EFFECT OF NITROGEN RATES AND SOURCES ON PLANT GROWTH, YIELD, QUALITY AND NITROGEN USE EFFICIENCY ON AFRICAN NIGHTSHADE VARIETIES (*Solanum spp*) IN KENYA

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Effect of Nitrogen Rates and Sources on Plant Growth, Yield, Quality and Nitrogen Use Efficiency on African Nightshade Varieties (*Solanum spp*) in Kenya

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DECLARATION

This thesis is my original work and has not been presented for a degree in any other University

Signature……………………………………………….Date……………………………

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DEDICATION

This thesis is dedicated to my daughter Ayanna and my late mother Pricilla Kerubo.

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ACYRONYMS AND ABBREVIATIONS

NUE	Nitrogen Use Efficiency
CAN	Calcium Ammonium Nitrate
AOAC	Association of Official Analytical Chemistry
Ds	Deci Siemens
EC	Electrical Conductivity
CEC	Cation Exchange Capacity
pH	Potential of Hydrogen

ABSTRACT

The increasing demand for African nightshade (*Solanum spp*) as a nutritious leafy vegetable in Kenya underscores the need for optimizing its agronomic practices to enhance production sustainability. Nitrogen, a critical nutrient, significantly influences plant growth, yield, and quality of African nightshade. However, the efficiency of nitrogen use and its impact vary depending on its rate and source. The objective of this study was to determine the effects of nitrogen rates and sources on growth, yield, vegetable quality and nitrogen use efficiency of African Nightshade (*Solanum villosum* and *Solanum Scabrum)* species. Understanding these dynamics will inform best practices for nitrogen management, thereby contributing to improved crop productivity and sustainable agricultural development. Two pot experiments were conducted in a green house at Jomo Kenyatta University of Agriculture and Technology between December 2016 and November 2017. The experiments were laid out in a complete randomized design with four treatments replicated four times.The first experiment was on the effects of Nitrogen rates on African nightshade growth ,yield and quality.The treatments included 0g, 0.9g, 1.8g and 3.6g of CAN fertilizer per plant. Data collection was on plant height, number of leaves, fresh leaf weight, protein content, nitrate content and nitrogen use efficiency of *Solanum villosum* and *Solanum Scabrum* species. Protein and nitrate content were determined at vegetative and reproductive stages using semi-micro kjeldal and colorimetric methods respectively. Statistical analysis on the data indicated that nitrogen fertilizer application rates had a significant effect on plant growth, yield, quality and NUE at $P \leq 0.05$ level of significance. At the vegetative stage plants supplied with 3.6 g per plant of CAN had a higher protein and nitrate content at 28.4% and 2.6 mg/kg in *Solanum villosum* and 29.3% and 2.3mg/kg in *Solanum scabrum* species. At the reproductive stage Statistical analysis on proteins and nitrates indicated that nitrogen significantly affected nitrate content at $P \leq 0.05$. NUE efficiency was calculated according to Moll et al 1982, statistical analysis on the data determined that plants in the control group were the most nutrient efficient at $P \leq 0.05$ followed closely by plants supplied with 0.9g of CAN. In the second experiment different nitrogen sources was seen to affect plant growth yield, quality and NUE of African nightshade. The treatments included 0g, 2.11g(Urea),3.6g(CAN) and 36.4g (organic fertilizer) per plant. Experimental layout and data collection parameters were similar to the first experiment. Statistical analysis conducted on the data indicated that nitrogen sources significantly affected plant growth, yield, quality and NUE of African Nightshade at $P \leq 0.05$ level of significance. There was a significant difference in NUE at $P \leq 0.05$ with plants in the control group and those supplied with organic fertilizer having a higher NUE. Based on results of this study it is recommended that farmers use nitrogen rate of 3.6 g of (CAN) per plant which is equivalent to 400kg of CAN per HA to improve the growth and yield of African nightshade. While increasing nitrogen rates boosts vegetable yield, it is crucial to strike a balance between yield and quality. In order to increase yields in a more sustainable way farmers should implement a split application strategy for nitrogen fertilizer and incorporate organic sources so as to improve NUE, reduce costs and vegetable maintain quality.

Keywords: Productivity, quality, nitrogen use efficiency, nitrates, protein.

CHAPTER ONE

INTRODUCTION

1.1 Background of the Study

Vegetable production plays a crucial role in global agriculture, contributing significantly to food security, nutrition, and economic stability (FAO et al., 2013). In recent years the vegetable farming industry has experienced considerable growth per capita production and consumption basis this growth is attributed to their nutritional status. Vegetables have a high content of vitamins, minerals, antioxidants, dietary fibers, low fats, soluble sugars and calorie content; hence they have become a significant component of the human diet (Rashmi & Pradeep, 2020). In Kenya, vegetable farming is a vital agricultural activity, supporting the livelihoods of millions of smallholder farmers and contributing to the national economy (MoALF&C, 2021).Among the diverse range of vegetables cultivated in Kenya, African nightshade holds a prominent place due to its nutritional value, adaptability to local growing conditions, and cultural significance (Maundu & Tegnas, 2005). It is rich in vitamins A and C, iron, calcium, and antioxidants, making it a valuable addition to diets in regions where nutritional deficiencies are prevalent (Onyango & Imungi, 2007).Indigenous vegetables like African nightshade are not only nutritious but also well-suited to local climatic conditions and play a crucial role in improving food security, reducing malnutrition, and increasing income for small-scale farmers (Habwe et al., 2009).

African nightshade is listed among the top priority African indigenous vegetables in Kenya (Abukutsa-Onyango & Onyango, 2005). The commonly grown species for vegetable consumption in Kenya is *S. scabrum,* and *S. villosum.* The consumption of 100g of fresh leaves by an adult is said to provide 100% of the recommended daily allowance for calcium, iron, B-carotene and 40% of proteins (Abukutsa-Onyango, 2003). The leaves and the fruits are utilized for medicinal purposes to treat ailments like stomachaches and diarrhea (Ekesa et al., 2009).

Despite its importance, the production of African nightshade in Kenya faces several challenges. These include limited access to high-quality seeds, inadequate knowledge of best agricultural practices and suboptimal agronomic practices particularly in nutrient management (Muhanji et al.,2011). Depletion in soil fertility particularly of nitrogen and soil erosion were identified as major root causes constraining the growth of agricultural output in Kenya (Mulinge et al., 2016). Under ideal agronomic practices African nightshade has a potential yield of 30-50 tonnes per hectare, however farmers have been reported to obtain dismal yields of between1.3-1.5 tonnes per hectares (Opala et al., 2013). Majority of farmers in rural Kenya are under the misconception that traditional vegetables are hardy and can perform well in areas with low fertility, drought and generally needs little to no care. It has however been proven that even in small amounts, improvement in farmer practices could improve the quality and quantity of vegetable yields obtained by small holder farmers (Erin et al., 2021).

Effective nitrogen management is essential for maximizing crop productivity and ensuring sustainable farming practices (Fageria & Baligar, 2005). The source and rate of nitrogen application can markedly affect the nitrogen use efficiency (NUE) of crops, impacting both the yield and the quality of the produce (Marschner, 2012). This is due to its ability to influence plant growth through increasing greenness of plants, CO2 assimilation rate, crop quality-yield and improving resistance to environmental stresses such as limited water availability and saline soil conditions (Chen et al.,2010). The plant`s nitrogen absorption and distribution are influenced by factors such as species variation, growth stage, nitrogen assimilation enzyme activity, nitrogen rates and nitrogen form (Sun et al., 2023). For maximum growth, leafy vegetables like African nightshade need varying amounts of nitrogen during the growing season however often times large amounts of nitrogen remain in the soil unutilized which leads to environmental contamination.

This research aims to investigate the effect of different nitrogen rates and sources on the growth, yield quality, and nitrogen use efficiency of African nightshade. The findings of this study are expected to provide insights into best nitrogen management practices for African nightshade this will contribute to improving productivity and sustainability in its cultivation. By optimizing nitrogen use, farmers can achieve higher yields and better-quality produce, which can lead to increased income and enhanced food security. Additionally, understanding the relationship between nitrogen application and NUE can help mitigate environmental impacts, promoting more sustainable agricultural practices (Raun & Johnson, 1999; Marschner, 2012).

1.2 Problem Statement

Despite African nightshade`s (*Solanum* species) importance as a leafy indigenous vegetable that is widely consumed in Africa, there is limited information on optimal nitrogen management practices for this crop. This information is crucial for maximizing yield, quality and nitrogen use efficiency. Inefficient nitrogen utilization not only limits crop productivity but also contributes to environmental degradation through nitrogen leaching and greenhouse gas emissions (Tilman et al., 2002). Currently agricultural practices often rely heavily on inorganic nitrogen fertilizers, which provide immediate nutrient availability and increases yield but can lead to rapid nutrient loss and reduced soil health over time (Drinkwater et al., 1998). Inappropriate nitrogen management can also affect the nutritional quality of the harvested leaves, impacting their vitamin and mineral content, and ultimately the health benefits to consumers .Organic nitrogen sources, on the other hand, offer benefits such as improved soil structure and long-term nutrient availability but may not meet the immediate nutrient demands of high-yielding crop (Rosen & Allan, 2007). Understanding the balance and interaction between organic and inorganic nitrogen sources is essential for developing sustainable fertilization strategies that enhance both productivity and environmental sustainability. Previous studies have demonstrated the importance of optimizing nitrogen rates and sources to improve NUE in various crops (Raun & Johnson, 1999; Ladha et al., 2005). However, specific research on African nightshade is lacking, particularly in terms of how different nitrogen management practices affect its growth, yield, quality, and NUE. This knowledge gap hinders the development of effective agronomic recommendations tailored to the needs of smallholder farmers, who are the primary cultivators of this crop (Vanlauwe et al., 2011). This study aims to fill the existing knowledge gap and provide evidence-based recommendations for sustainable nitrogen management practices that enhance both crop productivity and environmental health.

1.3 Main Objective

To establish the effect of nitrogen rates and sources on plant growth, yield, quality and nitrogen use efficiency of African nightshade (*Solanum spp and Villosum spp)* in Kenya.

1.3.1 Specific Objectives

i. To evaluate the effect of four nitrogen rates on plant growth (height, leaf number and leaf area), yield (marketable leaves) and nitrogen use efficiency (yield per unit of nitrogen applied) of *Solanum spp and Villosum spp.*
ii. To evaluate the effect of four nitrogen sources on plant growth (height, leaf number and leaf area), yield (marketable leaves) and nitrogen use efficiency (yield per unit of nitrogen applied) of *Solanum spp and Villosum spp.*
iii. To evaluate the effect of varying rates of nitrogen fertilizer and different nitrogen sources on vegetable quality (protein and nitrate content) at vegetative and reproductive stages of *Solanum spp and Villosum spp.*

1.4 Hypothesis

i. Nitrogen application rate does not affect plant growth, yield and nitrogen use efficiency of African nightshade.
ii. There is no significant difference in how organic and inorganic sources of nitrogen affect plant growth yield, quality and nitrogen use efficiency of African nightshade.
iii. Varying rates of inorganic fertilizer and different Nitrogen sources does not affect the vegetable quality of African Nightshade.

1.5 Justification

African nightshade (*Solanum* species) is an important traditional leafy vegetable that is valued for its nutritional and medicinal properties in various parts of Africa. However, optimizing its cultivation practices especially in terms of nitrogen management remains a key challenge. Nitrogen is a vital nutrient for plant growth but its misuse or overuse can lead to significant economic loss, compromise vegetable quality and cause environmental degradation (Cheng-wei et al., 2014). Determining the optimal rates and sources of nitrogen for African nightshade can help farmers apply the right amount of fertilizer. Most African nightshade farmers are primarily small-scale farmers who often face financial constraints hence efficient use of nitrogen can lower their input costs and increase profitability by improving yields with minimal cost implications (Vanlauwe et al., 2011). The use of excessive nitrogen has a high potential of causing environmental degradation due to leaching and greenhouse gas emissions. Understanding the optimal rates and sources of nitrogen can minimize the negative impact on the environment (Tilman et al., 2002). NUE is a key indicator of how well plants utilize available nitrogen, this brings about the need of establishing which sources and rates of nitrogen enhance the NUE of African nightshade. To attain sustainable production, agricultural practices must aim at intensifying productivity while protecting the environment, human and animal health. Improving nitrogen use efficiency (NUE) is a key element of this sustainable framework (Zhang et al., 2022).

These sustainable measures will decrease the reliance on intensive nitrogen application and mitigate the adverse environmental effects associated with nitrogen use (Govindasamy et al., 2023). The research on nitrogen management can provide valuable insights into the physiological responses of the plant to different nitrogen forms and rates. This knowledge can inform breeding programs aimed at developing varieties with enhanced NUE and resilience. According to Sustainable Development Solutions Network Crop nitrogen use efficiency is an indicator of progress towards our objective of attaining food security, enhancing nutrition, decreasing pollution and generally encouraging sustainable agriculture

CHAPTER TWO

LITERATURE REVIEW

2.1 Origin and Morphology of African Nightshade

African nightshades are several species of plants that are in the genus *Solanum* in the family *Solanaceae*. It is also called vegetable nightshade, black nightshade, garden nightshade and garden huckleberry. The center of diversity of most of the solanum species is believed to be South America (Ojiewo et al., 2013). In Africa there are four species of nightshade cultivated as leafy vegetables *S. scabrum*, *S. villosum*, *S. americanum* and *S. eldorettii.* (Schippers, 2002). In terms of production and consumption *S. scabrum and S. villosum*, are preferred as they show considerable diversity, vegetative features and fruits. *Solanum scabrum* is a tall species and can be annual or perennial it can have pubescence or be hairless. It has ovate leaves that are 7-12 cm long and 5-8cm wide, the flowers are purple and produce large fruits that are black when ripe. Among the four species of African nightshade it is considered to be the best in terms of productivity. *Solanum villosum* is an annual herb that is short and compact and grows up to about 70 cm and can be slightly or densely pubescent. The leaf blade is ovate, up to 8 cm long, 3–6 cm wide, the leaf margin can be either entire or shallowly lobed, and petioles are 4.5 cm long. The plant has clusters of 3–8 flowers in the inflorescence. *S. villosum* has white corolla, with dull yellow or orange globular berries which are 5–9 mm diameter. The berries of this plant have a characteristic of dropping from the pedicel when ripe.

2.2 Plant Distribution

African nightshade species can be found in the following regions; East, South and South East Asia, Tropical Africa, South Pacific and North America.

2.3 Ecological Requirements and Propagation

African nightshade thrives in well-drained sandy loam soil that is rich in organic matter, with a pH range of 6.0 to 6.5. It can grow at altitudes ranging from 0 to 2400 meters above sea level and requires an annual rainfall of 500mm to 1200mm. The optimal temperature range for its growth is between 15° C to 30 ° C (Lin et al., 2009). Propagation of solanum species is majorly done by seeds 1-3 seeds are sown in holes that are 30cm by 30 cm apart. To encourage stronger branches and extended harvest period a wider spacing is used if the vegetable is to be kept in cultivation for a long period. When cultivated commercially African nightshade is sown in nurseries and transplanted

2.4 Economic Importance of African Nightshade

African nightshade is one of the most important African vegetables in Solanaceae family grown in Kenya. Currently it is of high economic importance with a very viable market both in rural and urban areas. Initially, indigenous vegetables were perceived as food for poor people, however over the last few decades there has been a major shift in these perceptions leading to increased consumption. Promotion of these vegetables by Non-governmental Organization and through agricultural research, has propelled the cultivation of African nightshade from wild to semi-cultivation (Ali, 2005).The leaves and shoots of African nightshade are consumed as food in many parts of Kenya due to their high nutritional content, 100g of fresh weight contains 87.5g water, 1.0mg iron,4.3g protein,38k calories,15.7 carbohydrates,1.4g fibre,20mg ascorbic acid 442 mg calcium,75 mg phosphorous,3660ug beta carotene and 0.5mg riboflavin. They also contain phenolics such as nicotine quinine (Ojiewo et al., 2007). There has been an increasing trend in the use of this crop as local communities become more aware of its reputed health benefits. Various health-promoting bioactive phytochemicals such as phenolic compounds, carotenoids, and chlorophylls, have been recently identified in *S. scabrum*. This discovery could account for its growing popularity in urban areas and its longstanding use in traditional medicine (Odongo et al., 2018).

2.5 African nightshade nutritional requirements

African Nightshade require large amounts of nutrients and respond positively to increased soil fertility. Well-decomposed poultry, farmyard or compost manure at the rate of 5 - 10 t/ha should be added to the soil prior to planting. Where manure is unavailable, a compound fertilizer such as NPK should be incorporated at the rate of 120-150 kg/ha during sowing/transplanting. Nitrogen fertilizers such as urea and ammonium sulphate (60kg N/ha) are used as top/side dressings after every 2-3 harvests; and foliar sprays are used in commercial production (Mwai et al., 2013).

2.6 Effect of Nitrogen on Plant growth, Yield and Quality of African Nightshade

Nitrogen is an essential nutrient for plant growth and plays a major role in plant nutrition. It is a key constituent of proteins, amino acids, nucleic acids, enzymes, hormones and chlorophyll (Kalaji et al., 2016). In crop production, nitrogen is the second most limiting factor after water, its application has been shown to increase yields, biomass and protein content in plant tissue (Jurg et al., 2008). Several studies conducted on the effect of Nitrogen fertilizer on vegetable yield have shown that application of nitrogen does increase vegetable leaf yield. For instance, studies conducted on spinach showed that increasing nitrogen rates can significantly enhance the fresh leaf yield of spinach (Salehi & Kashi, 2001). The results of this study are similar to the results of a study by Sainju et al. (2017) where the results of the experiment indicated that nitrogen fertilization increased the yield and biomass of vegetables like tomatoes and peppers. Other studies conducted on nightshades showed that they require high amounts of nitrogen fertilizer (up to 5g N/plant) for higher leaf yield (Masinde et al., 2010). The results of this studies offer very important information to vegetable farmers on how they can potentially increase vegetative yield. In the absence of nitrogen plant growth is greatly impacted, nitrogen deficiency affects plant growth by reducing the photosynthetic assimilating area, this decreases light interception and dry matter production (Zhao et al., 2004). Another growth parameter that is greatly influenced by nitrogen availability is leaf growth, more influence is noticed when there had been scarcity of nitrogen. With an increased leaf area, vegetables can capture more sunlight, enhancing their photosynthetic capacity. This leads to higher energy production and supports overall plant growth and development (Zhu et., al 2010).

Mohamed (2011) indicated that increase in plant height with nitrogen fertilizer is due to its effect on cell division and expansion which increases the number of internodes and length of internodes which results in progressive increase in height. In most instances, more internodes usually mean more leaves, which increases the overall leaf area. This larger leaf area enhances the plant's ability to capture sunlight and conduct photosynthesis, leading to greater energy production (Hikosaka, 2004). More internodes and leaves also enhance the plant's ability to utilize absorbed nutrients effectively, leading to better growth and higher yields (Marschner, 2012).In field production nitrogen can promote root growth, allowing taller plants to develop deeper root systems that access nutrients and water from deeper soil layers (Lynch, 1995).

In spite of the existing literature on fertilizer use, African nightshade growers lack species specific guidelines on the amount of nitrogen to apply. Recent surveys indicate that most farmers in peri-urban areas, where production of Nightshade is more market-oriented, use fertilizers in varying rates. Without specific guidelines, farmers might be tempted to apply higher amounts of nitrogen to act as insurance against low yields (Mwai et al.,2013). McCall and Willumsen (1998), showed that high rates of nitrogen application in vegetables will unlikely translate to significant yield gain however the farmer will likely experience financial losses.

It is important to note that nutritional value of vegetables is highly influenced by farming practices and post-harvest handling. Farming practices like fertilizer application may not only influence the yield and quality of field vegetable crops, but also the chemical composition of the marketable product. Nitrogen fertilizer significantly influences the quality of vegetables particularly in terms of protein content and nitrate accumulation (Fageria & Baligar, 2005; Marschner, 2012; Raun & Johnson, 1999). According to Lawlor (2002) with inadequate nitrogen, the amino acid content of leaves falls, less protein is synthesized and growth is decreased. In most cases adequate nitrogen fertilization typically leads to a higher protein content in vegetables as nitrogen availability directly influences synthesis of amino acids and proteins. The increase in protein content due to nitrogen fertilization can vary among different vegetable species. Leafy greens such as spinach, kales and lettuce often show a more pronounced increase in protein content compared to fruiting vegetables (Watanabe & Osaki, 2002). Type of nitrogenous fertilizer used is also plays a key role in vegetable quality in terms of protein content. According to a study by Wang et al. (2014) application of urea on spinach resulted in higher protein content compared to composted manure. The rapid nitrogen availability from urea supported immediate protein synthesis, while the slow release from compost did not meet the plants' peak nitrogen demand as effectively. Another study by Xie et al. (2016) on lettuce *(Lactuca sativa)* found that the application of CAN during the rapid growth phase significantly increased protein content compared to the use of compost.

In addition to nitrogen fertilization, physiological age of the plant also plays a major role in terms of protein content. In the vegetative growth phase, plants actively assimilate nitrogen to support the development of leaves, stems, and roots. High protein content is typically observed in leafy vegetables harvested during this stage (Marschner, 2012). However, as plants transition to the reproductive stage, the allocation of nutrients, including nitrogen, shifts towards the production of flowers, fruits, and seeds. This means that protein synthesis may decrease in vegetative tissues as resources are redirected. This shift can lead to a reduction in protein content in leaves and stems (Mengel et al., 2001).

Many green leafy vegetables like nightshade are known to be accumulators of phytochemicals like nitrates hence their quality is significantly affected by the amount of fertilizer applied (Mazahar et al., 2015). A major drawback of fertilizer use, particularly in the case of nitrogen is excessive use beyond the crop's needs which leads to negative implications for the vegetable quality, environment, especially groundwater pollution and its associated health hazards (Tilman et al., 2002). When nitrogen is applied in excess of what the plant requires for protein production, it is accumulated as nitrates and stored predominantly in the green leafy part of the plant (Undie et al., 2012). Nitrate accumulation in vegetables often depends on genetics, environment and the amount and kind of nutrients present in the soil. It is especially closely related to the amount, time of application, and composition of the fertilizers applied (Kaymac, 2013). For instance, inorganic fertilizers like CAN and Urea lead to a higher accumulation of nitrates compared to organic fertilizer sources due to the immediate availability of nitrogen in inorganic sources. Urea in particular undergoes hydrolysis to form ammonium ($NH4+$), which is then converted to nitrate ($NO3-$) through the nitrification process. This conversion happens relatively quickly in the soil, leading to a rapid increase in nitrate levels (Feng et al., 2020).

According to a study on lettuce by Santamaria et al., (2006) the rapid availability of nitrogen from inorganic fertilizers exceeded the plants' immediate uptake capacity, resulting in higher nitrate levels in the plant tissues while the gradual nitrogen release from organic fertilizer matched the lettuce's nitrogen uptake patterns more effectively, reducing nitrate accumulation. Physiological age of the plant is also a key factor in nitrate accumulation with young actively growing plants having higher nitrate uptake and conversion rates due to the demands of cell division, elongation and setup of basic plant structure (Umar et al.,2007).

Nitrate by itself has been shown to be relatively non-toxic to humans however it may be transformed to nitrite endogenously, nitrite reacts with amines and amides to produce N-nitroso compounds. These compounds have been associated with increased susceptibility to diseases a factor which has led to increased concern about nitrates in vegetables (Santamaria, 2006).Expert Committee on Food Additives (JECFA) set the Acceptable Daily Intake for nitrate at 0 to 3.65 $mgkg^{-1}$body weight (Jana & Moktan, 2013).Consumption of diets having high nitrate contents has contributed to endogenous nitrosation, which could lead to thyroid condition, various kinds of human cancers, neural tube defects and diabetes (Modassir et al., 2017).It is therefore important for producers of leafy vegetables to understand the role of nitrogen and its metabolic interaction in crops as an important step toward increasing productivity without compromising on quality.

2.7 Effect of Nitrogen Sources and Rates on Nitrogen Use Efficiency

Production of African nightshade in Kenya is done using organic fertilizer, inorganic fertilizer or a combination of both. These are two distinct types of fertilizers that differ in their chemical composition, source and environmental impact. Most vegetable farmers prefer using inorganic fertilizers compared to organic fertilizers due its nutrient concentration, predictability, ease of use and cost effectiveness (Elliot, 2005). Studies conducted on the effect of inorganic *vs* organic fertilizers on vegetable yield have attributed the difference in yield to immediate availability and consistent supply of nitrogen from inorganic sources (Salomez et al., 2013). Inorganic fertilizers provide a quick and efficient source of nutrients because they are soluble and immediately available to plants, this allows for precise control over the nutrient supply to the plants leading to more predictable and uniform growth. There are several types of inorganic fertilizers that provide nitrogen to plants they include; Ammonium nitrate, Urea and Calcium ammonium nitrate.

When it comes to selection of inorganic nitrogenous fertilizer to use for production, there are several key parameters to consider so as to ensure optimal crop growth, yield and soil health. These factors include; nitrogen content of fertilizer, crop type, growth stage, soil type and pH, application method, environmental considerations and residual effect. In most cases the primary consideration is normally the nitrogen content of the fertilizer since it determines the amount of nitrogen available for the plant (Raun & Johnson, 1999). Different fertilizers contain varying percentages of nitrogen, and the choice depends on the crop's nitrogen requirement e.g. Urea (46% N), Ammonium nitrate (34% N) and CAN (27% N) (Havlin et al., 2013). Since crops vary in their nutritional needs, it necessary to factor this variation when selecting nitrogen fertilizer.For instance vegetables require more nitrogen compared to root crops and are able to positively utilize it and improve on marketable yield.According to studies conducted on spinach the results indicated that different N forms have an effect on vegetable growth, and most leafy vegetables preferred nitrate N to ammonium & Urea N in conclusion the application of nitrate fertilizer usually promoted plant growth (Stagnari et al., 2007).

Understanding the soil type being used and its characteristics is equally important because it helps in tailoring nitrogen fertilizer management practises. Soil type significantly affects efficiency and effectiveness of inorganic nitrogenous fertiliers due to variations in soil properties such as texture,organic matter,cation exchange capacity,Electrical Conductivity and pH (Rosen & Allan, 2007).For instance moderate CEC in soils suggests that the soil has a moderate ability to retain cations like ammonium ($NH_4^{+)}$, pottasium ($K^{+)}$, calcium ($Ca^{2+)}$ and magnesium (Mg^{2+}) this means that some of the nitrogen fertilizer applied inform of ammonium will be retained and utilized by plants. However inspite of this there is potential of leaching nitrate (NO_3^-) the form of nitrogen that's negatively charged and not held by the soil cation exchange sites (Havlin et al., 2013). Soil pH has the ability to influence the availability of nutrients such as phosphorous, calcium, magnesium, and nitrogen. In slightly acidic soils (pH of 6.0- 6.5), nitrogen fertilizer tends to be more available to plants compared to highly alkaline soils. This is because nitrogen in the form of ammonium tends to be more stable and less prone to volatilization in slightly acidic soils. Slightly acidic soils also favors nitrification process where ammonium is converted to nitrates which is a form of nitrogen that most plants take up. Nitrate based fertilizers are less acidifying compared to the ammonium based fertilizers (Weil, 2016). Low soil Electrical conductivity indicates the presence of minimal soluble salts in the soil this may cause reduced efficiency in applied nitrogenous fertilizers. The inefficiency is due to the lack of necessary ions to retain and deliver nitrogen to plant roots which reduces utilization leading to deficiency in the plant (Biederbeck et al., 1986). In the green house environment, high temperatures increase the risk of ammonia (NH_3) volatilization, which leads to nitrogen loss and consequently poor Nitrogen Use Efficiency (NUE). This is because at higher temperatures, the equilibrium between ammonium (NH4+) in the soil and ammonia gas (NH_3) shifts towards the gaseous form, increasing the rate of volatilization (Wang et al., 2017).

Alternatively if a farmer decides to use oganic fertilizer as a source of nitrogen, there are several factors to be considered to ensure effective nutrient management and optimal crop growth. There is a vast selection of organic N sources available and they vary in cost, availability and N content. Organic fertilizer work directly as a source of plant nutrients and indirectly influences the physical, biological and chemical properties of soil however they are comparatively low in nutrient content so larger quantities are required for plant growth (Roba, 2018).For vegetetable production, the most commonly used source of N are compost and cover crops since they are inexpensive,readily available and have positive benefits to the soil (Gaskell & Smith, 2007).A challenge for most organic farmers is synchronizing the N release pattern from organic ammendements with the N uptake demand during particular growth stages.The release of N from organic sources depends on the rate of decomposition and mineralization which are largely influenced by soil moisture, temperature and carbon to Nitrogen ratio of the organic material (Berry et al.,2006). As explained by Gaskell (2007), the initial burst of nitrogen from organic fertilizer may not coincide with peak demand of nitrogen for vegetables.As a result the nitrogen supplied by the organic source might be depleted by the time the plant reaches critical growth stages hence leading to potential nitrogen deficiencies and reduced yields.

Both organic and inorganic fertilizers influence Nitrogen Use efficiency differently due their distinct properties and modes of action. NUE is a measure of how effectively plants utilize available fertilizers for growth and development.It can be defined as the percent of the applied N fertilizer that is recovered by the current crop (de Jesus et al., 2024).Globally the average NUE for crops ranges between 50% to 80% this values vary from region to region primarily based on socio-economic factors. In Africa for example,due to the relatively low rates of N fertilizer used NUE reaches 80% this means that 20% of applied N fertilizer may be lost to the environment if the soil N levels have already reached a steady state (You et al., 2023). The NUE for vegetable production is estimated to be at 14% globally in comparison to maize and wheat at 46% and 40% respectively (Zhang et al., 2015).This estimation is in line with studies conducted on lettuce that indicated an NUE of 12% when single application of 180kg N/Ha and an NUE of 25% when receiving two split applications at 60kg N/Ha (Venezuela, 2024). The NUE index determines the ability of a crop to maintain productivity under limited supply of N to minimize losses from surplus N applications. In another experiment conducted on tomatoes the results indicated that under best management practices the NUE for vegetables may improve and reach 70% (Hochmuth & Hanlon, 2020).

There are three main components that determine the NUE of crops, these are N uptake efficiency (NUpE), N utilization efficiency (NUtE) and the harvest Index (NHI) (Bueren & Struick, 2017).These components are affected by the form of N uptake, either NO_3^- or NH_4^+, N translocation within the plant, the reduction of NO_3^-, and the assimilation of reduced N into organic compounds (Barker, 1989). The ability of crops to absorb nitrogen depends on various interconnected factors which include; soil fertility, cultivar type, soil moisture levels, temperature, seasonal timing, nitrogen uptake patterns, type of nitrogen fertilizer, pest and disease presence, farmer expertise among others.There are also physiological mechanisms of the crop that influence NUE they include ;root growth, N uptake pattern,leaf growth and N remobilization within the plant (Bueren & Struick, 2017).NUE is dependent on the physiological capacity of vegetable species to uptake, metabolize, and redistribute the necessary amount of N, during the appropriate stage of crop growth, to optimize yields. For leafy crops, N uptake patterns correspond to the periods of biomass accumulation because nitrogen is a critical nutrient that supports several physiological processes essential for plant growth and development (Lawlor, 2002).

It has been observed in several studies that higher N application rates especially above the optimum recommended rates results in lower NUE (Wortmann et al., 2011).Generally as insuarance against possible yield loss, farmers tend to apply N fertilizers above recommendated rates (Gianquinto et al., 2013).For instance vegetable growers in Spain and Florida tend to make N applications at above recommended rates in both greenhouse and field conditions resulting in environmental contamination (Prasad & Hochmuth, 2016).This practise not only reduced NUE but also compromised vegetable quality,led to yield depression in some varieties and also delayed vegetable maturity (Lan et al., 2021). In another study conducted on tomato production in California by Hartz et al. (2000) the results indicated that higher nitrogen application rates resulted in lower NUE because of increased leaching and volatilization losses. Due to plants limited capacity to absorb and utilize nitrogen, additional nitrogen does not translate to proportional increase in yield beyond a certain point. This saturation point results in diminishing returns for nitrogen uptake efficiency, lowering overall NUE at higher application rates (Moll et al., 1982).

Normally due to the rapid availability of nitrogen in inorganic fertilizers, nitrogen use efficiency (NUE) is often reduced. This is because the quick release of nitrogen increases the risk of nitrogen losses through leaching, volatilization, and denitrification, particularly if the timing of application is not well managed (Havlin, et al., 2013).According to a study conducted in Spain on tomatoes, it was observed that in the highest N application rate in the experiment about 90% of applied N resulted in environmental losses attributed to leaching or volatilisation (Jalpa & Mylavarapu, 2021). With such high potential chemical N losses, split application of inorganic N fertilizer is an effective strategy that can be used since it allows farmers to provide nitrogen when the crop's demand is highest, such as during rapid vegetative growth or just before repoductive growth (Tilman et al., 2002). Split fertilizer application also ensures that nitrogen fertilizer is not in excessive of what is required this is because the high nitrogen supply will lead to reduced Rubsco activity due to insufficient carbon dioxide supply caused by high nitrogen supply.Typically, high nitrogen supply increases the synthesis of Rubisco and other photosynthetic proteins which enhances the plants photosynthetic capacity, however if the carbon dioxide supply is insufficient to meet the demand, the rate of carbon fixation is limited leading to a buildup of unused nitrogen within the plant tissue. This results in decreased NUE as the nitrogen absorbed by the plant is not effectively converted to biomass (Xu et al., 2012).

An effective NUE management system should be designed to synchronize the levels of available N with the estimated demand during the different phenological stages of development. In comparison organic fertilizers tend to offer a better NUE in the long term due to the slow and steady release of the available N.According to the results of a study by Drinkwater et al. (1998) organic farming systems, which rely on organic nitrogen sources, had lower immediate NUE but better long-term soil fertility,sustainability and NUE compared to conventional systems using inorganic fertilizers.

Research on NUE has indicated that environmental losses may be reduced by 15-30% by adopting improved management strategies (Ciampitti & Vyn, 2012).Combining organic and inorganic fertilizers is one of the strategies used to optimize NUE by leveraging the slow release properties of the organic fertilizer and the immediate availability of the inorganic fertilizer. Organic nitrogen sources release nitrogen more slowly compared to inorganic fertilizers, as the nitrogen is bound in organic matter and must be mineralized by soil microorganisms before it becomes available to plants. This gradual release can result in lower initial NUE but potentially higher long-term NUE as nitrogen is released more consistently over the growing season and is less prone to losses (Zhou et al., 2017).Organic fertilizers also have a positive impact on soil health by enhacing soil structure, increasing organic matter content and promote microbial activity this improves root health and function leading to better nitrogen uptake (Tilman et al., 2002). Other recommended practices to improve NUE in commercial vegetable production include adopting the right source, rate, proper timing and N placement.Based on results from studies conducted on management strategies of NUE it was deduced that when farmers uniformly adopt particular management strategies, NUE tends to increase area-wide by 30-50% (Dobermann, 2007).An inmproved NUE helps to significantly reduce production costs, incidences of nutrient imbalance and environmental losses (Gaskell & Smith, 2007).

It is crucial for farmers to understand the impact of organic and inorganic sources of nitrogen on NUE, this will enable them to make informed decisions that enhance crop productivity, sustainability, profitabillity and will also address environmental and regulatory concerns.

CHAPTER THREE

MATERIALS AND METHODS

3.1 Description of the Research Location

The research was carried out in a greenhouse at Jomo Kenyatta University of Agriculture and Technology. The University is located in Juja Kiambu county at co-ordinates 1.1793° S 37.1184°E at an elevation of 1525M ASL, approximately 36 kilometers North East of the capital Nairobi. The average annual temperature of the area is 20°C - 22°C and it receives an average 800mm to 1200mm of rainfall annually. The study was conducted between October 2016 and May 2017.The first experiment was conducted between October 2016 to January 2017. The second experiment was conducted between January 2017 to April 2017. Analysis for proteins and nitrates was carried out in the Food Chemistry laboratory in Food Science Department at Jomo Kenyatta University of Agriculture and Technology.

3.2 Materials

Solanum scabrum species had three varieties; Abuku 1, Abuku 2 which were sourced from the African indigenous Vegetable project JKUAT and as standard Scabrum variety purchased from a local agrovet. *Solanum villosum* species had two varieties; Abuku 3 which was sourced from the African indigenous Vegetable project JKUAT and a standard villosum variety that was also purchased from a local agrovet were used as planting material for these experiments.

Plate 3.1: ***Solanum scabrum*** **species: A. Abuku1 and B. Abuku 2**

Plate 3.2: ***Solanum villosum*** **Species: C. Abuku 3**

Calcium ammonium nitrate (27% N), was used as a source of nitrogen for the first experiment. Triple Super Phosphate (46% Phosphate) and Muriate of Potash (60% Potash) were used a source of phosphorous and potassium respectively. In the second experiment, CAN (27% N), Urea (46%N) and processed organic fertilizer (3%N) were used as source of nitrogen.

3.3 Experimental Design

Completely randomized design was used in this study. In the first experiment there were four varying rates of CAN (0g,0.9g,1.8g,3.6g) and replicated four times. Five pots having a single plant each represented one treatment replication bringing the total number of pots per experiment to four hundred. In the second experiment there were three sources of nitrogen (control, CAN, Urea and processed organic fertilizer) replicated four times. Five pots having a single plant each represented one treatment replication bringing the total number of pots per experiment to four hundred.

Table 3.1: Nitrogen Rates Used in Objective 1

Treatment(T)	Weight of CAN (per plant)	description
T_1	0g	control
T_2	0.9g	Equivalent to 100kg CAN Ha
T_3	1.8g	Equivalent to 200kg CAN/Ha
T_4	3.6g	Equivalent to 400kg CAN/Ha

Table 3.2: Nitrogen Rates Used in Objective 2

Treatment(T)	Nitrogen source	description
T_1	No nitrogen	control
T_2 (3.6g)	Calcium ammonium nitrate	Equivalent to 400kg CAN/Ha
T_3 (2.11g)	urea	Equivalent to 234.4 kg urea/Ha
T_4 (36.4g)	Processed organic fertilizer	Equivalent to 4,044 kg organic /Ha

3.4 Methodology

3.4.1 Soil Analysis

The soil nitrogen content, EC and CEC of the potting media (red soil: sandy soil (3:1)) used during the experiment was determined before planting.

3.4.1.1 pH Determination

The pH content of the planting media was determined according to method 4.10.2 in Methods of soil Analysis (Sparks, et al., 1996)**.** 20 grams of soil sample was air dried, crushed and sieved through a 2mm sieve.10 grams of the airdried sieved soil sample was put into a clean beaker and 25ml of distilled water added to the soil creating a (1: 2.5 soil to water ratio). The mixture was thoroughly stirred to ensure that the soil particles were fully suspended in water and the mixture was allowed to stand for 30 minutes to equilibrate. The pH meter was first calibrated using a standard buffer solution with a pH of 7 followed by a buffer solution of pH 4.0. The electrode was rinsed with water between calibrations and blotted to dry using a tissue. The electrode was then fully immersed into the liquid part of the suspension and the pH value displayed on the meter recorded.

3.4.1.2 EC Determination

The EC content of the planting media was determined according to method 4.8.2 in methods of soil analysis (Sparks, et al., 1996)**.**Soil sample was air dried, crushed and sieved through a 2mm sieve. 20 grams of the air-dried sieved soil was put into a beaker and 40ml of distilled water added creating a 1:2 soil to water ratio. The soil mixture was stirred thoroughly for 30 minutes and left to settle. The EC meter was calibrated using known EC values and the conductivity cell rinsed using distilled water between calibrations, the conductivity cell was inserted into the clear soil extract and the EC value displayed on the screen recorded.

3.4.1.3 CEC Determination

The CEC content of the planting media was determined according to Schollenberger method using percolation apparatus (Schollenberger & Simon 1945). The bottom of the percolation tube was closed using cotton wool, filter paper and compressed with a glass stick.100 ml of 1M ammonium acetate was placed on the reservoir above the percolation tube. 5ml of 1M ammonium acetate in the reservoir was added to the stoppered percolation tube and 5g of soil slowly placed into the percolation tube. The walls of the percolation tube were rinsed with 1 M ammonium acetate. The percolation tube was connected to the reservoir tube and collecting flask was disconnected after 16 hours and the contents transferred to a 200 ml volumetric flask and made to volume with water. 50 ml of 80% ethanol was placed in the reservoir tube and percolation started to remove excess ammonium acetate and the extract was discarded. 100ml of 10% kcl was placed in the reservoir tube and the percolation process completed in 20 hours. The collecting flask was disconnected and the contents transferred into a 200 ml volumetric flask and made to volume with water. The distillate was titrated against dilute HCL and the titer volume recorded. The volume was used in the final calculation

The CEC was calculated based on the following formulae

$$CEC(cmol_c\, kg^{-1}) = 0.1 * f_{NaOH} * (v - b) * \frac{\left(\frac{200}{10} * \frac{1000}{w}\right)}{10 * f_m}$$

v = titer of the sample (mL)

b= titer of the blank (mL)

w= weight of air-dry soil sample (g)

f_{NaOH} = factor of 0.1M NaOH solution

f_{m}=Moisture correction factor of the soil sample

3.4.2 Propagation of African Nightshade

Plastic sleeves were filled with two kilograms of potting media which was a mixture of red soil and sand at the ratio of 3:1. Three seeds were planted per sleeve and later thinned to leave one plant per pot after two weeks.

3.4.3 Effect of Nitrogen Rates on Plant Growth, Yield, Quality and Nitrogen Use Efficiency

3.4.3.1 Crop Management

At planting each pot received phosphorus and potassium in the form of triple super phosphate (46% P_2O_5) 100kg/ha and Muriate of potash (60%K_2O) at 100kg/ha or 0.9 g per plant. At two weeks after planting varying rates of CAN was applied to each pot having one plant as per the treatment plan. The amount of water used for irrigation weekly was 400ml this amount was determined by measuring the saturation capacity of the planting media. Yellow sticky traps were used for white fly control and weeding was done regularly to reduce competition for the nutrients applied.

3.4.3.2 Measurement of Plant Height

The length of the main stem was taken using a meter rule from the base of the plant to the tip of longest leaf from three tagged plants. The first plant height was taken four weeks after emergence and subsequent measurements were taken at two-week interval until week fourteen.

3.4.3.3 Measurement of Number of Leaves

The number of leaves per plant was counted on tagged plants three weeks after emergence and subsequent counting done at a two-week interval and the counts recorded in a data sheet until the end of the experiment at fourteen weeks.

3.4.3.4 Measurement of Fresh Weight

The fresh weight of the leaves was measured using a digital balance electric balance (model LIBROR EB-3200D). The leaves were harvested at the end of the experiment which was fourteen weeks after planting and excess moisture blotted out and weighed immediately.

3.4.3.5 Measurement of Leaf Surface Area

The leaves were harvested at fourteen weeks after planting and surface area determined using a leaf area meter (LI-COR Li-3000) The results were recorded in a data book.

3.4.3.6 Measurement of Vegetable Quality

After vegetable harvest which was at fourteen weeks vegetable quality was analyzed. This was done by quantifying protein and nitrate content in the leaves based on the following procedures;

3.4.3.6.1 Protein Determination

Protein were determined using the semi-micro kjeldal method (AOAC, 1995), specification 950.46 method 20.87-37.1.22. Approximately 2 g of sample was weighed into a digestion flask together with a combined catalyst of 5 g potassium sulphate, 0.5 g of copper sulphate and 15 mL of Sulphuric acid. The mixture was heated in a fume hood till the digest color turned blue. This signified the end of the digestion process. The digest was cooled, transferred to 100 ml volumetric flask and topped up to the mark with deionized water. A blank digestion with the catalyst was also made. Exactly 10 mL of diluted digest was transferred into the distilling flask and washed with distilled water ,15 mL of 40% NaOH was added and washed with distilled water.

Distillation was done to a volume of about 60 mL distillate. The distillate was titrated using 0.02 N HCL to an orange color of the mixed indicator, which signified the end point. The nitrogen in the sample was calculated as

$$\% \, Nitrogen = \frac{(V1 - V2) \times N \times F \div (V \times 100)}{Sample\ wt}$$

Where: V1 is the titer for sample in ml, V2 is titer for blank in mL; N= normality of standard HCL (0.02); f= factor of std HCL solution; V= volume of diluted digest taken for distillation (10 mL); S= weight of sample taken for distillation (1 g). The protein content was calculated as;

$$\%Protein = Nitrogen \times Protein\ factor (6.25)$$

3.4.3.6.2 Method used for Nitrate Analysis

The nitrate content in the test samples was determined by the calorimetric method using salicylic acid (Cataldo et al., 1975). About 500 mg of fresh sample was weighed and put in a test tube, and 10 mL of hot (90-950C) distilled water added. The closed tubes were placed in a water bath at 800C for 30 minutes and shaken. The samples were cooled and centrifuged at 4500 rpm. Supernatant were decanted and weighed to determine the exact volume of extract. Chlorophyll in leaf extract were removed by adding 0.5 g $MgCO_3$ to the supernatant, and centrifuged again. The supernatant containing the nitrate extract was treated with 2 N NaOH and a combination of Salicylic acid and H_2SO_4 in a ratio 1:20 w: v. Standards were prepared using sodium nitrate and absorption was measured at 410 nm with UV-Vis spectrophotometer.

3.4.3.7 Measurement of Nitrogen Use efficiency

Nitrogen use efficiency was calculated at the end of the pot experiment according to Moll et al (1982) using data collected during the experiment. The data utilized in these calculations was total plant nitrogen at harvest, nitrogen rate (treatments) and leaf yield (harvest index).

NUE= (N uptake efficiency * N utilization efficiency)

Utilization efficiency= Gw/Nt

Uptake efficiency=Nt/Ns

Where Nt= total N in the plant at harvest

Ns=Nitrogen supply or rate of N fertilizer

GW=Leaf yield of the produce

3.4.3.7.1 Measurement of Total Plant Nitrogen at Harvest

Nitrogen content in the leaves at harvest was determined using the Kjeldahl method that involves digestion and distillation (AOAC,1995). The leaves were air dried and grinded into fine powder using mortar and pestle. 0.3 grams of the powdered leaf samples was put into a dry Kjeldahl digestion tube. The digestion mixture was prepared by adding 0.42g of Selenium powder and 14g lithium sulphate to 350 ml 30% hydrogen peroxide was added and mixed well. 420 ml of concentrated H_2SO_4 was added slowly while cooling in an ice bath and stored at 2^{o}C. 4.4 ml of digestion mixture was added to each tube and to 2 reagent blanks for each batch of sample**.** Digestion was done at 360^{o}C for 2 hours; after the solution turned colorless the contents were allowed to cool.25ml of distilled water was added and mixed well until no more sediment dissolved then allowed to cool. The sample was made up to 50 ml with distilled water and mixed well. The sample was allowed to settle so that a clear solution was taken from the top of the tube for analysis. 10 ml of the digested sample was put into the distillation chamber and 40% NaOH to the chamber. Ammonia was distilled into 5ml of 1% boric acid solution containing 4 drops of a mixed indicator. Distillation was continued until the indicator turned green. The distillate was titrated with 0.0073 N HCL until we got a definite pink.

% Nitrogen calculation

$$Nitrogen\ content\ (\%) = \frac{Volume\ of\ HCL\ used(ml) * Normality\ of\ HCL * 1.4}{Weight\ of\ sample\ (g)}$$

Where 1.4 is a conversion factor accounting for the atomic weight of nitrogen and the dilution factor.

3.4.4 Effect of Nitrogen Sources on Plant Growth, Yield, Quality and Nitrogen Use Efficiency

3.4.4.1 Crop Management

Two weeks after planting 3.6 g of CAN, 2.11 g of Urea and 36.4 g of organic fertilizer were applied to each pot having one plant. The grams of fertilizer applied was based on the results of the first experiment which indicated that 3.6 of CAN gave the best pant growth and yield. The varying nitrogen content in the different sources brought about the differences in quantities applied. Pots supplied with CAN and urea received phosphorus and potassium in the form of triple super phosphate (46% P_2O_5) 100kg/ha and Muriate of potash (60%K_2O) at 100kg/ha or 0.9 g per plant. Organic fertilizer had phosphorous and potassium content of more than 1%. The amount of water used for irrigation weekly was 400ml this amount was determined by measuring the saturation capacity of the planting media Yellow sticky traps were used for white fly control and weeding was done regularly to reduce competition for the nutrients applied.

3.4.4.2 Measurement of Plant Height

The length of the main stem was taken using a meter rule from the base of the plant to the tip of longest leaf from three tagged plants in each treatment replication. The first plant height was taken four weeks after emergence and subsequent measurements were taken at a two-week interval until week fourteen.

3.4.4.3 Measurement of Number of Leaves

The number of leaves per plant was counted on three tagged plants per each treatment replication three weeks after emergence and subsequent counting done at intervals of two week and the counts recorded in a data sheet until the end of the experiment at fourteen weeks.

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The fresh weight of the leaves was measured using a digital balance electric balance (model LIBROR EB-3200D). The leaves were harvested at the end of the experiment which was fourteen weeks after planting and excess moisture blotted out and weighed immediately.

3.4.4.5 Measurement of Leaf Surface Area

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3.4.4.6 Measurement of Vegetable Quality

After vegetable harvest at fourteen weeks vegetable quality was analyzed. This was done by quantifying protein and nitrate content in the leaves based on the following procedures;

3.4.4.6.1 Protein Determination

Percent protein was determined using the semi-micro kjeldal method (AOAC, 1995), specification 950.46 method 20.87-37.1.22. Approximately 2 g of sample was weighed into a digestion flask together with a combined catalyst of 5 g potassium sulphate, 0.5 g of copper sulphate and 15 mL of Sulphuric acid. The mixture was heated in a fume hood till the digest color turned blue. This signified the end of the digestion process. The digest was cooled, transferred to 100 mL volumetric flask and topped up to the mark with deionized water. A blank digestion with the catalyst was also made. Exactly 10 mL of diluted digest was transferred into the distilling flask and washed with distilled water, 15 mL of 40% NaOH was added and washed with distilled water. Distillation was done to a volume of about 60 mL distillate. The distillate was titrated using 0.02 N HCL to an orange color of the mixed indicator, which signified the end point. The nitrogen in the sample was calculated as

$$\% \, Nitrogen = \frac{(V1 - V2) \times N \times F \div (V \times 100)}{Sample \, wt}$$

Where: V1 is the titer for sample in ml, V2 is titer for blank in mL; N= normality of standard HCL (0.02); f= factor of std HCL solution; V= volume of diluted digest taken for distillation (10 mL); S= weight of sample taken for distillation (1 g). The protein content was calculated as;

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3.4.4.7 Measurement of Nitrogen Use Efficiency

Nitrogen use efficiency was calculated at the end of the pot experiment according to Moll et al (1982) using data collected during the experiment. The data utilized in these calculations was total plant nitrogen at harvest, nitrogen rate (treatments) and leaf yield (harvest index).

NUE= (N uptake efficiency * N utilization efficiency)

Utilization efficiency= Gw/Nt

Uptake efficiency=Nt/Ns

Where Nt= total N in the plant at harvest

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% Nitrogen calculation

$$Nitrogen\ content\ (\%) = \frac{Volume\ of\ HCL\ used(ml) * Normality\ of\ HCL * 1.4}{Weight\ of\ sample\ (g)}$$

Where 1.4 is a conversion factor accounting for the atomic weight of nitrogen and the dilution factor.

3.4.5 Data Analysis

Genstat statistical software version 14 was used for data analysis. Data from the growth parameters, vegetable quality and nitrogen use efficiency was analyzed using one-way analysis variance (one-way-ANOVA) to evaluate the effect of fertilizer rate and sources on African nightshade growth, yield, vegetable quality and NUE at 0.05 level of probability. Mean separation was done using turkey post hoc analysis test to evaluate the significance of the difference among the treatments.

CHAPTER FOUR

RESULTS

4.1 Soil Analysis

The soils used as potting media was slightly acidic and poor in total Nitrogen. The potting media had very low EC which indicated minimal soluble salts in the soil. The CEC of the potting media was moderate which influenced the nutrient retention capacity of the soils (Table 4.1).

Table 4.1: Chemical Properties of Potting Media Used in the Study

Parameter	Concentration
pH-H_2O (1:2.5 v/v)	6.02
Ec (dS/m) soil: water suspension 1:2:5	0.07
C.E.C (cmolc/kg)	14
Nitrogen (%)	0.05

4.2 Effect of Nitrogen Application Rates on Growth, Yield, Quality and NUE of African Nightshade

4.2.1 Effect of Nitrogen Application Rates on the Height of African Nightshade

There was no significant difference in height during the first, second and third height collection in both the *Solanum scabrum* and *Solanum villosum* species. At six weeks after planting a significant difference in height at $P \leq 0.05$ was observed only in the *Solanum villosum* species with plants in the control group having the least height (Table 4.2)

Table 4.2: Effect of Nitrogen Application Rates on Plant Height in *Solanum scabrum* and *Solanum villosum* Species Six Weeks after Treatment Application

Nitrogen rates (CAN g/plant)	Plant height (cm)	
	Solanum scabrum	*Solanum villosum*
0	29.0 a	20.82 a
0.9	33.0 a	29.43 b
1.8	31.4 a	30.52 b
3.6	33.0 a	31.65 b
$LSD_{0.05}$	7.06	7.18

Means followed by same letter(s) are not significantly different at ($P \leq 0.05$)

At fourteen weeks after treatment application, increasing the rate of CAN per plant was observed to significantly influence plant height at $P \leq 0.05$ in both *Solanum scabrum* and *Solanum villosum* species (Table 4.3). Plants supplied with 3.6 g of CAN in both species were taller in comparison to other treatments.

Table 4.3: Effect of Nitrogen Application Rates on Plant Height in *Solanum scabrum* and *Solanum villosum* Species Fourteen Weeks after Treatment Application

Nitrogen rates (CAN g/plant)	Plant height (cm)	
	Solanum scabrum	***Solanum villosum***
0	70.27a	42.08a
0.9	81.71b	62.69b
1.8	82.82b	63.91c
3.6	92.53c	80.06c
$LSD_{0.05}$	4.80	4.17

Means followed by same letter(s) are not significantly different at ($P \leq 0.05$)

4.2.3 Effect of Nitrogen Application Rates on African Nightshade Leaf Number

There was no significant difference in number of leaves at $P \leq 0.05$ due to nitrogen effect in the *Solanum scabrum* and *Solanum villosum* species at two, four and six weeks after treatment application. At fourteen weeks after treatment application, increasing the rate of CAN per plant was observed to significantly influence leaf number at $P \leq 0.05$ in both *Solanum scabrum* and *Solanum villosum* species (Table 4.4). Plants supplied with 3.6 g of CAN in both species had more leaves in comparison to other treatments.

Table 4.4: Effect of Nitrogen Application Rates on Number of Leaves in *Solanum scabrum* and *Solanum villosum* Species Fourteen Weeks after Treatment Application

Nitrogen rates (CAN g/plant)	No. of leaves	
	Solanum scabrum	*Solanum villosum*
0	77.4a	82.0a
0.9	83.5a	89.1a
1.8	92.4b	112.6b
3.6	117.2c	126.5c
$LSD_{0.05}$	9.08	10.72

Means followed by same letter(s) are not significantly different at ($P \leq 0.05$)

4.2.4 Effect of Nitrogen Application Rates on Yield of African Nightshade

Increasing nitrogen rate was seen to significantly increase leaf yield at $P \leq 0.05$ in both *Solanum scabrum* and *Solanum villosum* species. In both species, plants supplied with 3.6 g of CAN per plant had increased yield output at the end of the experiment (Fig 4.1)

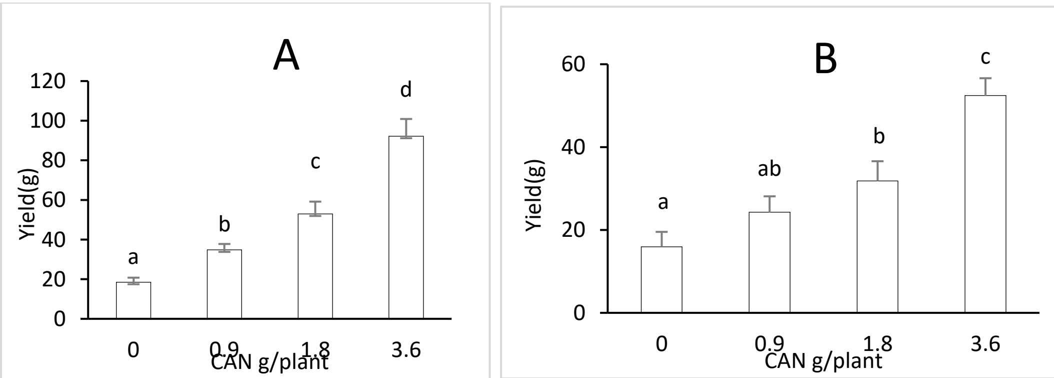

Figure 4.1: Yield response of African Nightshade Treated with Varied Rates of Nitrogen (A) *Solanum scabrum* (B) *Solanum villosum*

Vertical bars with same letter are not significantly different at ($P \leq 0.05$)

4.2.5 Effect of Nitrogen Application Rates on Leaf Area of African Nightshade

Increasing nitrogen rate was observed to significantly increase leaf area at $P \leq 0.05$ in both *Solanum scabrum* and *Solanum villosum* species. In *Solanum villosum* species plants supplied with 1.8 g of CAN per plant had the highest leaf area. In *Solanum* *scabrum* species plants supplied with 3.6 g of CAN were seen to have the largest leaf area (Table 4.5)

Table 4.5: Effect of Nitrogen Application Rates on Leaf Area in *Solanum scabrum* and *Solanum villosum* Species

Nitrogen rates (CAN g/plant)	**Leaf area (cm^2)**	
	Solanum villosum	***Solanum scabrum***
0	1046 a	528 a
0.9	1408 a	976 b
1.8	2285 b	1052b
3.6	1408 a	2200b
$LSD_{0.05}$	7.06	7.18

Means followed by same letter(s) are not significantly different at ($P \leq 0.05$)

4.2.6 Effect of Nitrogen application rates on Nitrogen Use Efficiency of African Nightshade

Increasing the rate of nitrogen application was seen to reduce NUE of African Nightshade. There was significant difference in NUE at $P \leq 0.05$ among *Solanum scabrum* and *Solanum villosum* varieties in the different nitrogen rates with plants supplied with 0 g of nitrogen being the most efficient (Fig 4.2).

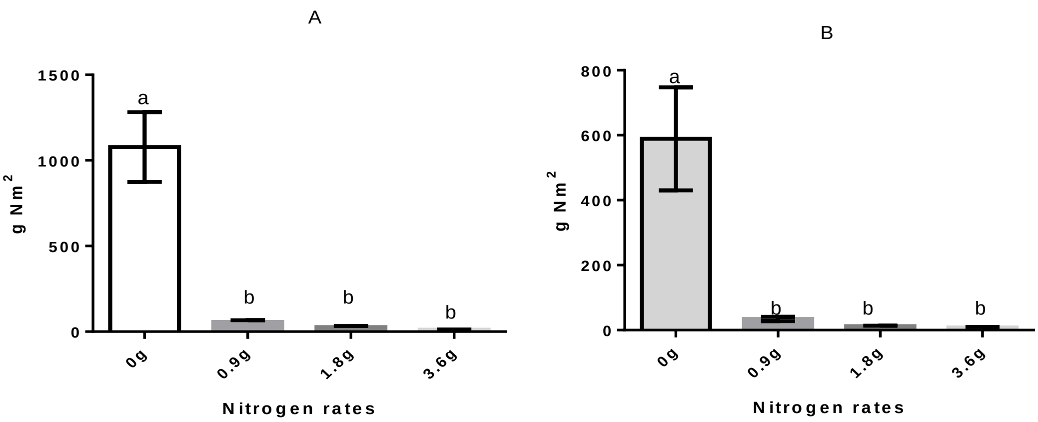

Figure 4.2: Nitrogen Use Efficiency of *Solanum scabrum* Spp (A) and *Solanum villosum* Spp (B) Treated with Varying Nitrogen Rates

Vertical bars with same letter are not significantly different at (P≤0.05)

4.2.7 Effect of Nitrogen Rates on Vegetable Quality of African Nightshade

4.2.7.1 Effect of Nitrogen Application Rate on Nitrate Content of African Nightshade at Vegetative Stage

Increasing nitrogen rate was seen not to have any significant effect on nitrate content level in the *Solanum scabrum* species. However, in the *Solanum villosum* species there was a significant difference in nitrate content at P≤0.05 level of significance due to varied rates of nitrogen (Fig 4.3)

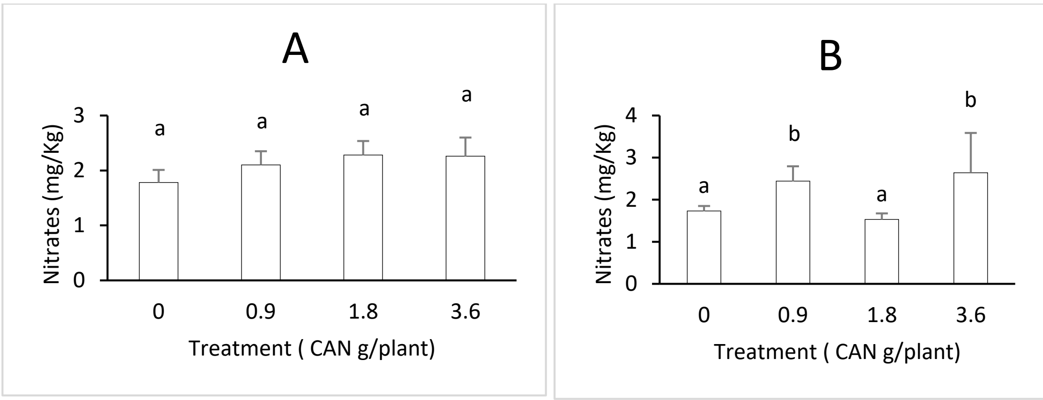

Figure 4.3: Nitrate content of African Nightshade at Vegetative Stage Treated with Varied Rates of Nitrogen (A) *Solanum scabrum* Spp (B) *Solanum villosum* Spp

Vertical bars with same letter are not significantly different at ($P \leq 0.05$)

4.2.7.2 Effect of Nitrogen Application Rate on Nitrate Content of African Nightshade at Reproductive Stage

At the reproductive stage there was significant difference in nitrate content among the plants at $P \leq 0.05$ with plants applied with 3.6 g N having the highest nitrate content in both *Solanum scabrum* and *Solanum villosum* species (Fig 4.4)

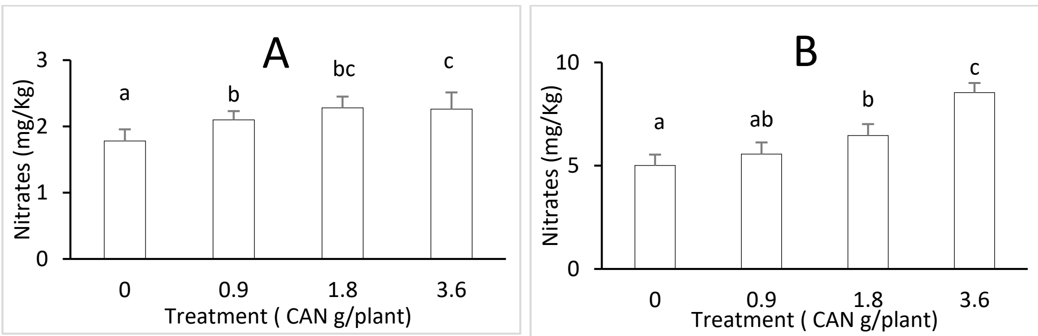

Figure 4.4: Nitrate Content of African Nightshade at Reproductive Stage Treated with Varied Rates of Nitrogen (A) *Solanum scabrum* Spp (B) *Solanum villosum* Spp

Vertical bars with same letter are not significantly different at ($P \leq 0.05$)

4.2.7.3 Effect of Nitrogen Application Rate on Protein Content of African Nightshade at the Vegetative Stage

Increasing nitrogen rate was seen to have a significant effect on protein content at $P \leq 0.05$ level of significance in the *Solanum scabrum* and *Solanum villosum* species (Table 4.6). In *Solanum villosum* species plants supplied with 3.6g of CAN had the most protein content. In *Solanum scabrum* species there was no significant difference in protein content at $P \leq 0.05$ between plants supplied with 1.8 g CAN and plants supplied with 3.6g CAN.

Table 4.6: Effect of Nitrogen Application Rates on Protein Content in *Solanum scabrum* and *Solanum villosum* Species at Vegetative Stage

Nitrogen rates (CAN g/plant)	**% Protein**	
	Solanum scabrum	*Solanum villosum*
0	9.11 a	11.24 a
0.9	18.90 b	17.65 b
1.8	23.98 c	23.02 c
3.6	29.27 c	28.43 d
$LSD_{0.05}$	2.11	3.20

Means followed by same letter(s) are not significantly different at ($P \leq 0.05$)

4.2.7.4 Effect of Nitrogen Application Rate on Protein Content of African Nightshade at the Reproductive Stage

There was a significant difference at $P \leq 0.05$ with plants supplied with 3.6 g of CAN per plant having higher protein content. A decrease in the amount of protein content was observed across the all the nitrogen rates at the reproductive stage (Table 4.7).

Table 4.7: Effect of Nitrogen Application Rates on Protein Content in *solanum scabrum* and *Solanum villosum* Species at Reproductive Stage

Nitrogen rates (CAN g/plant)	% Protein	
	Solanum scabrum	*Solanum villosum*
0	3.39 a	3.27 a
0.9	4.34 a	4.33 ab
1.8	5.88 b	6.23 b
3.6	8.27 c	8.60 c
$LSD_{0.05}$	1.17	4.29

Means followed by same letter(s) are not significantly different at ($P \leq 0.05$)

4.3 Effect of Sources of Nitrogen on Growth, Yield, Quality and NUE of African Nightshade

4.3.1 Effect of Sources of Nitrogen on the Height of African Nightshade

At the first, second and third collection on leaf number no significant difference in number of leaves due to nitrogen sources was observed among the *Solanum scabrum* and *Solanum villosum* species. At fourteen weeks after treatment application, source of nitrogen was seen to have a significant effect on plant height at $P \leq 0.05$ in comparison to the control (Table 4.8). However, the difference in height between the different nitrogen sources was not significantly different at $P \leq 0.05$.

Table 4.8*: Effect of Sources of Nitrogen on Protein Content in Solanum *scabrum* and Solanum *villosum* Species at Reproductive Stage*

Source of Nitrogen	Plant height (cm)	
	Solanum scabrum	***Solanum villosum***
Control	61.76 a	56.55 a
Organic	69.58 b	66.80 b
CAN	71.87 b	67.50 b
Urea	72.11 b	68.77 b
$LSD_{0.05}$	5.51	4.17

Means followed by same letter(s) are not significantly different at ($P \leq 0.05$)

4.3.2 Effect of Sources of Nitrogen on the Number of Leaves of African Nightshade

At the first, second and third collection on leaf number no significant difference in number of leaves due to nitrogen sources was observed among the *Solanum scabrum* and *Solanum villosum* species. At fourteen weeks after treatment application, source of nitrogen was seen to have a significant effect on plant height at $P \leq 0.05$ (Table 4.9). In both *Solanum scabrum* and *Solanum villosum* species plants supplied with CAN and urea had the highest leaf number these two treatments had no significant difference.

Table 4.9: Effect of Sources of Nitrogen on Number of Leaves in Solanum *scabrum* and Solanum *villosum* Species at Reproductive Stage

Source of Nitrogen	**No. of leaves**	
	Solanum scabrum	*Solanum villosum*
Control	64.08 a	58.00 a
Organic	69.92 a	68.33 a
CAN	74.00 b	73.67 b
Urea	77.08 b	79.17 b
$LSD_{0.05}$	5.811	12.38

Means followed by same letter(s) are not significantly different at ($P \leq 0.05$)

4.3.4 Effect of Sources of Nitrogen on the Leaf Area of African Nightshade

Sources of nitrogen significantly influenced leaf area at $P \leq 0.05$ among the *Solanum scabrum* and *Solanum villosum* species. Plants supplied with CAN and urea had larger leaf area compared to plants in the control group and those supplied with organic fertilizer at $P \leq 0.05$. There was however no significant difference in leaf area in plants supplied with CAN and Urea in both species (Fig 4.5).

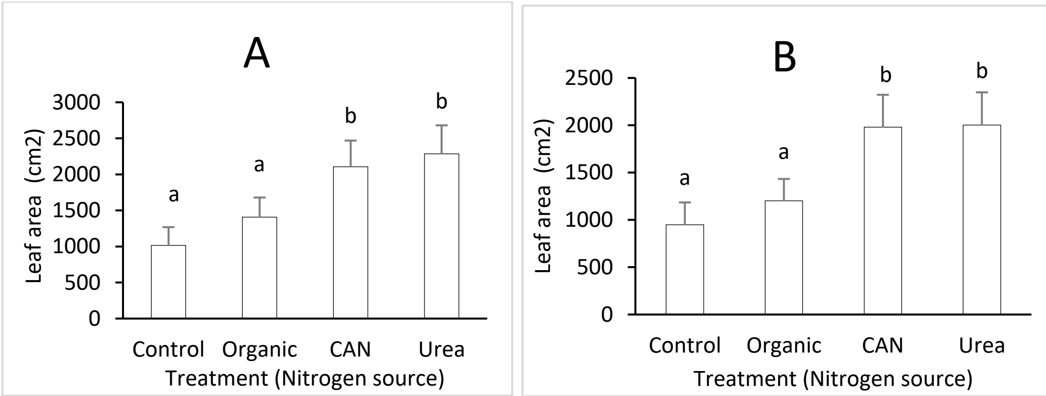

Figure 4.5: Leaf Area of African Nightshade Due to Sources of Nitrogen (A) *Solanum scabrum* Spp (B) *Solanum villosum* Spp

Vertical bars with same letter are not significantly different at ($P \leq 0.05$)

4.3.5 Effect of Sources of Nitrogen on the Yield of African Nightshade

Sources of nitrogen significantly influenced leaf yield at $P \leq 0.05$ in both *Solanum scabrum* and *Solanum villosum* species. Plants supplied with CAN and Urea had more yields in comparison to plants supplied with organic fertilizer and the control. The difference in yield between plants supplied with CAN and Urea was not significant at $P \leq 0.05$ in both species (Fig 4.6).

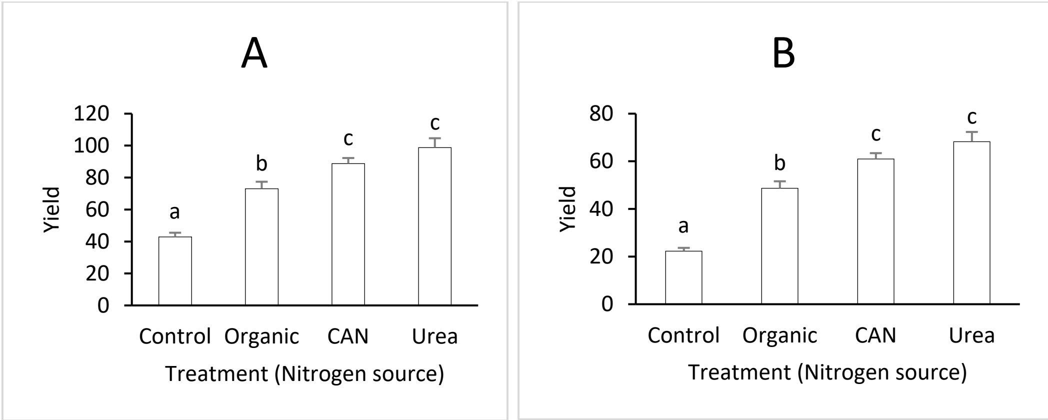

Figure 4.6: Leaf yield African Nightshade Due to Sources of Nitrogen (A) *Solanum scabrum* Spp (B) *Solanum villosum* Spp

Vertical bars with same letter are not significantly different ($P \leq 0.05$)

4.3.6 Effect of Sources of Nitrogen on the NUE of African Nightshade

Source of Nitrogen had an effect on the NUE of African nightshade. There was a significant difference in NUE at $P \leq 0.05$ in both *Solanum scabrum* and *Solanum villosum* species with plants in the control group and those supplied with organic fertilizer having a higher NUE compared to plants supplied with CAN and urea (Fig 4.7).

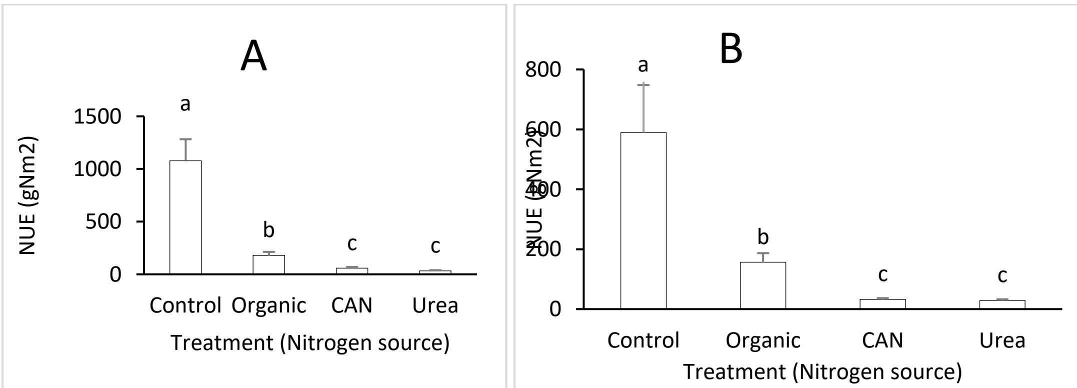

Figure 4.7: NUE of African Nightshade Due to Sources of Nitrogen (A) *Solanum scabrum* Spp (B) *Solanum villosum* Spp

Vertical bars with same letter are not significantly different ($P \leq 0.05$)

4.3.7 Effect of Sources of Nitrogen on Vegetable Quality of African Nightshade

4.3.7.1 Effect of Sources of Nitrogen on Nitrate Content of African Nightshade at Vegetative Stage

Source of nitrogen had an effect on the nitrate content in both *Solanum scabrum* and *Solanum villosum* species at the vegetative stage. In the *Solanum scabrum* species plants supplied with Urea fertilizer had significantly higher nitrate content at $P \leq 0.05$ compared to plants supplied with organic fertilizer and the control. The difference in nitrate content in plants supplied with organic fertilizer and plants supplied with CAN was not significant. In the *Solanum villosum* species plants supplied with urea had significantly higher nitrate content at $P \leq 0.05$ compared to plants supplied with CAN and organic fertilizer (Table 4.10).

Table 4.10: Effect of Sources of Nitrogen on Nitrate Content in *Solanum scabrum* and *Solanum villosum* Species at Vegetative Stage

Source of Nitrogen	Nitrate content (mg/kg)	
	Solanum scabrum	***Solanum villosum***
Control	3.87 a	5.01 a
Organic	4.48 b	6.46 b
CAN	4.81 bc	8.54 c
Urea	5.31 c	5.56 ab
$LSD_{0.05}$	0.56	1.39

Means followed by same letter(s) are not significantly different at ($P \leq 0.05$)

4.3.7.2 Effect of Sources of Nitrogen on Nitrate Content of African Nightshade at Reproductive Stage

Sources of nitrogen had an effect on the nitrate content in both *Solanum scabrum* and *Solanum villosum* species at the reproductive stage. The results indicated a significant difference at $P \leq 0.05$ in nitrate content among the *Solanum scabrum* and *Solanum villosum* species with plants supplied Urea having the highest nitrate content (Table 4.11). Generally, an increase in nitrate content was observed across all treatments at reproductive stage in comparison to the vegetative stage.

Table 4.11: Effect of Sources of Nitrogen on Nitrate Content in *Solanum scabrum* and *Solanum villosum* Species at Reproductive Stage

Source of Nitrogen	Nitrate content (mg/kg)	
	Solanum scabrum	***Solanum villosum***
Control	9.13 a	7.24 a
Organic	10.81 ab	8.62 b
CAN	12.68 b	9.89 c
Urea	9.89 a	8.17 ab
$LSD_{0.05}$	2.43	1.07

Means followed by same letter(s) are not significantly different at ($P \leq 0.05$)

4.3.7.3 Effect of Sources of Nitrogen on Protein Content of African Nightshade at Vegetative Stage

The source of Nitrogen had an effect on the protein quantities at the vegetative state in both African nightshade species. The results indicated a significant difference in protein content at $P \leq 0.05$ in the *Solanum scabrum* and *Solanum villosum* species due to source of nitrogen. In both species' plants supplied with Urea had a higher protein content at vegetative stage in comparison to the other treatments (Table 4.12)

Table 4.12: Effect of Sources of Nitrogen on Protein Content in *Solanum scabrum* and *Solanum villosum* Species at Reproductive Stage

Source of Nitrogen	% Protein	
	Solanum scabrum	***Solanum villosum***
Control	3.39 a	3.27 a
Organic	4.35 a	4.34 ab
CAN	5.88 b	6.23 b
Urea	8.20 c	8.60 c
$LSD_{0.05}$	1.16	1.92

Means followed by same letter(s) are not significantly different at ($P \leq 0.05$)

4.3.7.4 Effect of Sources of Nitrogen on Protein Content of African Nightshade at Reproductive Stage

The source of Nitrogen had an effect on the protein quantities at the reproductive state in both African nightshade species. The results indicated a significant difference in protein content at $P \leq 0.05$ in the *Solanum scabrum* and *Solanum villosum* species due to source of nitrogen. In both species' plants supplied with Urea had a higher protein content at vegetative stage in comparison to the other treatments (Table 4.13)

Table 4.13: Effect of Sources of Nitrogen on Protein Content in *Solanum scabrum* and *Solanum villosum* Species at Reproductive Stage

Source of Nitrogen	**% Protein**	
	Solanum scabrum	***Solanum villosum***
Control	1.90 a	1.64 a
Organic	2.56 a	2.52 ab
CAN	3.31 b	3.47 b
Urea	4.41 c	4.52 c
$LSD_{0.05}$	1.23	1.00

Means followed by same letter(s) are not significantly different at ($P \leq 0.05$)

CHAPTER FIVE

DISCUSSION

5.1 Effect of Nitrogen Application Rates on the Plant Growth and Yield of African Nightshade

Response of plant growth to different nitrogen rates is a topic that has been extensively studied by other researchers and the results indicate that increasing nitrogen rates has a positive impact on plant growth and development. Similarly, the results of this experiment indicated a positive response in plant growth with plants supplied with 3.6 g of CAN being taller, having more leaves, fresh leaf yield and a larger leaf area. Mohamed (2011) indicated that increase in plant height with nitrogen fertilizer is due to its effect on cell division and expansion this increases the number of internodes and length of internodes which results in progressive increase in height. In most instances, more internodes usually mean more leaves, which increases the overall leaf area. This larger leaf area enhances the plant's ability to capture sunlight and conduct photosynthesis, leading to greater energy production (Hikosaka, 2004).More internodes and leaves also enhance the plant's ability to utilize absorbed nutrients effectively, leading to better growth and higher yields (Marschner, 2012). It was observed that *Solanum scabrum* species that were supplied with 3.6g of CAN had an extensive root system. This observation was in agreement with Lynch (1995), on root development and architecture which indicated that nitrogen can promote root growth allowing taller plants to have a deeper root system.

From the results it is evident that increasing the rate of nitrogen application has a positive response in the number of leaves and consequently the fresh leaf yield at the end of the experiment. This is because adequate nitrogen availability promotes robust vegetative growth, including an increase in the number of leaves (Marschner, 2012). Plants that were in the control group and those supplied with 1.8g of CAN exhibited signs of chlorosis, stunted growth and premature senescence of leaves through out the experiment .This observation can be attributed to the lack of nitrogen, which is a critical nutrient that plays a mjor role in chlorophyll production that is essential for photosynthesis. Deficiency of nitrogen leads to stunted growth, as the plant lacks the necessary nutrients for cell division and expansion (Zhao et al., 2004). The effect of nitrogen fertilizer application on leaf area was evident in the *Solanum scabrum* species where plants supplied with 3.6g of CAN had the largest leaf area. With an increased leaf area, vegetables can capture more sunlight which enhances their photosynthetic capacity (Zhu et al., 2010). In *Solanum villosum* species, leaf area was largest at 1.8g of CAN which suggested a threshold beyond which additional nitrogen does not impact leaf growth in this species.

Plants supplied with 3.6g of CAN in both African Nightshade species had a higher leaf number which translated into high fresh leaf yields at the end of the experiment. This results are in agreement with studies conducted on spinach where the results showed that that increasing nitrogen rates can enhance the fresh leaf yield of spinach. Higher nitrogen availability promotes vegetative growth and leaf biomass (Salehi & Kashi, 2001). Plants supplied with 0.9g CAN and plants in the control group experienced premature leaf senescence which impacted its fresh leaf yield at the end of the experiment.This observation was expected because nitrogen deficiency affects plant growth by reducing the photosynthetic assimilating area, which decreases light interception and dry matter production (Zhao et al., 2004).

5.2 Effect of Nitrogen Application Rates on the Vegetable Quality of African Nightshade

Nitrogen fertilizer significantly influences the quality of vegetables particularly in terms of protein content and nitrate accumulation. In our study the results indicated that increasing nitrogen rates significantly increased protein content with plants supplied with 3.6g of CAN having a higher protein content in both the *Solanum scabrum* and *Solanum villosum* species at the vegetative stage. This observation can be attributed to nitrogen's role as a crucial component of amino acids the building blocks for proteins (Kalaji et al., 2016). Normally,adequate nitrogen fertilization leads to a higher protein content in vegetables as nitrogen availability directly influences synthesis of amino acids and proteins (Lawlor, 2002). Additionally, increase in protein content due to nitrogen fertilization can vary among different vegetable species with leafy vegetables showing a more pronounced increase in protein content compared to fruiting vegetables (Watanabe & Osaki, 2002). Although plants supplied with 3.6g of CAN still had the highest protein content in comparison to the other treatments at reproductive stage, a significant reduction in protein content was evident across all treatments from the vegetative to the reproductive stage. This is because during the vegetative growth phase, plants actively assimilate nitrogen to support the development of leaves, stems, and roots. High protein content is typically observed in leafy vegetables harvested during this stage (Marschner,2012). As plants transition to the reproductive stage, the allocation of nutrients, including nitrogen, shifts towards the production of flowers, fruits, and seeds. Protein synthesis may decrease in vegetative tissues as resources are redirected. This shift can lead to a reduction in protein content in leaves and stems (Mengel, et al., 2001).

Nitrate content of the plants in this study was significantly affected by increasing the nitrogen rates in both the *Solanum scabrum* and *Solanum villosum species* with plants supplied with 3.6 g of CAN a higher nitrate content at the vegetative stage.The nitrate content in all treatments at the vegetative stage was below the recommeded daily allowance of nitrate which is 3.7mg/kg which indicated good vegetable quality. Normally, during the vegetative stage, plants are highly active in photosynthesis and growth, which increases their demand for nitrogen. As a result, the nitrogen taken up is quickly assimilated into amino acids, proteins, and other nitrogen-containing compounds, leading to lower nitrate accumulation in plant tissues.

At the reproductive stage, nitrate content was highest in plants supplied with 3.6 g CAN however there was a visible increase in nitrate content across all treatments in the reproductive stage. Like most leafy vegetables African nightshade is prone to accumulation of phytochemicals like nitrates hence their quality is significantly affected by the amount of fertilizer applied (Mazahar et al 2015). Typically, when nitrogen is applied in excess of what the plant requires for protein production, it is accumulated as nitrates and stored predominantly in the green leafy part of the plant (Undie et al.,2012). Physiological age of the plants plays a huge role in nitrate accumulation with young actively growing plants having higher nitrate uptake and conversion rates due to the demands of cell division, elongation and setup of basic plant structure (Umar et al.,2007). Nitrate by itself has been shown to be relatively non-toxic to humans however, it can be transformed to nitrite endogenously, nitrite reacts with amines and amides to produce N-nitroso compounds. These compounds have been associated with increased susceptibility to diseases a factor which has led to increased concern about nitrates in vegetables (Santamaria, 2006)

5.3 Effect of Nitrogen Application Rates on the NUE of African Nightshade

The rate of nitrogen application significantly affected the nitrogen use efficiency of African nightshade as evidenced by the results of this study. The control group had the highest NUE followed by plants supplied with 0.9g of CAN. NUE is defined as the ratio of crop yield in relation to the amount of fertilizer applied. It indicates how effectively a plant converts nitrogen inputs into harvestable output (de Jesus et al., 2024). The high NUE observed in the control group indicates that plants in the control group were able to efficiently use the nitrogen available. The results of the initial potting media analysis indicated a pH of 6.02, CEC 14 cmolc/kg, EC 0.07 dS/m and soil nitrogen content of 0.05%. This soil parameters can significantly influence the NUE of applied fertilizers. The potting media`s CEC could have influenced the NUE of CAN since CAN has equal parts ammonium nitrogen and nitrate nitrogen hence the potting media only had a moderate ability to retain ammonium cations hence the nitrate anions had a potential to leach (Havlin et al., 2013).This means that part of the applied CAN could have potentially leached hence reducing the NUE. The low soil Electrical conductivity of potting media indicated the presence of minimal soluble salts in the soil this may have caused reduced efficiency in applied nitrogenous fertilizers. The inefficiency is due to the lack of necessary ions to retain and deliver nitrogen to plant roots which reduces utilization leading to deficiency in the plant (Biederbeck, et al 1986).High temperatures in the greenhouse could have also contributed to ammonia volatilization and increased nitrification, resulting in nitrogen loss and poor NUE. This is because at higher temperatures, the equilibrium between ammonium (NH_4^+) in the soil and ammonia gas (NH_3) shifts towards the gaseous form, increasing the rate of volatilization (Wang, et al., 2017).

Reduced NUE with increasing nitrogen supply as observed can also be due to the decreased Rubisco activity which is induced by relatively insufficient carbon-dioxide supply caused by high nitrogen supply. Normally, high nitrogen supply increases the synthesis of Rubisco and other photosynthetic proteins which enhances the plants photosynthetic capacity, however if the carbon dioxide supply is insufficient to meet the demand, the rate of carbon fixation is limited leading to a buildup of unused nitrogen within the plant tissue. This results in decreased NUE as the nitrogen absorbed by the plant is not effectively converted to biomass (Xu et al., 2012).

It has been observed in several studies that higher N application rates especially above the optimum recommended rates results in lower NUE (Wortmann, et al., 2011). This is because plants have a limited capacity to absorb and utilize nitrogen, hence additional nitrogen beyond a certain point does not translate to proportional increase in yield. This saturation point results in diminishing returns for nitrogen uptake efficiency, lowering overall NUE at higher application rates (Moll et al., 1982).

5.4 Effect of Nitrogen Source on the Plant Growth and Yield of African Nightshade

The choice between organic and inorganic nitrogen fertilizers can significantly impact vegetable growth. Both types of fertilizers provide essential nutrients, but they differ in nutrient content and release rates. In this study, it was observed that plants supplied with fertilizer in the form of urea and CAN had better plant growth compared to plants supplied with organic fertilizer and plants in the control group. This observation can be attributed to the fact that inorganic fertilizers such as urea and CAN are synthesized chemically hence they provide readily available nutrients leading to rapid responses. On the other hand the release of N from organic sources depends on the rate of decomposition and mineralization which are largely influenced by soil moisture, temperature and carbon to nitrogen ratio of the organic material (Berry et al., 2006). The release of nitrogen from inorganic sources is also predictable which allows for precise timing to match crop needs.In organic fertilizer, the initial burst of nitrogen may not coincide with the peak nitrogen demand during plant growth.As a result the nitrogen supplied by the organic source might have been depleted by the time the plant reached critical growth stages hence leading nitrogen deficiencies and reduced vegetable yields (Gaskell, 2007).

The results of this study are in agreement with the results of a similar study conducted in Spain on lettuce that demonstrated that inorganic fertilizer led to higher yields than organic amendments. The study attributed the yield difference to the immediate availability and consistent supply of nitrogen from inorganic sources which matched the crops nitrogen demand effectively (Salomez et al., 2013).

5.5 Effect of Nitrogen Source on Vegetable Quality of African Nightshade

The source of nitrogen used in vegetable farming can significantly impact the quality of the vegetables, particularly in terms of their protein content and nitrate accumulation. In this study plants supplied with urea in the *Solanum scabrum* and *Solanum villosum* species had the highest nitrate content. This observation can be attributed to the rapid conversion of urea to nitrate, higher nitrogen content of urea, and the nature of its transformation process which leads to higher nitrate accumulation in plants compared to the more balanced and gradual nitrogen release from CAN and organic fertilizers (Feng et al., 2020).

This result agrees with a similar study by Santamaria et al. (2006) where the results indicated that leafy vegetables like lettuce & spinach tend to accumulate more nitrates when supplied with inorganic nitrogen sources compared to organic sources. The rapid availability of nitrogen from inorganic fertilizers exceeded the plants' immediate uptake capacity, resulting in higher nitrate levels in the plant tissues while the gradual nitrogen release from organic fertilizer matched the lettuce's nitrogen uptake patterns more effectively, reducing the risk of nitrate accumulation. At the reproductive stage plants supplied with CAN had the highest nitrate content in both African nightshade species however an increase in nitrate content was observed across all treatments at the reproductive stage. This observation is attributed to the physiological age of the plant which is a key factor in nitrate accumulation with young actively growing plants having higher nitrate uptake and conversion rates due to the demands of cell division, elongation and setup of basic plant structure (Umar et al., 2007).

Protein content of the vegetables were seen to be highly influenced by source of nitrogen with plants supplied with urea having the highest protein content followed by CAN in both African nightshade species. This observation is in line with the results of a study on spinach (*Spinacia oleracea*) by Wang et al.(2014) where the application of urea resulted in higher protein content compared to composted manure. The rapid nitrogen availability from urea supported immediate protein synthesis, while the slow release from compost did not meet the plants' peak nitrogen demand as effectively. Another study by Xie et al. (2016) on lettuce *(Lactuca sativa)* found that the application of CAN during the rapid growth phase significantly increased protein content compared to the use of compost. It was observed that the timing of nitrogen availability from CAN matched the lettuce`s peak growth needs. At the reproductive stage there was a significant decrease in protein content across all treatments which was attributed to physiological age of the crop. As plants transitioned to the reproductive stage, the allocation of nutrients, including nitrogen, shifted towards the production of flowers, fruits, and seeds. Protein synthesis may have decreased in vegetative tissues as resources are redirected. This shift can lead to a reduction in protein content in leaves and stems (Mengel et al., 2001).

5.6 Effect of Nitrogen Source on NUE of African Nightshade

Nitrogen use efficiency varies significantly depending on the source of nitrogen used. In this study plants that were in the control group had the highest NUE followed by plants supplied with organic fertilizer. This means that the plants in the control group and plants supplied with organic fertilizer were able to efficiently utilize available nitrogen. Nitrogen use efficiency in inorganic fertilizers such as urea and CAN is often reduced due to the rapid availability of nitrogen in inorganic.This quick release of nitrogen lead to high initial NUE especially in crops with rapid nitrogen demand. However, due to inorganic nitrogen sources susceptibility to losses through volatilization, leaching, and denitrification, the overall NUE is reduced (Havlin et al., 2013).For instance urea which was one of the sources of nitrogen used in this study can volatilize as ammonia gas if not properly incorporated into the soil this can lead to reduced NUE. Also nitrate-based fertilizers like CAN are prone to leaching, especially in sandy soils or areas with high rainfall, leading to groundwater contamination and reduced NUE. In comparison organic nitrogen sources release nitrogen more slowly as the nitrogen is bound in organic matter and must be mineralized by soil microorganisms before it becomes available to plants. This gradual release can result in lower initial NUE but potentially higher long-term NUE as nitrogen is released more consistently over the growing season and is less prone to losses (Zhou et al., 2017).Organic fertilizers also add other benefits to the soil like improving soil structure. By increasing the soil organic matter content, the soil water-holding capacity, aeration, and root growth is improved which promotes more efficient nutrient uptake by plants hence increasing NUE.

Over the years, there are several studies that have compared the NUE of organic and inorganic nitrogen sources in various plants. For instance, a study by Drinkwater et al. (1998) reported that organic farming systems, which rely on organic nitrogen sources, had lower immediate NUE but better long-term soil fertility, sustainability and NUE compared to conventional systems using inorganic fertilizers. Another a study by Zhou et al. (2017) found that integrating organic and inorganic fertilizers resulted in higher NUE and crop yields compared to using either source alone. This synergistic effect is attributed to the complementary release patterns of nitrogen from the different sources, improving both immediate and long-term nitrogen availability.

CHAPTER SIX

CONCLUSIONS AND RECOMMENDATIONS

From this study it can be deduced that nitrogen application rates affects plant growth, yield and nitrogen use efficiency of African nightshade. High nitrogen rates (3.6g of CAN per plant) significantly enhanced plant growth parameters (plant height, leaf number, and leaf area) and yield (marketable leaves) in *Solanum spp* and *Villosum spp*. However, excessive nitrogen levels lead to reduced nitrogen use efficiency, as the yield does not proportionately increase with the applied nitrogen amount. This indicates that there is an optimal nitrogen rate beyond which the benefits diminish.

The source of nitrogen was also shown to have an effect on the plant growth yield and nitrogen use efficiency of African nightshade. The results indicate that plants supplied with inorganic nitrogen exhibited significantly higher growth and yield in the short term compared to those receiving organic nitrogen. Specifically, the inorganic nitrogen sources (CAN and urea) promoted rapid increases in plant height, leaf number, and leaf area, leading to a greater production of marketable leaves. This suggests that inorganic nitrogen is more readily available to plants, facilitating quicker uptake and immediate growth responses. However, while inorganic nitrogen enhances short-term yield and growth, its long-term effects on soil health and sustainability should be considered. Plants supplied with organic nitrogen exhibited higher NUE, meaning they produced more yield per unit of nitrogen applied. This improved efficiency is likely due to the gradual release of nutrients from organic sources, allowing for a more sustained uptake and utilization by the plants. These findings suggest that organic nitrogen sources are more effective in promoting sustainable and efficient nitrogen use in African nightshade cultivation, making them a preferable choice for enhancing productivity and environmental conservation.

Both the nitrogen rates and source of nitrogen have an effect on the nutritional quality of African nightshade (proteins and nitrates). High rates of nitrogen fertilizer application increase protein content but also raise nitrate levels in the vegetable tissue at both vegetative and reproductive stages of *Solanum scabrum* and *Solanum villosum* species. This is due to nitrogen`s role as building blocks for proteins, plants supplied with 3.6 g of CAN had the highest protein content at both vegetative and reproductive growth stages. However, due to the nitrate accumulating nature of leafy vegetables, plants supplied with 3.6 g of CAN also had the highest nitrate content at both vegetative and reproductive stages. The type of nitrogen source also impacts quality, with organic sources typically resulting in higher protein content and lower nitrate accumulation compared to synthetic sources. This highlights the importance of selecting appropriate nitrogen sources and managing application rates to optimize both nutritional value and safety.

Based on the results of this study it is recommended that farmers use nitrogen rate of 3.6 g of Calcium Ammonium Nitrate (CAN) per plant which is equivalent to 400kg of CAN per HA to improve the growth and yield of African nightshade. However, caution should be exercised due to increased nitrate levels and reduced nitrogen use efficiency. To improve NUE farmers should use split fertilizer application strategy instead of using one single application so as to supply small doses of nitrogen throughout the growing season. This reduces nitrogen losses and matches the plant's nitrogen demand more closely. Proper timing of nitrogen fertilizer application is also a key consideration since actively growing plants can absorb and utilize available nitrogen efficiently. Farmers should also incorporate organic nitrogen sources alongside CAN to improve NUE and reduce nitrate accumulation. Organic sources release nitrogen more slowly, promoting sustained plant growth and better nitrogen utilization. This approach leverages the immediate benefits of inorganic fertilizers and the long-term advantages of organic amendments.

6.1 Areas for Further Research

The study established that nitrogen fertilizer rates influences the yield and quality of African nightshade species. Therefore, there is need for further research to come up with species specific nitrogen fertilizer recommendations due to the morphological and physiological differences between *Solanum scabrum* and *Solanum villosum* species.

Since we have established that organic and inorganic sources of fertilizer affect NUE of African nightshade in a controlled environment, further research should be done to compare the results of the controlled environment to a field environment so as to validate findings and understand the influence of environmental factors on NUE.

Investigate the optimal timing for both organic and inorganic nitrogen applications so as to maximize nitrogen use efficiency (NUE) and yield of African nightshade. This includes examining the effects of split applications and developing season-specific strategies.

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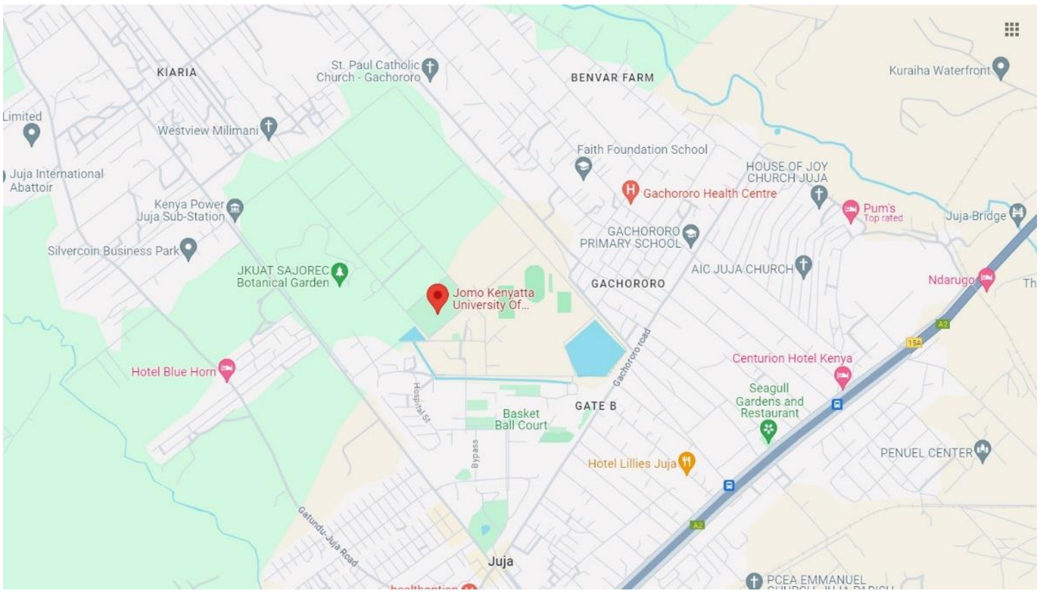
APPENDICES

Appendix I: Performance of African Indigenous Vegetables 2019-2020

Crop	2019			2020			% Of Total Value
	Area (Ha)	Volume (MT)	Value (KES)	Area (Ha)	Volume (MT)	Value (KES)	
Cowpea	79,535	159,386	3,512,308,830	36,018	113,666	3,348,701,203	**37.4**
African Nightshade	6,950	69,254	2,397,810,725	5,917	58,909	1,831,009,726	**20.5**
Spider Plant	4,280	35,295	1,229,098,895	3,949	36,445	1,315,530,681	**14.7**
Leaf Amarantha	3,996	54,813	1,322,286,150	3,237	38,172	831,076,886	**9.3**
Pumpkin Fruits	1,487	31,022	722,892,977	1,755	35,829	813,596,889	**9.1**
Sleder	355	7,107	350,836,860	841	5,605	260,730,596	**2.9**
Pumpkin Leaves	903	6,650	147,623,496	900	6,172	158,159,801	**1.8**
Jute Mallow	672	5,894	309,079,967	657	3,373	155,029,483	**1.7**
Grain Amarantha	453	3,020	178,728,617	511	2,459	127,453,939	**1.4**
Russian Confrey	75	644	19,460,000	163	1,354	50,321,660	**0.6**
Vine Spinash	193	811	33,530,230	217	1,030	29,476,001	**0.3**
Malabor	41	593	27,780,000	70	652	23,344,245	**0.3**
Total	**98,940**	**374,489**	**10,251,436,747**	**54,235**	**303,666**	**8,944,431,110**	**100.0**

Source: AFA-Horticulture Crops Directorate

Appendix II: Geographical Map of Study Site



Appendix III: Experimental Layout

$T_4V_5R_4$		$T_4V_2R_2$		$T_3V_3R_1$		$T_2V_4R_3$
$T_3V_2R_4$		$T_3V_2R_2$		$T_4V_2R_1$		$T_3V_1R_3$
$T_4V_2R_4$		$T_3V_4R_2$		$T_2V_2R_1$		$T_3V_3R_3$
$T_3V_5R_4$		$T_2V_3R_2$		$T_1V_3R_1$		$T_4V_1R_3$
$T_2V_3R_4$		$T_2V_5R_2$		$T_1V_4R_1$		$T_4V_3R_3$
$T_3V_3R_4$		$T_1V_3R_2$		$T_3V_2R_1$		$T_3V_2R_3$
$T_1V_4R_4$		$T_1V_5R_2$		$T_2V_3R_1$		$T_2V_3R_3$
$T_1V_2R_4$		$T_2V_4R_2$		$T_4V_3R_1$		$T_1V_2R_3$
$T_2V_4R_4$		$T_2V_1R_2$		$T_1V_2R_1$		$T_1V_4R_3$
$T_4V_1R_4$		$T_3V_3R_2$		$T_4V_4R_1$		$T_1V_5R_3$
$T_1V_1R_4$		$T_4V_1R_2$		$T_1V_1R_1$		$T_2V_5R_3$
$T_4V_3R_4$		$T_4V_3R_2$		$T_2V_1R_1$		$T_2V_1R_3$
$T_1V_3R_4$		$T_3V_5R_2$		$T_4V_5R_1$		$T_3V_4R_3$
$T_2V_1R_4$		$T_4V_5R_2$		$T_4V_1R_1$		$T_4V_2R_3$
$T_3V_IR_4$		$T_2V_2R_2$		$T_2V_4R_1$		$T_4V_4R_3$
$T_2V_5R_4$		$T_4V_4R_2$		$T_3V_4R_1$		$T_3V_5R_3$
$T_2V_2R_4$		$T_1V_4R_2$		$T_3V_1R_1$		$T_4V_5R_3$
$T_3V_4R_4$		$T_1V_1R_2$		$T_3V_5R_1$		$T_1V_3R_3$
$T_4V_4R_4$		$T_3V_1R_2$		$T_2V_5R_1$		$T_2V_2R_3$
$T_1V_5R_4$		$T_1V_2R_2$		$T_1V_5R_1$		$T_1V_1R_3$

Key

T= Treatment

V= Variety

R= Replication