application from 0 kg to 100 kg N ha⁻¹ and then decreased at 125 kg N ha⁻¹ (Table 4.2). However, in *A. cruentus* plant height results were not statistically significant.

Leaf number in *A. cruentus*, *C. olitorius* and *V. unguiculata* was not significantly ($P>0.05$) affected by nitrogen rates during the 2011/12 season although there was a tendency of leaf number to increase with increases in nitrogen application rates (Table 4.2). During the 2012/13 season, leaf number in *C. olitorius* increased significantly with approximately 40% from 0 kg N ha⁻¹ to 100 kg N ha⁻¹ and further increases up to 125 kg N ha⁻¹ led to significant decreases (Table 4.2). Leaf number in *V. unguiculata* increased significantly with approximately 45% ($P<0.05$) from 0 kg N ha⁻¹ to 50 kg N ha⁻¹ and then declined significantly as from 100 kg N ha⁻¹ to 125 kg N ha⁻¹ during the 2012/13 season (Table 4.2). Similarly to 2011/2012, the difference in leaf number of *A. cruentus* was not significant for the 2012/13 season (Table 4.2). A similar trend was observed by Olaniyi & Ajibola (2008) who also observed an increase in *C. olitorius* plant height, number of leaves in response to nitrogen application.

Table 4.2. Effect of nitrogen on vegetative growth of three selected African leafy vegetables for two seasons at ARC, Roodeplaat.

Crops	Parameters	Summer season 1 (2011/2012)				Summer season 2 (2012/2013)				
		N ha ⁻¹			LSD _(0.05)	N ha ⁻¹				LSD _(0.05)
		0 kg	44 kg	88 kg		0 kg	50 kg	100 kg	125 kg	
<i>A.cruentus</i>	Plant height (cm)	62.3 ^a	70.0 ^a	65.3 ^a	Ns	36.0 ^a	62.3 ^a	74.4 ^a	71.0 ^a	Ns
	Leaf number	145.4 ^a	198.7 ^a	205.0 ^a	Ns	200.6 ^a	115.0 ^a	137.8 ^a	133.9 ^a	Ns
	Leaf area index	2.1 ^b	2.2 ^b	3.2 ^a	0.91	1.4 ^b	1.8 ^a	2.2 ^a	1.8 ^a	0.50
<i>C.olitorius</i>	Plant height (cm)	89.3 ^a	85.0 ^a	93.3 ^a	Ns	85.6 ^b	97.4 ^{ab}	103.6 ^a	86.8 ^b	13.8
	Leaf number	90.22 ^a	104.44 ^a	84.44 ^a	Ns	126.9 ^b	149.7 ^b	252.6 ^a	133.1 ^b	52.13
	Leaf area index	1.9 ^a	2.6 ^a	2.9 ^a	Ns	2.1 ^b	2.2 ^b	2.8 ^a	3.1 ^a	0.54
<i>V.unguiculata</i>	Leaf number	69 ^a	58 ^a	116 ^a	Ns	93 ^b	157 ^a	128 ^{ab}	87 ^b	50.5
	Leaf area index	3.8 ^a	3.7 ^a	3.7 ^a	Ns	2.3 ^a	2.3 ^a	3.0 ^a	3.0 ^a	Ns

*Means followed by the same letters within a row are not significantly different according to Duncan's multiple range tests at P 0.05. CF was not recorded during the first season

In *V. unguiculata* results of leaf number concurs with those of Bluementhal *et al.* (1992) who showed an increase in growth parameters in response to nitrogen application. Increase in leaf number is due to increases in nutrients as the amount of N applied to the soil is increased. However, the sudden decline in higher N rates could imply that an optimum N rate is reached for these crops. Sonneveld & Voogt (2009) concluded that crop growth increases as the concentration of nutrients increases until an optimum is reached. Therefore, excess nitrogen may lead to reduced growth through oxidative damage to cellular biomolecules such as lipids, protein, DNA and RNA (Wei *et al.*, 2009).

4.4.1.2 Leaf area index (LAI).

In *A. cruentus* and *C. olitorius*, LAI increased from 0 kg N ha⁻¹ to 88 kg N ha⁻¹ during the 2011/12 season; however, the differences were only significant in *A. cruentus* (Table 4.2). During the 2012/13 season, LAI increased significantly as nitrogen application increased from 0 kg up to 100 kg N ha⁻¹ and declined at 125 kg N ha⁻¹ for both crops. In *V. unguiculata* nitrogen application reduced LAI during 2011/12 season (Table 4.2). During 2012/13 season LAI increased from 0 kg to 100 kg N ha⁻¹, however, the differences were not significant for both seasons. Increase in *A. cruentus* and *C. olitorius* LAI followed by a decline concurs with findings of studies of Abidin & Yasdar (1986) and Ainika *et al.* (2011) in *Amaranthus*. The initial increases in *A. cruentus* and *C. olitorius* LAI in response to nitrogen application could be due to increased nitrogen uptake by the plants due to its availability in the soil. Onyango (2002) reported that nitrogen promotes growth through cell division and expansion. Moreover, Abidin & Yasdar (1986) and Ainika *et al.* (2011) concluded that nitrogen application encourages above ground vegetative growth leading to higher leaf area index. The decline after an increase could be attributed to optimum nitrogen levels being reached.

4.4.2 Physiological parameters

4.4.2.1 Chlorophyll fluorescence (CF) and Stomatal conductance (SC)

Chlorophyll fluorescence (CF) and Stomatal conductance in *A. cruentus*, *V. unguiculata* and *C. olitorius* were not significantly ($P > 0.05$) affected by nitrogen treatments during both seasons (Table 4.3). Studies with maize have shown that nitrogen deficit can decrease (Dodd, 2003), increase or have no effect (Cechin & Fumis, 2004) on stomatal conductance.

Table 4.3. Effect of nitrogen on vegetative growth of three selected African leafy vegetables for two seasons at ARC, Roodeplaat.

Crops	Parameters	Summer season 1 (2011/2012)				Summer season 2 (2012/2013)				
		N ha ⁻¹			LSD _(0.05)	N ha ⁻¹				LSD _(0.05)
		0 kg	44 kg	88 kg		0 kg	50 kg	100 kg	125 kg	
<i>A. cruentus</i>	CCI	24.1 ^b	29.4 ^{ab}	33.0 ^a	5.8	23.5 ^b	30.3 ^a	32.7 ^a	31.6 ^a	3.64
	Stomatal conductance	194.4 ^a	196.9 ^a	204.8 ^a	Ns	175.3 ^a	175.1 ^a	172.8 ^a	179.7 ^a	Ns
	CF (Fv/Fm)	-	-	-		0.708 ^a	0.708 ^a	0.700 ^a	0.691 ^a	Ns
<i>C. olerarius</i>	CCI	33.0 ^a	37.3 ^a	37.6 ^a	Ns	32.6 ^b	35.9 ^b	46.6 ^a	44.9 ^{ab}	9.31
	Stomatal conductance	394.2 ^a	533.0 ^a	477.0 ^a	Ns	105.6 ^a	104.8 ^a	103.2 ^a	107.0 ^a	Ns
	CF (Fv/Fm)	-	-	-		0.707 ^a	0.724 ^a	0.714 ^a	0.741 ^a	Ns
<i>V. unguiculata</i>	CCI	33.1 ^a	37.4 ^a	37.1 ^a	Ns	47.9 ^a	55.5 ^a	55.3 ^a	51.2 ^a	Ns
	Stomatal conductance	395.9 ^a	400.9 ^a	458.9 ^a	Ns	96.9 ^a	108.7 ^a	111.0 ^a	109.1 ^a	Ns
	CF (Fv/Fm)	-	-	-		0.722 ^a	0.746 ^a	0.727 ^a	0.742 ^a	Ns

*Means followed by the same letters within a row are not significantly different according to Duncan's multiple range tests at P 0.05. CF was not recorded during the first season*Stomatal Conductance (mmol m⁻² s⁻¹)

4.4.2.2 Chlorophyll content index (CCI)

In *A. cruentus* and *C. olitorius*, chlorophyll content index (CCI) increased with increase in nitrogen application during the 2011/12 seasons; however, the differences were only significant in *A. cruentus* (Table 4.3). During the 2012/13 season, CCI increased with increases in nitrogen application for both crops; however, in *C. olitorius* there was a decline at 125 kg N ha⁻¹. In *V. unguiculata* there was no significant ($P>0.05$) responses in CCI due to nitrogen application for both seasons (Table 4.3). Similarly during 2012/13 season, there was no significant differences although CCI increased from 0 kg N ha⁻¹ to 50 kg N ha⁻¹ and declined from 100 kg N ha⁻¹ up to 125 kg N ha⁻¹ (Table 4.3). Higher CCI observed in *A. cruentus* and *C. olitorius* relative to the control is due to the role nitrogen in promoting chlorophyll synthesis. This is in agreement with other researchers that chlorophyll content is strongly related to nitrogen availability/concentration in the soil (Blackmer & Schepers, 1995; Sumeet *et al.*, 2009). Nitrogen application increases availability of nitrogen in the soil which is readily absorbed by crops. Furthermore, leaves with different nitrogen content differ greatly in chlorophyll content (Witt *et al.*, 2005). Nitrogen application increases availability of nitrogen in the soil which is readily absorbed by crops. Lack of significant differences among treatments in CCI of *V. unguiculata* may be attributed to the ability of this crop to fix nitrogen. Higher CCI in *A. cruentus* and *C. olitorius* relative to the control may imply higher degree in greenness of the leaf, leaf color is a quality attribute for marketing the crops (Hussain *et al.*, 2010).

4.4.3 Yield

4.4.3.1. Biomass accumulation per plant

Nitrogen application resulted in a significantly higher biomass accumulation per plant in *A. cruentus* and *C. olitorius* compared to the control during the 2011/12 and 2012/13 seasons (Table 4.4). During the 2011/12 season, biomass significantly increased from 0 kg N ha⁻¹ to 44 kg N ha⁻¹, however, further increases up 88 kg N ha⁻¹ led to a decline in biomass. During 2012/13 season, biomass per plant in *C. olitorius* and *A. cruentus* increased significantly with approximately 70% from 0 kg N ha⁻¹ to 100 kg N ha⁻¹ and remained constant thereafter.

Table 4.4. Effect of nitrogen on biomass accumulation per plant of three selected African leafy vegetables for two seasons at ARC, Roodeplaat.

Crops	Biomass kg/plant	Summer season 1 (2011/2012)			Summer season 2 (2012/2013)			
		N ha ⁻¹			N ha ⁻¹			
		0 kg	44 kg	88 kg	0 kg	50 kg	100 kg	125 kg
<i>A.cruentus</i>	FM stem+ leaves	0.16 ^b	0.26 ^a	0.22 ^{ab}	0.04 ^b	0.11 ^b	0.27 ^a	0.30 ^a
	FM leaves	0.10 ^a	0.16 ^a	0.13 ^a	0.02 ^c	0.05 ^b	0.08 ^a	0.07 ^{ab}
	FM stem	0.05 ^b	0.07 ^a	0.07 ^a	0.02 ^b	0.05 ^b	0.11 ^a	0.12 ^a
	DM leaves	0.02 ^b	0.03 ^a	0.02 ^{ab}	0.01 ^a	0.01 ^a	0.02 ^a	0.06 ^a
	DM stem	0.01 ^a	0.01 ^a	0.01 ^a	0.002 ^c	0.007 ^b	0.013 ^a	0.011 ^{ab}
<i>C.olitorius</i>	FM stem+ leaves	0.09 ^a	0.16 ^a	0.13 ^a	0.14 ^c	0.23 ^b	0.32 ^a	0.24 ^b
	FM leaves	0.06 ^a	0.09 ^a	0.08 ^a	0.05 ^c	0.07 ^b	0.09 ^a	0.07 ^b
	FM stem	0.03 ^b	0.08 ^a	0.06 ^{ab}	0.08 ^c	0.13 ^b	0.17 ^a	0.15 ^b
	DM leaves	0.01 ^a	0.01 ^a	0.01 ^a	0.01 ^a	0.01 ^a	0.02 ^a	0.08 ^a
	DM stem	0.01 ^a	0.01 ^a	0.01 ^a	0.010 ^c	0.019 ^{ab}	0.021 ^a	0.017 ^b
<i>V.unguiculata</i>	FM stem+ leaves	0.11 ^a	0.12 ^a	0.12 ^a	0.11 ^a	0.07 ^b	0.08 ^b	0.07 ^b
	FM leaves	0.06 ^a	0.06 ^a	0.07 ^a	0.04 ^a	0.05 ^a	0.06 ^a	0.05 ^a
	FM stem	0.11 ^a	0.11 ^a	0.11 ^a	0.03 ^a	0.02 ^b	0.02 ^b	0.02 ^b
	DM leaves	0.01 ^a	0.01 ^a	0.01 ^a	0.005 ^c	0.007 ^{bc}	0.011 ^a	0.008 ^b
	DM stem	0.01 ^a	0.01 ^a	0.04 ^a	0.001 ^c	0.001 ^{bc}	0.003 ^{ab}	0.004 ^a

*FM=Fresh Mass, DM= Dry Mass

In *V. unguiculata* there was no significant effect in biomass in response to nitrogen application (Table 4.4). During the 2012/13 season, nitrogen treatments at 50, 100 and 125 kg N ha⁻¹ reduced fresh biomass compared to the control, however, the difference was non-significant. Although fresh mass of *V. unguiculata* was reduced by nitrogen, dry matter content, on the other hand, was significantly increased from 0 kg up to 100 kg N ha⁻¹, with further increases leading to a decline in dry matter content (Table 4.4).

Results in *C. olitorius* are in agreement with the findings of Olaniyi & Ajibola (2008). These authors noted an increase in biomass due to nitrogen application in *C. olitorius*. In *A. cruentus* results were consistent with the findings of van Averbek *et al.* (2012) who reported yield increase up to a point in *Amaranthus*, with further increases causing a significant decline in yield. Increase in biomass per plant in *A. cruentus* and *C. olitorius* is attributed to nitrogen availability. Biomass of individual plants is important as it translates to yield (total or final) advantage. Studies have shown that *V. unguiculata* can produce 60% of maximum biomass in the absence of nitrogen which is attributed to nitrogen fixation (van Averbek *et al.*, 2012). Furthermore, an increase in nitrogen led to a significant decline in biomass. The negative effect of nitrogen in *V. unguiculata* is attributed to increase in nodule and root reductance activity (Sing & Nair, 1995).

4.4.3.2 Total fresh and dry yield

Nitrogen application significantly ($P < 0.05$) improved fresh and dry yield in *A. cruentus* and *C. olitorius* for both seasons (Table 4.5). During the 2011/12 season, fresh and dry yield components increased significantly from 0 kg N ha⁻¹ to 44 kg N ha⁻¹; however, further increases up to 88 kg N ha⁻¹ did not have any significant effect on yield. During the 2012/13 season, total fresh and dry yield components of *A. cruentus* and *C. olitorius* increased significantly ($P < .001$) with approximately 80% as nitrogen rates increased from 0 kg N ha⁻¹ up to 100 kg N ha⁻¹ then declined at 125 kg N ha⁻¹ (Table 4.5).

Table 4.5. Effect of nitrogen application on the yield of three selected African leafy vegetables obtained from two seasons at ARC, Roodeplaat.

Crops	Yield t ha ⁻¹	Summer season 1 (2011/2012)				Summer season 2 (2012/2013)				
		N ha ⁻¹				N ha ⁻¹				
		0 kg	44 kg	88 kg	LSD _(0.05)	0 kg	50 kg	100 kg	125 kg	LSD _(0.05)
<i>A. cruentus</i>	FM stem + leaves	10.360 ^b	17.150 ^a	14.900 ^{ab}	4.96	2.440 ^b	7.130 ^b	17.690 ^a	19.840 ^a	8.214
	FM leaves	6.432 ^a	10.844 ^a	8.593 ^a	Ns	1.389 ^c	3.426 ^b	5.583 ^a	4.560 ^{ab}	1.417
	FM stem	3.040 ^b	4.752 ^a	4.937 ^a	1.44	1.134 ^b	3.449 ^b	7.222 ^a	8.310 ^a	2.333
	DM leaves	1.021 ^b	1.830 ^a	1.329 ^{ab}	0.625	0.240 ^a	0.601 ^a	1.054 ^a	4.117 ^a	Ns
	DM stem	0.525 ^a	0.728 ^a	0.702 ^a	Ns	0.155 ^c	0.531 ^b	0.863 ^a	0.762 ^{ab}	0.279
<i>C. olerarius</i>	FM stem + leaves	5.898 ^a	10.602 ^a	8.657 ^a	Ns	9.120 ^c	15.000 ^b	21.900 ^a	16.250 ^b	2.588
	FM leaves	4.259 ^a	6.111 ^a	5.139 ^a	Ns	3.032 ^c	4.606 ^b	6.204 ^a	4.583 ^b	1.034
	FM stem	2.181 ^b	5.556 ^a	4.009 ^{ab}	1.964	5.208 ^c	8.981 ^b	11.389 ^a	9.773 ^b	1.452
	DM leaves	0.619 ^a	0.612 ^a	0.638 ^a	Ns	0.615 ^a	0.960 ^a	1.356 ^a	5.067 ^a	Ns
	DM stem	0.560 ^a	0.989 ^a	0.836 ^a	Ns	0.692 ^c	1.260 ^{ab}	1.432 ^a	1.115 ^b	0.291
<i>V. unguiculata</i>	FM stem + leaves	7.139 ^a	8.083 ^a	7.750 ^a	Ns	7.220 ^a	4.537 ^b	5.069 ^b	4.792 ^b	1.344
	FM leaves	4.028 ^a	4.861 ^a	3.889 ^a	Ns	2.824 ^a	3.662 ^a	3.912 ^a	3.588 ^a	Ns
	FM stem	1.944 ^a	1.944 ^a	1.944 ^a	Ns	2.098 ^a	0.995 ^b	1.065 ^b	1.028 ^b	0.345
	DM leaves	0.492 ^a	0.586 ^a	0.489 ^a	Ns	0.373 ^c	0.523 ^{bc}	0.752 ^a	0.571 ^b	0.157
	DM stem	0.092 ^a	0.1167 ^a	0.294 ^a	Ns	0.061 ^c	0.078 ^{bc}	0.174 ^{ab}	0.233 ^a	0.102

*Means followed by the same letters within a row are not significantly different according to Duncan's multiple range tests at P 0.05

Yield results in *A. cruentus* are in agreement with previous studies in *A. cruentus* who observed an increase in yield due to nitrogen application up to a maximum followed by a decline in *A. cruentus* (Manga, 2001; Olaniyi, 2008; Ainika *et al.*, 2011; van Averbek *et al.*, 2012). The increase in yield was attributed to the ability of nitrogen to promote yield of leafy vegetables by cell expansion/division and elongation (Onyango, 2002). Previous studies showed variations in yield response to nitrogen application rates in *A. cruentus*. Studies by Manga (2001) and Ainika *et al.* (2011) observed optimum yield at 50 kg N ha⁻¹ while Olaniyi (2008) observed highest dry matter yield in *A. cruentus* at 60 kg N ha⁻¹. Furthermore, van Averbek *et al.* (2012) reported increased yield in *A. cruentus* at 328 kg N ha⁻¹ and above. However, the latter study was undertaken in pots under greenhouse conditions which could explain the huge variation in results of van Averbek *et al.* (2012) study in comparison to the other two studies.

Yield results in *C. olerarius* are in agreement to previous studies that showed significant increases in yield at higher rates of 75 kg N ha⁻¹ (NIHORT, 1986) and 70 kg N ha⁻¹ (Ojo & Olufolaji, 1997) in *C. olerarius*. Studies by Onyango (2002) attributed the increase in yield to the role of nitrogen in promoting the yield of leafy vegetables through its effect on cell division, expansion and elongation. Lack of significance in most of the measured parameters in *C. olerarius* during the first season may imply that the conditions under which the experiment was carried were below optimum. Furthermore, during the second season there was a double increase in yield. Yield results in *A. cruentus* and *C. olerarius* showed that further applications of nitrogen above optimum had no benefits. This could have been attributed to the negative effects of nitrogen at higher rates which causes osmotic stress, leading to oxidative damage to many important cellular components, such as lipids, protein, DNA and RNA (Wei *et al.*, 2009). Results of yield in *C. olerarius* and *A. cruentus* were consistent with the results of growth, CCI and LAI.

Nitrogen application did not significantly increase fresh and dry yield in *V. unguiculata* during the 2011/12 season although there was a tendency of the yield to slightly increase from 0 kg N ha⁻¹ to 44 kg N ha⁻¹ and then decrease at 88 kg N ha⁻¹ (Table 4.5). During the 2012/13 season, FM stem + leaves and FM stem was significantly ($P < .001$) reduced by nitrogen application. The difference between the three treatments - 50, 100 and 125 kg N ha⁻¹ - was not significant. However, stem and leaf dry matter content increased significantly ($P < .001$) with the increase in nitrogen from 0 kg N ha⁻¹ up 100 kg N ha⁻¹ then it declined at

125 kg N ha⁻¹ (Table 4.5). Reduction in yield in *V. unguiculata* due to nitrogen fertiliser application may imply that the yield used in this study was above optimum. Studies by Chowdhury *et al.* (2000) showed that yield components in *V. unguiculata* generally increased with increasing nitrogen rates from 0, 15, 30 up to 45 kg N ha⁻¹, with the highest yield at 30 kg N ha⁻¹. Similar to results of dry matter in *V. unguiculata*, an increase in dry weight with an increasing rate of nitrogen fertiliser has been reported in lettuce leaves (Tei *et al.*, 2000). Increases of dry matter content in *V. unguiculata* treated with nitrogen may be attributed to the role of nitrogen in creating the plant dry matter as well as many energy-rich compounds which regulate photosynthesis and plant production (Wu *et al.*, 1998). Further increases of nitrogen up to 125 kg N ha⁻¹ had diminishing returns on dry matter content. Although application of 50 kg N ha⁻¹ promoted dry yield in *V. unguiculata*, net diminishing marginal returns observed on fresh yield, have commercial implications because commercial producers are interested mostly in fresh produce.

4.5 CONCLUSION

Results from 2011/12 summer season showed no significant differences in growth (plant height and leaf number) due to nitrogen application of the three plants tested, except for LAI or leaf size in *A. cruentus*. Similar findings were also observed in the 2012/13 season in *A. cruentus*, in which leaf size increased due to nitrogen application at the expense of leaf number and plant height. In *C. olerius*, during 2012/13 season, nitrogen application produced tall plant with more leaves although leaf size was compromised. In *V. unguiculata* nitrogen application improved leaf number and no significant effect was observed in LAI or leaf size.

During 2011/2012 season, no significant differences of nitrogen effects on physiological parameters such as stomatal conductance and Chlorophyll fluorescence was observed for the three crops tested. However, nitrogen application improved leaf color in terms of CCI for *A. cruentus* in the same season. Similar results were observed in the 2012/13 season and leaf colour was also improved in *A. cruentus* and *C. olerius*.

In *A. cruentus* nitrogen application at 44 kg N ha⁻¹ improved fresh and dry yield during 2011/2012 season except for leaf fresh mass and stem dry mass. Similarly in the 2012/13 season, nitrogen application at 100 kg N ha⁻¹ improved all measured parameters except for

leaf dry mass. During 2011/12 summer season, there were no significant benefits of applying nitrogen on fresh and dry mass in *C. olitorius* except for stem fresh mass. However, during 2012/13 season application of 100 kg N ha⁻¹ improved fresh and dry yield. In *V. unguiculata* there were no significant benefits of applying nitrogen on fresh and dry yield during 2011/12 and 2012/13 seasons.

Therefore the study concludes that growth, physiology and yield in *A. cruentus* was improved at both 44 and 100 kg N ha⁻¹ while in *C. olitorius* stem fresh yield improved at 44 kg N ha⁻¹ and all other measured parameters at 100 kg N ha⁻¹. There is need to explore the exact application rates in *A. cruentus* and *C. olitorius* because of the huge gap between the fertiliser treatments, 44 kg N ha⁻¹ and 100 kg N ha⁻¹. Furthermore, there is possibility that the optimum levels of these crops lies below 100 kg N ha⁻¹ and this can translate to low production. Therefore, the current study recommends that *C. olitorius* and *A. cruentus* could be commercialised at 44 kg N ha⁻¹ and 100 kg N ha⁻¹ which were lower nitrogen application rates than those recommended for *Amaranthaceae species*. In *V. unguiculata* nitrogen application rate of 50 kg N ha⁻¹ promoted growth, however, fresh yield was reduced. Therefore, farmers are recommended to use less nitrogen application rates than used in the current study in *V. unguiculata*.

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CHAPTER 5

Growth, physiology and yield responses of Chinese cabbage (*Brassica juncea*) to nitrogen, planting date, spacing and irrigation management

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5.1 ABSTRACT

A field trial was planted at Roodeplaat (25°35' S; 28°21' E; 1164 masl) over two winter seasons, from June to September, 2012 and 2013. The objective of this study was to evaluate non-heading Chinese cabbage (*Brassica juncea*) responses to varying plant densities, nitrogen levels, planting dates and irrigation frequency. The experimental design was a factorial (3*3*2*2*3) experiment, laid out in a split-split plot design, with three replications. The treatments were three plant densities of 133 333, 80 000, 50 000 plants ha⁻¹; three nitrogen levels (0, 50 and 100 kg N ha⁻¹); two irrigation frequencies applied once and three times a week respectively and two planting dates (1 June 2012 and 18 July 2013). Leaf number, plant height, stomatal conductance (SC), chlorophyll fluorescence (CF) and leaf yield were measured from four (4) weeks after transplanting. Crops irrigated thrice or once a week with 50 kg N ha⁻¹ combined with 50 000 plants ha⁻¹ produced tall plants and bigger leaves (LAI). Irrigating thrice a week combined with nitrogen application at 50/100 kg N ha⁻¹ improved CF. Irrigating once a week combined with 100 kg N ha⁻¹ increased CCI. Application of 50 and 100 kg N ha⁻¹ significantly (P<0.05) increased leaf number, plant height, CF and yield compared to the control (0 kg N ha⁻¹), early and late planting dates across both seasons. Irrigating three times a week led to a significant (P<0.05) increase in CF, plant height and yield for 18 July (2012) and 1 June (2013). Higher leaf quality parameter (CCI, LAI) indicates that *Brassica juncea* can be grown at densities of 50 000 plants ha⁻¹. However, plant density of 133 333 plants ha⁻¹ resulted in significantly (P<0.05) higher yield

in terms of leaf number and fresh mass for second planting date (18 July) in both seasons. The first planting date (1 June) was associated with higher yields and low aphid infestation compared to second planting date (18 July). Therefore, early planting, application of nitrogen at 50 kg N ha⁻¹, irrigating three times a week and utilising a spacing of 50 000 plants ha⁻¹ is recommended for *Brassica juncea*.

Keywords: non-heading Chinese cabbage, irrigation, nitrogen, plant density, planting date.

5.2 INTRODUCTION

Brassica juncea (non-heading Chinese cabbage-mustard spinach) is a leafy vegetable grown in southern Africa under the name leaf mustard (*B. juncea* ssp. 'Rugosa') or rape. Chinese cabbage, commonly known as *Mutshaina*, is an indigenized leafy vegetable in South Africa. Mustard greens are a good source of dietary fibre, provitamin A, vitamin C, vitamin K, thiamine, riboflavin, vitamin B6, folate and mineral nutrients (van Wyk, 2005). Studies of small holder farmers showed that agronomic management factors such as irrigation scheduling (van Averbeke & Netshithuthuni, 2010) and nitrogen application (van Averbeke *et al.*, 2006) could lead to improved production.

Nitrogen is important for plant growth and its uptake is influenced by temperature, soil water availability, microbial activities and soil chemistry (Splittstoesser, 1990). Nitrogen affects growth and yield of leafy vegetables through its effect on cell division, expansion, and elongation resulting in large stems and leaves, and enhanced quality (Onyango, 2002). Nitrogen deficiency results in a poor growth rate; earlier maturity and a shortened vegetative growth phase (Jasso-Chaverria *et al.*, 2005). Yoshizawa *et al.* (1981) cited by van Averbeke *et al.* (2007b) reported an optimum application rate of 120 kg N ha⁻¹ in fertile soils for non-heading Chinese cabbage under field conditions. Van Averbeke *et al.* (2007b) concluded that application rate of 188 kg N ha⁻¹ was optimum for non-heading Chinese cabbage planted in pots. However, results of pot trials do not always translate accurately under field conditions (van Averbeke *et al.*, 2012).

Scheduling water application is very critical to make the most efficient use of a drip irrigation system, as excessive irrigation reduces yield, while inadequate irrigation causes water stress

and reduces production (Yazgan *et al.*, 2008). Letsoalo & van Averbeke (2006) reported that small holder farmers needed two or three irrigations per week. In a recent study, van Averbeke & Netshithuthuni (2010) reported that non-heading Chinese cabbage needed to be irrigated at least twice per week to maintain the water content of the rooting zone close to field capacity and achieve maximum leaf yield when using canal irrigation.

Planting date management not only has a huge effect on crop growth, development, and yield but it also impacts insect pest management (Brown *et al.*, 1992). Chinese cabbage (non-heading) seedlings grow optimally at a temperature of 22°C (Ope a *et al.*, 1988). Sowing too early, and with soil temperature cooler than 19°C, has been reported to cause chilling damage leading to slow and incomplete emergence in cowpea (Ismail *et al.*, 1997). Studies on small holder farmers in Limpopo established that non-heading Chinese cabbage was mainly planted during April and May (van Averbeke *et al.*, 2007a). This earlier planting date had the advantage of superior market conditions, but the disadvantage of increasing the incidence of pests (van Averbeke *et al.*, 2007a).

Yield per unit area tends to increase as plant density increases up to a point and then declines (Akintoye *et al.*, 2009). Therefore, an optimum population level that provides the plant with the best environment to utilise soil moisture and nutrients more effectively but also avoids excessive competition among the plants must be established. Peirce (1987) recommended an intra-row spacing of 0.30 m to 0.38 m in non-heading Chinese cabbage. In eastern Asia, the average inter-row spacing is 0.60 m for non-heading Chinese cabbage (Matsumura, 1981). Plant density of 0.30 m to 0.38 m x 0.60 m results in planting densities ranging between 44 000 plants ha⁻¹ and 55 000 plants ha⁻¹ in non-heading Chinese cabbage.

South Africa faces challenges of water scarcity (Modi & Mabhaudhi, 2013) and farmers need to manage water efficiently. Drip irrigation is a modern irrigation method, which can readily establish a nearly constant water regime in the root zone (Beese *et al.*, 1982), and ensure plants grow under proper soil water for optimum yield. Successful commercialisation of *Brassica juncea* requires agronomic information such as planting population, planting time, irrigation frequency and nitrogen fertilizer application rates. However, this production information is limited for *Brassica juncea* (Chinese cabbage-mustard spinach). Therefore, it was hypothesized that planting density of 80 000 plants ha⁻¹, planting early in June, irrigating

thrice a week and application of 50 kg N ha⁻¹ will provide optimum yield in *Brassica juncea*. The objective of this study was to determine the combined effect of nitrogen, planting date, spacing and irrigation on physiology, growth and yield of Chinese cabbage using drip irrigation under field conditions in South Africa.

5.3. MATERIALS AND METHOD

5.3.1 Plant material

Chinese cabbage (variety Florida broadleaf) seeds were obtained from Starke Ayres Seed (Pty) Ltd.

5.3.2. Description of trial site

Field trials were carried out during the 2012 and 2013 winter seasons at Agricultural Research Council-Vegetable and Ornamental Plant Institute (ARC-VOPI) in Roodeplaat (25°35' S; 28°21' E; 1164 m a.s.l), Pretoria, South Africa. The area receives little or no rainfall during the winter season. The total amount of rainfall received at Roodeplaat during 2012 and 2013 winter seasons (June - September) was approximately 74.5 mm and 6.6 mm, respectively (ARC, Meteorological Station). Supplementary irrigation was 416 mm during 2012 and 417 mm during 2013 winter season. The soil type in the field is classified as Hutton clay loam (South African soil taxonomic system) and the composition is given in Table 5.1.

Table 5.1 Physical and chemical characteristics of the soil in the experimental field at ARC, Roodeplaat.

Parameter	2011/12 summer season	2012/13 summer season
P (mg kg ⁻¹)	1.3	20.2
K (mg kg ⁻¹)	17.5	8.6
Ca (mg kg ⁻¹)	566	900
Mg (mg kg ⁻¹)	204	313
Na (mg kg ⁻¹)	17.5	8.6
Exchangeable cation Ca (%)	56.7	60.7
Exchangeable cation Mg (%)	34	35.2
Exchangeable cation K (%)	7.8	3.6
Exchangeable cation Na (%)	1.5	0.5
pH	6.6	6.6
Clay (%)	25	25
Sand (%)	69	69

5.3.3. Experimental design and treatments

The experimental design was a factorial (3*3*2*2*3) experiment, laid out in a split-split plot design, with three replications. The treatments were: three plant densities (133 333, 80 000, 50 000 plants ha⁻¹), three nitrogen levels (0, 50 and 100 kg N ha⁻¹), two irrigation frequencies (applied once and three times a week, respectively) and two planting dates (1 June, first, and 18 July, second, 2012 and 2013 winter season). Nitrogen (limestone ammonium nitrate (LAN) 28% N) was applied according to results of soil fertility analysis for both seasons. Nitrogen was applied by banding in three split applications. The first application was at transplanting (50%), the second at four weeks after transplanting (25%) and the last at (25%) eight weeks after transplanting. Double super phosphate was applied at 88 kg (10.5% P) and 20 P kg ha⁻¹ at planting for the 2012 and 2013 winter seasons respectively. Potassium was applied at 215 kg ha⁻¹ during 2013 winter season while during the 2012 winter season it was deemed sufficient from soil analysis (ARC-ISCW) results.

The plot size was 42 m² (12 m x 3.5 m) for both 2012 and 2013 winter seasons. Each plot had three (3) crops, each crop occupied three (3) ridges per plot and plants were established on double rows per ridge for both seasons. The outer ridges or rows were meant to reduce border effects. During the first two weeks all treatments received the same amount of water to establish the plants and thereafter the irrigation treatments were imposed. Irrigation treatments were applied at a frequency of once a week and thrice a week for the same amount of water. Irrigation scheduling for both seasons was based on daily reference evapotranspiration (ET_o) which was obtained from an AWS at the experimental site. Reference evapotranspiration (ET_o) was then adjusted by a crop factor (K_c). The AWS calculates ET_o based on the FAO Penmann-Monteith method (Allen *et al.*, 1998). The drip irrigation system comprised of an electric powered pump, control unit (solenoid valves and controller), filter, water meters and polyethylene drip tape. The drippers were laterals at a spacing of 1 metre connected to a main line with a flow meter, a valve and a pressure gauge at the entrance of the plots to control the operating pressure and measure the irrigation volume. Thin-wall drip tape with emitters at 0.3 m intervals/spacing and a drip rate of approximately 2.0 hr⁻¹ at an operating pressure of 150 kPa was placed on the centre of the raised beds.

5.3.4. Agronomic practices

Soil samples were taken from the field prior to land preparation at a depth between 0.3 m to 0.6 m and submitted for soil fertility analyses at the Agricultural Research Council- Institute of Soil, Climate and Water (ARC-ISCW). Land preparation included ploughing, disking, rotovating and ridging using a tractor to achieve a fine seedbed. Seedlings were grown in 250 cavity polystyrene trays (Figure 5.1) filled with a commercial growing medium, Hygromix® (Hygrotech Seed Pty. Ltd., South Africa) and covered with vermiculate to minimize water losses from above the surface. Seedlings were transplanted at four weeks after sowing. Transplanting was done early in the morning to prevent transplanting stress (Figure 5.1).



Figure 5.1. *Brassica juncea* trial at Roodeplaat in 2012 winter season. Left: seedlings 250 cavity polystyrene trays. Right: transplanting of seedlings.

Routine weeding and scouting for pests and diseases were done to ensure best management practices for the trials. Aphids were controlled using 2 ml of Decis (Active ingredient, Deltamethrin) plus 5ml of Aqua-Right 5 as a sticker in 10 l of water per 1 600 m² and applied using a knapsack. Dilution rate was done by ARC Plant Protection Institute.

5.3.5. Data collection

Weather data (maximum and minimum air temperature, maximum and minimum relative humidity, rainfall, wind speed and reference evapotranspiration (ET_o) for the duration of the trials were monitored from an automatic weather station (AWS) situated within a 100 m radius from the field trials. Plant height, leaf number, stomatal conductance, chlorophyll

content index (CCI), chlorophyll fluorescence (CF) and leaf area index (LAI) were measured starting from four weeks after transplanting (WAT) and weekly thereafter. Data collection was done on the inner rows for both seasons to prevent border effects. A total of three (3) plants per replication were tagged for data collection for growth and physiology parameters. All measurements were done on leaves that had at least 50% green leaf area. Plant height was measured using a measuring tape from the ground level to the tip or apex of the tallest stem. Stomatal conductance measurements were done on the abaxial leaf surface using a steady state leaf porometer (Model SC-1, Decagon Devices, Inc., USA) (Figure 5.2).



Figure 5.2. Left: data collection of stomatal conductance using Decagon leaf porometer. Right: three data plants per each replicate were allowed to grow throughout the whole season at Roodeplaat in 2012 winter season.

The chlorophyll content index was determined on the adaxial surface using the CCM-200 *Plus* chlorophyll content meter (Opti-Sciences, Inc., USA). Leaf area index was measured using the LAI 2200 Canopy Analyzer (Li-Cor, USA). Photosynthetic efficiency as indicated by chlorophyll fluorescence (CF) was measured using a Handy Plant Efficiency Analyzer (PEA) chlorophyll fluorometer (Hansatech Instruments, U.K.). The measurement of CF was only done during the second season because the equipment was not available during the first season. Leaves were initially dark adapted (30 minutes) before measurements were taken. Values of F_v/F_m (the measurement of quantum yield potential of photosynthesis, or maximal photochemical efficiency of PSII) were recorded from the PEA and used for analysis. All measurements were done before irrigation and during mid-day.

Harvesting commenced at six (6) weeks after transplanting (WAT) and every two weeks thereafter. The sample size for yield was 1 m² for each replicate for both seasons. During each harvest, yields were determined by picking fresh marketable leaves. After each harvest the remaining leaves were plucked and allowed to re-grow for the next harvest (Figure 5.3). In order to obtain accurate results, the plants were weighed in the field to avoid loss of water. Dry matter content was obtained by oven drying at 70°C for 48 hours. Yield per hectare was obtained by conversion from measurements taken at 1 m² per replicate.



Figure 5.3. Left: Harvested *Brassica juncea* prepared to be oven dried. Right: after harvest plants were plucked and allowed to re-grow for the next harvest.

5.3.6. Statistical analyses

All data was subjected to statistical analysis using analysis of variance in GenStat® (Version 14, VSN International, UK) statistical package. Means were compared using Duncan's multiple range test (DMRT) at the 5% level of significance.

5.4 RESULTS AND DISCUSSION

5.4.1. Growth parameters

5.4.1.1. Plant height and leaf number

There was significant ($P < 0.001$) increase in plant height in response to interaction of irrigation, nitrogen and spacing on the first planting date (1 June) during the 2012 winter season (Figure 5.4). All interactions that had no nitrogen led to stunted growth in *Brassica*

juncea (Figure 5.5). This could be due to competition of crops for limited nutrients. Similarly, other researchers have observed the effect of nitrogen deficiency in terms of poor growth rate and shortened vegetative growth phase in cucumber (Jasso-Chaverria *et al.*, 2005).

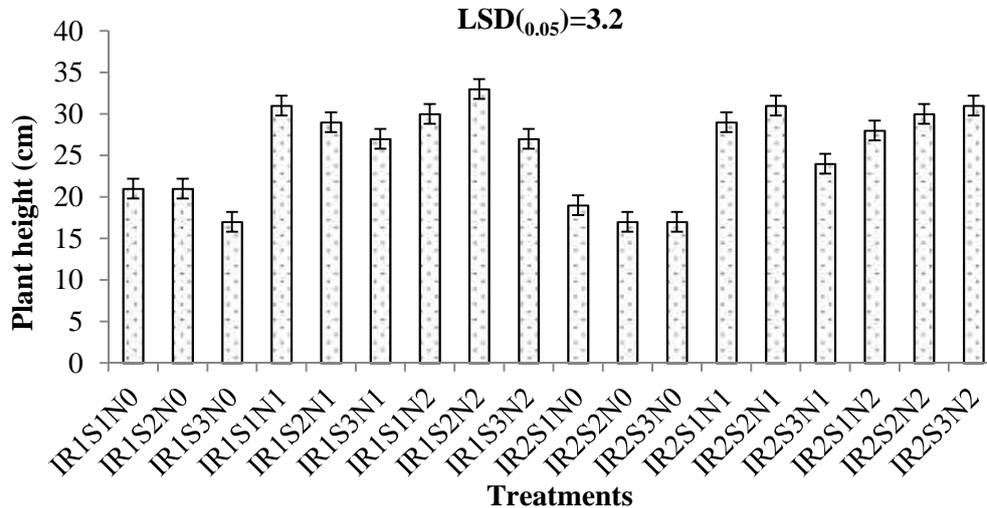


Figure 5.4. Interaction effect of irrigation, nitrogen and spacing on plant height of *Brassica juncea* on 1 June of 2012 season.

Key:

- | | |
|--|---|
| IR1 S1 N0= Irrigation once; 50 000 plants ha ⁻¹ ; 0 kg ha ⁻¹ | IR1 S2 N0= Irrigation once; 80 000 plants ha ⁻¹ ; 0 kg ha ⁻¹ |
| IR1 S3 N0= Irrigation once; 133 333 plants ha ⁻¹ ; 0 kg ha ⁻¹ | IR1 S1 N1= Irrigation once; 50 000 plants ha ⁻¹ ; 50 kg ha ⁻¹ |
| IR1 S2 N1= Irrigation once; 80 000 plants ha ⁻¹ ; 50 kg ha ⁻¹ | IR1 S3 N1= Irrigation once; 133 333 plants ha ⁻¹ ; 50 kg ha ⁻¹ |
| IR1 S1 N2= Irrigation once; 50 000 plants ha ⁻¹ ; 100 kg ha ⁻¹ | IR1 S2 N2= Irrigation once; 80 000 plants ha ⁻¹ ; 100 kg ha ⁻¹ |
| IR1 S3 N2= Irrigation once; 133 333 plants ha ⁻¹ ; 100 kg ha ⁻¹ | IR2 S1 N0= Irrigation thrice; 50 000 plants ha ⁻¹ ; 0 kg ha ⁻¹ |
| IR2 S2 N0= Irrigation thrice; 80 000 plants ha ⁻¹ ; 0 kg ha ⁻¹ | IR2 S3 N0= Irrigation thrice; 133 333 plants ha ⁻¹ ; 0 kg ha ⁻¹ |
| IR2 S1 N1= Irrigation thrice; 50 000 plants ha ⁻¹ ; 50 kg ha ⁻¹ | IR2 S2 N1= Irrigation thrice; 80 000 plants ha ⁻¹ ; 50 kg ha ⁻¹ |
| IR2 S3 N1= Irrigation thrice; 133 333 plants ha ⁻¹ ; 50 kg ha ⁻¹ | IR2 S1 N2= Irrigation thrice; 50 000 plants ha ⁻¹ ; 100 kg ha ⁻¹ |
| IR2 S2 N2= Irrigation thrice; 80 000 plants ha ⁻¹ ; 100 kg ha ⁻¹ | IR2 S3 N2= Irrigation thrice; 133 333 plants ha ⁻¹ ; 100 kg ha ⁻¹ |

Increasing nitrogen application from 50 kg N ha⁻¹ to 100 kg N ha⁻¹ in most of the treatment combinations did not significantly increase plant height in *Brassica juncea*. This could be due to the optimum levels of nitrogen reached at 50 kg N ha⁻¹. The treatment combinations that produced taller plants included irrigating thrice a week combined with 80 000 plants ha⁻¹ and 50 kg N ha⁻¹, and irrigating thrice a week combined with 133 333 plants ha⁻¹ and 50 kg N ha⁻¹.

Further increasing nitrogen up to 100 kg N ha⁻¹ in the previous treatment combinations did not increase plant height. Increases in plant height at higher plant density could be due to competition for nutrients. Also, it could be attributed to competition for light. As plants compete for light, they increase in height to intercept higher radiation (Yarnia, 2010). Irrigating once or thrice combined with 50 kg N ha⁻¹ and 50 000 plants ha⁻¹ also produced taller plants which were statistically similar to other treatment combinations which produced taller plants. Further increasing nitrogen up to 100 N ha⁻¹ in this treatment combination did not increase plant height. Taller plants obtained in this treatment could be due to less competition for available nutrients. Plants compete for resources such as solar radiation, water and nutrients at higher densities (Law & Egharevba, 2009).



Figure 5.5. *Brassica juncea* trial at Roodeplaat in 2012 winter season. Left: Plants with no nitrogen. Right: Plants with nitrogen fertilizer.

There was no significant interaction effect on the number of leaves per plant on the first planting date (1 June) and on the second planting date (18 July) in the 2012 season. Nitrogen application at 50 and 100 kg N ha⁻¹ significantly ($P < 0.05$) increased leaf number for both planting dates in the 2012 and 2013 seasons (Table 5.1). During the 2012 season, leaf number increased significantly ($P < 0.05$) with increase in nitrogen on the first planting date (1 June) while on the second planting date (18 July) the difference in leaf number between 50 and 100 kg N ha⁻¹ was not significant. Increase in leaf number in relation to nitrogen treatments could be due to availability of nutrients at 50 and 100 kg N ha⁻¹ compared to 0 kg N ha⁻¹.

Table 5.2. Leaf number of *Brassica juncea* as affected by irrigation, spacing and nitrogen for two planting dates over two winter seasons in Roodeplaat.

Treatment	2012 winter season		2013 winter season	
	Leaf number /plant		Leaf number/plant	
	1 June	18 July	1 June	18July
Irrigation				
Thrice	10 ^a	9 ^a	8 ^a	7.0 ^a
Once	10 ^a	9 ^a	8 ^a	7.0 ^a
LSD _{0.05}	Ns	ns	ns	Ns
Nitrogen				
100 kg ha ⁻¹	12 ^a	9 ^a	8 ^a	7.4 ^a
50 kg ha ⁻¹	10 ^b	9 ^a	8 ^a	7.4 ^a
0 kg ha ⁻¹	8 ^c	8 ^b	8 ^a	7.0 ^b
LSD _{0.05}	1.1	0.5	ns	0.1
Spacing/ plants ha⁻¹				
133 333	8 ^c	9 ^a	8 ^a	7.0 ^b
80 000	10 ^b	9 ^a	8 ^a	7.0 ^b
50 000	12 ^a	9 ^a	8 ^a	7.4 ^a
LSD _{0.05}	1.1	ns	ns	0.3

There was significant ($P < 0.05$) differences in leaf number in *Brassica juncea* due to varying plant densities for both the 2012 and the 2013 seasons (Table 5.2). During the 2012 season, increasing plant density from 50 000 plants ha⁻¹ to 133 333 plants ha⁻¹ significantly increased leaf number on the first planting date (1 June). There was no significant difference observed in leaf number due to varying plant densities on the second planting date (18 July) in the 2012 season. During the 2013 season lower plant density of 50 000 produced more leaves which was significant relative to 80 000 and 133 333 plants ha⁻¹; however, the differences between the two higher densities were not significant on the second planting date (18 July). Reduction of leaf number in higher densities could be due to competition for nutrients. Crops at higher densities compete for nutrients, physical spaces and water among plants at higher plant densities (Law & Egharevba, 2009).

Leaf number were significantly ($P < 0.05$) affected by irrigation frequency on the first planting date (1 June) and on the second planting date (18 July) in both the 2012 and the 2013 season (Table 5.1). Irrigating once a week reduced leaf number. This could be due to water stress. Inadequate irrigation has been reported to cause water stress and reduce production (Yazgan *et al.*, 2008). Studies have shown that water stress reduces plant height and leaf number in wild mustard (Mbatha & Modi, 2010) and wild water melon (Zulu & Modi, 2010).

5.4.1.2 Leaf Area Index (LAI)

There was a significant ($P < 0.001$) increase in LAI in response to the interaction of irrigation, nitrogen and spacing on LAI for the second planting date (18 July) in the 2012 season and on the second planting date (18 July) in the 2013 season (Figure 5.6 and 5.7).

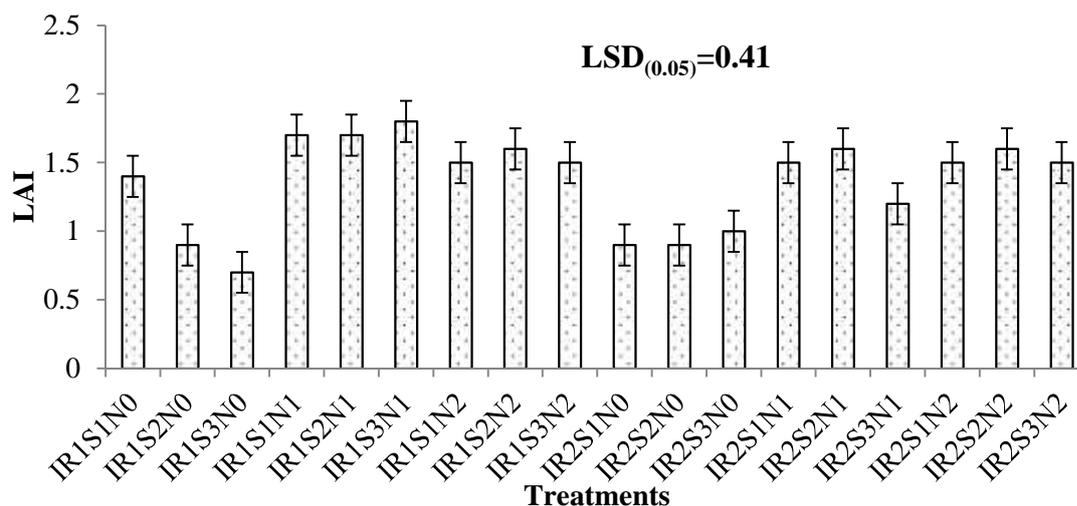


Figure 5.6. Interaction of irrigation, nitrogen and spacing on LAI of *Brassica juncea* on 1 June planting date during 2012 season.

Key:

IR1 S1 N0= Irrigation once; 50 000 plants ha⁻¹; 0 kg ha⁻¹
 IR1 S3 N0= Irrigation once; 133 333 plants ha⁻¹; 0 kg ha⁻¹
 IR1 S2 N1= Irrigation once; 80 000 plants ha⁻¹; 50 kg ha⁻¹
 IR1 S1 N2= Irrigation once; 50 000 plants ha⁻¹; 100 kg ha⁻¹
 IR1 S3 N2= Irrigation once; 133 333 plants ha⁻¹; 100 kg ha⁻¹
 IR2 S2 N0= Irrigation thrice; 80 000 plants ha⁻¹; 0 kg ha⁻¹
 IR2 S1 N1= Irrigation thrice; 50 000 plants ha⁻¹; 50 kg ha⁻¹
 IR2 S3 N1= Irrigation thrice; 133 333 plants ha⁻¹; 50 kg ha⁻¹
 IR2 S2 N2= Irrigation thrice; 80 000 plants ha⁻¹; 100 kg ha⁻¹

IR1 S2 N0= Irrigation once; 80 000 plants ha⁻¹; 0 kg ha⁻¹
 IR1 S1 N1= Irrigation once; 50 000 plants ha⁻¹; 50 kg ha⁻¹
 IR1 S3 N1= Irrigation once; 133 333 plants ha⁻¹; 50 kg ha⁻¹
 IR1 S2 N2= Irrigation once; 80 000 plants ha⁻¹; 100 kg ha⁻¹
 IR2 S1 N0= Irrigation thrice; 50 000 plants ha⁻¹; 0 kg ha⁻¹
 IR2 S3 N0= Irrigation thrice; 133 333 plants ha⁻¹; 0 kg ha⁻¹
 IR2 S2 N1= Irrigation thrice; 80 000 plants ha⁻¹; 50 kg ha⁻¹
 IR2 S1 N2= Irrigation thrice; 50 000 plants ha⁻¹; 100 kg ha⁻¹
 IR2 S3 N2= Irrigation thrice; 133 333 plants ha⁻¹; 100 kg ha⁻¹

During the 2012 and 2013 seasons, treatment combinations that had no nitrogen (0 kg N ha^{-1}) had significantly lower LAI compared to nitrogen (50 and 100 kg N ha^{-1}) treatments.

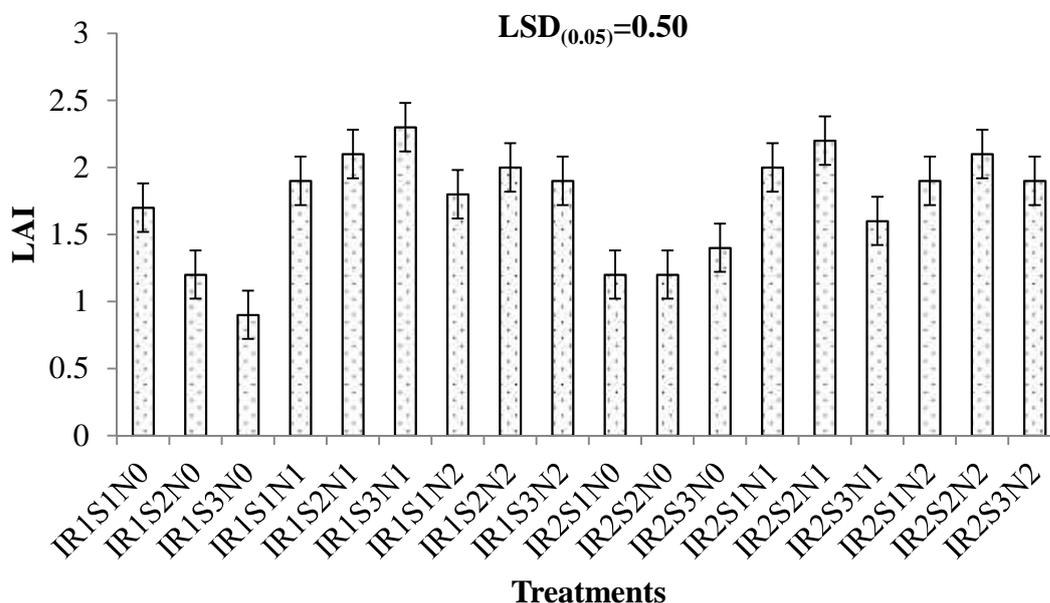


Figure 5.7. Interaction effect of irrigation, nitrogen and spacing on LAI of *Brassica juncea* for 1 June planting date during 2013 season.

Key:

- | | |
|--|---|
| IR1 S1 N0= Irrigation once; 50 000 plants ha^{-1} ; 0 kg ha^{-1} | IR1 S2 N0= Irrigation once; 80 000 plants ha^{-1} ; 0 kg ha^{-1} |
| IR1 S3 N0= Irrigation once; 133 333 plants ha^{-1} ; 0 kg ha^{-1} | IR1 S1 N1= Irrigation once; 50 000 plants ha^{-1} ; 50 kg ha^{-1} |
| IR1 S2 N1= Irrigation once; 80 000 plants ha^{-1} ; 50 kg ha^{-1} | IR1 S3 N1= Irrigation once; 133 333 plants ha^{-1} ; 50 kg ha^{-1} |
| IR1 S1 N2= Irrigation once; 50 000 plants ha^{-1} ; 100 kg ha^{-1} | IR1 S2 N2= Irrigation once; 80 000 plants ha^{-1} ; 100 kg ha^{-1} |
| IR1 S3 N2= Irrigation once; 133 333 plants ha^{-1} ; 100 kg ha^{-1} | IR2 S1 N0= Irrigation thrice; 50 000 plants ha^{-1} ; 0 kg ha^{-1} |
| IR2 S2 N0= Irrigation thrice; 80 000 plants ha^{-1} ; 0 kg ha^{-1} | IR2 S3 N0= Irrigation thrice; 133 333 plants ha^{-1} ; 0 kg ha^{-1} |
| IR2 S1 N1= Irrigation thrice; 50 000 plants ha^{-1} ; 50 kg ha^{-1} | IR2 S2 N1= Irrigation thrice; 80 000 plants ha^{-1} ; 50 kg ha^{-1} |
| IR2 S3 N1= Irrigation thrice; 133 333 plants ha^{-1} ; 50 kg ha^{-1} | IR2 S1 N2= Irrigation thrice; 50 000 plants ha^{-1} ; 100 kg ha^{-1} |
| IR2 S2 N2= Irrigation thrice; 80 000 plants ha^{-1} ; 100 kg ha^{-1} | IR2 S3 N2= Irrigation thrice; 133 333 plants ha^{-1} ; 100 kg ha^{-1} |

In the current study increasing nitrogen above 50 kg N ha^{-1} in any treatment combination did not increase LAI. Irrigating once or thrice combined with 50 kg N ha^{-1} and $50\,000 \text{ plants ha}^{-1}$ promoted LAI more than all the treatments. Further increasing nitrogen in this treatment combination did not increase LAI. Similarly increasing plant densities above $50\,000 \text{ plants ha}^{-1}$ did not have a significant effect on the LAI. Increase in LAI could be due to less competition for nutrients. Less competition increase availability of nitrogen in the soil which is readily absorbed by crops. Availability of nitrogen increases ground vegetative growth and

leaf area index (Abidin & Yasdar, 1986). Higher LAI may imply bigger leaf size of the vegetable crop which is the quality needed to market the produce. Results of the current study of bigger leaves (LAI) at lower densities of 50 000 plants ha⁻¹ concur with previous researchers (Matsumura, 1981; Pierce 1987). They observed highest leaf yield in non-heading Chinese cabbage at plant densities between 44 000 plants ha⁻¹ to 55 000 plants ha⁻¹

5.5. PHYSIOLOGICAL PARAMETERS

5.5.1. Chlorophyll fluorescence (CF)

There was significant ($P < 0.005$) increase in CF in response to interaction of irrigation and nitrogen on CF on the second planting date (18 July), 2012 season (Figure 5.6). Irrigating thrice a week combined with 50/100 kg N ha⁻¹ promoted CF compared to other treatment combinations. Treatment combinations that were irrigated thrice a week had significantly ($P < 0.005$) higher CF compared to treatments irrigated once a week. This could be attributed to excessive leaching of nitrogen as well as water losses to run off, drainage and soil evaporation in plants that were irrigated once a week. Furthermore, irrigating once a week limits water availability in the root zone.

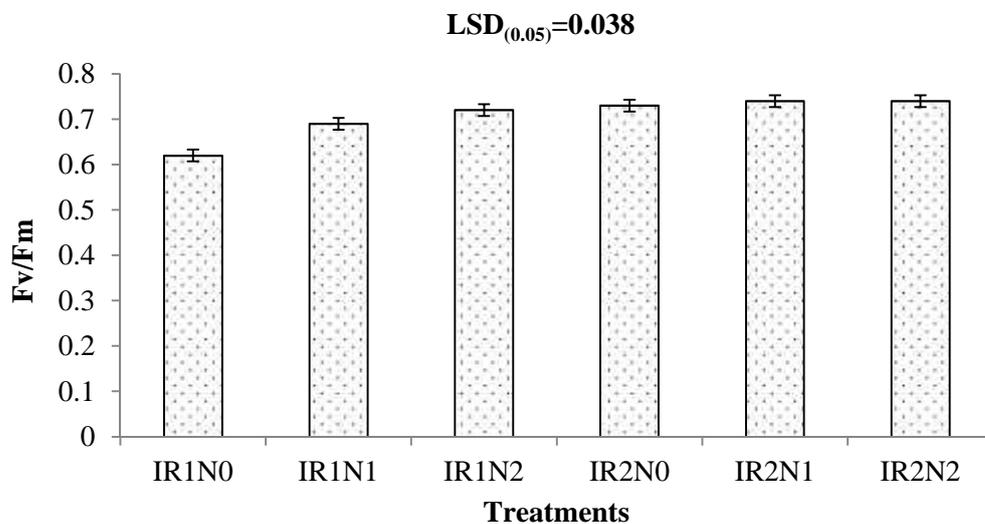


Figure 5.8. Interaction of irrigation and nitrogen on chlorophyll fluorescence (CF) of *Brassica juncea* for 18 July planting date during 2012 season.

Key:

IR1N0 = Irrigation once and 0 kg ha⁻¹;

IR1N2 = Irrigation once and 100 kg ha⁻¹;

IR2N1 = Irrigation thrice and 50 kg ha⁻¹;

IR1N1 = Irrigation once and 50 kg ha⁻¹;

IR2N0 = Irrigation thrice and 0 kg ha⁻¹;

IR2N2 = Irrigation thrice and 100 kg ha⁻¹.

Alternatively irrigating three times week increases water availability in the root zone due to frequent re-wetting of the top soil. A decrease in the rate of photosynthesis due to drought stress has been reported (Kawamitsu *et al.*, 2000). Chlorophyll fluorescence (CF) has been established to be a reliable stress indicator (Krause and Weis, 1991; Schreiber *et al.*, 1994). A decrease in CF by irrigating once a week can be attributed to leaching of nutrients and water stress. This implies that by irrigating once a week the quality of the yield was compromised. Similarly, Beletse *et al.* (2012) reported that yields obtained under water-stressed conditions may lack the quality needed to market the produce.

5.5.2 Stomatal conductance

There was no significant ($P>0.05$) interaction of irrigation, nitrogen and spacing on stomatal conductance on the first planting date (1 June) and on the second planting date (18 July) in the 2012 and 2013 seasons (Table 5.2). Similarly there was no significant ($P>0.05$) differences in stomatal conductance in response to varying nitrogen levels on the first planting date (1 June) and the late planting (18 July) during the 2012 season. During the 2013 season, on the first planting date (1 June) and on the second planting date (18 July), application of 100 kg N ha⁻¹ led to significantly higher stomatal conductance compared to 0 and 50 kg N ha⁻¹. Application of 50 kg N ha⁻¹ had higher stomatal conductance compared to 0 kg N ha⁻¹ although statistically they were the same. Similar reports with maize showed a decrease in stomatal conductance due to nitrogen deficit (Dodd, 2003). Studies have established that closure of stomata is the first response of almost all plants to water stress (Mansfield & Atkinson, 1990; Cornic & Massacci, 1996). This implies that at lower nitrogen levels plants were stressed. Stressed plants reduce the quality attributes of crops (Beletse *et al.*, 2012).

There was no significant ($P>0.05$) differences in stomatal conductance in response to varying plant densities for the 2012 and 2013 seasons (Table 5.3). The lack of differences for SC may also indicate that the experiments were conducted under optimum irrigation and fertilization conditions. The effect of irrigation frequency on stomatal conductance was significant on the first planting date (1 June) of the 2012 season (Table 5.3). Irrigating thrice a week induced stress compared to once a week on the first planting date (1 June). The results were not consistent for the 2013 season. The reason for this observation may be attributed to

environmental conditions, among other factors, that might have had an impact in SC. On the first planting date (1 June) the temperatures were low, hence there was less evaporation reducing the need to irrigate frequently.

Table 5.3. Effect of irrigation, spacing and nitrogen on the physiology of *Brassica juncea* for two different planting dates over two seasons.

Treatments	2012 winter season		2013 winter season	
	*SC		SC	
	1 June	18 July	1 June	18 July
Irrigation				
Thrice	166.9 ^b	181 ^a	534 ^a	488 ^a
Once	194.3 ^a	190 ^a	537 ^a	473 ^a
LSD 0.05	10.95	ns	ns	Ns
Nitrogen				
100 kg ha ⁻¹	179.3 ^a	181 ^a	575 ^a	523 ^a
50 kg ha ⁻¹	180.4 ^a	180 ^a	521 ^b	466 ^b
0 kg ha ⁻¹	182.2 ^a	195 ^a	511 ^b	452 ^b
LSD _{0.05}	ns	ns	46.7	51.5
Spacing				
133 333	176.9 ^a	185 ^a	509 ^a	494 ^a
80 000	188.5 ^a	186 ^a	523 ^a	469 ^a
50 000	176.5 ^a	187 ^a	574 ^a	478 ^a
LSD _{0.05}	ns	ns	46.7	Ns

*Stomatal Conductance -mmol m⁻² s⁻¹; *CF - (Fv/Fm).

5.5.3. Chlorophyll Content Index (CCI)

There was a significant (P<0.005) increase in CCI in response to the interaction of irrigation and nitrogen on the second planting date (18 July) in the 2012 season, on the first planting date (1 June) and on the second planting date (18 July) in the 2013 season (Figures 5.9 and 5.10).

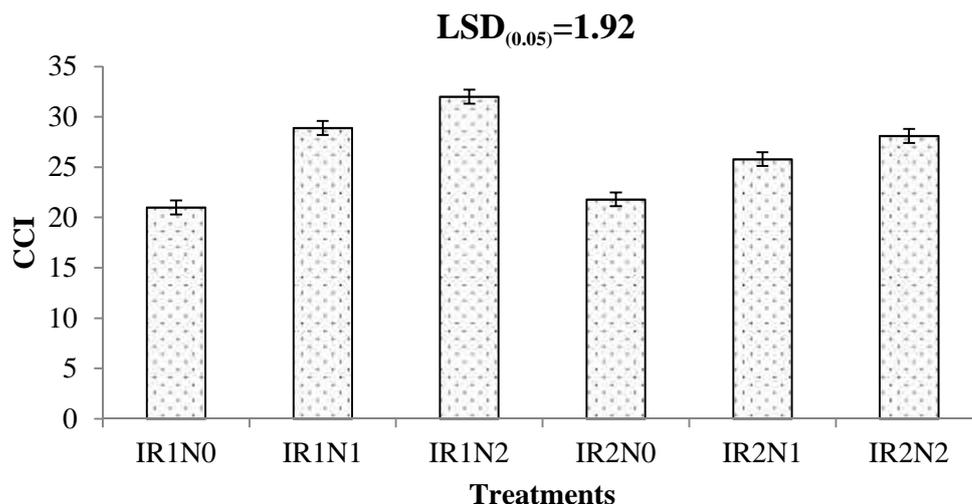


Figure 5.9. Interaction effect of irrigation and nitrogen on CCI of *Brassica juncea* for 18 July planting date during 2012 season.

Key:

IR1N0 = Irrigation once and 0 kg ha⁻¹;

IR1N2 = Irrigation once and 100 kg ha⁻¹;

IR2N1 = Irrigation thrice and 50 kg ha⁻¹;

IR1N1 = Irrigation once and 50 kg ha⁻¹;

IR2N0 = Irrigation thrice and 0 kg ha⁻¹;

IR2N2 = Irrigation thrice and 100 kg ha⁻¹.

For both the 2012 and 2013 seasons, interaction of irrigation with 0 kg N ha⁻¹ significantly reduced CCI compared to nitrogen treatments. The plants were yellowish in colour in the absence of nitrogen, which correlates with lower CCI at 0 kg N ha⁻¹. Chlorophyll is a green pigment present in plants, which captures radiation that is used in photosynthesis (Swain *et al.*, 2010). Nitrogen plays a role in chlorophyll synthesis (Jasso-Chaverria *et al.*, 2005). Leaves with different nitrogen content would, therefore, differ greatly in chlorophyll content (Witt *et al.*, 2005). Treatment combinations that had 100 kg N ha⁻¹ had significantly higher CCI compared to 50 kg N ha⁻¹. This concurs with findings of other researchers who reported that chlorophyll content was strongly related to nitrogen concentration in the soil (Blackmer and Schepers, 1995; Sumeet *et al.*, 2009).

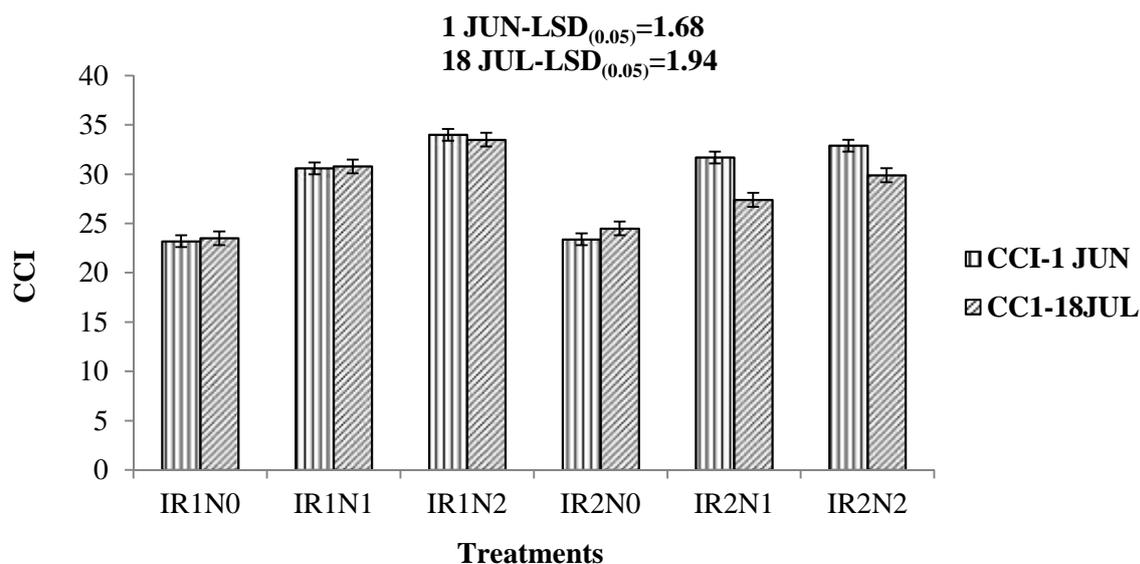


Figure 5.10. Interaction of irrigation and nitrogen on CCI of *Brassica juncea* for 1 June and 18 July planting dates during 2013 season.

Key:

IR1N0 = Irrigation once and 0 kg ha⁻¹;

IR1N1 = Irrigation once and 50 kg ha⁻¹;

IR1N2 = Irrigation once and 100 kg ha⁻¹;

IR2N0 = Irrigation thrice and 0 kg ha⁻¹;

IR2N1 = Irrigation thrice and 50 kg ha⁻¹;

IR2N2 = Irrigation thrice and 100 kg ha⁻¹.

For both seasons, irrigating once a week combined with 100 kg N ha⁻¹ had higher CCI compared to irrigating thrice a week. This could be due to higher concentration of nitrogen at 100 kg N ha⁻¹ which is available to the plant. The relatively high CCI adds value to the crop in that the market perceives the greenness of leafy vegetables as a quality index. Hussin *et al.* (2010) reported that for mustard spinach the most important quality attribute for marketing was colour.

5.6. Yield parameters

5.6.1. Total fresh and dry yield

There was no significant ($P > 0.05$) interaction between irrigation, nitrogen and plant spacing across both planting dates and seasons for yield. Application of nitrogen at 50 and 100 kg N ha⁻¹ led to significantly ($P < 0.05$) higher leaf number, fresh and dry mass compared to plants that had no nitrogen (0 kg N ha⁻¹) applied across both planting dates and seasons (Table 5.4). Results are in agreement with the report by van Averbek *et al.* (2006) that nitrogen increases

yield in Chinese cabbage. This reconfirms the role of nitrogen in promoting yield of leafy vegetables (Onyango, 2002). Results on nitrogen were consistent for both seasons and were consistent with results of LAI. In all planting dates across the two seasons, during the first harvest, there were no marketable leaves where nitrogen was not applied (Figure 5.11). Similar observations were made by van Averbeke *et al.* (2006) in non-heading Chinese cabbage. The observation of lack of marketable leaves in crops with no nitrogen concurs with studies that have established that *Brassica* species are heavy feeders of nitrogen (Thompson & Kelly, 1957).



Figure 5.11. Left: 18 July planting date in 2012 winter season. Right: 1 June planting date in 2012 winter season.

If farmers are to commercialize this crop and make a profit, they will need to apply fertilizer to improve the leafy quality in terms of size and appearance (Figure 8). From the current study, the differences between the levels 50 kg N ha^{-1} and 100 kg N ha^{-1} were not significant. Yoshizawa *et al.* (1981) reported that an application rate of 120 kg N ha^{-1} was optimum for Chinese cabbage under field conditions. Differences in results from our study might have been due to climatic factors and soil nutrient status, among other factors.

Higher plant density of $133\,333 \text{ plants ha}^{-1}$ resulted in significantly ($P < 0.05$) higher yield, approximately 40% in terms of leaf number on the second planting date (18 July) during the 2012 season relative to $50\,000$ and $80\,000 \text{ plants ha}^{-1}$ (Table 5.4). Similar observations were made during the 2013 season. Results for both seasons were consistent. Highest leaf yield at $133\,333 \text{ plants ha}^{-1}$ is contrary to previous work by researchers who observed the highest

yield between 44 000 plants ha⁻¹ to 55 000 plants ha⁻¹ (Matsumura, 1981; Pierce, 1987). Variation could be due to agro ecological regions and varieties used, among other factors. The ability to influence both fresh mass and number of leaves makes plant density of 133 333 plants ha⁻¹ an ideal plant density. This is because in some markets where leafy vegetables are sold, they are not sold on a mass basis but rather using a bunch (a certain number of leaves). In such instances, this suggests that at 133 333 plants ha⁻¹ yield (in this case leaf number) would produce more bunches. Results on leaf number per plant (Table 5.1) and of LAI (Table 5.2) showed that competition for nutrients was higher at 133 333 plants ha⁻¹ compared to 50 000 plants ha⁻¹. However, yield was highest at 133 333 plants ha⁻¹. This suggests that, although higher planting densities adversely affect growth and yield of individual plants, high yields can be attained due to increase in number of plants per unit area (Law & Egharevba, 2009).

Frequent irrigation, thrice a week, led to a significant ($P < 0.05$) increase of approximate 30% in fresh mass yield due to rapid growth on the second planting (18 July) of both seasons compared to irrigating once a week (Table 5.4). Results were consistent with the findings of Averbek & Netshithuthuni (2010) who reported that non-heading Chinese cabbage needed to be irrigated at least twice per week to maintain the water content of the rooting zone. As the season progressed, increase in temperatures may have led to increase in evaporation. Studies have reported that as evaporation rates increase, there is a need to irrigate frequently (Connor *et al.*, 1985; Whitfield *et al.*, 1986). Several experiments have shown positive responses in some crops to high frequency drip irrigation (Freeman *et al.*, 1976; Segal *et al.*, 2000; Sharmasarkar *et al.*, 2001).

Table 5.4. Effect of irrigation, spacing and nitrogen on the yield of *Brassica juncea* for 1 June and 18 July planting dates during the two winter seasons of 2012 and 2013 in Roodeplaat.

Treatment	2012 winter season						2013 winter season					
	FM Leaves (t ha ⁻¹)		DM leaves (t ha ⁻¹)		Leaf number m ⁻²		FM Leaves (t ha ⁻¹)		DM leaves (t ha ⁻¹)		Leaf number m ⁻²	
	1June	18July	1June	18 July	1June	18July	1June	18July	1June	18 July	1June	18July
Irrigation												
Thrice	7.25 ^a	8.44 ^a	0.66 ^a	0.717 ^a	25 ^a	29 ^a	8.066 ^a	5.781 ^a	0.930 ^a	0.511 ^a	40.0 ^a	35.0 ^a
Once	7.61 ^a	6.90 ^b	0.708 ^a	0.649 ^a	27 ^a	28 ^a	6.600 ^b	5.588 ^a	0.734 ^b	0.500 ^a	35.8 ^a	33.8 ^a
LSD _{0.05}	ns	1.3	ns	ns	ns	ns	0.9517	ns	0.1135	Ns	ns	ns
Nitrogen												
100 kg ha ⁻¹	8.06 ^a	9.84 ^a	0.736 ^a	0.827 ^a	29 ^a	36 ^a	8.338 ^a	7.347 ^a	0.896 ^a	0.708 ^a	38.6 ^a	42.0 ^a
50 kg ha ⁻¹	8.87 ^a	10.13 ^a	0.804 ^a	0.855 ^a	29 ^a	35 ^a	8.124 ^a	5.776 ^b	0.882 ^a	0.555 ^b	39.2 ^a	41.5 ^a
0 kg ha ⁻¹	5.36 ^b	3.04 ^b	0.512 ^b	0.367 ^b	23 ^a	15 ^b	5.538 ^b	3.930 ^c	0.719 ^b	0.254 ^c	35.9 ^a	19.6 ^b
LSD _{0.05}	1.004	1.586	0.083	0.094	ns	5.46	1.1656	0.9900	0.1391	0.0841	ns	5.85
Spacing plants/ha⁻¹												
133 333	7.80 ^a	8.488 ^a	0.715 ^a	0.704 ^a	29 ^a	35 ^a	7.221 ^a	6.881 ^a	0.799 ^a	0.579 ^a	36.1 ^a	44.2 ^a
80 000	7.33 ^a	6.941 ^a	0.663 ^a	0.625 ^a	27 ^a	25 ^b	7.314 ^a	5.352 ^b	0.822 ^a	0.501 ^a	37.0 ^a	33.1 ^b
50 000	7.15 ^a	7.584 ^a	0.675 ^a	0.721 ^a	22 ^a	26 ^b	7.466 ^a	4.819 ^b	0.875 ^a	0.437 ^b	40.6 ^a	25.8 ^c
LSD _{0.05}	ns	ns	ns	ns	ns	5.46	Ns	0.9900	ns	0.084	ns	5.85

5.7 CONCLUSION

Crops irrigated thrice or once a week with 50 kg N ha⁻¹ combined with 50 000 plants ha⁻¹ produced tall plants and bigger leaves (LAI) in *Brassica juncea* compared to all other treatments. Irrigating thrice a week combined with 50/100 kg N ha⁻¹ improved CF relative to all other treatments. Irrigating once a week combined with 100 kg N ha⁻¹ increased CCI compared to other treatments such as irrigating once with 50 kg N ha⁻¹ or 100 kg N ha⁻¹. Nitrogen application at 50/100 kg N ha⁻¹ improved leaf number and yield in *Brassica juncea* on the first planting date (1 June) and on the second planting date (18 July) in the 2012 and 2013 seasons compared to the control treatments. Higher plant density (133 333 plants ha⁻¹) resulted in higher yield on the first planting date (1 June) in both seasons relative to 50 000 and 80 000 plants ha⁻¹. Irrigating three times weekly led to higher yield for 18 July (2012) and 1 June (2013) compared to irrigating once a week.

Since 100/50 kg N ha⁻¹ performed similar in terms of LAI (leaf size), number of leaves and fresh mass of marketable leaves, farmers are recommended to apply 50 kg N ha⁻¹. This would in turn translate to cost savings for commercial producers in terms of fertilizer inputs. Farmers are recommended to apply 50 kg N ha⁻¹. Although yields in terms of fresh mass and leaf number were higher at 133 333 plants ha⁻¹ the quality (colour and size) needed to market the produce was compromised. Higher yields at 133 333 plants ha⁻¹ may increase production cost to a large scale drip commercial production in terms of inputs (e.g. seeds, fertiliser); and close spacing can reduce mechanization like weeding. Use of low plant densities will be suitable for mechanization as it will allow the use of implements during pesticide application and mechanical harvesting. It will also translate to cost saving in terms of seeds. Furthermore, the cost of seeds may be high due to low availability in the market. If farmers are to commercialise *Brassica juncea*, they are recommended to use 50 000 plants per ha⁻¹. As the winter season progresses, temperatures tend to increase, thus higher evaporation rates may necessitate increased frequency. Farmers are thus recommended to irrigate thrice a week as it increases leaf size (LAI) and fresh mass of marketable leaves.

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CHAPTER 6

6.1 GENERAL CONCLUSIONS

This study primarily focused on the response of traditional leafy vegetables to various agronomic factors under commercial production in South Africa. South Africa faces the challenges of water scarcity (Modi & Mabhaudhi, 2013), population growth (UNFP, 2011), and food and nutrition insecurity (de Klerk *et al.*, 2004). Traditional leafy vegetables can play a role towards food and nutrition security in the not so distant future. These challenges can be addressed in part by taking advantage of indigenous leafy vegetable cultivation. There is evidence that agronomic management factors such as irrigation, nitrogen, planting density, and planting date have an impact on growth, physiology and yield of crops (Chaves *et al.*, 2002; Onyango, 2002; Beletse *et al.*, 2012; van Averbek *et al.*, 2012). If farmers are to commercialize these crops and optimize yield, they should apply agronomic management factors with considerable precision.

The current agronomic study indicates that growth and yield in traditional leafy vegetables can be optimised through improved agronomic practices. The yield quality attributes such as colour, leaf size and fresh weight were improved with application of agronomic management factors. Agronomic practice such as spacing, irrigation and nitrogen led to improved plant growth in terms of leaf area index and, ultimately, improved canopy characteristics that translated to improved yield in *A. cruentus*, *C. olerius* and *Brassica juncea*. Results of growth and yield followed a similar trend with the results of physiological parameters. For example, in *A. cruentus* and *C. olerius* CCI increased with increases in nitrogen application followed by a decline. A similar pattern in plant growth and yield parameters were observed in response to increases in nitrogen application. The observations made in this study are in support of the view that states that alterations in a plant's behaviour initially occur at the molecular or physiological level before they manifest into morphological traits.

The results obtained in this study cannot be taken as standard growing procedures but as guidelines for these crops as agronomic factors such as irrigation, nitrogen, planting dates vary with locations, due to climatic factors such as rainfall and physical factors such as soil type. It is therefore, necessary that further studies should be done to investigate the response of *A. cruentus*, *C. olitorius*, *V. unguiculata* and *B. juncea*, to irrigation, nitrogen, plant density and planting date on various regions in South Africa.

6.2 REFERENCES

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APPENDIX 1 –LIST OF ANOVA FOR CHAPTER 3

Amaranthus -2011/2012 season

Variate: FM above ground

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	91.88	45.94	1.21	
Rep.*Units* stratum					
Treatment	2	6.09	3.05	0.08	0.923
Harvest	3	1335.70	445.23	11.72	<.001
Treatment.Harvest	6	175.34	29.22	0.77	0.602
Residual	22	836.06	38.00		
Total	35	2445.08			

Variate: FM leaves

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	38.23	19.11	1.20	
Rep.*Units* stratum					
Treatment	2	4.47	2.23	0.14	0.870
Harvest	3	771.08	257.03	16.08	<.001
Treatment.Harvest	6	67.56	11.26	0.70	0.649
Residual	22	351.76	15.99		
Total	35	1233.10			

Variate: FM stem

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	9.986	4.993	0.68	
Rep.*Units* stratum					
Treatment	2	6.664	3.332	0.46	0.640
Harvest	3	892.777	297.592	40.67	<.001
Treatment.Harvest	6	35.174	5.862	0.80	0.580
Residual	22	160.995	7.318		
Total	35	1105.596			

Variate: DM leaves

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	0.2437	0.1219	0.37	
Rep.*Units* stratum					
Treatment	2	0.0605	0.0302	0.09	0.913
Harvest	3	1.5166	0.5055	1.53	0.235
Treatment.Harvest	6	1.2788	0.2131	0.65	0.693
Residual	22	7.2666	0.3303		
Total	35	10.3662			

Variate: DM stem

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	0.4926	0.2463	0.74	
Rep.*Units* stratum					
Treatment	2	0.8575	0.4287	1.30	0.294
Harvest	3	9.6472	3.2157	9.72	<.001
Treatment.Harvest	6	1.2055	0.2009	0.61	0.722
Residual	22	7.2755	0.3307		
Total	35	19.4782			

Cowepea- 2012 season**Variate: FM above ground**

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	0.4519	0.2260	0.29	
Rep.*Units* stratum					
Treatment	2	0.6090	0.3045	0.40	0.680
Harvest	2	242.3759	121.1880	157.23	<.001
Treatment.Harvest	4	1.0224	0.2556	0.33	0.853
Residual	16	12.3322	0.7708		
Total	26	256.7915			

Variate: FM stem

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	0.6661	0.3331	0.62	
Rep.*Units* stratum					
Treatment	2	0.5640	0.2820	0.52	0.602
Harvest	2	6.5560	3.2780	6.08	0.011
Treatment.Harvest	4	1.2948	0.3237	0.60	0.668
Residual	16	8.6262	0.5391		
Total	26	17.7071			

Variate: FM leaves

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	0.1754	0.0877	0.44	
Rep.*Units* stratum					
Treatment	2	0.5056	0.2528	1.28	0.306
Harvest	2	44.8079	22.4039	113.02	<.001
Treatment.Harvest	4	0.1512	0.0378	0.19	0.940
Residual	16	3.1716	0.1982		
Total	26	48.8116			

Variate: DM leaves

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	0.03017	0.01509	1.22	
Rep.*Units* stratum					
Treatment	2	0.21715	0.10857	8.78	0.003
Harvest	2	0.54492	0.27246	22.04	<.001
Treatment.Harvest	4	0.13823	0.03456	2.80	0.062
Residual	16	0.19778	0.01236		
Total	26	1.12825			

Variate: DM stem

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	0.002550	0.001275	1.17	
Rep.*Units* stratum					
Treatment	2	0.023155	0.011578	10.63	0.001
Harvest	2	0.006251	0.003125	2.87	0.086
Treatment.Harvest	4	0.003503	0.000876	0.80	0.541
Residual	16	0.017434	0.001090		
Total	26	0.052894			

Corchorus- 2012 season

Variate: FM above ground

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	13.761	6.880	0.73	
Rep.*Units* stratum					
Treatment	2	19.704	9.852	1.05	0.385
Harvest	1	20.945	20.945	2.24	0.166
Treatment.Harvest	2	11.075	5.537	0.59	0.572
Residual	10	93.682	9.368		
Total	17	159.167			

Variate: FM stem

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	6.993	3.497	1.16	
Rep.*Units* stratum					
Treatment	2	2.923	1.461	0.48	0.630
Harvest	1	7.401	7.401	2.45	0.148
Treatment.Harvest	2	3.777	1.888	0.63	0.554
Residual	10	30.154	3.015		
Total	17	51.247			

Variate: FM leaves

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	2	4.871	2.436	0.81	0.466
Harvest	1	3.305	3.305	1.10	0.314
Treatment.Harvest	2	2.336	1.168	0.39	0.685
Residual	12	35.929	2.994		
Total	17	46.441			

Variate: DM leaves

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	0.26502	0.13251	1.70	
Rep.*Units* stratum					
Treatment	2	0.13331	0.06665	0.86	0.454
Harvest	1	0.31810	0.31810	4.08	0.071
Treatment.Harvest	2	0.01656	0.00828	0.11	0.900
Residual	10	0.77891	0.07789		
Total	17	1.51190			

Variate: DM stem

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	0.61296	0.30648	3.63	
Rep.*Units* stratum					
Treatment	2	0.26154	0.13077	1.55	0.260
Harvest	1	0.04875	0.04875	0.58	0.465
Treatment.Harvest	2	0.00060	0.00030	0.00	0.996
Residual	10	0.84535	0.08454		
Total	17	1.76921			

Amaranthus - 2013 season

Variate: FM above ground

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	8	178.88	22.36	1.62	
Rep.*Units* stratum					
Treat	2	384.53	192.27	13.92	<.001
Harvest	1	1328.63	1328.63	96.20	<.001
Treat.Harvest	2	412.82	206.41	14.95	<.001
Residual	40	552.44	13.81		
Total	53	2857.30			

Variate: FM leaves

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	8	14.366	1.796	1.22	
Rep.*Units* stratum					
Treat	2	28.067	14.033	9.55	<.001
Harvest	1	1.860	1.860	1.26	0.267
Treat.Harvest	2	10.077	5.038	3.43	0.042
Residual	40	58.808	1.470		
Total	53	113.178			

Variate: FM stem

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	8	48.299	6.037	1.34	
Rep.*Units* stratum					
Treat	2	93.050	46.525	10.33	<.001
Harvest	1	159.665	159.665	35.44	<.001
Treat.Harvest	2	79.532	39.766	8.83	<.001
Residual	40	180.207	4.505		
Total	53	560.753			

Variate: DM leaves

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	8	0.46873	0.05859	1.23	
Rep.*Units* stratum					
Treat	2	0.83821	0.41910	8.83	<.001
Harvest	1	1.43502	1.43502	30.24	<.001
Treat.Harvest	2	0.13907	0.06953	1.47	0.243
Residual	40	1.89799	0.04745		
Total	53	4.77901			

Variate: DM stem

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	8	0.6522	0.0815	0.76	
Rep.*Units* stratum					
Treat	2	1.0290	0.5145	4.82	0.013
Harvest	1	8.2316	8.2316	77.16	<.001
Treat.Harvest	2	1.4032	0.7016	6.58	0.003
Residual	40	4.2675	0.1067		
Total	53	15.5835			

Corchorus -2013 season

Variate: FM above ground

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	8	69.763	8.720	1.22	
Rep.*Units* stratum					
Treat	2	75.878	37.939	5.29	0.009
Harvest	1	917.641	917.641	127.97	<.001
Treat.Harvest	2	50.541	25.270	3.52	0.039
Residual	40	286.820	7.171		
Total	53	1400.643			

Variate: FM leaves

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	8	7.7356	0.9669	1.19	
Rep.*Units* stratum					
Treat	2	4.9680	2.4840	3.05	0.059
Harvest	1	1.5209	1.5209	1.87	0.180
Treat.Harvest	2	6.5430	3.2715	4.01	0.026
Residual	40	32.6016	0.8150		
Total	53	53.3691			

Variate: FM stem

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	8	34.677	4.335	1.31	
Rep.*Units* stratum					
Treat	2	32.481	16.240	4.89	0.013
Harvest	1	236.027	236.027	71.08	<.001
Treat.Harvest	2	7.297	3.649	1.10	0.343
Residual	40	132.824	3.321		
Total	53	443.306			

Variate: DM stem

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	8	2307.4	288.4	1.00	
Rep.*Units* stratum					
Treat	2	572.0	286.0	0.99	0.380
Harvest	1	167.5	167.5	0.58	0.450
Treat.Harvest	2	602.4	301.2	1.04	0.361
Residual	40	11532.9	288.3		
Total	53	15182.3			

Variate: DM leaves

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	8	0.20722	0.02590	0.48	
Rep.*Units* stratum					
Treat	2	0.44001	0.22000	4.08	0.024
Harvest	1	2.80414	2.80414	52.03	<.001
Treat.Harvest	2	0.20405	0.10203	1.89	0.164
Residual	40	2.15585	0.05390		
Total	53	5.81127			

Cowpea- 2013 season

Variate: FM above ground

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	8	25.314	3.164	1.29	
Rep.*Units* stratum					
Treat	2	149.511	74.756	30.54	<.001
Harvest	1	16.969	16.969	6.93	0.012
Treat.Harvest	2	8.329	4.165	1.70	0.195
Residual	40	97.923	2.448		
Total	53	298.046			

Variate: FM leaves

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	8	11.745	1.468	1.40	
Rep.*Units* stratum					
Treat	2	100.821	50.411	47.96	<.001
Harvest	1	5.326	5.326	5.07	0.030
Treat.Harvest	2	5.956	2.978	2.83	0.071
Residual	40	42.046	1.051		
Total	53	165.894			

Variate: FM stem

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	8	2.3132	0.2892	0.71	
Rep.*Units* stratum					
Treat	2	6.5790	3.2895	8.11	0.001
Harvest	1	0.7575	0.7575	1.87	0.179
Treat.Harvest	2	0.6190	0.3095	0.76	0.473
Residual	40	16.2324	0.4058		
Total	53	26.5012			

Variate: DM leaves

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	8	0.49552	0.06194	2.21	
Rep.*Units* stratum					
Treat	2	1.96073	0.98037	35.04	<.001
Harvest	1	0.56814	0.56814	20.31	<.001
Treat.Harvest	2	0.07695	0.03847	1.38	0.265
Residual	40	1.11916	0.02798		
Total	53	4.22051			

Variate: DM stem

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	8	0.09915	0.01239	0.58	
Rep.*Units* stratum					
Treat	2	0.35764	0.17882	8.33	<.001
Harvest	1	0.98603	0.98603	45.91	<.001
Treat.Harvest	2	0.10140	0.05070	2.36	0.107
Residual	40	0.85912	0.02148		
Total	53	2.40334			

APPENDIX 2: LIST OF ANOVAS FOR CHAPTER 4**Amaranthus- 2012/2013 season****Variate: FM above ground**

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	35.91	17.96	0.73	
Rep.*Units* stratum					
Treatment	2	214.93	107.47	4.35	0.031
Harvest	2	732.54	366.27	14.84	<.001
Treatment.Harvest	4	88.29	22.07	0.89	0.490
Residual	16	394.90	24.68		
Total	26	1466.57			

Variate: FM leaves

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	7.96	3.98	0.30	
Rep.*Units* stratum					
Treatment	2	87.59	43.80	3.28	0.064
Harvest	2	474.15	237.08	17.77	<.001
Treatment.Harvest	4	49.64	12.41	0.93	0.471
Residual	16	213.43	13.34		
Total	26	832.78			

Variate: FM stem

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	10.431	5.216	2.50	
Rep.*Units* stratum					
Treatment	2	19.685	9.842	4.72	0.025
Harvest	2	4.662	2.331	1.12	0.351
Treatment.Harvest	4	23.721	5.930	2.84	0.059
Residual	16	33.373	2.086		
Total	26	91.872			

Variate: DM leaves

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	0.0061	0.0031	0.01	
Rep.*Units* stratum					
Treatment	2	3.0035	1.5017	3.84	0.044
Harvest	2	11.4534	5.7267	14.63	<.001
Treatment.Harvest	4	1.5213	0.3803	0.97	0.450
Residual	16	6.2637	0.3915		
Total	26	22.2481			

Variate: DM stem

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	0.29007	0.14503	1.96	
Rep.*Units* stratum					
Treatment	2	0.22044	0.11022	1.49	0.255
Harvest	2	1.67086	0.83543	11.30	<.001
Treatment.Harvest	4	0.07464	0.01866	0.25	0.904
Residual	16	1.18249	0.07391		
Total	26	3.43850			

Corchorus -2012/2013 season

Variate: FM above ground

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	140.37	70.19	2.41	
Rep.*Units* stratum					
Treatment	2	100.56	50.28	1.73	0.209
Harvest	2	229.63	114.82	3.95	0.040
Treatment.Harvest	4	40.67	10.17	0.35	0.840
Residual	16	465.01	29.06		
Total	26	976.24			

Variate: FM stem

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	56.523	28.262	7.32	
Rep.*Units* stratum					
Treatment	2	51.377	25.689	6.65	0.008
Harvest	2	12.073	6.037	1.56	0.240
Treatment.Harvest	4	18.862	4.716	1.22	0.341
Residual	16	61.783	3.861		
Total	26	200.620			

Variate: FM leaves

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	81.46	40.73	3.79	
Rep.*Units* stratum					
Treatment	2	15.44	7.72	0.72	0.502
Harvest	2	211.07	105.54	9.82	0.002
Treatment.Harvest	4	15.44	3.86	0.36	0.834
Residual	16	171.93	10.75		
Total	26	495.34			

Variate: DM leaves

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	1.3758	0.6879	2.58	
Rep.*Units* stratum					
Treatment	2	0.0022	0.0011	0.00	0.996
Harvest	2	0.3829	0.1914	0.72	0.502
Treatment.Harvest	4	0.1406	0.0351	0.13	0.968
Residual	16	4.2581	0.2661		
Total	26	6.1596			

Variate: DM stem

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	1.3576	0.6788	2.22	
Rep.*Units* stratum					
Treatment	2	0.8512	0.4256	1.39	0.277
Harvest	2	4.4778	2.2389	7.33	0.006
Treatment.Harvest	4	0.1254	0.0314	0.10	0.980
Residual	16	4.8878	0.3055		
Total	26	11.6998			

Cowpea -2012/2013 seasons**Variate: FM above ground**

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	10.873	5.437	1.50	
Rep.*Units* stratum					
Treat	2	2.753	1.377	0.38	0.693
Harvest	1	5.372	5.372	1.48	0.251
Treat.Harvest	2	1.346	0.673	0.19	0.833
Residual	10	36.182	3.618		
Total	17	56.526			

Variate: FM stem

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	1.6204	0.8102	2.69	
Rep.*Units* stratum					
Treat	2	0.0000	0.0000	0.00	1.000
Harvest	1	0.6173	0.6173	2.05	0.183
Treat.Harvest	2	0.3086	0.1543	0.51	0.614
Residual	10	3.0093	0.3009		
Total	17	5.5556			

Variate: FM leaves

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	2.8549	1.4275	1.48	
Rep.*Units* stratum					
Treat	2	3.3179	1.6590	1.72	0.228
Harvest	1	0.6173	0.6173	0.64	0.442
Treat.Harvest	2	1.4660	0.7330	0.76	0.493
Residual	10	9.6451	0.9645		
Total	17	17.9012			

Variate: DM leaves

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	0.00481	0.00241	0.10	
Rep.*Units* stratum					
Treat	2	0.03676	0.01838	0.78	0.484
Harvest	1	0.06722	0.06722	2.85	0.122
Treat.Harvest	2	0.00731	0.00366	0.16	0.858
Residual	10	0.23556	0.02356		
Total	17	0.35167			

Variate: DM stem

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	0.12077	0.06039	0.88	
Rep.*Units* stratum					
Treat	2	0.14670	0.07335	1.07	0.380
Harvest	1	0.03705	0.03705	0.54	0.480
Treat.Harvest	2	0.12077	0.06039	0.88	0.445
Residual	10	0.68719	0.06872		
Total	17	1.11248			

Amaranthus -2012/2013 season**Variate: FM above ground**

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	8	1050.3	131.3	0.87	
Rep.*Units* stratum					
Treat	3	3757.6	1252.5	8.28	<.001
Harvest	1	592.7	592.7	3.92	0.053
Treat. Harvest	3	1148.6	382.9	2.53	0.066
Residual	56	8473.3	151.3		
Total	71	15022.5			

Variate FM leaves

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	8	24.727	3.091	0.69	
Rep.*Units* stratum					
Treat	3	174.545	58.182	12.92	<.001
Harvest	1	1.104	1.104	0.25	0.622
Treat.Harvest	3	13.807	4.602	1.02	0.390
Residual	56	252.232	4.504		
Total	71	466.416			

Variate: : FM stem

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	8	14.20	1.77	0.15	
Rep.*Units* stratum					
Treat	3	598.35	199.45	16.34	<.001
Harvest	1	42.65	42.65	3.49	0.067
Treat.Harvest	3	164.82	54.94	4.50	0.007
Residual	56	683.56	12.21		
Total	71	1503.59			

Variate: : DM stem

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	8	0.6284	0.0785	0.45	
Rep.*Units* stratum					
Treat	3	5.3426	1.7809	10.17	<.001
Harvest	1	0.4249	0.4249	2.43	0.125
Treat.Harvest	3	0.4367	0.1456	0.83	0.482
Residual	56	9.8062	0.1751		
Total	71	16.6387			

Corchorus -2012/2013 season

Variate: FM above ground

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	8	88.99	11.12	0.74	
Rep.*Units* stratum					
Treat	3	1483.75	494.58	32.92	<.001
Harvest	1	0.09	0.09	0.01	0.940
Treat.Harvest	3	294.32	98.11	6.53	<.001
Residual	56	841.29	15.02		
Total	71	2708.44			

Variate: FM leaves

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	8	12.548	1.569	0.65	
Rep.*Units* stratum					
Treat	3	90.529	30.176	12.58	<.001
Harvest	1	16.213	16.213	6.76	0.012
Treat.Harvest	3	23.081	7.694	3.21	0.030
Residual	56	134.327	2.399		
Total	71	276.698			

Variate: FM stem

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	8	77.287	9.661	2.04	
Rep.*Units* stratum					
Treat	3	370.379	123.460	26.12	<.001
Harvest	1	0.204	0.204	0.04	0.836
Treat.Harvest	3	113.625	37.875	8.01	<.001
Residual	56	264.726	4.727		
Total	71	826.221			

Variate: DM leaves

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	8	590.55	73.82	0.98	
Rep.*Units* stratum					
Treat	3	230.77	76.92	1.02	0.389
Harvest	1	65.17	65.17	0.87	0.355
Treat.Harvest	3	215.61	71.87	0.96	0.419
Residual	56	4204.00	75.07		
Total	71	5306.10			

Variate: DM stem

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	8	1.9043	0.2380	1.25	
Rep.*Units* stratum					
Treat	3	5.4048	1.8016	9.50	<.001
Harvest	1	1.2745	1.2745	6.72	0.012
Treat.Harvest	3	4.5266	1.5089	7.95	<.001
Residual	56	10.6232	0.1897		
Total	71	23.7334			

Cowpea-2012/2013 season

Variate: FM above ground

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	8	64.821	8.103	2.00	
Rep.*Units* stratum					
Treat	3	81.679	27.226	6.72	<.001
Harvest	1	69.620	69.620	17.19	<.001
Treat.Harvest	3	186.061	62.020	15.31	<.001
Residual	56	226.848	4.051		
Total	71	629.028			

Variate: FM stem

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	8	2.4929	0.3116	1.17	
Rep.*Units* stratum					
Treat	3	15.4524	5.1508	19.33	<.001
Harvest	1	10.8954	10.8954	40.88	<.001
Treat.Harvest	3	20.4058	6.8019	25.52	<.001
Residual	56	14.9256	0.2665		
Total	71	64.1721			

Variate: FM leaves

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	8	25.323	3.165	1.43	
Rep.*Units* stratum					
Treat	3	11.891	3.964	1.80	0.158
Harvest	1	1.188	1.188	0.54	0.466
Treat.Harvest	3	18.561	6.187	2.80	0.048
Residual	56	123.557	2.206		
Total	71	180.520			

Variate: DM leaves

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	8	0.49613	0.06202	1.12	
Rep.*Units* stratum					
Treat	3	1.32111	0.44037	7.98	<.001
Harvest	1	0.04828	0.04828	0.88	0.354
Treat.Harvest	3	0.85000	0.28333	5.14	0.003
Residual	56	3.08880	0.05516		
Total	71	5.80433			

Variate: DM stem

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	8	0.12272	0.01534	0.65	
Rep.*Units* stratum					
Treat	3	0.35698	0.11899	5.08	0.004
Harvest	1	0.00359	0.00359	0.15	0.697
Treat.Harvest	3	0.46301	0.15434	6.59	<.001
Residual	56	1.31246	0.02344		
Total	71	2.25877			

APPENDIX 3: LIST OF ANOVAS FOR CHAPTER 5

2012 season

Variate: CCI-1 June

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	82.01	41.00	1.58	
Rep.*Units* stratum					
Irrigation	1	0.37	0.37	0.01	0.905
Fertiliser	2	8219.48	4109.74	158.19	<.001
Spacing	2	956.07	478.04	18.40	<.001
WAT	8	934.91	116.86	4.50	<.001
Irrigation.Fertiliser	2	202.76	101.38	3.90	0.021
Irrigation.Spacing	2	77.95	38.97	1.50	0.225
Fertiliser.Spacing	4	197.29	49.32	1.90	0.110
Irrigation.WAT	8	180.71	22.59	0.87	0.543
Fertiliser.WAT	16	934.08	58.38	2.25	0.004
Spacing.WAT	16	495.87	30.99	1.19	0.272
Irrigation.Fertiliser.Spacing	4	89.15	22.29	0.86	0.490
Irrigation.Fertiliser.WAT	16	292.29	18.27	0.70	0.791
Irrigation.Spacing.WAT	16	511.28	31.96	1.23	0.243
Fertiliser.Spacing.WAT	32	643.53	20.11	0.77	0.808
Irrigation.Fertiliser.Spacing.WAT	32	853.52	26.67	1.03	0.432
Residual	322	8365.62	25.98		
Total	485	23036.90			

Variate: LAI-1 June

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	8.0070	4.0035	7.75	
Rep.*Units* stratum					
Irrigation	1	1.2897	1.2897	2.50	0.115
Fertiliser	2	31.5509	15.7754	30.52	<.001
Spacing	2	1.2447	0.6223	1.20	0.301
WAT	7	383.7996	54.8285	106.08	<.001
Irrigation.Fertiliser	2	1.8774	0.9387	1.82	0.165
Irrigation.Spacing	2	0.6313	0.3156	0.61	0.544
Fertiliser.Spacing	4	2.0326	0.5081	0.98	0.417
Irrigation.WAT	7	26.4986	3.7855	7.32	<.001
Fertiliser.WAT	14	20.1932	1.4424	2.79	<.001
Spacing.WAT	14	3.8057	0.2718	0.53	0.917
Irrigation.Fertiliser.Spacing					

Irrigation.Fertiliser.WAT	4	5.1694	1.2923	2.50	0.043
Irrigation.Spacing.WAT	14	3.0492	0.2178	0.42	0.967
Fertiliser.Spacing.WAT	14	5.4523	0.3895	0.75	0.719
Irrigation.Fertiliser.Spacing.WAT	28	21.0581	0.7521	1.46	0.069
Residual	28	14.9381	0.5335	1.03	0.425
Total	286	147.8221	0.5169		
Total	431	678.4198			

Variate: CONDUCTANCE-1 June

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	19853.	9927.	1.97	
Rep.*Units* stratum					
Irrigation	1	121652.	121652.	24.20	<.001
Fertiliser	2	975.	487.	0.10	0.908
Spacing	2	19936.	9968.	1.98	0.139
WAT	11	4529826.	411802.	81.93	<.001
Irrigation.Fertiliser	2	24605.	12303.	2.45	0.088
Irrigation.Spacing	2	9154.	4577.	0.91	0.403
Fertiliser.Spacing	4	20454.	5113.	1.02	0.398
Irrigation.WAT	11	240845.	21895.	4.36	<.001
Fertiliser.WAT	22	142862.	6494.	1.29	0.171
Spacing.WAT	22	68699.	3123.	0.62	0.910
Irrigation.Fertiliser.Spacing	4	26430.	6607.	1.31	0.264
Irrigation.Fertiliser.WAT	22	110433.	5020.	1.00	0.465
Irrigation.Spacing.WAT	22	78009.	3546.	0.71	0.835
Fertiliser.Spacing.WAT	44	193372.	4395.	0.87	0.701
Irrigation.Fertiliser.Spacing.WAT	44	196581.	4468.	0.89	0.676
Residual	430	2161267.	5026.		
Total	647	7964952.			

Variate: CCI-18 July

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	280.39	140.20	4.66	
Rep.*Units* stratum					
Irrigation	1	405.83	405.83	13.50	<.001
Fertiliser	2	4951.40	2475.70	82.35	<.001
Spacing	2	536.54	268.27	8.92	<.001
WAT	6	6193.60	1032.27	34.34	<.001

Irrigation.Fertiliser	2	385.19	192.59	6.41	0.002
Irrigation.Spacing	2	50.15	25.08	0.83	0.435
Fertiliser.Spacing	4	30.56	7.64	0.25	0.907
Irrigation.WAT	6	396.40	66.07	2.20	0.044
Fertiliser.WAT	12	1257.95	104.83	3.49	<.001
Spacing.WAT	12	202.90	16.91	0.56	0.871
Irrigation.Fertiliser.Spacing	4	95.49	23.87	0.79	0.530
Irrigation.Fertiliser.WAT	12	200.89	16.74	0.56	0.875
Irrigation.Spacing.WAT	12	212.25	17.69	0.59	0.851
Fertiliser.Spacing.WAT	24	597.35	24.89	0.83	0.700
Irrigation.Fertiliser.Spacing.WAT	24	610.88	25.45	0.85	0.675
Residual	250	7515.96	30.06		
Total	377	23923.75			

Variate: LAI-18 July

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	4.410	2.205	1.62	
Rep.*Units* stratum					
Irrigation	1	84.498	84.498	62.25	<.001
Fertiliser	2	67.256	33.628	24.77	<.001
Spacing	2	4.770	2.385	1.76	0.176
WAT	4	253.755	63.439	46.73	<.001
Irrigation.Fertiliser	2	4.389	2.194	1.62	0.202
Irrigation.Spacing	2	4.430	2.215	1.63	0.199
Fertiliser.Spacing	4	4.810	1.202	0.89	0.474
Irrigation.WAT	4	91.723	22.931	16.89	<.001
Fertiliser.WAT	8	9.504	1.188	0.88	0.539
Spacing.WAT	8	33.065	4.133	3.04	0.003
Irrigation.Fertiliser.Spacing	4	2.183	0.546	0.40	0.807
Irrigation.Fertiliser.WAT	8	10.501	1.313	0.97	0.463
Irrigation.Spacing.WAT	8	3.210	0.401	0.30	0.967
Fertiliser.Spacing.WAT	16	16.412	1.026	0.76	0.734
Irrigation.Fertiliser.Spacing.WAT	16	25.236	1.577	1.16	0.303
Residual	178	241.626	1.357		
Total	269	861.777			

Variate: CONDUCTANCE-18 July

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	26744.	13372.	3.29	
Rep.*Units* stratum					
Irrigation	1	6777.	6777.	1.67	0.198
Fertiliser	2	17000.	8500.	2.09	0.126
Spacing	2	186.	93.	0.02	0.977
WAT	6	393948.	65658.	16.14	<.001
Irrigation.Fertiliser	2	11009.	5504.	1.35	0.260
Irrigation.Spacing	2	4845.	2423.	0.60	0.552
Fertiliser.Spacing	4	10833.	2708.	0.67	0.616
Irrigation.WAT	6	63084.	10514.	2.58	0.019
Fertiliser.WAT	12	119937.	9995.	2.46	0.005
Spacing.WAT	12	40551.	3379.	0.83	0.619
Irrigation.Fertiliser.Spacing	4	10341.	2585.	0.64	0.638
Irrigation.Fertiliser.WAT	12	22170.	1848.	0.45	0.939
Irrigation.Spacing.WAT	12	33997.	2833.	0.70	0.755
Fertiliser.Spacing.WAT	24	66884.	2787.	0.68	0.865
Irrigation.Fertiliser.Spacing.WAT	24	59137.	2464.	0.61	0.928
Residual	250	1017167.	4069.		
Total	377	1904610.			

Variate: FM leaves -18 July

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	44.14	22.07	1.94	
Rep.*Units* stratum					
Irrigation	1	63.66	63.66	5.59	0.021
Fertiliser	2	1161.71	580.86	50.99	<.001
Spacing	2	43.46	21.73	1.91	0.156
Harvest	1	1913.01	1913.01	167.93	<.001
Irrigation.Fertiliser	2	32.03	16.01	1.41	0.252
Irrigation.Spacing	2	3.98	1.99	0.17	0.840
Fertiliser.Spacing	4	42.39	10.60	0.93	0.451
Irrigation.Harvest	1	93.42	93.42	8.20	0.006
Fertiliser.Harvest	2	74.54	37.27	3.27	0.044
Spacing.Harvest	2	19.73	9.86	0.87	0.425
Irrigation.Fertiliser.Spacing	4	47.79	11.95	1.05	0.388
Irrigation.Fertiliser.Harvest	2	50.26	25.13	2.21	0.118
Irrigation.Spacing.Harvest					

Fertiliser.Spacing.Harvest	2	1.96	0.98	0.09	0.918
Irrigation.Fertiliser.Spacing.Harvest	4	44.28	11.07	0.97	0.429
Residual	4	49.94	12.49	1.10	0.365
	70	797.41	11.39		
Total	107	4483.71			

Variate: DM leaves tha 1

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	0.17233	0.08617	2.16	
Rep.*Units* stratum					
Irrigation	1	0.12169	0.12169	3.05	0.085
Fertiliser	2	5.41037	2.70518	67.88	<.001
Spacing	2	0.18891	0.09445	2.37	0.101
Harvest	1	14.80815	14.80815	371.59	<.001
Irrigation.Fertiliser	2	0.04676	0.02338	0.59	0.559
Irrigation.Spacing	2	0.07243	0.03622	0.91	0.408
Fertiliser.Spacing	4	0.21877	0.05469	1.37	0.252
Irrigation.Harvest	1	0.21053	0.21053	5.28	0.025
Fertiliser.Harvest	2	0.02013	0.01007	0.25	0.777
Spacing.Harvest	2	0.25415	0.12707	3.19	0.047
Irrigation.Fertiliser.Spacing	4	0.15731	0.03933	0.99	0.420
Irrigation.Fertiliser.Harvest	2	0.10746	0.05373	1.35	0.266
Irrigation.Spacing.Harvest	2	0.04639	0.02320	0.58	0.561
Fertiliser.Spacing.Harvest	4	0.11120	0.02780	0.70	0.596
Irrigation.Fertiliser.Spacing.Harvest	4	0.16406	0.04101	1.03	0.398
Residual	70	2.78955	0.03985		
Total	107	24.90019			

Variate: leaf number-18 July

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	109.0	54.5	0.40	
Rep.*Units* stratum					
Irrigation	1	66.9	66.9	0.50	0.484
Fertiliser	2	10189.2	5094.6	37.79	<.001
Spacing	2	2130.6	1065.3	7.90	<.001
Harvest	1	5278.0	5278.0	39.15	<.001

Irrigation.Fertiliser	2	304.2	152.1	1.13	0.329
Irrigation.Spacing	2	129.1	64.6	0.48	0.621
Fertiliser.Spacing	4	1152.3	288.1	2.14	0.085
Irrigation.Harvest	1	374.1	374.1	2.77	0.100
Fertiliser.Harvest	2	3326.1	1663.1	12.34	<.001
Spacing.Harvest	2	36.4	18.2	0.13	0.874
Irrigation.Fertiliser.Spacing	4	88.1	22.0	0.16	0.956
Irrigation.Fertiliser.Harvest	2	605.7	302.9	2.25	0.113
Irrigation.Spacing.Harvest	2	99.1	49.5	0.37	0.694
Fertiliser.Spacing.Harvest	4	396.9	99.2	0.74	0.570
Irrigation.Fertiliser.Spacing.Harvest	4	366.2	91.6	0.68	0.609
Residual	70	9437.0	134.8		
Total	107	34089.0			

2013 season

Variate: height-1 June

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	105.29	52.64	2.73	
Rep.*Units* stratum					
Spacing	2	106.77	53.39	2.77	0.070
Irrigation	1	98.87	98.87	5.13	0.027
Fertiliser	2	48.16	24.08	1.25	0.293
WAT	1	632.36	632.36	32.80	<.001
Spacing.Irrigation	2	30.67	15.34	0.80	0.455
Spacing.Fertiliser	4	137.43	34.36	1.78	0.142
Irrigation.Fertiliser	2	89.02	44.51	2.31	0.107
Spacing.WAT	2	1.09	0.55	0.03	0.972
Irrigation.WAT	1	3.23	3.23	0.17	0.684
Fertiliser.WAT	2	4.01	2.00	0.10	0.901
Spacing.Irrigation.Fertiliser	4	80.20	20.05	1.04	0.393
Spacing.Irrigation.WAT	2	0.65	0.33	0.02	0.983
Spacing.Fertiliser.WAT	4	18.81	4.70	0.24	0.912
Irrigation.Fertiliser.WAT	2	10.70	5.35	0.28	0.759
Spacing.Irrigation.Fertiliser.WAT	4	15.41	3.85	0.20	0.938
Residual	70	1349.53	19.28		
Total	107	2732.19			

Variate: leaves-1 June

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	0.321	0.160	0.13	
Rep.*Units* stratum					
Spacing	2	3.352	1.676	1.36	0.264
Irrigation	1	0.124	0.124	0.10	0.752
Fertiliser	2	2.858	1.429	1.16	0.320
WAT	1	67.952	67.952	55.04	<.001
Spacing.Irrigation	2	5.558	2.779	2.25	0.113
Spacing.Fertiliser	4	10.216	2.554	2.07	0.094
Irrigation.Fertiliser	2	7.224	3.612	2.93	0.060
Spacing.WAT	2	0.508	0.254	0.21	0.814
Irrigation.WAT	1	2.676	2.676	2.17	0.145
Fertiliser.WAT	2	0.706	0.353	0.29	0.752
Spacing.Irrigation.Fertiliser	4	0.862	0.216	0.17	0.951
Spacing.Irrigation.WAT	2	0.451	0.225	0.18	0.834
Spacing.Fertiliser.WAT	4	1.751	0.438	0.35	0.840
Irrigation.Fertiliser.WAT	2	4.265	2.133	1.73	0.185
Spacing.Irrigation.Fertiliser.WAT	4	1.302	0.326	0.26	0.900
Residual	70	86.420	1.235		
Total	107	196.546			

Variate: Fv Fm-1 June

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	0.003578	0.001789	1.01	
Rep.*Units* stratum					
Spacing	2	0.005426	0.002713	1.53	0.220
Irrigation	1	0.012546	0.012546	7.09	0.009
Fertiliser	2	0.008461	0.004230	2.39	0.096
WAT	2	0.030194	0.015097	8.54	<.001
Spacing.Irrigation	2	0.010061	0.005030	2.84	0.063
Spacing.Fertiliser	4	0.004064	0.001016	0.57	0.682
Irrigation.Fertiliser	2	0.004328	0.002164	1.22	0.298
Spacing.WAT	4	0.000303	0.000076	0.04	0.996
Irrigation.WAT	2	0.009172	0.004586	2.59	0.080
Fertiliser.WAT	4	0.005547	0.001387	0.78	0.538
Spacing.Irrigation.Fertiliser	4	0.003677	0.000919	0.52	0.721
Spacing.Irrigation.WAT	4	0.004159	0.001040	0.59	0.672
Spacing.Fertiliser.WAT	8	0.016471	0.002059	1.16	0.328
Irrigation.Fertiliser.WAT	4	0.011481	0.002870	1.62	0.174

Spacing.Irrigation.Fertiliser.WAT	8	0.010089	0.001261	0.71	0.680
Residual	106	0.187492	0.001769		
Total	161	0.327048			

Variate: height-18 July

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	20.767	10.384	1.74	
Rep.*Units* stratum					
Spacing	2	47.570	23.785	3.99	0.023
Irrigation	1	29.037	29.037	4.87	0.031
Fertiliser	2	586.459	293.229	49.20	<.001
WAT	1	327.259	327.259	54.91	<.001
Spacing.Irrigation	2	12.340	6.170	1.04	0.360
Spacing.Fertiliser	4	39.344	9.836	1.65	0.171
Irrigation.Fertiliser	2	2.167	1.083	0.18	0.834
Spacing.WAT	2	5.377	2.688	0.45	0.639
Irrigation.WAT	1	2.177	2.177	0.37	0.548
Fertiliser.WAT	2	8.130	4.065	0.68	0.509
Spacing.Irrigation.Fertiliser	4	22.623	5.656	0.95	0.441
Spacing.Irrigation.WAT	2	0.039	0.020	0.00	0.997
Spacing.Fertiliser.WAT	4	6.660	1.665	0.28	0.890
Irrigation.Fertiliser.WAT	2	0.669	0.334	0.06	0.945
Spacing.Irrigation.Fertiliser.WAT	4	2.763	0.691	0.12	0.976
Residual	70	417.158	5.959		
Total	107	1530.539			

Variate: leaves-18 July

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	3.8416	1.9208	6.11	
Rep.*Units* stratum					
Spacing	2	2.9774	1.4887	4.73	0.012
Irrigation	1	0.2973	0.2973	0.95	0.334
Fertiliser	2	5.3848	2.6924	8.56	<.001
WAT	1	9.6800	9.6800	30.79	<.001
Spacing.Irrigation	2	0.2737	0.1368	0.44	0.649
Spacing.Fertiliser	4	0.6584	0.1646	0.52	0.719

Irrigation.Fertiliser	2	0.3724	0.1862	0.59	0.556
Spacing.WAT	2	0.7428	0.3714	1.18	0.313
Irrigation.WAT	1	0.0010	0.0010	0.00	0.955
Fertiliser.WAT	2	0.3477	0.1739	0.55	0.578
Spacing.Irrigation.Fertiliser	4	1.2510	0.3128	0.99	0.416
Spacing.Irrigation.WAT	2	0.1872	0.0936	0.30	0.743
Spacing.Fertiliser.WAT	4	0.9053	0.2263	0.72	0.581
Irrigation.Fertiliser.WAT	2	0.1008	0.0504	0.16	0.852
Spacing.Irrigation.Fertiliser.WAT	4	0.6831	0.1708	0.54	0.705
Residual	70	22.0103	0.3144		
Total	107	49.7150			

Variate: FM leaves tha-18 July

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	11.507	5.754	0.94	
Rep.*Units* stratum					
Spacing	2	1.102	0.551	0.09	0.914
Irrigation	1	58.028	58.028	9.44	0.003
Harvest	1	332.421	332.421	54.07	<.001
Fertiliser	2	174.860	87.430	14.22	<.001
Spacing.Irrigation	2	10.523	5.261	0.86	0.429
Spacing.Harvest	2	34.148	17.074	2.78	0.069
Irrigation.Harvest	1	1.274	1.274	0.21	0.650
Spacing.Fertiliser	4	28.850	7.213	1.17	0.330
Irrigation.Fertiliser	2	8.473	4.237	0.69	0.505
Harvest.Fertiliser	2	72.099	36.049	5.86	0.004
Spacing.Irrigation.Harvest	2	10.952	5.476	0.89	0.415
Spacing.Irrigation.Fertiliser	4	54.473	13.618	2.22	0.076
Spacing.Harvest.Fertiliser	4	20.547	5.137	0.84	0.507
Irrigation.Harvest.Fertiliser	2	0.938	0.469	0.08	0.927
Spacing.Irrigation.Harvest.Fertiliser	4	12.108	3.027	0.49	0.741
Residual	70	430.329	6.148		
Total	107	1262.631			

Variate: DM t ha-18 July

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	0.07450	0.03725	0.43	
Rep.*Units* stratum					
Spacing	2	0.11016	0.05508	0.63	0.536
Irrigation	1	1.03512	1.03512	11.83	<.001
Harvest	1	2.15539	2.15539	24.63	<.001
Fertiliser	2	0.69887	0.34943	3.99	0.023
Spacing.Irrigation	2	0.48893	0.24446	2.79	0.068
Spacing.Harvest	2	0.42496	0.21248	2.43	0.096
Irrigation.Harvest	1	0.00035	0.00035	0.00	0.950
Spacing.Fertiliser	4	0.21716	0.05429	0.62	0.649
Irrigation.Fertiliser	2	0.10328	0.05164	0.59	0.557
Harvest.Fertiliser	2	0.61458	0.30729	3.51	0.035
Spacing.Irrigation.Harvest	2	0.44009	0.22005	2.51	0.088
Spacing.Irrigation.Fertiliser	4	0.27308	0.06827	0.78	0.542
Spacing.Harvest.Fertiliser	4	0.78800	0.19700	2.25	0.072
Irrigation.Harvest.Fertiliser	2	0.11125	0.05563	0.64	0.533
Spacing.Irrigation.Harvest.Fertiliser	4	0.50780	0.12695	1.45	0.227
Residual	70	6.12598	0.08751		
Total	107	14.16950			

Variate: no of leaves-18 July

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	37.9	18.9	0.08	
Rep.*Units* stratum					
Spacing	2	416.2	208.1	0.89	0.416
Irrigation	1	489.8	489.8	2.09	0.153
Harvest	1	13962.8	13962.8	59.55	<.001
Fertiliser	2	213.6	106.8	0.46	0.636
Spacing.Irrigation	2	841.7	420.8	1.79	0.174
Spacing.Harvest	2	530.0	265.0	1.13	0.329
Irrigation.Harvest	1	3.7	3.7	0.02	0.900
Spacing.Fertiliser	4	1098.9	274.7	1.17	0.331
Irrigation.Fertiliser	2	770.3	385.1	1.64	0.201
Harvest.Fertiliser	2	776.1	388.0	1.66	0.198
Spacing.Irrigation.Harvest	2	361.2	180.6	0.77	0.467

Spacing.Irrigation.Fertiliser	4	373.9	93.5	0.40	0.809
Spacing.Harvest.Fertiliser	4	1466.8	366.7	1.56	0.194
Irrigation.Harvest.Fertiliser	2	555.6	277.8	1.18	0.312
Spacing.Irrigation.Harvest.Fertiliser	4	286.4	71.6	0.31	0.873
Residual	70	16412.1	234.5		
Total	107	38597.1			