THE EFFECT OF NITROGEN FERTILISATION ON THE GROWTH, YIELD AND QUALITY OF SWISS CHARD (*Beta vulgaris* var. *cicla*)

by

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Submitted in fulfilment of the requirements for the degree of Magister Scientiae Agriculturae

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May 2008

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DECLARATION

I declare that this dissertation hereby submitted by me for the Magister Scientiae Agriculturae degree at the University of the Free State is my own independent work and has not previously been submitted by me at another university. I further more cede copyright of the dissertation in favor of the University of the Free State.

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ABSTRACT

THE EFFECT OF NITROGEN FERTILISATION ON THE GROWTH, YIELD AND QUALITY OF SWISS CHARD (*Beta vulgaris* var. *cicla*)

The sustainability of subsistence farming associated with the health of rural communities' necessitated research on Swiss chard as it forms an integral part of food consumed by the poor in developing countries. Two separate pot experiments were carried out during the 2005/06 and 2006/07 seasons in the glasshouse of the Department of Soil, Crop and Climate Sciences at the University of the Free State. The objective of this study was to determine the effect of nitrogen fertiliser on growth, yield and quality of Swiss chard.

The first pot trial was conducted to evaluate the effect of five nitrogen levels (0, 50, 100, 200, 400 kg N ha^{-1}) and four application times on the growth, yield and quality of two Swiss chard cultivars ('Fordhook Giant' and 'Rhubarb'). Two Swiss chard seedlings were planted per pot, filled with topsoil of the fine sandy loam Bainsvlei form. Two weeks after planting plants were thinned to one seedling per pot. Different nitrogen levels were applied to the relevant pots as follows: once every second, fourth, sixth or eighth week. It was only the total dry mass per plant and total nitrogen content per leaf of 'Rhubarb' plants that was significant higher than that of 'Fordhook Giant'. The other parameters measured for the two cultivars did not differ significantly from each other. Nitrogen levels positively influenced the early growth, yield and quality parameters measured. It was the highest nitrogen level (400 kg N ha^{-1}) that resulted in the highest number of leaves harvested, leaf fresh and dry mass, leaf area and leaf nitrogen content. Nitrogen application times significantly influenced only the early growth of Swiss chard plants and the nitrogen content of leaves. Early plant growth reacted better where nitrogen was split into three equal applications (T_4). The total nitrogen content of Swiss chard leaves was significantly higher where nitrogen was split into five equal (T_2) or three equal (T_4) applications.

In the second pot trial the effect of different nitrogen sources applied at different levels on the growth, yield and quality of Swiss chard cultivars was determined. The response of Swiss chard plants to nine nitrogen levels (0, 100, 200, 300, 400, 500, 600, 700, 800 kg N ha^{-1}) from six different nitrogen sources (ammonium nitrate, potassium nitrate, calcium nitrate, ammonium sulphate, urea ammonium nitrate and urea) were studied. Based on the findings of this study, nitrogen significantly influenced growth, number of leaves harvested, leaf fresh and dry mass, leaf area and leaf nitrate content of 'Fordhook Giant' plants with best results obtained at 800 kg N ha^{-1}. Nitrogen sources did not influence either the total number of leaves harvested nor the fresh mass of harvested Swiss chard leaves. In both cases, ammonium nitrate gave the best results and calcium nitrate the poorest. Urea influenced the leaf area positively followed by urea ammonium nitrate, with calcium nitrate resulting in the smallest leaf area per plant. Dry mass of Swiss chard leaves was also significantly higher where urea was used as nitrogen source compared to where calcium nitrate was used. No significant differences amongst the other nitrogen sources. Ammonium nitrate and potassium nitrate significantly stimulated the accumulation of nitrate in Swiss chard leaves, whereas the other nitrogen sources did not play any role in nitrate accumulation in the leaves of Swiss chard.

Keywords: nitrogen application level, nitrogen application time, leaf number, leaf area, leaf fresh mass, leaf dry mass, leaf nitrogen content, leaf nitrate content

UITTREKSEL

DIE EFFEK VAN STIKSTOFBEMESTING OP DIE GROEI, OPBRENGS EN KWALITEIT VAN SNYBEET (*Beta vulgaris* var. *cicla*)

Die volhoubaarheid van bestaansboerdery, tesame met die gesondheid van landelike gemeenskappe, noodsaak navorsing op snybeet juis omdat dit so 'n integrale deel van arm gemeenskappe se voedselbehoefte uitmaak. Twee afsonderlike potproewe is gedurende die 2005/06 en 2006/07 seisoen in die glashuise van die Departement Grond-, Gewas- en Klimaatwetenskappe by die Universiteit van die Vrystaat uitgevoer. Die doel van die studie was om die invloed van stikstofbemesting op die groei, opbrengs en kwaliteit van snybeet te ondersoek.

Die eerste potproef is uitgevoer om die invloed van vyf stikstofpeile (0, 50, 100, 200, 400 kg N ha^{-1}) en vier toedieningstye op die groei, opbrengs en kwaliteit van twee snybeet cultivars ('Fordhook Giant' en 'Rhubarb') te ondersoek. Twee saailinge is aanvanklik per pot, gevul met grond (fyn sandleem Bainsvleivorm), geplant. Na twee weke is die saailinge uitgedun tot een per pot. Verskillende stikstofpeile is as volg aan die relevante potte toegedien: een keer elke tweede, vierde, sesde of agste week. Dit is slegs die totale droë massa per plant en die totale stikstofinhoud per blaar van 'Rhubarb' wat betekenisvol verskil het van 'Fordhook Giant'. Die ander parameters het nie betekenisvol verskil tussen die twee cultivars nie. Die vroeë groei (eerste agt weke na plant), opbrengs en kwaliteit van snybeet is positief deur die verskillende stikstofpeile beïnvloed. Die hoogste stikstofvlak (400 kg N ha^{-1}) het die beste resultate gelewer vir die aantal blare geoes, vars- en droë massa van die blare, blaaroppervlak en stikstofinhoud van die blare. Die toedieningstye van stikstof het die vroeë groei van snybeet asook die stikstofinhoud van die blare betekenisvol beïnvloed. Vroeë groei van snybeet het beter gereageer waar stikstof toegedien is in minder paaiemente (T_4). Die totale stikstofinhoud van snybeetblare was betekenisvol hoër waar stikstof opgedeel is in vyf (T_2) of drie (T_4) gelyke toedienings.

Die tweede potproef is uitgevoer om die invloed van verskillende stikstofbronne, toegedien teen verskillende peile, op die groei, opbrengs en kwaliteit van snybeet te bepaal. Die reaksie van ses stikstofbronne (ammoniumnitraat, kaliumnitraat, kalsiumnitraat, ammoniumsulfaat, ureumammoniumnitraat en ureum) toegedien teen nege verskillende stikstofpeile (0, 100, 200, 300, 400, 500, 600, 700, 800 kg N ha^{-1}) is ondersoek. Resultate van die studie dui duidelik daarop dat stikstof ‘n positiewe invloed op groei, aantal blare geoes, blaarvars en –droë massa, blaaroppervlak, asook die nitraatinhoud van die blare gehad het. Die beste resultate is verkry waar 800 kg N ha^{-1} toegedien is. Stikstofbronne het nie die totale aantal blare geoes of die varsmassa van die blare van snybeet betekenisvol beïnvloed nie. In beide gevalle het ammoniumnitraat die beste resultate gegee en kalsiumnitraat die swakste. Ureum het die blaaroppervlak van snybeet positief beïnvloed gevolg deur ureumammoniumnitraat terwyl kalsiumnitraat die swakste gevaar het. Droë massa van snybeet se blare was betekenisvol hoër waar ureum toegedien is as waar kalsiumnitraat toegedien is as stikstofbron. Ammoniumnitraat en kaliumnitraat het die akkumulasie van nitraat in die blare van snybeet betekenisvol gestimuleer terwyl die ander stikstofbronne nie ‘n betekenisvolle rol gespeel het nie.

DEDICATION

This dissertation is dedicated to my father,

***Motseki Emmanuel Motseki**, who taught me that even the*

largest task can be successfully complete

if it is done one step at a time.

I also wish to dedicate this dissertation to my late mother,
***`Mapontśo Francina Motseki**. She taught me to persevere and*
prepared me to face challenges with faith and humility. She was a constant
source of inspiration to my life. Although she is not here to give me strength
and support I always feel her presence, which used to urge
me to strive to achieve my goals in life.

ACKNOWLEDGEMENTS

The academic program that culminated into this dissertation and the successful completion thereof would not have been possible without the numerous assistance - moral, financial and academic - of some people whom I would love to express my gratitude to.

I would like to acknowledge the following people for their contribution towards the successful completion of this study:

- First, my gratitude goes to God almighty whose guidance and grace have kept me in the process of life and its endeavors. Everything flowed out smoothly because I kept GOD first, with GOD everything is possible.

- Many thanks to my supervisor Dr. Gesine Engelbrecht, who read my numerous drafts, assisted making some sense of the confusion and also for her unconditional support, patience and positive contributions during the study. Without her support this study would not have been possible. When it seemed very tough to continue, her voice of reassurance was a great source of energy. Thank you again Dr. Engelbrecht.

- My co-supervisor, Dr. Gert Ceronio for his valuable contributions throughout the study. He is greatly acknowledged for his constructive corrections and suggestions for improvements.

- The staff of Soil, Crop and Climate Sciences Department are worthy of my thanks. I wouldn't have made it without their contribution. From the inception of the program to its completion it was marked by their constant support and good advice. In this category, I wish to express my profound gratitude especially to Dr. Elmarie Van der Watt.

- Thanks to the University of Free State for providing me with the financial means to complete my studies. The head of the Department of Soil, Crop and Climate Sciences, Prof. CC Du Preez is warmly acknowledged for all the support he provided. I will always be grateful to you.

- My family, especially my father, who always encouraged me during my studies, my brother and three sisters for being so understanding and supporting me emotionally. My late mother – My source of inspiration. My little angel daughter `Nteboheleng` this is for you.

- My family, relatives, and friends deserve my gratitude for their encouragements during this work which sometimes felt like a never ending story.

- And finally, a special thank you to my big friend, Mr. Sello Paul Rasello for being so understanding and also bringing humour during difficult times and his inspiring words when things really got tough. *Kea leboha Motaung!*

***To everyone who assisted me in any other form to make this project a reality, whose name did not appear here, I still express my appreciation.**

Pontśo Christina Motseki

May 2008

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CHAPTER 1

MOTIVATION AND OBJECTIVES

1.1 MOTIVATION

South Africa is self-sufficient with regard to vegetable production and also exports both, fresh and processed vegetables (Olivier, 1974). However, hunger and malnutrition are still found in many rural and urban areas. It has been estimated that in South Africa at least 3 million people under the age of 15 suffer from malnutrition (Louw, 1992). Vegetables are of great importance in alleviating malnutrition as they contribute significantly to the number of calories and nutrients in daily diets. The scarcity of vegetables in the diet is a major cause of vitamin A deficiency, which causes blindness and even death in young children throughout the semi-arid and arid areas of Africa (AVRDC, 1990).

Vegetables are produced in most parts of the South Africa. However, in certain areas farmers tend to concentrate on specific crops; for example, green beans are mainly grown in the Tzaneen. From 2004/05 to 2005/06 (July-June), the total production of vegetables (excluding potatoes) decreased by 1.0%, from 2 206 431 to 2 184 763 tons. Concerning the major vegetable types in terms of volumes produced, increases occurred in the case of carrots, pumpkins and onions, which increased by 3.0, 2.7 and 1.0%, respectively (Directorate Agricultural Information, 2006). The largest decrease, 6.7%, was found in the production of cabbages which was followed by tomatoes with 2.8% for the same period. Approximately 53% of the volume of vegetables produced is traded on the major South African fresh produce markets. The total volume of vegetables (excluding potatoes) sold on these markets during 2005/06 amounted to 1 160 151 t, while 1 173 277 t were sold during 2004/05, which presents a decrease of 1.1% (Directorate Agricultural Information, 2006).

Swiss chard (*Beta vulgaris* var. *cicla*) belongs to the family Chenopodiaceae. Horticultural history indicates that Swiss chard was cultivated as early as 350 B.C.

(MacGillivray, 1953; Splittstoesser, 1990). It is a dicotyledonous biennial crop, generally treated as an annual and it can be harvested continually for a period of four to five months. Swiss chard is grown for its large crisp and fleshy leaves and is found to be gaining in popularity as either a baby or a mature vegetable. The leaves which are a rich green colour are extremely nutritious and high in fibre (MacGillivray, 1953; Splittstoesser, 1990).

Swiss chard is a hardy, cool season crop (MacGillivray, 1953) and a very nutritively demanding crop (Santamaria *et al.*, 1999a). The mineral content of Swiss chard leaves is influenced by the amount, frequency and method of fertilisation (Santamaria *et al.*, 1999a). The nutritive value of Swiss chard also differs significantly between different cultivars (Pokluda & Kuben, 2002). Swiss chard is characterised by high sodium and oxalates levels (1678-6031 mg kg^{-1} fresh mass) (Santamaria *et al.*, 1999b). The mean sodium content of 13 Swiss chard cultivar leaves has been reported as 2100 mg kg^{-1} fresh mass, potassium 4198 mg kg^{-1} fresh mass, calcium 481 mg kg^{-1} fresh mass and magnesium 361 mg kg^{-1} fresh mass (Pokluda & Kuben, 2002).

It is highly relevant to consider factors that might help in maintaining Swiss chard production. Correct cultural practices such as the adequate application of fertilisers have to be adhered to and carried out in order to obtain good yields (Everaarts, 1993). Fertilisation is one of the methods used to increase yield and nitrogen is the most commonly used nutrient. The usage of fertiliser has increased considerably over the years (MacGillivray, 1953; Bidwell, 1979; Splittstoesser, 1979; Goh & Vityakon, 1983; 1986). Concomitant with this increased fertiliser application is the need to establish optimum application levels of fertilisers for growing vegetables.

Bearing in mind that nitrogen fertilisation plays an important role in the production of vegetables, and even more so in leafy vegetables such as Swiss chard, and that agricultural crop production has to increase considerably to attain feeding the growing world population. Efforts should therefore be focused on increasing crop yields per hectare rather than increasing the area for agricultural production.

1.2 HYPOTHESIS

- The growth, yield and quality of Swiss chard will increase by increasing nitrogen levels.
- The growth, yield and quality of Swiss chard will increase with more nitrogen application times.
- The growth, yield and quality of Swiss chard will differ with different nitrogen sources.

1.3 MAIN OBJECTIVE

The main objective of this study was to quantify the effect of nitrogen on the growth, yield and quality of Swiss chard (*Beta vulgaris* var. *cicla*).

1.3.1 Sub-objectives

- To determine the effect of different nitrogen levels applied at different times on the growth, yield and quality of two Swiss chard cultivars.
- To determine the effect of different nitrogen sources on growth, yield and quality of Swiss chard.

CHAPTER 2

LITERATURE REVIEW

2.1 INTRODUCTION

Beta vulgaris var. *cicla* (Swiss chard) is important in the human diet, especially in poorer South African communities. As was mentioned earlier, Swiss chard belongs to the Chenopodiaceae family. This family of vegetables is nutritionally low in fat and cholesterol, yet is a rich source of protein and contains all the essential amino acids. It is also a valuable source of vitamin A, C, E, K and iron (Table 2.1). Ten milligrams of iron per day is recommended for humans as spinach is high in iron, but the high oxalates in spinach may reduce iron intake. These vegetables are also surprisingly high in other minerals such as calcium, magnesium, phosphorus, copper, manganese and potassium. On the other hand, Swiss chard specifically, is very high in sodium, and a large portion of the calories is from sugars (USDA SR20, 2007).

To produce optimum yields of good quality Swiss chard, often high amounts of nitrogen fertiliser are applied. The recommended total amount of nitrogen fertiliser for Swiss chard is 160 to 260 kg ha^{-1} (FSSA, 2007). In reality, the amount of nitrogen fertiliser used is probably higher as farmers may apply more fertiliser than recommended to secure yields. Nitrogen is an element required for plant growth and is an important component of proteins, enzymes and vitamins in plants. Furthermore, it is a central part of the essential photosynthetic molecule, chlorophyll. It is present in plant alkaloids and thousands of other substances that are of great social and economic importance in our society (Bidwell, 1979).

Plants absorb nitrogen in the form of nitrate ions (NO_3^-) and ammonium ions (NH_4^+) through their roots. The quantity of nitrogen absorbed by a plant depends on many variables, including the stage of plant growth, the concentration and balances of other nutrients in the soil, the availability of soil water, and climate conditions. Most crops take up nitrate in greater amounts than ammonium.

Table 2.1: Water and nutrient content of fresh leaves per 100 g of Swiss chard (USDA SR20, 2007)

Nutrition Factor	Content per measure
Macro components	
Total lipids (Fat) (g)	0.28
Total carbohydrates (g)	3.74
Protein (g)	0.30
Calories (kcal)	19.0
Carbohydrates components	
Dietary fiber (g)	3.70
Amino acids	
Tryptophan (g)	0.02
Leucine (g)	0.13
Lysine (g)	0.10
Histidine (g)	0.04
Fats	
Cholesterol (mg)	0.0
Fatty acids, total saturated	0.06
Fatty acids, total monounsaturated	0.08
Fatty acids, total polyunsaturated	0.14
Vitamins	
Vit. A (IU)	6116
Vit. C (Ascorbic Acid) (mg)	30.0
Thiamin (mg)	0.04
Riboflavin (mg)	0.09
Folate total ((mcg)	14.0
Choline total (mg)	18.0
Betaine (mg)	0.30
Vitamin E (mg)	1.89
Vitamine K (mcg)	830.0
Niacin (mg)	0.40
Minerals	
Calcium (mg)	51
Iron (mg)	1.8
Potassium (mg)	379
Magnesium (mg)	81
Sodium (mg)	213
Zinc (mg)	0.36
Phosphorous (mg)	46
Copper (mg)	0.179
Manganese (mg)	0.366
Selenium (mcg)	0.9
Other	
Water (%)	92.66
Carotene, Beta (mcg)	3647

Nitrate, unlike ammonium, accumulates in plant tissue, which cause nitrogen to be available in greater amounts than required for optimal growth. While nitrate is easily leached from soils by percolating water, ammonium is normally converted to nitrate in the soil before the plant can use it (Bidwell, 1979; Makovic & Djurovka, 1990).

Nitrogen is necessary to produce a reliable and optimal yield of quality vegetables. It is however, the most difficult element to manage in a fertilisation system in order to ensure an adequate, yet not excessive amount of available nitrogen within the rhizosphere from planting to harvest (Peck, 1981). One key to efficient fertilisation is to avoid over-fertilisation. A crop that is over-fertilised with nitrogen may be more susceptible to diseases than those that are not, or may have elevated nitrate levels in vegetable tissues (Everaarts, 1994). Elevated nitrate levels influence the quality of vegetables in a variety of ways. Other vegetable crops such as Brussel sprouts have been found to taste even more bitter when over-fertilised with nitrogen, producing undesirable, elongated sprouts. Vitamin C levels in vegetables drop as nitrate levels increase. Besides the detrimental effects of nitrogen over-fertilisation of crops also causes water pollution through leaching of nitrate (Babik, Rumpel & Elkner, 1996).

Nitrogen is no more important to plant survival than any other essential element. However, it is required in a much greater quantity than most other nutrients, so cropping practices often call for large applications of nitrogen fertiliser to maximise yields (Splittstoesser, 1990).

2.2 NITROGEN FERTILISATION AND CROP GROWTH

There is general agreement that of all the improvements or corrections that have been made to soil the application of nitrogen fertiliser has the greatest effect in terms of increasing crop production. As the supply or availability of growth factors such as water and mineral nutrients increase, the growth rate and yield increase. Nitrogen is found to be the most important growth limiting factor in numerous field experiments that have been carried out in the past (Mengel & Kirby, 1987; Wiesler & Horst, 1992).

Healthy crop growth is one of the best preventions against nitrate leaching because a healthy crop can grow fast and absorb nitrogen from the soil. The yield response to nitrogen fertilisation depends greatly on moisture. Improved moisture conditions usually translate into higher yields up to a point where other limiting factors come into play. Excess moisture can reduce yield due to leaching losses of nitrate, as well as loss of nitrates by conversion (denitrification) to gases that escape from the soil. High levels of available soil nitrogen early on in the growing season can promote excessive vegetative growth and high water use (Wiesler & Horst, 1992).

2.2.1 Crop nutrition with special reference to nitrogen fertilisation

Plants use inorganic minerals for nutrition, whether grown in the field or in a container. Many different chemical elements are found in plants, but only sixteen commonly occurring have been found, of which some are essential. The essential mineral elements may be classified as major elements or macro-nutrients that are required in relatively large amounts (Bidwell, 1979). Nitrogen, phosphorus and potassium are the three major nutrients of concern to producers. Nitrogen is usually more responsible for increasing the growth of plants than any other element. It is a component of proteins and is therefore involved in regulating most processes that occur in plants.

Nitrogen deficiency causes poor growth, stunted plants and low yields (Mengel & Kirby, 1987; Splittstoesser, 1990). Because nitrogen is a component of chlorophyll, a yellow colour beginning with the lower leaves, is a common symptom of nitrogen deficiency. Nitrogen tends to promote vegetative growth relatively more than reproductive growth as it is the key factor in vegetable growth and yield. Plants given excess nitrogen tend to be tall with weak stems and under certain conditions an oversupply of nitrogen can cause lodging (Bidwell, 1979; Mengel & Kirby, 1987). Nitrogen is usually deficient in soils when provided to plants as inorganic fertilisers, but is also present in the air although plants cannot directly utilise it.

All vegetables have different requirements, especially with regard to nitrogen application (Goodlass *et al.*, 1997). When plants are given a large amount of nitrogen fertiliser, the plants produce large amounts of vegetative growth. Leafy greens such as mustard, cabbage and spinach are heavy users of nitrogen. Broccoli and sweet corn also require more nitrogen than some other vegetables. Therefore, green leafy vegetables are usually fertilised with nitrogen to obtain high yields. Phosphorus and potassium are important to the proper development of roots and seeds. Legumes obtain nitrogen from the atmosphere and do not require heavy nitrogen fertilisation. Excessive nitrogen in some plants may lead to luxury consumption and nitrate accumulation. The consumption of vegetables high in nitrate may be dangerous to consumer health (Vieira, Vasconcelos & Monteiro, 1998), due to the possibility of methaemoglobinaemia, as well as the conversion of nitrate into nitrite in saliva which is thought to lead to the formation of carcinogenic nitrosamines in the intestinal tract (Vulsteke & Biston, 1978; Van Eysinga, 1984; Vogtmann *et al*., 1984; Santamaria *et al*., 1999a; Turan & Sevimli, 2005; Santamaria, 2006). Thus, nitrate in drinking water is considered to contribute to an increased cancer risk of the urinary tract, bladder and oesophagus due to endogenous nitrate reduction to nitrite (Goebell *et al*., 2004; Anjana & Muhammad, 2006).

Proper nutrition is essential for satisfactory crop growth and production. Efficient application of the correct types and amounts of fertilisers and time of application is important in achieving profitable yields. Plants require nitrogen in relatively large quantities and in forms that are readily available. Most nursery producers use large quantities of nitrogen fertilisers to meet the needs of their crops. However, a thorough understanding of nitrogen fertiliser can be useful in optimising both the level and form of nitrogen best suited for the plant species, stage of growth, time of year and production objectives (Marschner, 1986; Mengel & Kirby, 1987).

The quantitative nitrogen requirements of vegetable crops consist of the amount of nitrogen that will actually be taken up by the plant and integrated into its biomass and a quantity of nitrogen that must nevertheless be present in the soil in order for the crop to achieve its full potential yield. The addition of these requirements provides the value of overall plant nitrogen needs (Bidwell, 1979).

2.2.2 Crop response to nitrogen fertilisation

Although crops usually respond to fertilisers, this is not always the case. A crop's response to nitrogen depends on soil conditions, crop species in particular, the amount of nitrogen availabity in the soil and the amount of nitrogen that will become available during the growing season or period (Mengel & Kirby, 1987). The response is generally poorer when the level of nitrogen available in soil is high. In an extended field trial performed by Zebarth, Freyman & Kowalenko (1991), high levels of nitrogen have been found to influence yields of cabbage. A positive yield response when nitrogen was increased to 500 kg N ha^{-1} was observed. Peck (1981) also observed a yield increase of cabbage of about 4 kg m^{-2} fresh mass, compared to cabbage plants where nitrogen was not applied.

Sorensen, Johansen & Poulsen (1994) reported an optimum yield of marketable crisphead lettuce at a level of 150 kg N ha^{-1} and a decrease in the incidence of tipburn with increasing nitrogen fertiliser. From a nutritional point of view, crisphead lettuce grown at low nitrogen levels and harvested at an early stage is to be preferred due to a high content of nutrients, especially vitamin C.

High nitrogen levels have often been found to influence optimum yields in cabbage. Significantly higher yields of cabbage at high nitrogen levels (200 kg N ha^{-1}) were reported than yields at lower levels (0, 50 and 100 kg N ha^{-1}) (Ghanti *et al.*, 1982; Gupta, 1987; Everaarts & De Moel, 1998; Parmar *et al.*, 1999). The increase in yield was attributed to the fact that higher nitrogen levels resulted in larger leaf areas.

2.3 CROP NUTRIENT MANAGEMENT

Nutrients must be applied at levels necessary to achieve realistic crop yields and timing of nutrient application should also be improved. Agronomic crop production technology should be used to increase nutrient use efficiency (Marschner, 1986).

2.3.1 Nitrogen sources

The nitrogen forms which are readily taken up by plants are ammonium (NH_4^+) and nitrate (NO_3^-). Nitrogen applications increase soil acidity and therefore it requires liming. General nitrogen fertilisers are ammonia (82% N), urea (46% N), limestone ammonium nitrate (28% N), urea ammonium nitrate (32% N), ammonium sulfate nitrate (27% N) and ammonium nitrate (35% N) (FSSA, 2007).

Anhydrous ammonia is the slowest of all nitrogen fertiliser forms to convert to nitrate. Therefore, it would have the least chance of nitrogen loss due to leaching or denitrification. It must be injected into the soil; therefore, it would have no loss due to surface volatilisation. The disadvantage of anhydrous ammonia is that it is hazardous to handle. Urea converts rapidly to nitrate nitrogen, usually in less than two weeks during spring. Denitrification on wet or compacted soils can be serious. Leaching can be a problem in coarse soils. In no-till situations, surface volatilisation can be a problem if the urea is not placed in contact with the soil and the weather is dry for several days after spreading (FSSA, 2007).

Limestone ammonium nitrate is not a homogenous salt but is a mixture of limestone, mainly dolomitic lime and ammonium nitrate. Urea ammonium nitrate is usually made up of urea and ammonium nitrate. The nitrate in both these products is subjected to leaching and denitrification occurs from the time it is placed in the field. Ammonium sulfate nitrate is a nitrogen source with little or no surface volatilisation loss when applied to most soils. Ammonium sulfate nitrate is also a good source of sulphur when needed. It is a physical mixture of ammonium sulphate and ammonium nitrate. Its disadvantage is that it is the most acidifying form of nitrogen fertiliser requiring approximately 2 to 3 times as much lime to neutralise the same amount of acidity as formed by other common nitrogen carriers. Ammonium nitrate is another nitrogen source but it quickly converts to nitrate. For soils subjected to leaching or denitrification, ammonium nitrate would not be preferred or is not suitable for such soils. Ammonium nitrate may also not be used as a fertiliser in South Africa because it is highly explosive (FSSA, 2007).

2.3.2 Method of application

For financial survival of the producer the efficient and effective management of a fertiliser programme, including the application method is important. One important factor to consider in the efficient use of fertilisers is the placement of the material in relation to the plant. Factors to be considered in the placement of fertilisers include crop root characteristics, crop requirements at various growth stages, applied fertiliser characteristics, moisture availability, the climate when fertiliser is to be applied and the time of application. Fertilisers can be applied in several ways. The most important point to remember is to apply fertiliser at the proper level, as over-application can result in plant damage or death (Marschner, 1986; Grubinger, 1999; FSSA, 2007).

The fertiliser should be placed in the correct zone in the soil where it will serve the plant to its best advantage. Fertilisers, therefore, should be placed in such a way that nutrients are available to the plants at all times during its growth. The correct amount of nutrients should also be made available to the developing crop. Pre-plant fertilisation is normally accomplished by broadcast and incorporation over the entire field or over the crop beds and is best suited to large volumes of material not having a tendency to leach, and on soils with a significant shortage of nutrients. Formerly, fertilisers were applied broadcast and ploughed into the soil. When phosphorus and potassium are applied in this way they are fixed by the soil and much of it is not available to plant roots. Recently, application of plant nutrients in bands near the seed and plants has been practised (AVRDC, 1990; FSSA, 2007).

Different placement methods can ensure that the nutrient is immediately available to rapidly growing plants; for example, banded below the seed at planting or applied gradually over a lengthy growing period. Placement will also affect the degree of interaction between the fertiliser and the soil, which is particularly important where nutrients can become unavailable due to reactions with soil minerals such as nitrogen immobilisation. The following are methods used when applying fertiliser (Grubinger, 1999; FSSA, 2007):

2.3.2.1 Band-placement versus broadcasting

Banding fertiliser refers to the application of fertiliser at planting, thus, placing the fertiliser to either one or both sides and below the seed at planting. Care should be taken as placement too close to the seed or at too high a level can cause fertiliser burn and inhibit germination. Broadcasting of fertilisers refers to the uniform application of fertilisers across the entire soil surface. This may be done before the field is ploughed, immediately before planting, or while the crop is growing. Broadcasting is efficient and often the method of choice in areas with perennial plants (Grubinger, 1999; FSSA, 2007).

Comparison of these two application methods as far as application levels and the corresponding yields are concerned, is determined by the fertility level of the soil. Band placement of fertiliser is usually more effective than broadcasting in soils with low soil fertility and low application levels. As application levels increase there is a point where yields will actually begin to decrease in the case of band-placement and the efficiency of broadcast application will exceed that of band-placement while the yield still increases. In high fertility soils there are much smaller difference between these two application methods at low fertilisation levels (FSSA, 2007).

2.3.2.2 Side-dressing or top-dressing

Side-dressing is the post-emergence application of fertiliser alongside the crop row or to closely-spaced crops. This assists in supplying nitrogen in a readily available form to growing plants (FSSA, 2007).

2.3.2.3 Fertigation

Fertigation is the application of soluble fertiliser through an irrigation system. This method is relatively new in South Africa and will affect the accessibility of applied nutrients (Grubinger, 1999). Application of chemicals through irrigation should be safe for field use, should not reduce yield and should be soluble and compatible (FSSA, 2007).

2.3.2.4 Foliar application

Foliar application refers to the spraying of leaves of growing plants with fertiliser solutions. The foliar application of mineral nutrients by means of sprays offers a method of supplying nutrients to plants more rapidly than methods involving root application. This method can be an effective remedy for a crop suffering from a nutrient deficiency. These solutions may be prepared in a low concentration to supply any one plant with a nutrient or a combination of nutrients. Foliar fertilisers are diluted solutions applied directly to leaves and should not be relied upon to supply the total nitrogen, phosphorus, and potassium needs of plants. Foliars can be used to supplement soil applications of these nutrients (Marschner, 1986; Archer, 1988; Grubinger, 1999). The most efficient way to apply nitrogen is by soil application. Foliar application of nitrogen should be viewed as a temporary or emergency solution only (FSSA, 2007).

2.3.3 Frequency/ Timing of application

Crop, soil and nutrient type influence the time of fertilisation. The development pattern of vegetable crops differs therefore nutrient needs vary. Rainfall and temperature influences the availability of nutrients to plants, from the time they are applied, to when they are used by the plant. Generally, vegetable fertilisers are applied before planting, at planting or during the entire growth season as side or top-dressings (Cooke, 1982; AVRDC, 1990; FSSA, 2007).

Fertiliser should be applied when plants need it, when it will be most effective, and when plants can readily take it up. The best way to ensure that added nutrients are used efficiently by plants and to reduce the risk of nutrient loss to the environment is to match nutrient availability to plant demand over time. Annual crops, perennial crops and pastures all have different patterns of nutrient demand over time, and respond differently according to soil moisture status and temperature. These factors should be considered in planning fertiliser applications (Archer, 1988; AVRDC, 1990).

Mobile nutrients such as nitrogen or potassium are most effectively used when split application are applied frequently during crop growth. This is usually preferable to one large application. However, crops may have short periods of very high nutrient demand and so a larger application will be required just prior to that period. Fertigation systems (adding nutrients in irrigation water) provide flexibility in applying nutrients to meet plant demand but regular top-dressing or side-dressing of fertiliser can have similar effects, provided that there is sufficient moisture to move nutrients into the soil (AVRDC, 1990; FSSA, 2007).

2.3.4 Nitrogen loss control

Many intensive systems of field vegetable production are not sustainable because they lose excessive amounts of nitrogen to the environment. Processes in the nitrogen cycle of agricultural systems include assimilation, mineralisation/immobilisation, nitrification, denitrification, ammonia volatilisation, nitrate leaching, runoff and erosion. Emission of nitrogen from agriculture may affect the quality of the atmosphere, ground and surface waters. In field vegetable production, nitrate leaching is the dominant process affecting the environment. Often, large amounts of nitrogen, including residual soil mineral nitrogen and the nitrogen present in crop residues, remain in the soil after harvesting the crop. Both sources of nitrogen may affect groundwater quality through nitrate leaching (Nielsen, 2006). Timing of application should take these risks into account (Mengel & Kirby, 1987). The nitrogen source a farmer chooses should depend on how serious a problem he has with the mentioned nitrogen losses. The cost of nitrogen is another consideration when choosing a fertiliser source.

2.3.4.1 Denitrification

Denitrification occurs when nitrate (NO_3^-) is present in a soil and there is not enough oxygen present to supply the needs of bacteria and micro-organisms in the soil. Nitrogen losses by denitrification may be higher with NO_3^- than with NH_4^+. If oxygen levels are low, bacteria and micro-organisms strip the oxygen from the nitrate and the end result is the production of nitrogen gas, which volatilises from the soil. The three conditions that create an environment that promotes denitrification are wet (waterlogged) soils, compaction and warm temperatures (>20°C) (Nielsen, 2006). However, if the waterlogging is only temporary, the denitrification process will stop when the soil dries (Baker & Mills, 1980).

2.3.4.2 Leaching

Nitrate is very mobile in the soil. For this reason it is susceptible to leaching down the soil profile with excessive rain or irrigation water on soil already at field capacity. Being an easily leached substance that is widely used as fertiliser, it can cause water pollution if improperly managed (Marschner, 1986). Leaching losses of nitrogen occur when soils have more incoming water (rain or irrigation) than the soil can hold. As water moves through the soil, nitrates in the soil solution are picked up and moved with the water. Ammonium forms of nitrogen have a positive charge and are held by the negative sites on the clay in the soil; therefore, ammonium forms of nitrogen leach very little. In sands where there is very little clay, ammonium forms of nitrogen do leach. Relatively coarse soils such as sands are the only ones in which significant leaching of nitrogen appears important (Mengel & Kirby, 1987; Nielsen, 2006). One way to minimise nitrogen leaching and denitrification is to minimise the time nitrogen is in the soil before plant uptake.

2.3.4.3 Volatilisation

Volatilisation of nitrogen happens when urea forms of nitrogen break down and form ammonia gases and where there is little soil water to absorb them. This condition occurs when urea forms of nitrogen are placed in the field but not in direct contact with the soil. This situation can occur when urea is spread on plant residues. The level of surface volatilisation depends on the moisture level, temperature and surface pH of the soil. If the soil surface is moist, water evaporates into the air. Ammonia released from the urea is picked up in the water vapour and lost. On dry soil surfaces, less urea is lost. Applying urea fertilisers when weather is cooler also slows down nitrogen loss. If the surface of the soil has been limed within the past three months with two tons or more of lime per hectare, urea-based fertilisers should not be applied unless they can be incorporated into the soil (Baker & Mills, 1980; Mengel & Kirby, 1987; Nielsen, 2006). To stop ammonia volatilisation from urea, the urea should be tied up by the soil. Enough rain water is important to wash the urea from the residue in the soil, or the farmer should place the urea-based fertiliser in direct contact with soil by tillage, banding or dribbling.

2.3.4.4 Immobilisation

Immobilisation is the fourth nitrogen loss mechanism but it is temporary in nature. When nitrogen fertiliser is applied to soil, some of the nitrogen is taken up by micro-organisms in the soil, in a and the process known as immobilisation. The immobilised nitrogen is incorporated into proteins, nucleic acids, and other organic nitrogen constituents of microbial cells. As such, it becomes part of the biomass of the plant (Mulvaney, Azam & Simmons, 1993). As the microbes die and decay, some of the "biomass nitrogen" is released as ammonium through the process of mineralisation, and the remainder undergoes conversion to more stable organic nitrogen compounds, ultimately becoming part of soil organic matter.

2.3.4 Nitrogen balance

Agriculture is a major contributor to nitrate contamination of groundwater. Therefore, farmers are asked to reduce the impact of nitrogen on the environment. Crops may increase yields but decrease nitrogen use efficiency when the supply of nitrogen is increased. If it has not been volatilised or denitrified, nitrogen not utilised by the crop can accumulate in the soil and, in consequence, increases the risk of leaching with corresponding environmental consequences. Nitrogen balances (nitrogen fertilisation minus nitrogen uptake by the harvest products) at field scale or in larger areas are often used to estimate the leaching risk. However, there is much evidence that, in the short term, the link between fertiliser use (except excessive amounts) and nitrate in water is not very direct. A nutrient surplus in itself may not be sufficient to quantitatively determine the amount of nutrient lost via various pathways, because of the interaction with other environmental parameters. On the other hand, nitrogen balances can give an indication of the risks that are associated with specific farming practices, especially in the wider environment and if integrated over a relatively long period (Sieling & Kage, 2006).

2.4 PRODUCE QUALITY

Due to the low energy content and high dietary fiber, vitamins and minerals an increased intake of vegetable food products are recommended to improve human health. However, concerns about how different growing conditions influence product quality, especially the content of nitrate and vitamins, are still present.

The consumer demand for high nutritive quality may conflict with the growers wish for high marketable quality. Nitrogen fertiliser is one of the most important growth factors influencing yield and the chemical composition of vegetables, as it has been identified as the major factor that influences the nitrate content in vegetables. Excessive amounts of nitrogenous fertiliser are applied to crops, as it is a reasonable insurance against yield losses and their economic consequences (Huang, 2002). Nevertheless, when nitrogen input exceeds the demand, plants are no longer able to absorb it and subsequently nitrogen starts to build up in the soil mostly as nitrate. This will cause imbalances of nutrients in the soil and increase the nitrate level in ground water supplies, which influences the nitrate content of plants, especially leafy vegetables. There is conflicting evidence regarding the potential long-term health risks associated with nitrate levels encountered in the human diet. Therefore, high nitrate accumulation in vegetables is a concern because it presents health hazards for humans (Goh & Vityakon, 1983; 1986; Lairon *et al.,* 1984; Sorensen *et al.*, 1994).

Factors responsible for nitrate accumulation in plants are mainly nutritional, environmental and physiological. Nitrogen fertilisation is found to be the major factor that influences the nitrate content in vegetables. Appropriate strategies should be adopted and the role of individual physiological factors should be determined to limit accumulation of nitrate in vegetables and the use of nitrogen fertiliser should be optimised (Blom-Zanstra,1989).

Literature indicates that, for successful vegetable production nitrogen fertiliser is required and needs to be applied in order to increase the growth and yield of vegetable plants. The balance between application levels, yield and quality should therefore be determined.

CHAPTER 3

MATERIAL AND METHODS

3.1 GENERAL

Two pot trials were conducted in glasshouses of the Department of Soil, Crop and Climate Sciences at the University of the Free State, Bloemfontein in 2005 and 2006. Soil analysis was done at the laboratory of the Free State Department of Agriculture at Glen while the leaf analysis was done in the laboratories of the Departments of Soil, Crop and Climate Sciences and the Institute for Groundwater Studies at the University of the Free State in Bloemfontein.

3.2 SOIL COLLECTION AND PREPARATION

Topsoil of the fine sandy loam Bainsvlei form (Soil Classification Working Group, 1991) was used in these pot trials. Soil was dried at room temperature, sieved through a 5 mm screen and mixed manually several times and stored until needed. Enough soil was collected for both pot trials and analysed for nutrient deficiencies. The fertility status of the soil was in general, excellent, according to local guidelines (FSSA, 2007). Some physical and chemical properties of the soil are indicated in Table 3.1.

Phosphorus (9 kg P ha^{-1}) and potassium (30 kg K ha^{-1}) fertiliser were applied before planting according to the withdrawal amounts and an expected yield of 20 ton ha^{-1}. Phosphoric acid and potassium chloride were used as sources of phosphorous and potassium, respectively.

Table 3.1: Physical and chemical properties of the topsoil used in both pot trials

Property*	
Particles size distribution (%)	
Sand (0.02-2 mm)	84
Clay and Silt (<0.002-0.02)	16
pH $_{(KCl)}$	5.6
EC (mS m^{-1})	41
Nutrients (mg kg^{-1})	
P (Olsen)	38.0
Ca (NH_4OA_C)	480.1
Mg (NH_4OA_C)	93.3
K (NH_4OA_C)	206.3
Na (NH_4OA_C)	63.3
Zn (HCl)	18.7

***Determined with standard procedure (The Non-Affiliated Soil Analysis Working Committee, 1990)**

3.3 EXPERIMENTAL DESIGN AND TREATMENTS

A randomised complete block design was used for both pot trials conducted in this study. The treatments however differed between the trials in agreement with the study's objectives.

In the first pot trial the response of two Swiss chard cultivars to nitrogen levels and nitrogen application times was investigated and was conducted in 2005. A total of forty treatment combinations were applied for the trial, including two cultivars, five nitrogen levels and four nitrogen application times (Table 3.2). All treatment combinations were replicated four times.

The two Swiss chard cultivars selected for the trial were 'Fordhook Giant' and 'Rhubarb' (Table 3.2). 'Fordhook Giant' reaches maturity in approximately 45 to 55 days. It has large dark green slightly crumpled leaves with broad glossy ribs and its petiole is broad and white. 'Rhubarb' reaches maturity in 50 to 60 days and is a cultivar with dark green leaves which are slightly crinkled. The leaf petiole and veins are bright red and the petiole is slightly flattened.

Table 3.2: Summary of the treatments applied in 2005 to investigate the response of Swiss chard to nitrogen levels and application times

Cultivar	
C_1 **C_2**	Fordhook Giant Rhubarb
Nitrogen levels (NH_4NO_3) (kg ha^{-1})	
N_0 **N_1** **N_2** **N_3** **N_4**	0 100 200 300 400
Application times	
T_1 **T_2** **T_3** **T_4**	Once every 2nd week (11 times) Once every 4th week (5 times) Once every 6th week (4 times) Once every 8th week (3 times)

In the second pot trial the response of Swiss chard to nitrogen sources and nitrogen levels was investigated in 2006 (Table 3.3). The pot trial involved one Swiss chard cultivar ('Fordhook Giant'), six nitrogen sources and nine nitrogen levels. A total of fifty-six treatment combinations were applied and each combination was replicated four times.

For each treatment combination 4 L pots were filled with soil. Before transplanting the Swiss chard seedlings in June the soil was watered to field capacity. Three seedlings were planted per pot. Two weeks after planting the seedlings were thinned to one plant per pot. Pots were kept at field capacity using a drip irrigation system with a capacity of 4 L h^{-1}. The pots were, manually kept free from weeds during the growth season.

Table 3.3: Summary of the treatments applied in 2006 to investigate the response of Swiss chard to different nitrogen sources and nitrogen levels

Nitrogen levels (kg ha^{-1})	
N_0	0
N_1	100
N_2	200
N_3	300
N_4	400
N_5	500
N_6	600
N_7	700
N_8	800
Nitrogen sources	
S_1	Ammonium nitrate
S_2	Calcium nitrate
S_3	Potassium nitrate
S_4	Ammonium sulphate
S_5	Urea ammonium nitrate
S_6	Urea

Fertilisation treatments were carried out by applying the appropriate amounts of nitrogen in solution to the pots. The relevant nutrient solution was poured evenly on the soil surface of each pot whereafter the pots were irrigated. Ammonium nitrate was used as source of nitrogen in the first pot trial, while in the second pot trial different nitrogen sources were used as indicated in Table 3.3. In the first pot trial the required nitrogen was applied at different times during the growth season as indicated in Table 3.2, but for the second pot trial nitrogen was applied every second week (11 applications), starting with planting and then up to 18 weeks after planting.

In order to simulate the natural conditions in which Swiss chard plants grow, the glasshouse temperatures were kept at 22°C (± 1°C) during the day and 15°C (±1°C) during the night.

3.4 COLLECTION OF DATA

3.4.1 Growth parameters

Number of leaves

For the first pot trial (2005) the number of leaves per plant was counted once every two weeks from week 2 to 6 after planting. Only leaves that were fully developed were considered for counting.

3.4.2 Yield and quality parameters

Number of leaves harvested

Leaves were harvested once every 4 weeks from 9 up to 21 weeks after planting in the first pot trial (2005) and once every 3 or 4 weeks from 8 up to 21 weeks after planting in the second pot trial (2006). All leaves longer than 15 cm were harvested and counted.

Leaf area

Leaf area (cm^2 $leaf^{-1}$) of all harvested leaves was measured using a LiCor belt driven leaf area meter (model LI 3100). This was done every fourth week from 9 up to 21 weeks after planting in 2005 and in 2006 every third or fourth week from 8 up to 21 weeks after planting.

Leaf fresh and dry mass

At harvest the leaf fresh mass (g) with attached petiole, was measured. The leaf blades were kept in brown paper bags and dried in an oven at 60°C for 7 days. After drying, the leaf blades were weighed to determine the dry mass. This was done for both pot trials (2005 and 2006).

Leaf nitrogen status

For the first pot trial the harvested dried leaves were milled and the nitrogen content analysed using standard procedures. Steam distillation was used to determine the nitrogen after digestion of the samples with sulphuric acid (Agrilasa, 2002).

Leaf nitrate status

For the second trial the dried leaves were milled and analysed for nitrate. Nitrate was extracted from the leaves by using the hot water extraction method where a sample of 0.2 g ground plant material was shaken in 50 ml hot water for a period of 3 hours (Goh & Vityakon, 1986). Nitrate content was determined by ion chromatography which is a standard method for the examination of water and wastewater (Eaton *et al*., 2005). The Dionex system was used in the laboratory with AG-14 as a guard column and AS-14 as the analytical column and conductivity was determined and used as detector.

3.5 STATISTICAL ANALYSIS

An analysis of variance was done on all measured parameters to determine the significance of differences between means of treatments using the NCSS 2000 statistical program (Hintze, 1999) and Turkey's test for the LSD ≤ 0.05.

CHAPTER 4

NITROGEN FERTILISATION AFFECTING SWISS CHARD PRODUCTION

4.1 INTRODUCTION

Swiss chard is one of the most neglected vegetables in South Africa and the area under Swiss chard production in South Africa is not commercially important. This vegetable offers an important increase in the current vegetable assortment and it can also play an important role in human diet, especially in poorer communities, which makes it worthwhile to investigate. Swiss chard is often referred to as greens or leafy vegetables and is known to have a high demand for fertiliser (Pokluda & Kuben, 2002).

Appropriate production practices may increase the growth rate and yield of crops and one of these production factors is fertilisation. The influence of fertilisation on the growth and yield of vegetables is of great importance. Fertilisers are used extensively to produce high yields and its usage has increased over the years (Goh & Vityakon, 1983). Along with increased fertiliser applications there is a need to determine the optimum application rates for different vegetables (Kansal *et al.*, 1981; Goh & Vityakon, 1983).

Nitrogen is one of the important plant nutrients and is required in rather large amounts compared to other essential nutrients and plants especially leafy vegetables, e.g. cabbage, respond quickly to nitrogen fertiliser. Nitrogen is an essential component of chlorophyll, proteins and enzymes. It stimulates root and vegetative growth, as well as the intake of other essential nutrients (Bidwell, 1979; Kansal *et al.*, 1981; Goh & Vityakon, 1983). Adequate application of nitrogen fertiliser promotes vegetative growth and green colour of leafy vegetables which, again, is of great importance to yield and quality (Ware & McCollum, 1980; Peck, 1981; Splittstoesser, 1990; Hadfield, 1995).

Growers often tend to apply large amounts of nitrogen fertiliser to obtain high yields of good quality (Neeteson, 1997). From an economic perspective this may be sound but not from an environmental perspective. A considerable portion of the applied nitrogen may remain in the soil after harvest. This nitrogen includes residual soil mineral nitrogen and nitrogen present in crop residue (Neeteson, 1997). Both of these nitrogen sources may have a harmful effect on the environment. The quality of the ground water may be negatively affected through leaching, as well as air quality through nitrous oxide emission (Huang, 2002). According to Huang (2002) agricultural use of chemical and organic nitrogen fertiliser is a major contributor of non-point source pollutants leading to a variety of water quality problems in the US.

It is important for growers to adopt the best nitrogen management plan to reduce the negative impact of agricultural production on the environment (Huang, 2002). For sustainable vegetable production, improved efficiency of nitrogen management may be possible if the correct nitrogen levels are applied at the time of maximum crop need (Neeteson, 1997; Sawyer, 2001; Huang, 2002). This would also avoid excessive application of nitrogen and reduce the amount of residual nitrogen lost to the environment. Nitrogen deficient plants are identified by poor growth and poor colour. Excessive nitrogen generally leads to excessive vegetative growth (Bidwell, 1979). Over-fertilisation with nitrogen results in lower yields and a poor quality vegetable, such as in Swiss chard and carrots. The response of vegetables to different nitrogen rates is well documented. Nitrogen optimises the yield of broccoli, cauliflower (Dufault & Waters, 1985), Chinese vegetables (Hill, 1990), lettuce (Gardener & Pew, 1972), spinach (Briemer, 1982) and Swiss chard (Goh & Vityakon, 1986).

Nitrogen applied before planting is more vulnerable to losses than when applied during the growing season (Huang, 2002). Therefore, the early application of nitrogen, especially in the nitrate form, should be avoided (Sawyer, 2001). Split applications or a single application of nitrogen fertiliser during the growing season can match the crop's nitrogen needs without a reduction in yield. This may be the least costly practice for nitrogen fertiliser application (Huang, 2002). Welch, Tyler & Ririe (1985) reported that split application of nitrogen was more efficient than a single application for celery, cauliflower and cabbage as the yield increase was significantly higher, especially in areas where leaching of nutrients was high. However, Cleaver *et al*. (1971) reported that top-dressing was not important for cabbage, provided sufficient nitrogen was applied at planting. The practical and economic implications of split application should always be kept in mind. Unfavourable weather conditions during the growth season can stop the grower from entering the field to apply nitrogen fertiliser and the lack of nitrogen can again reduce crop yield and cause loss of income for the grower.

Based on this background, an experiment was conducted in the glasshouse to determine the response of two Swiss chard cultivars to different combinations of nitrogen levels and different application times.

4.2 RESULTS AND DISCUSSION

4.2.1 Number of leaves

A summary of the analyses of variance that was done to determine the effects of different nitrogen levels and application times on the number of leaves of 'Fordhook Giant' and 'Rhubarb' plants from 2 up to 6 weeks after planting is given in Table 4.1.

Table 4.1: Summary of the analyses of variances showing the significant effects of nitrogen levels and nitrogen application times on the number of leaves of 'Fordhook Giant' and 'Rhubarb' plants from 2 up to 6 weeks after planting

Weeks after planting	Cultivar (C)	Nitrogen level (NL)	Nitrogen application time (NA)	C x NL	NL x NA	C x NA
2	ns	*	*	ns	ns	ns
4	ns	*	*	ns	ns	ns
6	ns	*	*	ns	*	ns

LSD $_{(T \leq 0.05)}$
ns = no significant differences
* = significant differences

Inspection of this table showed that neither the interaction between cultivar and nitrogen levels nor cultivar and nitrogen application times significantly influenced the number of leaves for Swiss chard plants. There was also no significant difference between the number of leaves counted for 'Fordhook Giant' and 'Rhubarb' plants.

The interaction between nitrogen levels and nitrogen application times influenced significantly the number of leaves counted only at 6 weeks after planting (Table 4.2). At all four nitrogen application times the number of leaves increased significantly with an increase in nitrogen levels. It was only with the T_3 treatment where the number of leaves did not increase when the nitrogen level increased from 200 to 400 kg N ha^{-1}. The number of leaves increased significantly from 2.3 at 0 kg N ha^{-1} to 5.6 at 400 kg N ha^{-1} with the T_4 treatment. The same tendency was observed for the T_1, T_2 and T_3 treatments (Table 4.2).

Table 4.2: Effect of nitrogen levels and application times on the number of leaves counted of Swiss chard plants 6 weeks after planting

Nitrogen levels (NL) kg ha^{-1}	**Nitrogen application times (NA)**			
	T_1	**T_2**	**T_3**	**T_4**
0	2.0	2.3	2.1	2.3
50	2.6	3.0	2.9	3.3
100	3.4	3.8	4.0	4.0
200	4.3	4.4	4.5	4.5
400	4.8	4.6	4.5	5.6
LSD$_{(T\leq0.05)}$ NL X NA	**0.2**			
% Nitrogen received of the total required	27.3	40	25	33.3
Last nitrogen application (Weeks after planning)	4	4	0	0

Although not always significant the number of leaves counted for Swiss chard plants that received low nitrogen levels (0, 50, 100 and 200 kg N ha^{-1}) was higeher in the T_2, T_3 and T_4 treatments than with the T_1 treatment (Table 4.2). The reason for this may be attributed to the fact that the T_1 treated plants received only 27.3% of the total required nitrogen, with the last application 4 weeks after planting, compared to the T_3 and T_4 treated plants that received 25 and 33.3% respectively of the required nitrogen already with planting. The T_2 treated plants received their last nitrogen application 4 weeks after planting but at that stage these plants received 40% of the required nitrogen compared to the 27.3% of the T_1 treated plants.

However, where a high nitrogen level (400 kg N ha^{-1}) was applied the number of leaves counted for the T_1 and T_4 treatments were significantly more than for the T_2 and T_3 treatments (Table 4.2). At this stage the T_4 treated plants received 33.3% (all with planting) and the T_1 treated plants 27.3% of the required nitrogen (last nitrogen application 4 weeks after planting) compared to the T_3 treated plants that received 25% (all with planting) and the T_2 treated plants 40% (last nitrogen application 4 weeks after planting) of the required nitrogen. The reason for this phenomenon was not clear.

As shown in Table 4.3 the number of leaves counted increased significantly with increasing levels of nitrogen for all three counting times. Significantly, more leaves were counted where 400 kg N ha^{-1} was applied than in all the other nitrogen treatments over all three counting times. The number of leaves increased significantly from 2.16 where no nitrogen was applied to 4.91 where 400 kg N ha^{-1} was applied at 6 weeks after planting. This was also true for week 2 and 4 after planting.

Table 4.3: Effect of nitrogen levels on the number of fully expanded leaves of Swiss chard plants from week 2 up to week 6 after planting

Weeks after planting	**Nitrogen levels (NL) (kg ha^{-1})**					**$LSD_{(T \leq 0.05)}$ NL**
	0	**50**	**100**	**200**	**400**	
2	0.25	0.97	1.81	2.40	2.75	**0.09**
4	1.22	1.97	2.81	3.43	3.75	**0.09**
6	2.16	2.94	3.78	4.40	4.91	**0.08**

Nitrogen application times significantly influenced the number of leaves counted from 2 to 6 weeks after planting as shown in Table 4.4. The number of leaves counted was significantly more for the T_4 treatment than for the T_1, T_2 and T_3 treatments at all three counting times. During week 4 and 6 after planting the number of leaves counted for the T_2 and T_3 treatments was also significantly more than the T_1 treatment.

Table 4.4: Effect of nitrogen application times on the number of fully expanded leaves of Swiss chard plants from week 2 up to week 6 after planting

Weeks after planting	Nitrogen application times (NA)								$LSD_{(T \leq 0.05)}$ NA
	T_1		T_2		T_3		T_4		
2	1.53	(9.09)*	1.58	(20)	1.6	(25)	1.85	(33.3)	**0.08**
4	2.50	(18.18)	2.58	(20)	2.63	(25)	2.85	(33.3)	**0.07**
6	3.40	(27.27)	3.60	(40)	3.60	(25)	3.95	(33.3)	**0.07**

***Values in brackets indicate the % nitrogen the plants received of the total nitrogen required.**

At six weeks after planting, the number of leaves counted for the T_4 (33.3%) treatment was still significantly more than the T_2 (40%) treatment. This may be attributed to the fact that the T_2 treated plants received 20% of the 40% nitrogen only two weeks before counting the leaves compared to the T_4 treated plants that received 33.3% nitrogen already with planting.

From these results it is clear that nitrogen levels and nitrogen application times significantly influenced the early growth (first 6 weeks after planting) of Swiss chard. The application of nitrogen promoted early growth of Swiss chard plants when leaf number serves as index, irrespective of cultivar or nitrogen application time. Biemond, Vos & Struik (1995) also reported that larger available amounts of nitrogen led to an earlier start of sprout growth in Brussel sprouts. Swiss chard plants formed significantly more leaves where 400 kg N ha^{-1} was applied. Results obtained by Khuzwayo (2004) on cabbage plants also showed that nitrogen fertiliser increased the number of cabbage leaves during the first 54 days after sowing compared to plants that did not receive any nitrogen.

Nitrogen application time also influenced the early growth of Swiss chard plants. Swiss chard plants responded better to nitrogen applications that were split into either 4 or 3 applications (T_3 and T_4) than where nitrogen was split into 11 applications (T_1). The implication of this is that a higher percentage of the required nitrogen was applied with planting namely; 25% and 33.3% for the T_3 and T_4 treatments respectively, compared to 9.09% for the T_1 treatment. More nitrogen is therefore available for early growth of Swiss chard plants with the T_3 and T_4 application times.

4.2.2 Number of leaves harvested

A summary of the analyses of variance that was done to determine the effects of different nitrogen levels and application times on the number of leaves harvested every four weeks of 'Fordhook Giant' and 'Rhubarb' plants from 9 up to 21 weeks after planting is given in Table 4.5.

Table 4.5: Summary of the analyses of variances showing the significant effects of nitrogen levels and nitrogen application times on the number of leaves harvested per plant every four weeks of 'Fordhook Giant' and 'Rhubarb' plants from 9 up to 21 weeks after planting

Weeks after planting	Cultivar (C)	Nitrogen level (NL)	Nitrogen application time (NA)	C × NL	NL × NA	C × NA
9	*	*	ns	ns	ns	ns
13	*	*	ns	*	ns	ns
17	ns	*	*	ns	ns	ns
21	*	*	*	ns	*	ns
Total number of leaves harvested plant^{-1}	*	*	ns	*	ns	ns

LSD $_{(T \leq 0.05)}$
ns = no significant differences
*** = significant differences**

As shown in Table 4.5, the interaction between cultivar and nitrogen application times was not significant at all four harvesting times. The interaction between cultivar and nitrogen levels significantly influenced the number of leaves harvested per plant at 13 weeks after planting. Although not always significant, the number of leaves harvested per plant increased with increasing nitrogen levels for both cultivars (Table 4.6). The number of leaves harvested for 'Fordhook Giant' increased significantly from 0.13 at 0 kg N ha^{-1} to 1.81 and 3.19 at 200 and 400 kg N ha^{-1}, respectively. 'Rhubarb' showed the same tendency where leaves harvested increased significantly from 0.19 at 0 kg N ha^{-1} to 1.44, 2.75 and 4.13 at 100, 200 and 400 kg N ha^{-1}, respectively. Although not significant, the number of leaves harvested per 'Rhubarb' plant was more than for 'Fordhook Giant' at all the nitrogen levels except where 50 kg N ha^{-1} was applied.

Table 4.6: Effect of nitrogen levels on the number of leaves harvested per plant of 'Fordhook Giant' and 'Rhubarb' plants 13 weeks after planting

Nitrogen levels (NL) (kg ha^{-1})	Cultivar (C)	
	Fordhook Giant	Rhubarb
0	0.13	0.19
50	0.63	0.44
100	0.81	1.44
200	1.81	2.75
400	3.19	4.13
$LSD_{(T\leq0.05)}$ C X NL	**1.04**	

The number of leaves harvested per Swiss chard plant from the T_1 and T_3 treated plants that received 400 kg N ha^{-1} was significantly higher than in all the other treatments at 21 weeks after planting (Table 4.7). At 21 weeks after planting all the plants had received all (100%) the required nitrogen. The phenomenon that the T_1 and T_3 treated plants yielded significantly more leaves than the other treatments at this late growth stage, may be attributed to the fact that these plants received their last nitrogen application shortly before harvesting. The T_1 treated plants received their last two nitrogen applications, 18 and 20 weeks after planting (9.09% per application); the T_3 treated plants 18 weeks after planting (25% per application) and the T_2 and T_4 treated plants 16 weeks after planting (40% and 33.3% per application, respectively). These results indicate that not only high nitrogen levels but nitrogen applied late during the growth season (week 18 and 20) significantly influenced the number of leaves harvested for Swiss chard plants in the late stages of harvesting (21 weeks after planting).

Table 4.7: Effect of nitrogen levels and application times on the number of leaves harvested per plant of Swiss chard plants 21 weeks after planting

Nitrogen levels (NL) (kg ha^{-1})	Nitrogen application times (NA)			
	T_1	T_2	T_3	T_4
0	0.13	0.13	0.80	0.00
50	0.38	0.00	0.13	0.00
100	0.13	0.25	0.25	0.50
200	0.38	0.25	1.38	0.25
400	4.13	1.88	2.25	1.38
$LSD_{(T\leq0.05)}$ NL X NA	**1.89**			
% Nitrogen received per application	9.09	20	25	33.3
Last nitrogen application (weeks after planting)	20	16	18	16

The number of leaves harvested for 'Rhubarb' was significantly higher than for 'Fordhook Giant' from 9 up to 21 weeks after planting, except at 17 weeks after planting (Table 4.8). At 9 weeks after planting 4.96 leaves were harvested from 'Fordhook Giant' plants compared to 6.56 from 'Rhubarb' plants.

Table 4.8: Number of leaves harvested per plant of 'Fordhook Giant' and 'Rhubarb' from 9 up to 21 weeks after planting

Weeks after Planting	Cultivar (C)		$LSD_{(T\leq0.05)}$ C
	Fordhook Giant	Rhubarb	
9	4.96	6.56	**0.62**
13	1.31	1.79	**0.29**
17	1.04	1.23	**ns**
21	0.44	0.94	**0.33**

Inspection of Table 4.9 reveals that the number of leaves harvested from 9 up to 21 weeks after planting increased with increasing levels of nitrogen. Significantly higher leaves were harvested at 400 kg N ha^{-1} than at 0 and 50 kg N ha^{-1} over all the harvesting times. At 9 weeks after planting the number of Swiss chard leaves harvested increased from 3.66 at 0 kg N ha^{-1} to 8.31 at 400 kg N ha^{-1}. Leaves harvested 9 and 13 weeks after planting were also significantly more at 100 and 200 kg N ha^{-1} than at 0 kg N ha^{-1}. Nitrogen applied at 200 kg N ha^{-1} 17 weeks after planting also significantly increased the number of leaves harvested per plant compared to 0, 50 and 100 kg N ha^{-1}.

Table 4.9: Effect of nitrogen levels on the number of leaves harvested per plant of Swiss chard plants from 9 up to 21 weeks after planting

Weeks after planting	**Nitrogen levels (NL) (kg ha^{-1})**					**$LSD_{(T \leq 0.05)}$ NL**
	0	**50**	**100**	**200**	**400**	
9	3.66	4.47	5.69	6.69	8.31	**1.37**
13	0.16	0.53	1.13	2.28	3.66	**0.63**
17	0.06	0.53	0.84	1.78	2.44	**0.80**
21	0.06	0.13	0.28	0.56	2.41	**0.72**

Nitrogen application time did not influence the number of leaves harvested per plant during the early weeks of harvesting (9 and 13 weeks after planting) as shown in Table 4.10. At 17 weeks after planting the number of leaves harvested from the T_1, T_2 and T_4 treated plants was significantly more than those harvested from the T_3 treated plants. At this stage the T_2 and T_4 treated plants received all (100%) the required nitrogen whereas the T_1 and T_3 plants received only 72.72 and 75% of the required nitrogen respectively. The T_3 plants received their last application already 12 weeks after planting whereas the other treatments received their last application four weeks later (16 weeks after planting) which may explain the reason for the difference in leaf numbers harvested.

Table 4.10: Effect of nitrogen application times on the number of leaves harvested of Swiss chard plants from 9 up to 21 weeks after planting

Weeks after planting	Nitrogen application times (NA)												$LSD_{(T\leq 0.05)}$ NA
	T_1			T_2			T_3			T_4			
9	5.8	(8)*	[36.36]**	6.0	(8)	[40]	5.8	(6)	[50]	5.8	(8)	[33.33]	**ns**
13	1.8	(12)	[63.63]	1.6	(12)	[80]	1.5	(12)	[75]	1.3	(8)	[66.66]	**ns**
17	1.4	(16)	[72.72]	1.3	(16)	[100]	0.5	(12)	[75]	1.3	(16)	[100]	**0.7**
21	1.0	(20)	[100]	0.5	(16)	[100]	0.8	(18)	[100]	0.4	(16)	[100]	**0.6**

***Values in ( ) brackets indicate the last application time of nitrogen (weeks after planting)**
****Value in [ ] brackets indicate the % nitrogen the plants received of the total nitrogen required**

Twenty-one weeks after planting all plants had received 100% of the required nitrogen and the number of leaves harvested for the T_1 treated plants was still significantly more than the T_4 plants. The T_1 treated plants received their last nitrogen 20 weeks after planting compared to the T_4 treated plants that received their last application 16 weeks after planting. Although not significantly more leaves were harvested for the T_1 and T_3 treated plants than the T_2 and T_4 treated plants, both the T_2 and T_4 treated plants received their last nitrogen already 16 weeks after planting compared to 18 and 20 weeks after planting for the T_3 and T_1 treated plants, respectively.

The total number of leaves harvested per plant over the period of 12 weeks was significantly influenced by the interaction between cultivar and nitrogen levels (Table 4.5). As shown in Figure 4.1 the total number of leaves harvested per plant increased with increasing nitrogen levels for both ‘Fordhook Giant’ and ‘Rhubarb’ plants. The total number of leaves harvested for ‘Fordhook Giant’ increased from 3.1 to 15.1 when the nitrogen level increased from 0 to 400 kg N ha^{-1}. This was also true for ‘Rhubarb’ where the total number of leaves harvested increased from 5.0 at 0 kg N ha^{-1} to 18.5 at 400 kg N ha^{-1}. Although not always significant, the total number of leaves harvested for ‘Rhubarb’ was more than for ‘Fordhook Giant’ over all the different nitrogen levels. Nitrogen application times did not significantly influence the total number of leaves harvested (Table 4.5).

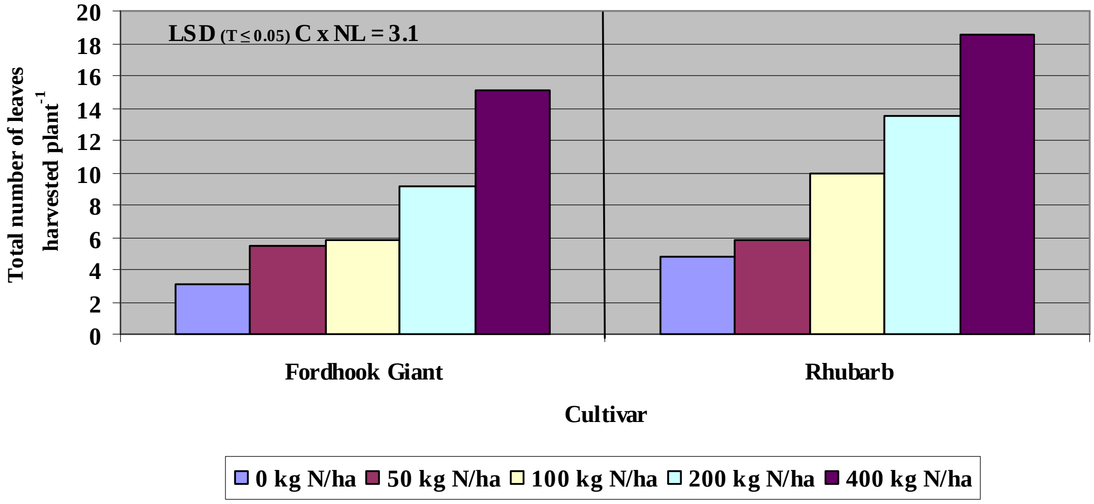

Figure 4.1: Effect of nitrogen levels on the total number of leaves harvested per plant for 'Fordhook Giant' and 'Rhubarb' plants

As was expected these results indicate that different nitrogen levels had a pronounced influence on the number of leaves harvested per Swiss chard plant. As the nitrogen levels increased the number of leaves harvested per plant increased irrespective of the cultivar or time of nitrogen application. In this study the highest number of leaves was obtained where 400 kg N ha^{-1} was applied. These results are similar to those of Goh & Vityakon (1983; 1986) who also obtained higher leaf numbers for spinach where 400 and 450 kg N ha^{-1} were applied. Gülser (2005) however, reported that different nitrogen levels did increase the number of spinach leaves harvested but not significantly. The reason for this may be attributed to the fact that much lower nitrogen levels, between 30 and 150 kg N ha^{-1}, were used in that study. Other crops, such as cabbage, also produced larger heads when 400 kg N ha^{-1} was applied (Turan & Sevimli, 2005).

Time of nitrogen application did not significantly influence the total number of leaves harvested for Swiss chard plants. However, nitrogen that was split into 11 applications had a significantly positive effect on the number of leaves harvested at the later stages of harvesting (weeks 17 and 21 after planting). Biemond (1995a) also reported only a small difference in the total number of spinach leaves where nitrogen was split into five, instead of two nitrogen applications.

4.2.3 Leaf fresh mass

A summary of the analyses of variance that was done to determine the effects of different nitrogen levels and application times on the fresh mass per leaf of harvested leaves of 'Fordhook Giant' and 'Rhubarb' plants from 9 up to 21 weeks after planting is given in Table 4.11. Leaf fresh mass, as indicated in this section, is the total of the leaf blade and the petiole, and it is expressed as fresh mass per leaf harvested.

Table 4.11: Summary of the analyses of variances showing the significant effects of nitrogen levels and nitrogen application times on the fresh mass of 'Fordhook Giant' and 'Rhubarb' plants from 9 up to 21 weeks after planting

Weeks after planting	Cultivar (C)	Nitrogen level (NL)	Nitrogen application time (NA)	C × NL	NL × NA	C × NA
9	*	*	ns	ns	ns	ns
13	ns	*	*	ns	ns	ns
17	ns	*	*	ns	*	ns
21	*	*	ns	ns	ns	ns
Total of leaf fresh mass plant^{-1}	ns	*	ns	ns	ns	ns

LSD $_{(T \leq 0.05)}$
ns = no significant differences
*** = significant differences**

As shown in Table 4.11 the interaction between cultivar and nitrogen levels, as well as between cultivar and nitrogen application times did not significantly influence fresh mass of leaves harvested. The interaction between nitrogen levels and nitrogen application times significantly influenced leaf fresh mass at 17 weeks after planting. As was expected, fresh mass increased with increasing nitrogen levels at all the four application times (Table 4.12). Fresh mass of leaves harvested from plants that received nitrogen at a level of 200 or 400 kg N ha^{-1} was significantly higher than where 0, 50 or 100 kg N ha^{-1} was applied in 11 applications (T_1). Fresh mass increased from 0.0 g at 0 kg N ha^{-1} to

16.4 g at 400 kg N ha^{-1}. The same tendency was observed for the T_2, T_3 and T_4 treatments (Table 4.12). Leaf fresh mass of the T_1 treated plants that received 400 kg N ha^{-1} was also significantly higher than the T_3 and T_4 treated plants.

Table 4.12: Effect of nitrogen levels and application times on fresh mass (g) per leaf of harvested Swiss chard plants 17 weeks after planting

Nitrogen levels (NL) (kg ha^{-1})	Nitrogen application times (NA)			
	T_1	T_2	T_3	T_4
0	0.0	0.9	0.0	1.1
50	1.9	2.2	1.7	3.0
100	3.1	1.8	2.6	4.1
200	12.7	4.2	6.3	9.1
400	16.4	11.6	8.1	8.5
$LSD_{(T \leq 0.05)}$ NL X NA	**7.94**			
% Nitrogen received of the total required	72.72	100	75	100
Last nitrogen application (weeks after planting)	16	16	12	16

At 17 weeks after planting, both the T_2 and T_4 treated plants had received 100% of the total nitrogen required compared to 72.72% of the T_1 and 75% of the T_3 treated plants (Table 4.12). The reason why the fresh mass of the T_1 treated leaves was higher than the other treatments at 400 kg N ha^{-1} may be attributed to the fact that the nitrogen was split into 11 applications and was applied at regular intervals every second week, compared to the other treatments where the nitrogen was split into 5 (T_2); 4 (T_3) and 3 (T_4) applications.

Leaf fresh mass of 'Fordhook Giant' was significantly greater than that of 'Rhubarb' leaves 9 weeks after planting, while at 21 weeks after planting the opposite was true (Table 4.13). This may be attributed to the fact that 'Fordhook Giant' reached maturity two weeks later than 'Rhubarb'.

Table 4.13: Fresh mass (g) per leaf of harvested 'Fordhook Giant' and 'Rhubarb' plants from 9 up to 21 weeks after planting

Weeks after planting	Cultivar (C)		$LSD_{(T \leq 0.05)}$ C
	Fordhook Giant	Rhubarb	
9	20.3	14.8	**2.3**
13	10.6	9.7	**ns**
17	5.0	5.0	**ns**
21	1.8	3.7	**1.4**

As shown in Table 4.14 fresh mass of leaves harvested increased with increasing levels of nitrogen from 9 up to 17 weeks after planting. Fresh mass of leaves harvested from plants that received 100, 200 or 400 kg N ha^{-1} was significantly higher than plants that did not receive any nitrogen at 9 and 13 weeks after planting. Seventeen weeks after planting it was only the leaf fresh mass of plants that received 200 kg N ha^{-1} and 400 kg N ha^{-1} and at 21 weeks after planting plants that received 400 kg N ha^{-1} that were significantly higher than plants that did not receive any nitrogen. Fresh mass of leaves harvested 21 weeks after planting increased from 0.4 g to 7.9 g where no or 400 kg N ha^{-1} was applied respectively. The same tendency was observed at the other harvesting times.

Table 4.14: Effect of nitrogen levels fresh mass (g) per leaf of harvested Swiss chard plants from 9 up to 21 weeks after planting

Weeks after planting	Nitrogen levels (NL) (kg ha^{-1})					$LSD_{(T \leq 0.05)}$ NL
	0	50	100	200	400	
9	9.2	12.8	17.4	20.1	28.1	**5.1**
13	1.1	4.1	7.6	15.7	22.5	**3.8**
17	0.4	1.7	2.8	8.1	11.1	**3.0**
21	0.4	0.6	2.0	2.8	7.9	**3.0**

Inspection of Table 4.15 reveals that nitrogen application time did not influence fresh mass during the first and the last weeks (9 and 21 weeks after planting) of harvesting. At 13 weeks after planting fresh mass of leaves harvested from the T_1 treated plants was significantly more than the T_2 treated plants, although the T_1 treated plants received less nitrogen (63.63%) than the T_2 treated plants (80%). Leaf fresh mass, 17 weeks after planting, was significantly more for the T_1 treatment than for the T_2 and T_3 treatments. This may be attributed to the fact that T_3 treated plants received 75% of the required nitrogen already at 12 weeks after planting compared to the T_1 treated plants that received 72.72% of the required nitrogen 16 weeks after planting. Although the T_2 treated plants received 100% of the required nitrogen (16 weeks after planting) the leaf fresh mass was still significantly lower than the T_1 treatment. There is a clear trend that fresh mass of leaves harvested from Swiss chard plants reacted positively to nitrogen applied in small amounts at regular intervals (11 applications) over the entire growth season.

Table 4.15: Effect of nitrogen application times on the fresh mass (g) per leaf of after planting harvested Swiss chard plants from 9 up to 21 weeks

Weeks after planting	Nitrogen application times (NA)												LSD ($T \leq 0.05$) NA
	T_1			T_2			T_3			T_4			
9	18.2	(8)*	[36.36]**	16.9	(8)	[40]	16.4	(6)	[50]	18.5	(8)	[33.33]	**ns**
13	12.1	(12)	[63.63]	8.7	(12)	[80]	10.4	(12)	[75]	9.4	(8)	[66.66]	**3.2**
17	6.8	(16)	[72.72]	4.1	(16)	[100]	3.3	(12)	[75]	5.1	(16)	[100]	**2.6**
21	3.6	(18)	[100]	2.7	(16)	[100]	3.1	(18)	[100]	1.7	(16)	[100]	**ns**

***Values in ( ) brackets indicate the last application time of nitrogen (weeks after planting)**
****Value in [ ] brackets indicate the % nitrogen the plants received of the total nitrogen required**

Nitrogen levels significantly influenced the total fresh mass of harvested leaves for Swiss chard plants, irrespective of the cultivar or nitrogen application time (Table 4.11). The total fresh mass of leaves harvested per Swiss chard plant increased from 26.2 g at 0 kg N ha^{-1} to 207.7 g at 400 kg N ha^{-1} (Table 4.16).

Table 4.16: Effect of nitrogen levels on the total leaf fresh mass (g) per plant harvested over a period of 21 weeks of Swiss chard plants

Nitrogen levels (NL) (kg ha^{-1})	Leaf fresh mass (g plant^{-1})
0	26.2
50	46.6
100	72.1
200	116.7
400	207.7
LSD$_{(T\leq 0.05)}$ NL	**20.7**

From this data it is clear that nitrogen fertilisation had a significant influence on the fresh mass of leaves harvested from Swiss chard plants. As nitrogen levels increased leaf fresh mass increased, irrespective of cultivar and application times. The highest leaf fresh mass was obtained at 400 kg N ha^{-1}. Sonetra, Borin & Preston (2005) and Hammad *et al.* (2007) also reported that fresh mass of spinach increased when nitrogen levels increased. Similar results on cabbage plants were obtained by Ghanti *et al.* (1982), where yield contributing characteristics such as fresh mass, head diameter and number of marketable heads increased with an increase in nitrogen levels up to 200 kg N ha^{-1}. This positive effect of nitrogen on fresh mass was also reported for other crops such as turnip greens (Vieira *et al.*, 1998), lettuce (Lairon *et al.*, 1984) and cauliflower (Kaniszewski & Rumpel, 1998).

Time of nitrogen application did not significantly influence the total fresh mass of leaves harvested from Swiss chard plants. Nitrogen split into 11 applications over the growing season influenced fresh mass of leaves harvested during week 13 and 17 after planting. The yield of sweet onions increased where nitrogen was applied more frequently (33% of the total nitrogen in the first 12 weeks of the growth period plus three applications of 22% each in the second 12 weeks of the growth period) (Batal *et al.*, 1994). In a study by Millard & Robinson (1990), late application of foliar sprays of urea slightly increased fresh mass (tuber yield) of potatoes as compared to a single application at planting.

4.2.4 Leaf area

A summary of the analyses of variance that was done to determine the effects of different nitrogen levels and application times on the leaf area per leaf of 'Fordhook Giant' and 'Rhubarb' plants from 9 up to 21 weeks after planting is given in Table 4.17. Leaf area of Swiss chard in this section refers to the leaf area (cm^2) per harvested leaf of only the blades without the petiole.

Table 4.17: Summary of the analyses of variances showing the significant effects of nitrogen levels and nitrogen application times on leaf area of harvested leaves of 'Fordhook Giant' and 'Rhubarb' plants from 9 up to 21 weeks after planting

Weeks after planting	Cultivar (C)	Nitrogen level (NL)	Nitrogen application time (NA)	C × NL	NL × NA	C × NA
9	*	*	ns	ns	ns	ns
13	ns	*	ns	ns	ns	ns
17	ns	*	*	ns	*	ns
21	*	*	ns	ns	ns	ns
Total leaf area plant^{-1}	ns	*	ns	ns	ns	ns

LSD $_{(T \leq 0.05)}$
ns = no significant differences
*** = significant differences**

As shown in Table 4.17 the interaction between cultivar and nitrogen levels and also between cultivar and nitrogen application times did not significantly influence leaf area. The interaction between nitrogen levels and nitrogen application times significantly influenced leaf area of Swiss chard at 17 weeks after planting. As was expected, leaf area increased with increasing nitrogen levels at all four application times (Table 4.18). Leaf area of leaves harvested from plants that received 400 kg N ha^{-1} that was split into 11 applications (T_1) was significantly larger than all the other treatment combinations except where 200 kg N ha^{-1} was applied for the same nitrogen application time (T_1). Leaf area increased from 0.0 cm^2 at 0 kg N ha^{-1} to 235.8 cm^2 at 400 kg N ha^{-1} for the T_1 treatment. The same tendency was observed for the T_2, T_3 and T_4 treatments (Table 4.18).

Table 4.18: Effect of nitrogen levels and application times on the leaf area (cm^2) per leaf harvested for Swiss chard plants 17 weeks after planting

Nitrogen levels (NL) ($kg\ ha^{-1}$)	Nitrogen application times (NA)			
	T_1	T_2	T_3	T_4
0	0.0	14.6	0.0	12.5
50	24.5	35.0	0.0	39.0
100	44.7	24.5	36.2	55.5
200	189.9	67.1	73.6	136.7
400	235.8	163.8	128.6	132.0
$LSD_{(T\leq0.05)}$ NL X NA	**106.9**			
% Nitrogen received of the total required	72.72	100	75	100
Last nitrogen applied (weeks after planting)	16	16	12	16

At 17 weeks after planting both the T_2 and T_4 treatments received 100%, whilst the T_1 treatment 72.72% and the T_3 treatment 75% of the total nitrogen required (Table 4.18). The reason why the leaf area of the T_1 treated leaves was larger than the other treatments at 400 kg N ha^{-1} may be attributed to the fact that the T_1 treated plants received their nitrogen in 11 applications (once every second week) as compared to the other treatments that received nitrogen at fewer intervals but with more nitrogen at a time.

Leaf area of both cultivars decreased as the harvesting period progressed (Table 4.19). The leaf area of ‘Fordhook Giant’ plants was significantly larger than ‘Rhubarb’ plants at 9 weeks after planting. At 13 weeks after planting the leaf area of ‘Fordhook Giant’ was also larger than that of ‘Rhubarb’ but not significantly. The reason why the leaf area of ‘Rhubarb’ was larger than that of ‘Fordhook Giant’ at 17 and 21 weeks after planting may be that ‘Rhubarb’ reached maturity two weeks later than ‘Fordhook Giant’.

Table 4.19: Leaf area (cm^2) per leaf harvested of 'Fordhook Giant' and 'Rhubarb' plants from 9 up to 21 weeks after planting

Weeks after planting	Cultivar (C)		$LSD_{(T \leq 0.05)}$ C
	Fordhook Giant	Rhubarb	
9	279.8	246.1	**29.9**
13	200.0	189.4	**ns**
17	62.8	78.7	**ns**
21	25.8	59.2	**21.2**

As shown in Table 4.20 leaf area increased with increasing levels of nitrogen at all four harvesting times, although not always significantly. At 9 and 13 weeks after planting the leaf area from plants that received 100, 200 or 400 kg N ha^{-1} was significantly larger than leaves of plants that received 0 or 50 kg N ha^{-1}. During the later stages of harvesting, i.e. at 17 weeks after planting the leaf area of plants that received 200 or 400 kg N ha^{-1} and also at 21 weeks after planting plants that received 400 kg N ha^{-1} was significantly larger than plants that did not receive any nitrogen fertiliser.

Table 4.20: Effect of nitrogen levels on the leaf area (cm^2) per leaf harvested of Swiss chard 9 up to 21 weeks after planting

Weeks after planting	Nitrogen levels (NL) (kg ha^{-1})					$LSD_{(T \leq 0.05)}$ NL
	0	50	100	200	400	
9	127.2	181.8	263.5	315.1	427.3	**66.1**
13	18.0	66.2	139.9	305.6	443.8	**69.9**
17	6.8	24.7	40.2	116.9	165.1	**40.9**
21	6.4	5.8	28.5	37.2	134.7	**46.9**

Nitrogen application time only influenced leaf area during week 17 after planting as shown in Table 4.21. The leaf area measured 17 weeks after planting was significantly larger for the T_1 treatment than for the T_2 and T_3 treatments, while leaf area for the T_2, T_3 and T_4 treatments did not differ significantly from one another.

Table 4.21: Effect of nitrogen application times on the leaf area (cm^2) per leaf harvested of Swiss chard plants from 9 up to 21 weeks after planting

Weeks after planting	Nitrogen application times (NA)												LSD ($T \leq 0.05$) NA
	T_1			T_2			T_3			T_4			
9	267	(8)*	[36.36]**	261	(8)	[40]	238	(6)	[50]	284	(8)	[33.33]	**ns**
13	223	(12)	[63.63]	176	(12)	[80]	193	(12)	[75]	185	(8)	[66.66]	**ns**
17	99	(16)	[72.72]	61	(16)	[100]	47	(12)	[75]	75	(16)	[100]	**34.4**
21	51	(20)	[100]	42	(16)	[100]	48	(18)	[100]	27	(16)	[100]	**ns**

***Values in ( ) brackets indicate the last application time of nitrogen (weeks after planting)**
****Value in [ ] brackets indicate the % nitrogen the plants received of the total nitrogen required**

The same trend observed for leaf area was observed for number of leaves harvested. Although the T_1 treated plants had received less nitrogen than the other plants at 17 weeks after planting, these plants reacted better to nitrogen that was applied more frequently during the growing season than where nitrogen was applied less frequently but in larger amounts at a time. At twenty one weeks after planting, all plants received 100% of the total required nitrogen and although not significant, the leaf area of the T_1 treatment was still larger than the other treatments.

The total leaf area of harvested leaves for Swiss chard plants over a period of 21 weeks was only significantly influenced by nitrogen levels (Table 4.17). The total leaf area per plant increased from 158.3 cm^2 at 0 N kg ha^{-1} to 1170.9 cm^2 at 400 kg N ha^{-1} (Table 4.22).

Table 4.22: Effect of nitrogen levels on the total leaf area (cm^2) per plant harvested of Swiss chard plants from 9 up to 21 weeks after planting

Nitrogen levels (NL) (kg ha^{-1})	**Total leaf area (cm^2 $plant^{-1}$)**
0	158.3
50	278.4
100	472.2
200	774.7
400	1170.9
$LSD_{(T\leq0.05)}$ NL	**136.9**

Nitrogen fertilisation increased the leaf area of harvested leaves regardless of the cultivar or nitrogen application time. The largest leaf area was obtained where 400 kg N ha^{-1} was applied. These results concur with results of Gülser (2005) who reported an increase in spinach leaf area with increasing nitrogen from 0 to 150 kg N ha^{-1}. The same results were also reported for other vegetable crops such as broccoli, sweet pepper (Tremblay & Senécal, 1988; Masson, Tremblay & Gosselin, 1991) and tomatoes (Melton & Dufault, 1991).

Application time of nitrogen did not influence the total leaf area of leaves harvested from Swiss chard plants. Biemond (1995b) also reported that leaf area of mature leek plants was not influenced by either two or six equal applications of nitrogen fertiliser. However, leaf area of Swiss chard leaves harvested 17 weeks after planting benefited from nitrogen fertiliser that was split into 11 equal applications (applied every second week).

4.2.5 Leaf dry mass

A summary of the analyses of variance that was done to determine the effects of different nitrogen levels and application times on the dry mass of ‘Fordhook Giant’ and ‘Rhubarb’ leaves from 9 up to 21 weeks after planting is given in Table 4.23. Leaf dry mass in this section refers to the dry mass of the leaf blade without the petiole.

Table 4.23: Summary of the analyses of variances showing the significant effects of nitrogen levels and nitrogen application times on the dry mass of leaves of 'Fordhook Giant' and 'Rhubarb' plants from 9 up to 21 weeks after planting

Weeks after planting	Cultivar (C)	Nitrogen level (NL)	Nitrogen application time (NA)	C × NL	NL × NA	C × NA
9	ns	*	ns	ns	ns	ns
13	*	*	ns	ns	ns	ns
17	*	*	*	ns	*	ns
21	*	*	ns	ns	ns	ns
Total leaf dry mass plant^{-1}	*	*	ns	ns	ns	ns

LSD $_{(T \leq 0.05)}$
ns = no significant differences
*** = significant differences**

As shown in Table 4.23 the interaction between cultivar and nitrogen levels and also between cultivar and nitrogen application times was not significant at all four harvesting times. The interaction between nitrogen levels and nitrogen application times significantly influenced the leaf dry mass at 17 weeks after planting (Table 4.23). Leaf dry mass increased with increasing nitrogen levels at all four application times (Table 4.24) except for the T_2 treated plants. Leaf dry mass of harvested Swiss chard plants that received 200 or 400 kg N ha^{-1} was significantly higher than where 0, 50 or 100 kg N ha^{-1} was applied at the T_1 treatment. For the T_1 treatment the leaf dry mass increased from 0.0 g at 0 kg N ha^{-1} to 3.1 g at 400 kg N ha^{-1}. The same tendency was observed fo the T_2, T_3 and T_4 treatments (Table 4.24). The leaf dry mass of the T_1 treated plants that received 400 kg N ha^{-1} was also significantly higher than the T_2, T_3 and T_4 treated plants.

At 17 weeks after planting, both the T_2 and T_4 treated plants received 100% of the total nitrogen required compared to 72.72% for the T_1 and 75% for the T_3 treated plants (Table 4.24). Leaf dry mass of Swiss chard plants reacted positively to nitrogen applied every second week (T_1) at a level of 400 kg ha^{-1} compared to the other nitrogen application times. This was also true for leaf fresh mass (Section 4.2.3) and leaf area (Section 4.2.4).

Table 4.24: Effect of nitrogen levels and application times on leaf dry mass (g) of harvested leaves of Swiss chard plants 17 weeks after planting

Nitrogen levels (NL) ($kg\ ha^{-1}$)	Nitrogen application times (NA)			
	T_1	T_2	T_3	T_4
0	0.0	0.1	0.0	0.1
50	0.2	0.3	0.0	0.4
100	0.7	0.2	0.3	0.6
200	2.0	0.7	1.0	1.0
400	3.1	2.1	1.6	1.7
$LSD_{(T \leq 0.05)}$ NL X NA	**0.6**			
% Nitrogen received of the total required	72.72	100	75	100
Last nitrogen application (weeks after planting)	16	16	12	16

Leaf dry mass of 'Rhubarb' was significantly higher than of 'Fordhook Giant' from 13 up to 21 weeks after planting, except at 9 weeks after planting (Table 4.25).

Table 4.25: Dry mass (g) of harvested leaves of 'Fordhook Giant' and 'Rhubarb' plants from 9 up to 21 weeks after planting

Weeks after planting	Cultivar (C)		$LSD_{(T \leq 0.05)}$ C
	Fordhook Giant	Rhubarb	
9	5.0	5.0	**ns**
13	1.7	2.3	**0.2**
17	0.7	1.0	**0.1**
21	0.3	0.6	**0.1**

Leaf dry mass increased with increasing levels of nitrogen from 9 up to 21 weeks after planting (Table 4.26). The dry mass of leaves harvested from Swiss chard plants that received 100, 200 and 400 kg N ha^{-1} was significantly higher than plants that received no nitrogen at 9 and 13 weeks after planting. Seventeen weeks after planting the leaf dry mass of plants that received 100, 200 and 400 kg N ha^{-1} was significantly higher than plants that received 0 and 50 kg N ha^{-1}. Leaf dry mass of Swiss chard plants that received 400 kg N ha^{-1} was significantly higher than leaves from plants that received no nitrogen 21 weeks after planting. At 21 weeks after planting the dry mass of leaves increased from 0.0 g where no nitrogen was applied to 1.4 g where 400 kg N ha^{-1} was applied. The same tendency was observed for the other harvesting times (Table 4.26).

Table 4.26: Effect of nitrogen levels on the dry mass (g) of harvested leaves of Swiss chard from 9 up to 21 weeks after planting

Weeks after planting	Nitrogen levels (NL) (kg ha^{-1})					$LSD_{(T \leq 0.05)}$ NL
	0	**50**	**100**	**200**	**400**	
9	1.7	2.3	4.0	6.2	11.0	**0.9**
13	0.2	0.6	1.2	2.9	5.2	**0.4**
17	0.1	0.2	0.5	1.3	2.1	**0.3**
21	0.0	0.2	0.2	0.3	1.4	**0.3**

Nitrogen application time significantly influenced dry mass at week 17 after planting (Table 4.27). Leaf dry mass of harvested leaves of Swiss chard plants was significantly more for the T_1 treatment than the other nitrogen application treatments. This may be attributed to the fact that T_3 treated plants received 75% of the required nitrogen at 12 weeks after planting compared to the T_1 treated plants that received 72.72% of the required nitrogen 16 weeks after planting. Although the T_2 and T_4 treatments received 100% of the required nitrogen 16 weeks after planting, the leaf dry mass was still significantly lower than the T_1 treatment. There is a clear indication that Swiss chard plants reacted positively to nitrogen applied in small quantities at regular intervals over the entire growth season.

Table 4.27: Effect of nitrogen application times on leaf dry mass (g) per leaf harvested of Swiss chard plants from 9 up to 21 weeks after planting

Weeks after planting	Nitrogen application times (NA)												LSD $_{(T\leq0.05)}$ NA
	T_1			T_2			T_3			T_4			
9	5.1	(8)*	[36.36]**	4.7	(8)	[40]	5.2	(6)	[50]	4.9	(8)	[33.33]	**ns**
13	2.1	(12)	[63.63]	1.9	(12)	[80]	2.1	(12)	[75]	1.9	(8)	[66.66]	**ns**
17	1.2	(16)	[72.72]	1.0	(16)	[100]	1.0	(12)	[75]	1.0	(16)	[100]	**0.2**
21	0.5	(20)	[100]	0.4	(16)	[100]	0.5	(18)	[100]	0.3	(16)	[100]	**ns**

***Values in ( ) brackets indicate the last application time of nitrogen (weeks after planting)**
****Value in [ ] brackets indicate the % nitrogen the plants received of the total nitrogen required**

The total leaf dry mass of 'Rhubarb' and 'Fordhook Giant' differed significantly from each other (Table 4.23). The total leaf dry mass for harvested leaves of 'Rhubarb' plants (9.0 g) was significantly more than for 'Fordhook Giant' plants (7.7 g) ($LSD_{(T\leq0.05)}$ = 2.3). Increasing nitrogen levels increased the total dry mass of harvested Swiss chard leaves regardless of the cultivar or nitrogen application time. The total leaf dry mass increased from 2.0 g at 0 kg N ha^{-1} to 19.2 g at 400 kg N ha^{-1} (Table 4.28).

Table 4.28: Effect of nitrogen levels on the total leaf dry mass (g) per plant of Swiss chard plants harvested over a period of 21 weeks

Nitrogen levels (NL) (kg ha^{-1})	Dry mass (g $plant^{-1}$)
0	2.0
50	3.4
100	5.9
200	10.7
400	19.2
$LSD_{(T\leq0.05)}$ NL	**2.3**

As the nitrogen levels increased the dry mass of leaves harvested per plant increased irrespective of the cultivar or time of nitrogen application. In this study the highest dry mass of leaves harvested was obtained where 400 kg N ha^{-1} was applied. These findings were similar to findings of other researchers on crops such as spinach (Hammad *et al*.,

2007) celery, lettuce, broccoli, tomatoes (Masson *et al*., 1991), turnip greens (Vieira *et al*., 1998), peppers (Dufault & Schultheis, 1994) and Brussel sprouts (Biemond, Vos & Struik 1996). Lairon *et al*. (1984) reported that the dry matter of lettuce was not significantly influenced by nitrogen application levels. The reason for these conflicting results may be attributed to the low nitrogen levels used on lettuce (200 kg N ha^{-1}) compared to levels used on Swiss chard (400 kg N ha^{-1}).

Results showed that nitrogen application time did not influence the total dry mass of leaves harvested; however, where nitrogen was split into 11 applications (T_1) over the growing season, the time of the nitrogen application had a positive effect on the dry mass of Swiss chard leaves harvested 17 weeks after planting. Different results were found for other vegetable crops such as Brussel sprouts. Biemond *et al*. (1995) observed that nitrogen applied in three equal applications affected dry matter of Brussel sprouts more positively than nine equal applications. Reeves, Wood & Touchton (1993) reported greater dry matter production of maize (*Zea mays* L.) when nitrogen was applied at planting (100%) as a single application compared with split application of nitrogen at planting (33.33%) and the remainder of the nitrogen (66%) applied as a side-dress application.

4.2.6 Leaf nitrogen content

A summary of the analyses of variance that was done to determine the effects of different nitrogen levels and application times on the nitrogen content of 'Fordhook Giant' and 'Rhubarb' harvested leaves is given in Table 4.29.

Table 4.29: Summary of the analyses of variances showing the significant effects of nitrogen levels and nitrogen application times on the nitrogen content (%) of 'Fordhook Giant' and 'Rhubarb' harvested leaves

Nutrient	Cultivar (C)	Nitrogen level (NL)	Nitrogen application time (NA)	C x NL	NL x NA	C x NA
N	*	*	*	ns	ns	*

LSD $_{(T \leq 0.05)}$
ns = no significant differences
*** = significant differences**

As shown in Table 4.29 the interaction between cultivar and nitrogen levels and also between nitrogen levels and nitrogen application times did not significantly influence the nitrogen content of Swiss chard leaves. The interaction between cultivar and nitrogen application times significantly influenced the nitrogen content of Swiss chard plants. The percentage nitrogen in 'Rhubarb' leaves was more than in 'Fordhook Giant' leaves, regardless of the nitrogen application time, except where nitrogen was split into 4 applications (T_3). The nitrogen content of 'Rhubarb' leaves from the T_1, T_2 and T_4 treated plants was significantly more than leaves from the T_3 treated plants. 'Fordhook Giant' leaves harvested from the T_2, T_3 and T_4 treated plants contained significantly more nitrogen than leaves from the T_1 treated plants (Table 4.30).

Table 4.30: Effect of nitrogen application times on the leaf nitrogen content (%) of 'Fordhook Giant' and 'Rhubarb' plants

Nitrogen application times	**Cultivar (C)**	
	Fordhook Giant	**Rhubarb**
T_1	2.1	2.5
T_2	2.5	2.6
T_3	2.3	2.2
T_4	2.5	2.6
$LSD_{(T\leq 0.05)}$ C x NA	**0.3**	

The nitrogen content of 'Rhubarb' plants (2.5%) was significantly more than that of 'Fordhook Giant' plants (2.3%) (Table 4.31).

Table 4.31: Nitrogen content (%) of 'Fordhook Giant' and 'Rhubarb' leaves harvested

Cultivar	**Nitrogen (%)**
Fordhook Giant	2.3
Rhubarb	2.5
$LSD_{(T\leq 0.05)}$ C	**0.1**

Inspection of Table 4.32 shows that the nitrogen content of Swiss chard leaves increased significantly with increasing levels of nitrogen. Leaf nitrogen content of Swiss chard was significantly more where 100, 200 or 400 kg N ha^{-1} was applied compared to where 0 kg N ha^{-1} was applied. As nitrogen levels increased from 0 to 400 kg N ha^{-1}, the nitrogen content of Swiss chard leaves increased from 2.0% to 3.3%.

Table 4.32: Effect of nitrogen levels on the nitrogen content (%) of 'Fordhook Giant' and 'Rhubarb' leaves harvested

Nitrogen levels (NL) ($kg\ ha^{-1}$)	Nitrogen (%)
0	2.0
50	2.1
100	2.2
200	2.5
400	3.3
$LSD_{(T\leq 0.05)}$ NL	**0.2**

Inspection of Table 4.33 shows that nitrogen application times significantly influenced the nitrogen content of Swiss chard leaves. The nitrogen content of Swiss chard leaves was significantly higher for the T_2 (5 equal applications) and T_4 (3 equal applications) treatments than the T_1 (11 equal applications) and T_3 (4 equal applications) treatments.

Table 4.33: Effect of nitrogen application times on the nitrogen content (%) of 'Fordhook Giant' and 'Rhubarb' leaves harvested

Nitrogen application times (NA)	Nitrogen (%)
T_1	2.3
T_2	2.6
T_3	2.2
T_4	2.5
$LSD_{(T\leq 0.05)}$ NA	**0.2**

Nitrogen fertilisation increased the nitrogen content of Swiss chard leaves regardless of the cultivar or nitrogen application time. The highest nitrogen content was recorded where 400 kg N ha^{-1} was applied. Nitrogen content of leaves of several crops increased with increasing nitrogen levels such as spinach (Cantliffe, 1973a; Hammad *et al.*, 2007), beet (Cantliffe, 1973a), lettuce (Soundy *et al.*, 2001), cabbage, Brussel sprouts, broccoli (Sanderson & Ivany, 1999) and turnip greens (Vieira *et al.*, 1998).

4.3 CONCLUSIONS

Results from this study emphasise the importance of sustainable nitrogen management which aims to supply enough nitrogen for optimum growth and yield, while losses to the environment are kept to a minimum. This can only be done by matching the demand of the crop for nitrogen with the application time, as well as taking into account the practical and financial implications thereof.

From this research it is clear that nitrogen fertiliser has a beneficial effect on the growth (number of leaves during the first 6 weeks after planting), yield (number of leaves harvested and leaf fresh mass) and quality (leaf area, leaf dry mass and nitrogen content of the leaf) of Swiss chard plants. In this study, the optimum nitrogen application level for all the parameters measured was 400 kg N ha^{-1}. The positive effect of nitrogen fertiliser may be due to the physiological effect it has on the vegetative growth and plant development. Cultivar differences should also be taken into consideration when managing nitrogen fertilisation. In this study 'Rhubarb' yielded more leaves than 'Fordhook Giant'.

Nitrogen application time did significantly influence the early growth (first 6 weeks after planting) of Swiss chard plants. It is clear that where more nitrogen was available (T_2, T_3 and T_4 treatments) during the first six weeks after planting, the better the growth of the plants when the number of leaves were used as an index. Although the application time of nitrogen did not influence the total yield (number of leaves harvested, leaf area and leaf fresh mass) of Swiss chard, it is clear that nitrogen applied late in the growth season had a positive influence on the harvest (T_1 treatment). The total nitrogen content of the leaves was however, negatively influenced by the T_1 treatment.

CHAPTER 5

NITROGEN SOURCES AFFECTING SWISS CHARD PRODUCTION

5.1 INTRODUCTION

Commercial vegetable production represents one of the most intensive uses of land and it often results in the rapid depletion of soil fertility (Goh & Vityakon, 1983). In arid and semi-arid regions both water and nitrogen are the main limiting factors for crop production. Vegetables are mostly planted under irrigation and water is then normally not a limiting factor for yield, but nitrogen requirements of these crops are high and nitrogen is then the most limiting factor (Qawasmi *et al.*, 1999).

Swiss chard is planted for its dark green succulent leaves. Growers tend to apply large quantities of fertiliser, especially nitrogen to leafy greens to maximise yield and quality, as well as to gain greater economic returns (Cantliffe, 1973b; Baker, 1980; Goh & Vityakon, 1983). This can also been seen in the increased use of fertiliser over the past years. Nitrogen fertiliser used in excess is often wrongly considered as insurance against yield losses. Excessive use of nitrogen fertilisation can lead to environmental contamination due to leaching, volatilisation, denitrification and surface run off, as well as to accumulation of nitrate in leafy vegetables beyond safe limits (Umar, Iqbal & Abrol, 2007). For most crops recommendations of nitrogen fertiliser application rates are available, but a large variation in the optimum values exists due to environmental conditions (Van Keulen & Stols, 1991).

The nitrate forms of nitrogen fertiliser are widely used in vegetable production resulting in accumulation of nitrates in plants if the rate of their intake exceeds the rate of reduction to ammonium (Luo, Lion & Yan, 1993). The use of nitrogen fertiliser in the ammonium form or as a mixture of nitrate and ammonium can reduce the nitrate content in plants (Inal & Tarakcioglu, 2001; Santamaria *et al.*, 2001). Ammonium and nitrate fertilisation of plants directly influence the yield but also the chemical composition of vegetables whereby the latter may affect the health of the consumer (Goh & Vityakon, 1983). Nitrogen levels and the timing of nitrogen application can lead to nitrate accumulation in plants. The quantity of available nitrogen is one of the most important factors that may lead to high nitrate levels in the plant (Schuphan *et al*., 1967; Baker & Maynard, 1971; Peck *et al.*, 1971; Maynard *et al*., 1976).

It is well known that spinach accumulates nitrate in response to excess nitrogen (Briemer, 1982; Goh & Vityakon, 1986). Vegetables play an important role in human diet and an even greater part in the nutrient supply for some groups such as vegetarians in developed countries or among the poor in developing countries (Lairon *et al*., 1984). The nutritional value of the food that humans consume is determined by the composition of the raw materials which is influenced by cultivar selection, growing conditions and post harvest treatments (Schuphan *et al.,* 1967). The main sources of nitrates for humans are plants and drinking water. High nitrate and nitrite levels can be toxic to humans. The maximum level (limit) for nitrate in spinach according to the European commission regulation No. 563/2002 for fresh spinach is 2500-3000 mg kg^{-1} fresh mass and for preserved, deep frozen or frozen spinach is 2000 mg kg^{-1} fresh mass (European Commission, 2002).

Excessive transformation of nitrates into nitrite in organisms leads to methemoglobinemia. This intoxication results from a reaction between nitrite and hemoglobine and formation of methemoglobinemia, which cannot bind and exchange oxygen in the blood. A high level of methemoglobin can even cause death (Schuphan *et al*., 1967; Vulsteke & Biston, 1978). Although spinach is claimed to be a valuable vegetable, it has also, from time to time, been claimed as a potential hazard for infants due to its nitrate content, which under certain conditions, might give rise to toxic amounts of nitrate (Schuphan *et al*., 1967). Therefore, efforts to minimise the accumulation of nitrates in leafy vegetables are needed (Umar & Iqbal, 2006) that can add value to vegetable products already popular for their nutritional and therapeutic properties (Santamaria, 2006). The leaf blades of spinach are known to accumulate fewer nitrates than the petioles but more than the midrib, and the nitrate content tends to be higher in autumn-winter than in the spring (Santamaria *et al.*, 1999b). Researchers suggest that by cutting the spinach higher from the soil surface might improve the quality of spinach (Elia, Santamaria & Serio, 1998). The response of Swiss chard plants to nitrogen varies in different cultivars, as well as from different agricultural practices. It appears that the availability of different nitrogen fertilisers to Swiss chard plants may have a determining effect on leaf growth, yield and quality. The study was aimed to evaluate the effect of different nitrogen sources applied at different levels on the growth, yield and quality of Swiss chard.

5.2 RESULTS AND DISCUSSION

5.2.1 Number of leaves harvested

A summary of the analyses of variance that was done to determine the effects of different nitrogen sources and levels on the number of leaves harvested every three or four weeks of 'Fordhook Giant' plants from 8 up to 21 weeks after planting is given in Table 5.1.

Table 5.1: Summary of the analyses of variances showing the significant effects of nitrogen sources and levels on the number of leaves harvested per plant every three or four weeks of 'Fordhook Giant' from 8 up to 21 weeks after planting

Weeks after planting	Nitrogen source (NS)	Nitrogen level (NL)	NS× NL
8	ns	*	*
11	*	*	ns
14	ns	*	ns
17	ns	*	ns
21	ns	*	*
Total number of leaves harvested plant^{-1}	ns	*	*

LSD $_{(T \leq 0.05)}$
ns = no significant differences
*** = significant differences**

As shown in Table 5.1 the interaction between nitrogen source and nitrogen level significantly influenced the number of leaves harvested per plant at 8 and 21 weeks after planting. Although not significant, more leaves were harvested from Swiss chard plants that received ammonium nitrate (7.5), potassium nitrate (7.0) or urea (7.0) than plants that received calcium nitrate (6.5), ammonium sulphate (6.5) or urea ammonium nitrate (6.5) as nitrogen source (800 kg N ha^{-1}) eight weeks after planting (Table 5.2). As was expected, the number of Swiss chard leaves harvested increased with increasing nitrogen levels irrespective of the nitrogen source. At the same nitrogen level, there was no significant difference amongst the different nitrogen sources except where 700 kg N ha^{-1} was applied as urea ammonium nitrate. The number of leaves harvested from these plants was significantly lower than from plants that received other nitrogen sources. The highest number of leaves was obtained where 800 kg N ha^{-1} was applied irrespective of the nitrogen source.

Plants that received ammonium nitrate, potassium nitrate, ammonium sulphate or urea ammonium nitrate as nitrogen source at 500 kg N ha^{-1} and higher, yielded significantly more leaves than plants that did not receive any nitrogen. Plants that received calcium nitrate or urea yielded significantly more leaves already from 400 kg N ha^{-1} compared to plants that did not receive any nitrogen (8 weeks after planting).

Table 5.2: Effect of nitrogen sources and levels on the number of leaves harvested per plant of 'Fordhook Giant' 8 weeks after planting

Nitrogen level (NL) (kg ha^{-1})	Nitrogen source (NS)					
	NH_4 NO_3	KNO_3	$Ca(NO_3)_2$	$(NH_4)_2$ SO_4	$CO(NH_2)_2$. NH_4NO_3	CO $(NH_2)_2$
0	0.0	0.0	0.0	0.0	0.3	0.5
100	0.8	2.0	0.0	2.0	0.5	2.0
200	1.5	1.8	1.8	1.0	2.0	1.5
300	2.0	1.8	2.5	1.5	1.3	2.0
400	2.0	2.8	3.0	1.5	3.0	3.5
500	4.3	3.3	4.5	4.3	3.5	3.5
600	4.0	4.3	3.8	4.8	3.8	3.8
700	6.3	6.5	6.8	5.8	3.8	7.0
800	7.5	7.0	6.5	6.5	6.5	7.0
$LSD_{(T\leq0.05)}$ NS X NL	**2.9**					

At twenty-one weeks after planting, the reaction of Swiss chard plants to different nitrogen sources and levels was not consistent like it was at eight weeks after planting (Table 5.3). Although not significant, plants that received ammonium sulphate or urea ammonium nitrate produced more leaves than plants that received other nitrogen sources at application rates higher than 500 kg N ha^{-1}. The number of Swiss chard leaves harvested at a specific nitrogen level was also not significantly influenced by the different nitrogen sources which were also true at 8 weeks after planting. As the nitrogen level increased, the number of leaves harvested also tended to increase but this trend was not so clear for all the nitrogen sources at this late stage of harvesting (21 weeks after planting).

Table 5.3: Effect of nitrogen sources and levels on the number of leaves harvested per plant of 'Fordhook Giant' 21 weeks after planting

Nitrogen level (NL) (kg ha^{-1})	Nitrogen source (NS)					
	NH_4 NO_3	KNO_3	$Ca(NO_3)_2$	$(NH_4)_2$ SO_4	$CO(NH_2)_2$. NH_4NO_3	CO $(NH_2)_2$
0	0.0	0.0	0.0	0.0	0.0	0.0
100	0.0	1.0	0.0	3.5	2.8	3.8
200	2.3	5.3	5.0	1.0	3.3	3.8
300	5.8	3.0	4.0	3.5	5.5	5.3
400	5.5	6.0	5.0	3.5	2.8	5.3
500	6.5	5.3	5.5	4.3	5.3	5.3
600	4.8	5.0	6.3	7.0	6.8	4.5
700	4.8	6.3	5.5	7.2	7.8	6.8
800	6.8	5.3	4.3	7.3	7.0	6.0
$LSD_{(T \leq 0.05)}$ NS X NL	**5.1**					

Nitrogen source influenced the number of Swiss chard leaves harvested at 11 weeks after planting (Table 5.4). Significantly more leaves were harvested from plants that received urea than from plants that received urea ammonium nitrate as nitrogen source. No significant differences existed amongst the other nitrogen sources.

Table 5.4: Effect of nitrogen sources on the number of leaves harvested per plant of 'Fordhook Giant' from 8 up to 21 weeks after planting

Weeks after planting	Nitrogen source (NS)						$LSD_{(T \leq 0.05)}$ NS
	NH_4 NO_3	KNO_3	$Ca(NO_3)_2$	$(NH_4)_2SO_4$	$CO(NH_2)_2$. NH_4NO_3	CO $(NH_2)_2$	
8	3.1	3.3	3.2	3.0	2.7	3.4	**ns**
11	3.9	3.7	3.6	3.6	3.2	4.4	**1.0**
14	4.8	4.4	3.9	4.5	4.8	5.0	**ns**
17	4.4	4.2	3.3	4.4	4.3	4.5	**ns**
21	4.1	4.1	3.9	4.2	4.5	4.5	**ns**

As shown in Table 5.5 it is clear that nitrogen level significantly influenced the number of leaves harvested at all five harvesting times irrespective of the nitrogen source. At 8, 14 and 21 weeks after planting all the plants that received nitrogen yielded significantly more leaves than plants that did not receive any nitrogen.

Table 5.5: Effect of nitrogen levels on the number of leaves harvested per plant of 'Fordhook Giant' from 8 up to 21 weeks after planting

Weeks after planting	Nitrogen level (NL) (kg ha^{-1})									$LSD_{(T\leq0.05)}$ NL
	0	100	200	300	400	500	600	700	800	
8	0.1	1.2	1.6	1.8	2.6	3.9	4.0	6.0	6.8	**0.9**
11	0.1	1.1	3.0	3.1	3.3	4.8	4.8	5.9	7.3	**1.4**
14	0.2	2.7	3.6	4.2	4.8	5.4	6.4	6.9	7.0	**1.7**
17	0.0	1.5	3.3	4.6	5.0	5.3	5.8	6.0	6.8	**1.6**
21	0.0	1.8	3.5	4.5	4.7	5.3	5.7	6.9	6.2	**1.6**

The same trend was observed at 11 and 17 weeks after planting but it was only plants that received 200 kg N ha^{-1} and more that yielded significantly more leaves than plants that received no or 100 kg N ha^{-1}. The highest nitrogen level (800 kg ha^{-1}) yielded the most number of leaves of all the harvesting times except at 21 weeks after planting when plants that received 700 kg N ha^{-1} yielded the most number of leaves.

The total number of leaves harvested per plant over the period of 21 weeks was significantly influenced by the interaction between nitrogen source and nitrogen level (Table 5.6). As shown in Table 5.6 the total number of Swiss chard leaves harvested per plant increased with increasing nitrogen levels for all nitrogen sources. The total number of leaves harvested for Swiss chard plants where ammonium nitrate was applied increased from 1.8 to 37.0 per plant when the nitrogen level was increased from 0 to 800 kg ha^{-1}. The same trend was observed for the other nitrogen sources except for potassium nitrate that showed a decrease in the number of leaves harvested when nitrogen was increased from 700 kg ha^{-1} (32.5) to 800 kg ha^{-1} (30.3). Although not always significant, the most number of leaves (37) were harvested where ammonium nitrate was applied, followed by ammonium sulphate (36), urea ammonium nitrate (35) and urea (34.8) at the same nitrogen level.

Table 5.6: Effect of nitrogen sources and levels on the total number of leaves harvested per plant of 'Fordhook Giant'

Nitrogen level (NL) ($kg\ ha^{-1}$)	Nitrogen source (NS)					
	$NH_4\ NO_3$	KNO_3	$Ca(NO_3)_2$	$(NH_4)_2\ SO_4$	$CO(NH_2)_2.\ NH_4NO_3$	$CO\ (NH_2)_2$
0	1.8	0.0	0.0	0.0	0.2	0.5
100	2.5	7.8	1.0	15.8	7.3	15.5
200	13.0	18.8	17.5	6.0	18.8	16.0
300	21.3	14.0	17.0	17.5	18.0	22.0
400	20.3	25.0	19.3	16.8	15.5	26.0
500	28.3	21.5	25.0	23.3	25.0	25.5
600	27.3	27.0	26.8	29.0	27.3	23.3
700	31.0	32.5	27.5	33.6	29.0	34.3
800	37.0	30.3	28.0	36.0	35.0	34.8
$LSD_{(T\leq0.05)}$ NS X NL	**16.5**					

It is clear from these results that different nitrogen sources did not significantly influence the total number of Swiss chard leaves harvested. Different nitrogen sources only differed in the level at which they started to significantly influence the number of leaves harvested. On the other hand, different nitrogen levels did significantly influence the number of leaves harvested from Swiss chard plants, which corresponded with results that were obtained earlier in Chapter 4, Section 4.2.2. In both trials, the highest number of leaves harvested was obtained at the highest nitrogen level applied, namely 400 or 800 $kg\ ha^{-1}$. Gülser (2005) also studied the effect of urea and ammonium sulphate on the yield components of spinach. No significant differences were reported between the two nitrogen sources, as well as the different nitrogen levels on the number of spinach leaves harvested. The reason for these conflicting results may be attributed to the low nitrogen levels applied (150 $kg\ ha^{-1}$) to spinach. Results from this current study show that significant differences were only distinct from 200 kg N ha^{-1} applied as urea and 300 kg N ha^{-1} applied as ammonium sulphate to Swiss chard plants.

Surprisingly, there was no sign of a decrease in the total number of Swiss chard leaves harvested where high levels of nitrogen (800 kg ha^{-1}). This do not correspond with data from Goh & Vityakon (1983) who reported a yield reduction at a nitrogen application rate of 600 kg ha^{-1} for spinach.

5.2.2 Leaf fresh mass

A summary of the analyses of variance that was done to determine the effects of different nitrogen sources and levels on the fresh mass of leaves of 'Fordhook Giant' plants harvested from 8 up to 21 weeks after planting is given in Table 5.7. Leaf fresh mass, as indicated in this section, is the total of the leaf blade and the petiole, and it is expressed as fresh mass per leaf harvested.

Table 5.7: Summary of the analyses of variances showing the significant effects of nitrogen sources and levels on the leaf fresh mass of 'Fordhook Giant' plants harvested from 8 up to 21 weeks after planting

Weeks after planting	Nitrogen source (NS)	Nitrogen level (NL)	NS × NL
8	*	*	*
11	*	*	ns
14	ns	*	ns
17	ns	*	ns
21	ns	*	ns
Total leaf fresh mass $plant^{-1}$	ns	*	ns

LSD $_{(T \leq 0.05)}$
ns = no significant differences
*** = significant differences**

As shown in Table 5.7 the interaction between nitrogen source and nitrogen level significantly influenced leaf fresh mass at 8 weeks after planting. Nitrogen source did not significantly influence the fresh mass of leaves. It was only the nitrogen level where the different nitrogen sources started to influence the fresh mass significantly (Table 5.8) that differed. 'Fordhook Giant' plants that received potassium nitrate, urea ammonium nitrate or urea as nitrogen source at 400 kg N ha^{-1} and more, produced leaves with significantly higher fresh mass than plants that did not receive any nitrogen. Plants that received calcium nitrate and ammonium nitrate at 300 kg N ha^{-1} and more yielded significantly heavier leaves than plants that did not receive any nitrogen. Nitrogen applied as ammonium sulphate only started to influence the fresh mass of Swiss chard leaves significantly from 500 kg N ha^{-1}. As was expected leaf fresh mass of 'Fordhook Giant' increased with increasing nitrogen levels irrespective of the nitrogen source. The greatest leaf fresh mass was measured where 800 kg N ha^{-1} was applied irrespective of the nitrogen source.

Table 5.8: Effect of nitrogen sources and levels on fresh mass (g) per leaf harvested of 'Fordhook Giant' 8 weeks after planting

Nitrogen level (NL) (kg ha^{-1})	**Nitrogen source (NS)**					
	NH_4 NO_3	**KNO_3**	**$Ca(NO_3)_2$**	**$(NH_4)_2$ SO_4**	**$CO(NH_2)_2$. NH_4NO_3**	**CO $(NH_2)_2$**
0	0.0	0.0	0.0	0.0	1.4	3.8
100	4.5	10.4	2.3	9.8	2.9	8.6
200	11.1	9.9	9.2	6.1	8.2	8.1
300	13.0	8.9	13.6	10.0	4.2	8.5
400	10.2	15.3	15.2	7.8	13.5	13.4
500	18.8	13.2	16.9	16.7	13.4	14.6
600	19.5	20.0	18.2	16.5	16.3	14.9
700	22.5	22.2	19.5	19.5	14.0	15.5
800	24.5	24.1	21.9	24.7	17.4	16.9
$LSD_{(T\leq0.05)}$ NS x NL	**11.3**					

Nitrogen source significantly influenced the fresh mass of leaves at 8 and 11 weeks after planting (Table 5.9). At 8 weeks after planting, the leaf fresh mass was significantly greater for plants that received ammonium nitrate (13.8 g) or potassium nitrate (13.7 g) as nitrogen source than plants that received urea ammonium nitrate (10.2 g). Leaf fresh mass of Swiss chard plants that received urea (15 g), ammonium nitrate (14.6 g) or potassium nitrate (14.7 g) as nitrogen source was significantly greater than plants that received urea ammonium nitrate (11.4 g) at 11 weeks after planting.

Table 5.9: Effect of nitrogen sources on the fresh mass (g) per leaf harvested of 'Fordhook Giant' from 8 up to 21 weeks after planting

Weeks after planting	Nitrogen source (NS)						$LSD_{(T \leq 0.05)}$ NS
	NH_4NO_3	KNO_3	$Ca(NO_3)_2$	$(NH_4)_2SO_4$	$CO(NH_2)_2.NH_4NO_3$	$CO(NH_2)_2$	
8	13.8	13.7	12.7	12.3	10.2	11.6	**2.7**
11	14.6	14.7	13.4	13.9	11.4	15.0	**3.2**
14	16.1	14.9	12.9	15.4	15.6	15.8	**ns**
17	15.2	15.1	12.6	15.2	15.8	15.7	**ns**
21	16.1	15.4	14.5	15.1	16.8	16.8	**ns**

Nitrogen level significantly influenced the fresh mass of Swiss chard leaves harvested at all five harvesting times irrespective of the nitrogen source (Table 5.10). At 8 and 14 weeks after planting, all plants that received nitrogen produced significantly heavier leaves than plants that did not receive any nitrogen. The greatest leaf fresh mass was recorded where 800 kg N ha^{-1} was applied at all the harvesting times.

Table 5.10: Effect of nitrogen levels on the fresh mass (g) per leaf harvested of 'Fordhook Giant' plants from 8 up to 21 weeks after planting

Weeks after planting	Nitrogen level (NL) (kg ha^{-1})									$LSD_{(T \leq 0.05)}$ NL
	0	100	200	300	400	500	600	700	800	
8	0.8	6.0	8.8	9.7	12.6	15.6	17.6	18.8	21.6	**3.6**
11	0.4	3.3	9.3	10.0	12.1	17.3	21.0	23.2	27.8	**4.2**
14	0.4	7.9	10.3	13.1	14.4	17.5	21.3	24.3	26.7	**5.4**
17	0.0	4.9	10.0	14.1	14.7	17.7	21.6	24.6	26.8	**5.3**
21	0.0	5.0	10.2	13.7	16.7	18.3	23.4	25.9	27.8	**5.5**

Nitrogen level significantly influenced the total fresh mass of harvested leaves for Swiss chard plants, irrespective of nitrogen source (Table 5.7). The total leaf fresh mass harvested per 'Fordhook Giant' plant increased from 1.7 g at 0 kg N ha^{-1} to 130.7 g at 800 kg N ha^{-1} (Table 5.11). Leaf fresh mass obtained earlier and discussed in Chapter 4, Section 4.2.3, corresponded with these results and the greatest leaf fresh mass was also obtained at the highest nitrogen level applied (400 kg ha^{-1}). Similar, results were obtained for spinach plants by Goh & Vityakon (1983), Elia *et al.* (1998) and Hammad *et al*. (2007). The nitrogen levels where the maximum spinach yield (expressed as fresh mass) was measured was between 300 and 450 kg N ha^{-1} (Goh & Vityakon, 1983). Higher nitrogen levels (600 kg ha^{-1}) significantly depressed the yield of spinach where nitrogen was applied as potassium nitrate or urea in a silt loam soil. These conflicting results may be atttributed to differences in soil type and the initial fertility of the soil used in the different pot trials. The fresh mass of Swiss chard did not show any decrease even at very high nitrogen levels (800 kg ha^{-1}). The same trend was observed for cauliflower where nitrogen increased fresh mass when 0-675 kg N ha^{-1} was applied but it decreased again when nitrogen was further increased to 900 kg ha^{-1} (Kaniszewski & Rumpel, 1998).

Table 5.11: Effect of nitrogen levels on the total leaf fresh mass (g) per plant harvested over a period of 21 weeks of 'Fordhook Giant' plants

Nitrogen level (NL) (kg ha^{-1})	**Leaf fresh mass (g)**
0	1.7
100	27.2
200	46.6
300	60.7
400	70.5
500	86.4
600	105.0
700	116.9
800	130.7
$LSD_{(T\leq 0.05)}$ NL	**18.4**

As shown in Table 5.7 nitrogen source did not significantly influence the total fresh mass of Swiss chard leaves harvested. It is only in the early stages of harvest (8 and 11 weeks after planting) where ammonium nitrate, potassium nitrate and urea significantly influenced the fresh mass of Swiss chard leaves, but this was not evident in the later stages of harvesting. The yield of butterhead lettuce was also not influenced by the different organic (castor-oil seed cake) and inorganic nitrogen sources (ammonium nitrate and Chilean nitrate soda) (Lairon *et al*., 1984). Goh & Vityakon (1983) on the other hand, reported that the yield of spinach plants expressed as top growth (g plant^{-1}) was significantly influenced by potassium nitrate compared to ammonium nitrate, ammonium sulphate+N-serve, urea and ammonium sulphate+N-serve+potassium nitrate used in a pot trial. The yield of shallot where ammonium nitrate or urea was used as nitrogen source was higher than where ammonium sulphate was used (Woldetsadik, Gertsson & Ascard, 2002). Both fennel and celery did not respond to different nitrogen sources concerning yield features (Santamaria *et al*., 1999a). Fresh mass of cauliflower leaves was significantly greater where calcium nitrate was used as a nitrogen source followed by ammonium sulphate, ammonium nitrate and then urea (Kaniszewski & Rumpel, 1998).

5.2.3 Leaf area

A summary of the analyses of variance that was done to determine the effects of different nitrogen sources and levels on the leaf area per leaf of ‘Fordhook Giant’ plants from 8 up to 21 weeks after planting is given in Table 5.12. Leaf area of Swiss chard in this section refers to the leaf area (cm^2) per harvested leaf of only the blades without the petiole.

Table 5.12: Summary of the analyses of variances showing the significant effects of nitrogen sources and nitrogen levels on the leaf area of harvested leaves of 'Fordhook Giant' plants from 8 up to 21 weeks after planting

Weeks after planting	Nitrogen source (NS)	Nitrogen level (NL)	NS × NL
8	ns	*	ns
11	*	*	*
14	ns	*	ns
17	ns	*	ns
21	*	*	*
Total leaf area $plant^{-1}$	*	*	*

LSD $_{(T \leq 0.05)}$
ns = no significant differences
*** = significant differences**

As shown in Table 5.12 the interaction between nitrogen source and nitrogen level significantly influenced the leaf area of 'Fordhook Giant' plants at 11 weeks after planting. At the same nitrogen level, leaf area of Swiss chard plants did not differ significantly amongst the different nitrogen sources (Table 5.13). Significantly larger leaf areas were measured from plants where ≥200 kg N ha^{-1} was applied as urea or urea ammonium nitrate, ≥300 kg N ha^{-1} as ammonium nitrate, ≥400 kg N ha^{-1} as potassium nitrate and ≥500 kg N ha^{-1} as ammonium sulphate than of plants that did not receive any nitrogen. Leaf area increased with increasing levels of nitrogen for all the nitrogen sources and the largest leaf area was measured at 800 kg N ha^{-1} for most nitrogen sources except potassium nitrate, ammonium sulphate and calcium nitrate. Leaf area of plants that received these three nitrogen sources tended to decrease when the nitrogen level increased from 700 to 800 kg ha^{-1}.

Table 5.13: Effect of nitrogen sources and levels on the leaf area (cm^2) per leaf harvested for 'Fordhook Giant' plants 11 weeks after planting

Nitrogen level (NL) ($kg\ ha^{-1}$)	Nitrogen source (NS)					
	$NH_4\ NO_3$	KNO_3	$Ca(NO_3)_2$	$(NH_4)_2\ SO_4$	$CO(NH_2)_2.NH_4NO_3$	$CO\ (NH_2)_2$
0	23	0	0	0	0	0
100	34	77	0	110	0	141
200	144	218	114	121	245	255
300	261	133	195	198	65	266
400	133	268	203	136	273	250
500	278	202	230	266	288	281
600	287	307	287	253	277	288
700	287	398	403	392	273	401
800	402	381	332	380	396	410
$LSD_{(T\leq0.05)}$ NS x NL	**221**					

The interaction between nitrogen sources and nitrogen levels significantly influenced the leaf area of leaves harvested of 'Fordhook Giant' plants at 21 weeks after planting (Table 5.14). At this late growing stage (21 weeks after planting) the same trends were observed as were earlier (11 weeks after planting). Nitrogen sources did not significantly influence the leaf area at the same nitrogen level. It was only at different nitrogen levels at which the nitrogen sources started to influence the leaf area significantly, that differed. Furthermore, leaf area increased with increasing nitrogen levels and the largest leaf area was measured where 800 kg N ha^{-1} was applied for most nitrogen sources except for potassium nitrate and ammonium sulphate that tended to decrease when nitrogen increased from 700 to 800 kg ha^{-1}.

Table 5.14: Effect of nitrogen sources and levels on the leaf area (cm^2) per leaf harvested for 'Fordhook Giant' plants 21 weeks after planting

Nitrogen levels (NL) ($kg\ ha^{-1}$)	Nitrogen source (NS)					
	$NH_4\ NO_3$	KNO_3	$Ca(NO_3)_2$	$(NH_4)_2\ SO_4$	$CO(NH_2)_2.NH_4NO_3$	$CO\ (NH_2)_2$
0	0	0	0	0	0	0
100	0	34	3	164	121	149
200	99	226	240	60	253	264
300	271	201	202	206	270	276
400	276	277	280	209	283	260
500	286	211	239	211	281	289
600	295	314	320	257	300	297
700	364	407	314	409	379	411
800	411	293	344	394	410	420
$LSD_{(T\leq0.05)}$ NS X NL	**207**					

Nitrogen source significantly influenced the leaf area at 11 and 21 weeks after planting as shown in Table 5.15. At 11 weeks after planting, significantly larger leaf areas were measured where urea (254 cm^2) was applied as nitrogen source compared to where calcium nitrate (196 cm^2) or urea ammonium nitrate (193 cm^2) was applied as nitrogen source. There was however, no significant difference amongst the other nitrogen sources. Significantly larger leaf areas were measured for plants that received urea (263 cm^2) compared to ammonium sulphate (212 cm^2) at 21 weeks after planting. Although not significant, larger leaf areas were also obtained where urea was applied as nitrogen source, compared to the other nitrogen sources at 8, 14 and 17 weeks after planting.

Table 5.15: Effect of nitrogen sources on the leaf area (cm^2) per leaf harvested of 'Fordhook Giant' from 8 up to 21 weeks after planting

Weeks after planting	Nitrogen source (NS)						$LSD_{(T \leq 0.05)}$ NS
	NH_4NO_3	KNO_3	$Ca(NO_3)_2$	$(NH_4)_2SO_4$	$CO(NH_2)_2 . NH_4NO_3$	$CO(NH_2)_2$	
8	219	216	217	213	212	246	**ns**
11	213	220	196	206	193	254	**52**
14	224	193	194	213	223	242	**ns**
17	218	211	189	212	230	244	**ns**
21	222	218	215	212	255	263	**49**

As shown in Table 5.16 the leaf area measured increased with increasing levels of nitrogen at all five harvesting times. At 8, 14 and 21 weeks after planting, all the plants that received nitrogen produced significantly larger leaf areas than plants that did not receive any nitrogen. At 11 and 17 weeks after planting the same trend was observed except that it was only from 200 kg N ha^{-1} that leaf area was significantly larger than that of plants that received no or 100 kg N ha^{-1}. The largest leaf area was recorded where 800 kg N ha^{-1} was applied at 8, 11 and 14 weeks after planting. At 17 and 21 weeks after planting, leaf area tended to decrease when nitrogen levels increased from 700 to 800 kg ha^{-1}.

Table 5.16: Effect of nitrogen levels on the leaf area (cm^2) per leaf harvested of 'Fordhook Giant' from 8 up to 21 weeks after planting

Weeks after planting	Nitrogen level (NL) (kg ha^{-1})									$LSD_{(T\leq0.05)}$ NL
	0	**100**	**200**	**300**	**400**	**500**	**600**	**700**	**800**	
8	16	102	175	208	243	235	270	320	415	**70**
11	4	61	183	186	210	244	283	371	384	**69**
14	4	103	169	225	228	250	272	324	356	**77**
17	0	71	162	244	225	251	275	366	360	**76**
21	0	78	190	237	264	253	297	381	379	**65**

The total leaf area per plant was significantly influenced by the interaction between nitrogen source and nitrogen level over the 21 week period (Table 5.12). As shown in Table 5.17 the total leaf area of 'Fordhook Giant' plants increased with increasing nitrogen level for most of the nitrogen sources. The total leaf area for 'Fordhook Giant' plants where urea was supplied increased from 68 to 2976 cm^2 when the nitrogen level increased from 0 to 800 kg ha^{-1}. The same trend was observed for the other nitrogen sources except for potassium nitrate which showed a decrease when nitrogen increased from 700 kg ha^{-1} (1810 cm^2) to 800 kg ha^{-1} (1661 cm^2). At the same nitrogen level the different nitrogen sources did not significantly influence the total leaf area, except at 800 kg ha^{-1} with urea as nitrogen source where the leaf area (2976 cm^2) was significantly larger than plants that received nitrogen as calcium nitrate (1659 cm^2). The nitrogen level at which the nitrogen sources started to influence the total leaf area of Swiss chard plants also differed amongst the sources. Ammonium sulphate significantly influenced the leaf area already at 100 kg N ha^{-1}. Potassium nitrate, calcium nitrate, urea ammonium nitrate and urea started influencing the leaf area significantly at 200 kg N ha^{-1} and ammonium nitrate at 300 kg N ha^{-1}.

Table 5.17: Effect of nitrogen sources and levels on the total leaf area (cm^2) per 'Fordhook Giant' plant

Nitrogen level (NL) (kg ha^{-1})	**Nitrogen source (NS)**					
	NH_4 NO_3	**KNO_3**	**$Ca(NO_3)_2$**	**$(NH_4)_2$ SO_4**	**$CO(NH_2)_2$. NH_4NO_3**	**CO $(NH_2)_2$**
0	45	0	0	0	31	68
100	126	352	47	804	358	802
200	653	1034	865	416	1197	1094
300	1322	854	1039	1128	991	1284
400	1165	1363	1170	894	1114	1326
500	1399	962	1265	1169	1269	1346
600	1379	1505	1468	1327	1434	1318
700	1777	1810	1609	1819	1618	1953
800	1998	1661	1659	1960	2023	2976
$LSD_{(T\leq0.05)}$ NS X NL	**786**					

Different nitrogen sources did not have a consistent influence on the leaf area of Swiss chard plants. In general, there was no significant trend and it was only at 800 kg N ha^{-1} where urea did influence the leaf area significantly, compared to calcium nitrate, while no significant differences existed amongst the other nitrogen sources.

Again, the nitrogen level influenced the leaf area significantly, which is in agreement with earlier results discussed in Chapter 4, Section 4.2.4 where the highest nitrogen level produced leaves with the largest leaf area. Gülser (2005) also reported an increase in leaf area of spinach with an increase in nitrogen level. At nitrogen levels as low as 150 kg ha^{-1} leaf area was significantly influenced where ammonium sulphate and urea were used as nitrogen source and this corresponded with the results of this study where significant reaction was obtained for Swiss chard plants at 100 kg N ha^{-1} for these two nitrogen sources. Nitrogen applied as ammonium sulphate and urea also significantly influenced leaf area of spinach plants (Gülser, 2005). Leaf area of spinach plants that received urea as nitrogen source was significantly larger than the leaf area of plants that received ammonium sulphate. Although not significant, the same trend was observed in this study for most nitrogen levels.

5.2.4 Leaf dry mass

A summary of the analyses of variance that was done to determine the effects of different nitrogen sources and levels on the dry mass of 'Fordhook Giant' leaves from 8 up to 21 weeks after planting is given in Table 5.18. Leaf dry mass in this section refers to the dry mass of the leaf blade without the petiole.

Table 5.18: Summary of the analyses of variances showing the significant effects of nitrogen sources and nitrogen levels on leaf dry mass 'Fordhook Giant' plants from 8 up to 21 weeks after planting

Weeks after planting	Nitrogen source (NS)	Nitrogen level (NL)	NS × NL
8	ns	*	*
11	*	*	*
14	ns	*	ns
17	*	*	*
21	ns	*	ns
Total leaf dry mass plant^{-1}	*	*	ns

LSD $_{(T \leq 0.05)}$
ns = no significant differences
*** = significant differences**

The interaction between nitrogen source and nitrogen level significantly influenced the leaf dry mass at weeks 8, 11 and 17 after planting. It is clear from Tables 5.19, 5.20 and 5.21 that nitrogen increased the dry mass of Swiss chard leaves harvested irrespective of the nitrogen source. The greatest dry mass for Swiss chard was obtained where 800 kg N ha^{-1} was applied and there was no evidence of a decrease in dry mass even at very high nitrogen levels (800 kg ha^{-1}).

Table 5.19: Effect of nitrogen sources and nitrogen levels on the dry mass (g) of harvested leaves of 'Fordhook Giant' plants 8 weeks after planting

Nitrogen level (NL) (kg ha^{-1})	Nitrogen source (NS)					
	NH_4 NO_3	KNO_3	$Ca(NO_3)_2$	$(NH_4)_2$ SO_4	$CO(NH_2)_2$. NH_4NO_3	CO $(NH_2)_2$
0	0.0	0.0	0.0	0.0	0.2	0.7
100	0.6	1.0	0.0	1.0	0.3	1.0
200	1.8	1.3	1.3	0.5	1.3	0.8
300	2.0	1.0	1.8	1.0	0.5	1.5
400	2.0	3.0	2.3	1.5	2.5	2.5
500	3.8	2.8	3.3	3.8	2.3	3.5
600	4.3	4.5	4.8	4.0	4.8	4.3
700	5.3	4.5	4.8	5.0	5.8	5.3
800	6.3	7.0	6.5	7.0	7.5	8.0
$LSD_{(T\leq0.05)}$ NS x NL	**1.9**					

Nitrogen source significantly influenced the dry mass of harvested leaves only at 11 and 17 weeks after planting (Table 5.20 and 5.21). At 11 weeks after planting the dry mass of harvested leaves was significantly higher where nitrogen at 800 kg ha^{-1} was applied as potassium nitrate (8.3 g) than where urea ammonium nitrate (6.0 g) was. At 17 weeks after planting, leaf dry mass of plants that received ammonium sulphate (7.5 g) was significantly higher than where potassium nitrate (5.0 g) or calcium nitrate (5.0 g) was applied.

Table 5.20: Effect of nitrogen sources and nitrogen levels on the dry mass (g) of harvested leaves of 'Fordhook Giant' plants 11 weeks after planting

Nitrogen level (NL) ($kg\ ha^{-1}$)	Nitrogen source (NS)					
	$NH_4\ NO_3$	KNO_3	$Ca(NO_3)_2$	$(NH_4)_2\ SO_4$	$CO(NH_2)_2.NH_4NO_3$	$CO\ (NH_2)_2$
0	0.3	0.0	0.0	0.0	0.0	0.0
100	0.3	0.5	4.9	0.5	4.8	0.3
200	1.3	1.5	1.3	0.8	1.3	1.8
300	2.0	0.3	1.0	1.3	0.3	1.5
400	1.0	3.0	1.8	1.3	1.3	2.0
500	2.3	2.0	2.3	2.8	2.0	4.0
600	4.0	3.5	3.3	4.5	4.5	4.5
700	4.8	4.8	4.5	4.5	4.5	5.0
800	7.0	8.3	7.5	6.8	6.0	7.0
$LSD_{(T\leq0.05)}$ NS x NL	**2.1**					

The nitrogen level at which the different nitrogen sources started to significantly influence the leaf dry mass was not consistent at 8, 11 and 17 weeks after planting. Ammonium sulphate was the only source that started to influence the dry mass significantly from 500 kg N ha^{-1} at all three mentioned harvesting times. Ammonium nitrate influenced the dry mass significantly from 300 kg N ha^{-1} at 8 weeks after planting and from 600 kg N ha^{-1} at 11 and 17 weeks after planting. Potassium nitrate, calcium nitrate, urea and urea ammonium nitrate all started to influence the dry mass significantly at 400 kg N ha^{-1} at 8 weeks after planting (Table 5.19) and at 11 and 17 weeks after planting the nitrogen level had increased to between 500 and 600 kg ha^{-1} (Tables 5.20 and 5.21).

Table 5.21: Effect of nitrogen sources and nitrogen levels on the dry mass (g) of harvested leaves of 'Fordhook Giant' plants 17 weeks after planting

Nitrogen level (NL) ($kg\ ha^{-1}$)	Nitrogen source (NS)					
	$NH_4\ NO_3$	KNO_3	$Ca(NO_3)_2$	$(NH_4)_2\ SO_4$	$CO(NH_2)_2.NH_4NO_3$	$CO\ (NH_2)_2$
0	0.0	0.0	0.0	0.0	0.0	0.0
100	0.0	0.3	0.0	0.8	0.5	0.8
200	0.5	2.0	1.3	0.5	1.8	1.0
300	2.0	1.5	0.8	2.0	1.3	1.0
400	1.8	3.0	1.5	2.3	1.3	3.0
500	2.0	1.5	3.0	3.0	3.5	3.5
600	4.3	5.0	4.0	5.0	4.8	4.8
700	5.8	4.5	4.5	4.8	4.8	5.0
800	6.3	5.0	5.0	7.5	6.5	5.8
$LSD_{(T\leq0.05)}$ NS X NL	**2.4**					

As shown in Table 5.22, nitrogen source only influenced the dry mass of 'Fordhook Giant' leaves harvested at 11 and 17 weeks after planting. At 11 weeks after planting, a significantly greater dry mass was measured for 'Fordhook Giant' leaves that received potassium nitrate or urea than plants that received urea ammonium nitrate. The dry mass of Swiss chard leaves that received ammonium sulphate or urea as nitrogen source was significantly greater than leaves from plants that received calcium nitrate (17 weeks after planting). Although not significant, the greatest leaf dry mass was obtained where urea was applied as nitrogen source for most harvesting times except at 17 weeks after planting where the greatest dry mass was obtained from leaves that received ammonium sulphate.

Table 5.22: Effect of nitrogen sources on the dry mass (g) per leaf harvested of 'Fordhook Giant' plants from 8 up to 21 weeks after planting

Weeks after planting	Nitrogen source (NS)						$LSD_{(T \leq 0.05)}$ NS
	$NH_4 NO_3$	KNO_3	$Ca(NO_3)_2$	$(NH_4)_2 SO_4$	$CO(NH_2)_2 . NH_4NO_3$	$CO (NH_2)_2$	
8	2.8	2.7	2.7	2.6	2.7	3.8	**ns**
11	2.6	2.7	2.4	2.5	2.2	2.9	**0.5**
14	2.5	2.2	2.2	2.4	2.6	2.7	**ns**
17	2.5	2.5	2.2	2.9	2.5	2.8	**0.6**
21	2.6	2.6	2.5	2.6	2.8	2.9	**ns**

As shown in Table 5.23, the leaf dry mass increased with increasing nitrogen level at all five harvesting times irrespective of the nitrogen source. At all harvesting times the dry mass of plants that received 200 kg N ha^{-1} or more was significantly higher than plants that did not receive any nitrogen. The greatest dry mass at all five harvesting times was measured where 800 kg N ha^{-1} was applied.

Table 5.23: Effect of nitrogen levels on the dry mass (g) of harvested leaves of 'Fordhook Giant' from 8 up to 21 weeks after planting

Weeks after planting	Nitrogen level (NL) (kg ha^{-1})									$LSD_{(T \leq 0.05)}$ NL
	0	**100**	**200**	**300**	**400**	**500**	**600**	**700**	**800**	
8	0.1	0.6	1.1	1.3	2.3	3.2	4.4	5.0	7.0	**0.6**
11	0.0	0.3	1.1	1.3	1.7	2.6	4.0	4.7	7.0	**0.7**
14	0.0	0.6	1.2	1.5	2.5	2.9	3.7	4.4	5.3	**0.8**
17	0.0	0.4	1.2	1.4	2.1	2.8	4.6	4.9	6.0	**0.8**
21	0.0	0.6	1.4	1.7	2.3	2.4	3.9	5.4	6.4	**1.0**

Nitrogen source significantly influenced the total dry mass of harvested leaves of Swiss chard plants (Table 5.18). The total dry mass of leaves harvested where urea (14.4 g) was applied was significantly greater than where calcium nitrate (11.9 g) was applied (Table 5.24). No significant differences existsed amogst the other nitrogen sources.

Table 5.24: Effect of nitrogen sources on the total leaf dry mass (g) per plant harvested of 'Fordhook Giant' plants over a period of 21 weeks

Nitrogen source (NS)	Leaf dry mass (g)
NH_4 NO_3	13.1
KNO_3	12.8
$Ca(NO_3)_2$	11.9
$(NH_4)_2$ SO_4	13.1
$CO(NH_2)_2$. NH_4NO_3	13.0
CO $(NH_2)_2$	14.4
$LSD_{(T\leq 0.05)}$ NS	**2.0**

Nitrogen level significantly influenced the total dry mass of harvested leaves of 'Fordhook Giant' plants (Table 5.18). The total dry mass of leaves harvested increased from 0.2 g at 0 kg N ha^{-1} to 31.8 g at 800 kg N ha^{-1} (Table 5.25). Plants that received nitrogen at 200 kg N ha^{-1} and more, produced leaves with a significantly greater total dry mass than plants that did not receive any nitrogen or 100 kg N ha^{-1}. Again high nitrogen levels did not negatively influence the total dry mass of Swiss chard leaves.

Table 5.25: Effect of nitrogen levels on the total leaf dry mass (g) per plant harvested of 'Fordhook Giant' plants over a period of 21 weeks

Nitrogen level (NL) (kg ha^{-1})	Leaf dry mass (g)
0	0.2
100	2.6
200	6.2
300	7.1
400	10.8
500	13.8
600	20.8
700	24.3
800	31.8
$LSD_{(T\leq 0.05)}$ NL	**2.6**

Nitrogen source influenced the dry mass of harvested leaves of 'Fordhook Giant' plants only at 11 and 17 weeks after planting. Increased nitrogen levels significantly increased the dry mass of 'Fordhook Giant' leaves. These results are similar to the earlier results discussed in Chapter 4, Section 4.2.5. In both pot trials the highest nitrogen level applied (400 or 800 kg ha^{-1}) resulted in the greatest dry mass of leaves. These results emphasise the essential role of nitrogen in plant growth and quality. Hammad *et al*. (2007) also studied the influence of ammonium nitrate applied at four levels (0, 40, 60 and 80 kg N ha^{-1}), and also different ratios of ammonium and nitrate combined together on spinach. The author stated that nitrogen source and nitrogen level influenced dry mass of spinach leaves. Dry mass of lettuce was also influenced by nitrogen level and nitrogen source. The greatest dry mass for lettuce was obtained where ammonium sulphate was applied as nitrogen source followed by calcium nitrate and then by urea (Abu-Rayyan, Kharawish & Al-Ismail, 2004).

5.2.5 Leaf nitrate content

A summary of the analyses of variance that was done to determine the effects of different nitrogen sources and levels on the nitrate content (mg kg^{-1}) of 'Fordhook Giant' leaf blades based on dry mass (DM) is given in Table 5.26.

Table 5.26: Summary of the analyses of variances showing the significant effects of nitrogen sources and nitrogen levels on the nitrate content (mg kg^{-1} DM) of harvested leaves of 'Fordhook Giant'

Nutrient	Nitrogen source (NS)	Nitrogen level (NL)	NS x NL
Nitrate (NO_3)	*	*	*

LSD $_{(T \leq 0.05)}$
ns = no significant differences
*** = significant differences**

As shown in Table 5.26, the interaction between nitrogen source and nitrogen level significantly influenced the nitrate content of Swiss chard leaves. Ammonium nitrate and potassium nitrate were the only two nitrogen sources that significantly influenced the nitrate content of the leaves (Table 5.27 and Table 5.28). The nitrate content of leaves that received ammonium nitrate increased from 2.0 mg kg^{-1} where no nitrogen was applied to 783.3 mg kg^{-1} where 800 kg N ha^{-1} was applied. Where potassium nitrate was applied the nitrate increased from 2.0 mg kg^{-1} to 613.3 mg kg^{-1} at the same nitrogen level.

Interestingly, the nitrate content of plants that received calcium nitrate increased only from 1.3 mg kg^{-1} where no nitrogen was applied to 8.0 mg kg^{-1} where 800 kg N ha^{-1} was applied. Furthermore, no significant differences occurred where nitrogen was applied as ammonium sulphate, urea ammonium nitrate or urea, irrespective of the nitrogen level. Although not significant, the nitrate content of plants that received ammonium sulphate, urea ammonium nitrate or urea as nitrogen source tended to increase with an increase in nitrogen but started to decrease if the nitrogen level was increased further. The highest nitrate content for ammonium sulphate was observed at 500 kg N ha^{-1}, urea ammonium nitrate at 700 kg N ha^{-1} and urea at 300 kg N ha^{-1}. Nitrate content of leaves significantly increased when ammonium nitrate or potassium nitrate was applied at 500 kg N ha^{-1} compared to plants that did not receive any nitrogen.

Table 5.27: Effect of nitrogen sources and nitrogen levels on the nitrate content (mg kg^{-1} DM) of the leaf blades of 'Fordhook Giant' plants

Nitrogen level (NL) (kg ha^{-1})	Nitrogen source (NS) (mg kg^{-1})					
	NH_4NO_3	KNO_3	$Ca(NO_3)_2$	$(NH_4)_2SO_4$	$CO(NH_2)_2.NH_4NO_3$	$CO(NH_2)_2$
0	2.0	2.0	1.3	2.8	1.3	2.8
100	88.5	95.0	4.3	5.8	3.3	2.6
200	163.9	116.2	3.6	5.3	3.3	2.0
300	172.3	181.0	3.4	4.3	2.4	6.0
400	276.1	114.8	3.1	2.5	3.7	2.8
500	364.8	497.5	2.7	6.3	2.4	3.4
600	306.4	516.9	6.1	3.7	2.7	3.1
700	395.3	425.5	7.5	2.3	5.3	3.1
800	783.3	613.3	8.0	2.8	2.3	2.8
$LSD_{(T\leq0.05)}$NS x NL	**280.8**					

Different nitrogen sources significantly influenced the accumulation of nitrate in spinach plants especially when the nitrate content in the nitrogen source increased (Hammad *et al*., 2007). Results obtained by Vulsteke & Biston (1978) showed no significant difference in the nitrate content of canned celery between different nitrogen sources (ammonium nitrate, calcium nitrate and sodium nitrate) which do not correspond with data of this study on Swiss chard where ammonium nitrate and calcium nitrate significantly influences the nitrate content compare with the other nitrogen sources. Venter & Fritz (1979) interestingly, also found no differences in the nitrate content of kohlrabi tubers that received calcium nitrate or ammonium sulphate as nitrogen source which correspond with our results. The nitrate content of lettuce was found to be the highest where calcium nitrate was used as a nitrogen source followed by urea and then ammonium sulphate (Abu-Rayyan *et al*., 2004). The nitrate content of cauliflower curds was also significantly lower where urea was applied as nitrogen source compared to ammonium sulphate, ammonium nitrate and calcium nitrate (Kaniszewski & Rumpel, 1998).

Table 5.28: Effect of nitrogen sources on the nitrate content (mg kg^{-1} DM) of the leaf blades of 'Fordhook Giant' plants

Nitrogen source (NS)	**Nitrate content (mg kg^{-1} DM)**
NH_4NO_3	283.6
KNO_3	284.7
$Ca(NO_3)_2$	4.4
$(NH_4)_2SO_4$	4.0
$CO(NH_2)_2.NH_4NO_3$	3.0
$CO(NH_2)_2$	3.2
$LSD_{(T\leq0.05)}$ NS	**65.4**

The nitrate content of Swiss chard leaves increased with increasing level of nitrogen (Table 5.29). Schuphan *et al.* (1967); Cantliffe (1973a), Voghtmann *et al.* (1984), Elia *et al*. (1998) and Hammad *et al*. (2007) all reported an increase in the nitrate content with increase in nitrogen levels for spinach leaves and so did Vulsteke & Biston (1978) for the pods of snap beans and canned celery. This was also true for the nitrate content of lettuce (Lairon *et al*., 1984), cabbage (Turan & Sevimli, 2005) and turnip greens (Vieira *et al*., 1998). Nádasy (2002) reported that ammonium nitrate and calcium nitrate and to a lesser extent ammonium sulphate increased the nitrate content of the leafy stems of green peas at maturity. Turan & Sevimli (2005) reported that potassium nitrate and ammonium nitrate increased the nitrate content of cabbage much more than ammonium sulphate and urea which correspond with the results of this study.

Table 5.29: Effect of nitrogen levels on the nitrate content (mg kg^{-1} DM) of the leaf blades of 'Fordhook Giant' plants

Nitrogen level (NL) (kg ha^{-1})	Nitrate content (mg kg^{-1} DM)
0	2.0
100	33.3
200	49.0
300	61.6
400	67.2
500	146.2
600	139.8
700	139.8
800	235.4
$LSD_{(T\leq0.05)}$ NL	**87.4**

5.3 CONCLUSIONS

Nitrogen fertiliser did have a distinct influence on the yield and quality of 'Fordhook Giant' plants. Higher nitrogen levels increased the number of leaves harvested, leaf fresh and dry mass, leaf area and the nitrate content of the leave. Surprisingly, all these parameters increased with increasing nitrogen levels and there were no signs of a decrease in any of these parameters at the high nitrogen levels used in this trial. For all the measured parameters, 800 kg N ha^{-1} gave the best results for Swiss chard plants.

The different nitrogen sources used did not differ significantly from one another at the same nitrogen level. It was in most cases only the nitrogen level where where the different sources started to significantly influence the leaf area and the total number of leaves harvested that differed. Although not significant, the most number of leaves per plant was harvested where ammonium nitrate was used as nitrogen source followed by ammonium sulphate, urea ammonium nitrate, and urea, whereas potassium nitrate and calcium nitrate produced the least number of leaves per plant.

Leaf fresh mass was not significantly influenced by the nitrogen source but the highest leaf mass was measured where ammonium nitrate was applied as nitrogen source followed by urea, potassium nitrate, ammonium sulphate, urea ammonium nitrate and lastly calcium nitrate. It was only the dry mass of leaves that received urea as nitrogen source that was significantly greater than leaves that received calcium nitrate.

The greatest total leaf area per plant was measured where urea was applied as nitrogen source followed by urea ammonium nitrate, ammonium nitrate, ammonium sulphate, potassium nitrate and then finally the least leaf area was measured for calcium nitrate.

Different nitrogen sources significantly influenced the nitrate accumulation in Swiss chard leaves where ammonium nitrate or potassium nitrate was applied. Urea, urea ammonium nitrate, ammonium sulfate, potassium nitrate and calcium nitrate did not stimulate the accumulation of nitrate in the leaves of Swiss chard.

CHAPTER 6

SUMMARY AND RECOMMENDATIONS

6.1 SUMMARY

Swiss chard is known as a vegetable with high nutritional value and one of the most underestimated vegetables that could play a much more important role in the diets of both people in developed countries, as well as the poor in developing countries in Africa. Swiss chard is grown for its bright green leaves and is known as a very nutritional crop.

Leafy vegetables are known to react positively to fertiliser, especially nitrogen. Nitrogen plays an essential role in the green colour and vegetative growth of crops. For this reason growers tend to apply much more nitrogen than the optimum recommended for ensuring high yields. Excess nitrogen does not only influence the environment negatively but also impacts on the quality of Swiss chard because it stimulates the accumulation of nitrate in the plants. The quality of Swiss chard is not only associated with the fresh mass of the leaves leaf size (leaf area) and dry mass, but is also influenced by the nitrate content of the leaves. High nitrate levels in Swiss chard leaves are hazardous to the health of humans. The quality of Swiss chard can be improved by avoiding the petioles of the leaves and using only the blades because the petioles accumulate more nitrate than the blades.

The management of nitrogen fertilisation for sustainable vegetable production is very important. This does not only include the optimum nitrogen level for high yields of good quality, but also the choice of nitrogen source that may influence the quality of the produce. The timing of nitrogen application is also an important part of fertilisation management which allows for the availability of fertiliser when the crop needs it and prevents the unnecessary losses to the environment, influencing the quality of ground water negatively. Cultivars may also react differently to fertilisation and need to be taken into account when managing fertilisation.

The objective of Swiss chard production should thus be to attain high yields of a good quality by applying optimum nitrogen and not in excess, to prevent the accumulation of nitrates in the plant as well as not to influence the environment negatively. The objective therefore of this study was to quantify the effect of nitrogen fertilisation on the growth, yield and quality of Swiss chard. In order to achieve this, two pot trials were conducted in the greenhouse.

First pot trial

This trial was conducted to determine the response of two Swiss chard cultivars namely: 'Fordhook Giant' and 'Rhubarb' to five different nitrogen levels (0, 50, 100, 200 and 400 kg ha^{-1}) applied at four different application times (T_1, T_2, T_3 and T_4).

The two Swiss chard cultivars did not differ in their response to nitrogen level and application time during the first six weeks after planting. Total number of leaves harvested per plant, total leaf fresh mass per plant and also the total leaf area per plant did not differ between the two cultivars. It was only the total dry mass per plant and total nitrogen content per leaf of 'Rhubarb' plants that was greater than that of 'Fordhook Giant'. Differences observed between the two cultivars can be attributed to the longer growth period of 'Rhubarb' compared with that of 'Fordhook Giant'.

Nitrogen levels significantly influenced the early growth, number of leaves harvested, leaf area, fresh and dry mass of harvested leaves and also the nitrogen content of Swiss chard leaves. Higher nitrogen levels positively influenced all the yield and quality parameters measured.

Interestingly, nitrogen application times significantly influenced only the early growth of Swiss chard plants and the nitrogen content of leaves . Early plant growth reacted better where nitrogen was split into fewer applications (T_4). The number of leaves, leaf fresh and dry mass and leaf area in the later stages of harvest (13 and 17 weeks after planting) seem to react better to nitrogen that was split into 11 equal applications (T_1). The total nitrogen content of Swiss chard leaves was significantly higher where nitrogen was split up into five equal (T_2) or three equal (T_4) applications.

Second pot trial

This trial was conducted to determine the response of Swiss chard cultivar 'Fordhook Giant' to six different nitrogen sources (ammonium nitrate, calcium nitrate, potassium nitrate, ammonium sulphate, urea ammonium nitrate and urea) applied at nine different nitrogen levels (0, 100, 200, 300, 400, 500, 600, 700 and 800 kg ha^{-1}).

Nitrogen fertiliser positively influenced the number of leaves harvested, leaf fresh and dry mass, leaf area and nitrate accumulations positively. All parameters measured increased with increasing nitrogen level and no data indicated any negative influence on any one of the parameters measured, even under the very high nitrogen levels applied in this pot trial.

Nitrogen source did not influence either the total number of leaves harvested nor the fresh mass of harvested Swiss chard leaves. In both cases, ammonium nitrate gave the best results and calcium nitrate the poorest. Urea influenced the leaf area positively followed by urea ammonium nitrate, with calcium nitrate resulting in the smallest leaf area per plant. Dry mass was also significantly greater where urea was used as nitrogen source compared to where calcium nitrate was used. No significant differences occurred amongst the other nitrogen sources.

The nitrate concentration of leaves was significantly influenced by the nitrogen source. Ammonium nitrate and potassium nitrate stimulated the accumulation of nitrate in Swiss chard leaves, whereas the other nitrogen sources did not play any role in nitrate accumulation in the blades of Swiss chard leaves.

6.2 RECOMMENDATIONS

Based on results of these two pot trials the following recommendations can be made with respect to nitrogen fertilisation of Swiss chard plants:

- Characteristics of Swiss chard cultivars such as the length of the growth period should be taken in to account when nitrogen application decisions are made.

- Considering the different responses of the different parameters measured for nitrogen fertilisation, it seems that the best results are obtained at nitrogen levels between 700 and 800 kg ha^{-1} taking environmental and economic factors into account.

- The best response to nitrogen fertilisation was obtained where nitrogen was split into three or five equal applications throughout the growing season of Swiss chard.

- In an attempt to improve the quality of Swiss chard leaves and also taking into consideration the response of different parameters to different nitrogen sources, it is recommended that using ammonium nitrate or potassium nitrate as nitrogen source should be avoided and that instead urea, ammonium sulphate or urea ammonium nitrate be used.

- Yield parameters did not respond well to calcium nitrate and it is not recommended for use on Swiss chard.

In retrospect, only a few aspects of the fertilisation of Swiss chard were addressed in this study and the following warrants further investigation:

- The response of Swiss chard to even higher nitrogen levels to obtain a proper crop response curve to establish an optimum nitrogen level.

- The response of Swiss chard to different ratios of ammonium to nitrate applications.
- The effect of the interaction between nitrogen and phosphorus, as well as nitrogen and potassium on Swiss chard growth.

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