

# COMPARATIVE EFFECTS OF BIOSLURRY AND CHEMICAL FERTILIZER ON SOIL PROPERTIES AND PERFORMANCE OF SPIDER PLANT (Cleome gynandra L).

A Dissertation submitted in partial fulfillment of the requirement for the award of a Master of Science Degree in Crop Science (Horticulture)

By

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Dean of Faculty's name and signature	Date

## **STATEMENT OF ORIGINALITY**

I declare that the materials included in this dissertation was put together by the author at the Botswana University of Agriculture and Natural Resources between 2021 and 2023. As far as I am aware, this dissertation is not submitted to any other university or organisation for the award of any certificate, diploma or degree and every source of information utilised to compile this dissertation has been duly credited.

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Author's signature

Date

# DEDICATION

This dissertation is devoted to my mother, siblings and friends for their endless prayers and support throughout this journey. To my young cousins, nephews and nieces, you are my greatest love, may you always believe in the boundless possibilities that await you. This achievement is dedicated to you.

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	Table of Contents
CERT	rificationi
APPF	ii iii
STAT	TEMENT OF ORIGINALITYiii
DEDI	ICATIONiv
ACK	NOWLEDMENTS v
List o	f Acronyms x
ABST	r <b>RACT</b> xi
СНАРТ	<b>ER 1</b> 1
INTRO	<b>DUCTION</b> 1
1.1	Background1
1.2	Statement of the problem6
1.2.	1 Justification7
Resea	arch question8
1.3	Objective9
1.3.	<i>1 Main objective9</i>
1.4	Study hypotheses9
СНАРТ	' <b>ER 2</b>
LITERA	ATURE RIVIEW
2.1	Introduction11
2.2	Crop Description11
2.2.	<i>Environmental factors affecting the growth of Cleome gynandra L.</i> 13
2.3	Nutrient depletion15

# **Table of Contents**

2.4	Nutrient reple	enishment	16
2.5	Integrated nut	trient management	18
2.6 Nitr	ogen Use Efficie	ency (NUE)	19
2.7	7.1 Nitrogen -		20
2.6	5.2 Phosphori	US	22
2.6	5.3 Potassium	1	22
2.8	Use of fertilize	ers in crop production	24
2.9	Limitations of	Bioslurry as a fertilizer	43
CHAP	<b>FER 3</b>		44
MATE	RIALS AND ME	ETHODS	44
3.1	Study site desc	cription	44
3.1	.1 Planting n	naterials and treatments	44
3.2	Experimental	description	45
3.2	.1 Greenhou	se experiment	45
3.2	2.2 Field Expe	eriment	46
3.3 D	ATA COLLECT	ГІОN	47
3.3	2.1 Climate of the	experimental site	47
3.3	3. Soil sampl	ling and Analysis	48
3.3	2.4 Soil carbon see	questration and carbon dioxide $(CO_2)$ emission	determination51
3.3	.6 Plant nutritio	onal analysis	54
3.4	Statistical ana	lysis	55
СНА	PTER 4		56

<b>RESULTS</b> 56
4.1. Effect of Bioslurry and Urea fertiliser on Soil Physiochemical Properties59
4.1.1 Effect of Bioslurry and Urea fertiliser on Soil Chemical Properties after spider plant
harvest59
4.2. Quantitative traits of spider plant as influenced by bioslurry and urea fertiliser.
67
4.2.1. Influence of Bioslurry and Urea fertiliser on plant height, leaf number and leaf with
petiole length70
4.2.2. Number of days to 50% emergence and 50% flowering as affected by Bioslurry and
Urea application71
4.2.3. Bioslurry and Urea fertiliser effects on the chlorophyll content and stomatal
conductance of spider plant73
4.2.4. Effects of Bioslurry and Urea fertiliser on Nitrogen Use Efficiency of Spider plant
Accessions76
4.3 CO <sub>2</sub> Emissions as affected by Bioslurry and Urea fertiliser application77
4.4 Effects of Bioslurry and Urea fertiliser on Plant mineral content79
4.5 Assessment of optimum application rates of Bioslurry and urea fertiliser using
different growth and yield components of spider plant83
CHAPTER 585
DISCUSSION85
5.1. Soils of the study areas85
5.2. Bioslurry nutrient composition analysis86

5.3. Quantitative traits of spider plant as influenced by bioslurry and urea fertilis	er.
	86
5.4. Effect Bioslurry and Urea on CO2 emissions1	00
5.5 Effect of Bioslurry and Urea fertiliser on Soil Physiochemical Properties 1	04
5.6 Effect of fertilisation on Nutrient Content of Spider plant	11
5.7 Bioslurry and Urea application rates for optimum growth and yield of Spid	ler
<b>plant</b> 1	13
<b>CHAPTER 6</b> 1	15
CONCLUSION AND RECOMMENDATIONS	15
<b>6.1 Conclusion</b> 1	15
6.2 Recommendations 1	15
CHAPTER 7 1	16
REFERENCE1	16
Appendix 11	75

CSA	- Climate Smart Agriculture
FAO	- Food and Agricultural Organisation of the United Nations
GHG	- Green House Gases
ISFM	- Integrated Soil Fertility Management
K	- Potassium
Ν	- Nitrogen
Р	- Phosphorus
UNDP -	United Nations Development Programme
UNEP -	United Nations Environmental Programme
FAO	- Food Agricultural Organisation
NUE	- Nitrogen Use Efficiency
BS	- Bioslurry
CF	- Chemical Fertilizer
SOC	- Soil Organic Carbon
TN	- Total Nitrogen
ALVs	- African Leafy Vegetables
GDP	-Gross Domestic Product

# List of Acronyms

#### ABSTRACT

Spider plant (*Cleome gynandra* L.) is a promising indigenous leafy vegetable in the diet of many African rural populations. However, soil fertility for production of spider plant remains an unexploited area leading to low crop yields. Bioslurry (BS) is a by-product of biogas production rich in organic matter, plant nutrients and a potential substitute for chemical fertilizer (CF). It can also be utilized as a soil amendment to restore soil health. Pot and field experiments were conducted to compare the effects of BS and CF on soil properties and performance of spider plant. A factorial combination of two landraces of spider plant were laid out in CRD and RCBD with three replications in the greenhouse and field, respectively. Treatments were landraces (Tot 89-26 and Rothwe), and N applied as Urea at 0, 60, 120, and 180kg N/ha and bioslurry at 0, 10, 20 and 30 ton/ha using a sandy loam soil.

Results revealed that fertilization with BS and CF significantly (p<0.05) improved plant growth parameters and the effect between the two fertilizers was not statistically different. The highest plant height (27.04cm), leaf number (23.39) and leaf petiole length (9.43cm) were recorded from BS20, CF60 and BS20 respectively compared to the control. Accession alone influenced leaf petiole length and leaf number. All fertilizer treatments significantly increased yield parameters. Furthermore, application rate of 120kg N /ha and 20 ton/ha BS recorded the highest chlorophyll content while high stomatal conductance was recorded from 30 ton/ha BS and the results were at par with BS20, CF60, CF120 and CF180. Spider plant efficiently used N when low rates of bioslurry and urea at 10 ton/ha and 60kg N/ha respectively were applied. Results showed a highly significant ( $p \le 0.001$ ) effect of fertilizer application on CO<sub>2</sub> emissions and carbon sequestration. High CO<sub>2</sub> emissions and low carbon sequestration were recorded from 30 ton/ha of Urea while low emissions and high carbon sequestration were recorded from 30 ton/ha of bioslurry. Various application rates of bioslurry and urea significantly influenced soil chemical properties while no significant effect was observed on soil physical properties. Overall, the findings of this study revealed that the impact of bioslurry and urea on growth and yield of spider plant were not significantly different. Thus, bioslurry can be used as an alternate nitrogen supply for spider plant cultivation. Bioslurry significantly improved soil properties, reduced  $CO_2$  emissions through carbon sequestration, hence promoting sustainable crop production.

Keywords: Carbon sequestration, Bioslurry, Soil properties, Spider plant

# CHAPTER 1 INTRODUCTION

#### 1.1 Background

The global population is anticipated to reach 9.8billion in 2050 (UN, 2017) and Botswana's population is expected to reach 2.8 million by 2030 and 3.4 million by the year 2050 (world bank group, 2021) and with this increasing population need to be fed. Agriculture is one of the economic sectors that is responsible for feeding this ever-increasing population. At the time of independence in 1966, the agriculture sector had a share of 42.7% of the Gross Domestic Product (GDP), but over the years, that contribution has steadily went down, to less than 2% as of now (Ministry of Finance and Development Planning, 2010). However, agriculture is a significant sector of Botswana's economy because it generates jobs, revenue, and food, primarily for rural area dwellers. It continues to have a negligible effect on the nation's ability to develop meaningfully and to feed its growing population, forcing farmers to cultivate their land intensively to increase crop yields per unit of land. It has experienced low performance attributed to environmental problems including land degradation driven by soil loss and nutrient depletion and impact on climate change. Furthermore, the fact that 70% of Botswana's landscape consists of desert (Botswana Economic and Political Overview, 2022) and 85% of the soil is sand, making poor soils extremely leached and lacking in nitrogen, phosphorus, calcium, and magnesium concentrations (Pule-Meulenberg and Dakora, 2007). The sector's poor performance adds to the difficulty of combating poverty. Soil fertility and climate are the main restraining factors in arable farming (CSO, 2008).

Climate change impose a significant risk to Botswana's economic development. The country vulnerability to the impacts of changes in climate across economic sectors is indicative from magnitude of stress on major drivers of GDP like biodiversity, water and agriculture (UNDP, 2010). Therefore, it is important for the country to develop its capacity in dealing with present

and future impacts of changes in climate to sustain its development and attain its poverty reduction goals. Managed agricultural soils have the capacity to retain carbon and thus help reduce greenhouse gases (GHG) emissions and elevate organic carbon in soils which also profit agricultural output through enhancing environmental quality and soil health by lowering soil runoff. Acting on carbon sequestration will not only stimulate important changes in land management but will also through improvement in organic matter levels have a notable direct impact on soil properties and a positive effect on environmental or agricultural quality and biodiversity. These consequences include increased soil fertility, land productivity for food production and food security (FAO, 2001). Mechanisms of stabilizing carbon in soils include utilization of reused organic materials such as biochar, biosolids and compost to boost soil organic matter reserves (Smith, 2008). Biogas digesters can lower GHG emissions by reducing the collection and burning of charcoal and firewood. Furthermore, soil organic matter losses through soil organic carbon decomposition and deforestation are reduced when biogas is used. Also, bio-digesters produce fertiliser in a way that leads to less emission than manufacture of synthetic fertilisers.

Food insecurity and malnutrition are a challenge in rural communities due to lack of resources to adopt improved crop production technologies such as application of fertilizers to enhance poor soils for crop production and drought stress (Legwaila et al., 2013). furthermore, medical personnel and nutritionist in Botswana showed a concern that decline in the utilization of indigenous edible plants lead to nutritional deficits (Madisa and Tshamekang, 1997). In Botswana rural communities rely on a variety of local food plants (indigenous edible plants) to support their lives to escape hunger in times of food crisis as these species provide alternative source of nutrition, medicine and cash income. According to Legwaila et al. (2013) majority of native food plants are comparable to their alien or domesticated counterparts as they have more nutrient content and are abundant in zinc, vitamin A, C, E, minerals, iron, carbohydrates and

proteins. They can also perform better under cultivation with relatively low input levels. However, these are non-commodity crops that are mostly disregarded by research and development programs, and they are also neglected and underutilized. Legwaila et al. (2013) reported that about 150 palatable uncultivated and semi-cultivated plant species are utilized by rural dwellers on regular basis or during times of food scarcity. Indigenous edible plants that are mostly consumed in Botswana include leafy vegetables like African nightshade (*Solanum* spp.), pigweed (*Amaranthus* spp.), cowpea leaves (*Vigna* spp.), spider plant (*Cleome gynandra* L.) and pumpkin leaves (*Cucurbita* spp.). Among these less consumed leafy indigenous vegetables, the study will focus on a useful food crop *Cleome gynandra* because despite its nutritional and medicinal benefits it is still unrecognised even-though it can support the fulfilment of food security, nutrition and welfare.

Spider plant (*Cleome gynandra*) is a fast growing and promising local green vegetable that is crucial particularly in the diet of many African rural populations including our country Botswana. It is from the Cleomaceae family, and it is an upright annual crop with spider shaped white to pink-purple flowers which grows naturally in the wild (Mishra et al., 2011). It is widely spread in Southern Africa and currently distributed throughout tropical and sub-tropical regions of the world (Mishra et al., 2011). This plant is drought tolerant and can survive in drier and hot environments like in Botswana and in regions where drought occurrences are anticipated to rise due to climate change. Crops that are currently classified as minor crops such as *C. gynandra* and adapted to marginal climate conditions deserve to be promoted, developed and evaluated. Therefore, it is essential to advocate for the production and utilization of this functional crop by establishing adequately outlined horticultural production combinations regarding fertilizer application to ensure nutritional security.

Most farmers in Botswana are smallholders who require ongoing assistance with economic growth to make agriculture profit oriented. Thus, the government acknowledges the

significance of this smallholder sub-sector and therefore, has implemented several initiatives to avail various farming technologies to farmers to boost their production output hence, reducing the incidence of rural poverty and the wide productivity gap. These technologies include use of hybrid seeds, farming machinery, modern farming practices and fertilizers use. The latter is one of the key initiatives implemented by the government of Botswana to improve crop yield. Applying the right type and quantity of fertilizer can increase the production output and revenue of farm households. However, the fertilizer is imported from other countries and thus the amount and price of these fertilizers is increasing from time to time. In 2020 NPK fertilizer import for Botswana was 5388 tonnes and its use per unit of arable land was 51.3 kgha<sup>-1</sup>. Additionally, the cost of these imported fertilizers, 2022; Knoema. com/ Botswana/ topics/ Agricultural fertilizer- import, 2022). This results in scarcity of the supply and costly full levels of the recommended chemical fertilizer application by smallholder farmers due to their poor economic capacity even though subsidized.

On the other hand, heavy application of inorganic fertilizers has negative impact on the ecosystem and their continuous application decrease land productivity due to nutrient mining, build-up of salts and other harmful elements. Inorganic fertilizers contain more plant nutrients than their counterparts, organic fertilizers but despite that, the availability of compounds that promote growth in organic fertilizers make them crucial for the improvement of soil fertility and production (Sanwal et al., 2007; Adeleye et al., 2010). Therefore, mobilization of organic nutrients sources that are economically viable and environmentally prudent is important for farmers to improve their yields and income as well as protecting the environment.

Bioslurry is the liquid left over captured after mixed anaerobic digestion of organic matter such as livestock manure (Amon et al., 2006). When fermentation without oxygen occurs, 25 to 30 percent of organic matter is turned into biogas and the other portion is transformed into bioslurry. It contains significant amounts of both organic and inorganic salt and is rich in vital nutrients such as carbon, phosphate, ammonium salt and sylvine (Zhao et al., 2015). According to Amrit (2006), bioslurry contains 25-40% organic carbon, 1.6%N, 1.55%P and 1.0% K whereas farm manure only has an NPK value of 0.8, 0.7 and 0.7 percent while NPK level of decomposed manure is 1.0%, 0.6% and 1.2% respectively. Bioslurry has greater nutrient levels and nitrogen lost through leaching is lower in contrast to inorganic fertilizer, compost and manure (Islam, 2006). Cow dung, crop residues, household, industrial and slaughterhouse wastes are the most popular used feedstocks particularly in the sub-Saharan Africa (Parawira, 2009).

Since bioslurry is an abundant supply of plant nutrients and organic matter, it has the capability to be amongst the leading organic fertilizers to strengthen soils because it can improve the nutrient, biological and physical quality of the soil in addition to granting crops both macroand micronutrients (Guojun et al., 2008). Moreover, its large surface area enhances the soil physical characteristics by improving enzymatic activities, retaining water as well as managing soil fertility, soil environment gas and heat. Incorporating bioslurry into the soil enhances yield and quality of vegetables like size and shapes (Biramo, 2017). Other benefits of bioslurry include seed pelleting, due to the abundance of amino acids, growth hormones and antibiotics in it. It can also be used as a biological pesticide, and it can mitigate climate change effects through carbon sequestration in soil (Warnars, 2013). Furthermore, bioslurry also averts detrimental environmental effects of waste disposal. Given the aforementioned information, bioslurry can reduce farmers production costs by up to 50% by decreasing their utilization of chemical fertilizers (Islam, 2006) thus it can be a solution to farming challenges especially to the smallholder and rural communities. This technology is of profound importance for reaching the millennium development goals at it prevents drastic climate change, eliminate extreme poverty, and ensure ecological sustainability. However, the literature is very scarce reporting

the influence of bioslurry as a sustainable and eco-friendly technology promoting sustainable agriculture relative to chemical fertilizers in Botswana.

#### **1.2** Statement of the problem

Land degradation, climate change and the prevalent poor soil fertility are major constraints of agricultural productivity in Botswana. Farmers depend on the utilization of inorganic fertilizers and soil amendments to optimize yields with the primary goal of supplementing depleted nutrients in the soil. Although inorganic fertilizers have immediate effects on plant productivity, their continuous use cause the soil condition to deteriorate, and their manufacture contribute to climate change. In addition, inorganic fertilizers are costly, and majority of small-scale farmers cannot afford them. Despite all these challenges brought by inorganic fertilizers, food demanded by the increasing population increases exponentially. Therefore, there is need to find other alternatives that are economically and environmentally friendly such as organic fertilizers. Bioslurry is one such inexpensive organic fertilizer that could be used by farmers to reduce dependence on inorganic fertilizers. It can be produced on farm using locally available materials. However, information about the application of bioslurry as a fertilizer and soil conditioner is limited in Botswana.

Climate in southeast Botswana is semi-arid with variable rainfall patterns and frequent dry spells during cropping seasons therefore, drought is a recuring factor and results in occasional crop failures (FAO, 2014). Hence, it is essential to come up with crops that are suited to the local prevailing conditions to diversify and increase crop yields. Indigenous vegetables such as spider plant (*Cleome gynandra* L.) are highly nutritious and drought tolerant hence have the potential to increase and diversify crop yield. Their introduction in farming systems may reduce reliance on exotic vegetables and hence reduce import bill. Several studies have been conducted on spider plant worldwide. However, information on the agronomic management of

this plant species is limited in Botswana. This information is crucial for its adaptation by local farmers.

Hence, the objective of the study was to evaluate the use of bioslurry as a nutrient recycling and eco-friendly practice for efficient crop-soil nutrient management and assess its impact on performance of (*Cleome gynandra* L.) spider plant, and soil attributes in contrast to the current farmers' practice (chemical fertilization).

#### 1.2.1 Justification

The use of bioslurry could be an alternative to chemical fertilizer in enhancing soil fertility and improving crop output in developing nations (Alberdi et al., 2018) including Botswana. Bioslurry is a by-product obtained after anaerobic fermentation of organic materials such as crop residues, animal wastes among others during biogas production. Biogas installation can reduce GHG's emissions into the atmosphere and, lower pollution from surface runoff and leaching by replacing synthetic fertilizers and changing traditional manure management technologies (Debebe and Itana, 2016). These direct benefits can assist the farming community in achieving GHG's reduction goals. Reduced odours and weed seeds are two other significant environmental benefits associated with utilizing bioslurry as an organic fertilizer instead of inorganic fertilizers and untreated manures. Thus, bioslurry is a cheap source of nutrients and a way of waste management. In addition, biogas production programme release gas that provide energy for multiple uses which can solve fuel crisis in rural households in Botswana. Like the rest of the Sub-Saharan Africa (SSA), access to food and meeting food requirements in Botswana continues to be a challenge under this challenging climate and poor soils. Spider plant is one of the native crops that can be researched for wild plant domestication and crop diversification. It is a multifaceted crop that can be utilized as a cosmetic and vegetable (Mishra et al., 2017). It also contributes towards household food and nutritional security. It is a drought and heat-tolerant crop capable of growing under hot and dry environments prevailing in

Botswana. It uses a photosynthesis C4 pathway, which aids in its ability to survive in hot and arid environments (Mishra et al., 2017). There is a continuous collection of spider plant vegetable and seeds of accessions by rural communities, farmers, and gene bank curators from the wild and arable fields where it grows as a weed. The findings of this study will expand knowledge amongst the farming community on the use of bioslurry as an environmentally friendly fertilizer and soil conditioner and establish optimal bioslurry application rates for *Cleome gynandra* L. production under Botswana conditions for its domestication. It is anticipated that the use of bio-slurry by farmers will be beneficial to them by improving soil fertility and yields and reducing costs associated with the purchase of inorganic fertilizers. They will also be able to adapt spider plants in their cropping systems.

### **Research question**

Can bioslurry improve soil fertility, soil structure, crop productivity and increase carbon sequestration in the soil?

# 1.3 Objective

#### 1.3.1 Main objective

To evaluate the effects of inorganic fertilizer and bioslurry on soil properties and performance of *C. gynandra*.

### 1.3.2 Specific Objectives

- I. Investigate the effect of inorganic nitrogen fertilizer and bioslurry on soil properties and
  *C. gynandra* performance.
- II. Compare nitrogen use efficiency (NUE) of *C. gynandra* under inorganic fertilizer and bio slurry.
- III. To assess CO<sub>2</sub> emission and carbon sequestration on soils treated with bioslurry and inorganic fertilizers.

### 1.4 Study hypotheses

#### Hypothesis 1

H<sub>0</sub>- The effects of bioslurry and inorganic nitrogen fertilizers on soil properties and *cleome* gynandra L. performance is the same.

H<sub>A</sub>- The effects of bioslurry and inorganic nitrogen fertilizers on soil properties and *cleome gynandra* L. performance is different.

# Hypothesis 2

Ho- NUE of C. gynandra under inorganic nitrogen fertilizer and bio slurry is the same.

H<sub>A</sub>- NUE of C. gynandra under inorganic nitrogen fertilizer and bio slurry is different.

### Hypothesis 3

Ho- There is no difference in CO<sub>2</sub> emission and carbon sequestration on soils treated with bio slurry and inorganic nitrogen fertilizers.

Ha- There is a difference in CO<sub>2</sub> emission and carbon sequestration on soils treated with bio slurry and inorganic nitrogen fertilizers.

### **CHAPTER 2**

#### LITERATURE RIVIEW

#### 2.1 Introduction

Huge amounts of bioslurry pose disposal challenge which may end up causing contamination to the environment and climate change challenge due to release of greenhouse gases in the atmosphere. High nutrients that are readily available give a golden opportunity for bioslurry to be utilized as an organic fertilizer to enhance food security of rural communities and smallholder farmers who lack access to chemical fertilizers due to their economic challenges thus producing less produce with low quality. Its utilization can be beneficial in the sense that it aids in improving soil quality. In comparison to mostly used nutrient sources such as compost manure and chemical fertilizers, the worth of bioslurry and its optimal application rate for maximum results are necessary to be assessed. Bioslurry has received little attention as an alternative fertilizer and soil amendment source in Botswana and its utilization is still overlooked because of inadequate knowledge about the value of the product hence essential to evaluate this product as a nutrient cycling and eco-friendly alternative fertilizer relative to chemical fertilizers which will increase knowledge of bioslurry and raise awareness to farmers bioslurry technology in Botswana.

## 2.2 Crop Description

Spider plant (*Cleome gynandra* L.) is a fast growing and promising native green vegetable that is crucial to the diets of many African rural populations including our country Botswana. It is from the Cleomaceae family, and it is an erect yearly plant with spider shaped white to pinkpurple flowers which grows naturally in the wild (Mishra et al., 2011). *Cleome gynandra* L. is much branched and grows up to 250-600mm tall and becomes woody as it ages. It is widely dispersed in Southern Africa and currently widespread all over the sub-tropical and tropical regions of the world (Mishra et al., 2011). According to Rao and Rejendrudu (1989) C. gynandra species use C4 photosynthetic route mechanism as a means of adaptation that permits it to endure in withered and hotter conditions. It grows best in summer from November to February on soil provided with organic manure. Spider plant is not a heavy nutrient feeder like many other foliage vegetables, it requires little inputs for optimal production compared to their exotic counterparts (Habwe and Walingo, 2008) hence with the production of spider plant farmers will be able to produce more yields with best quality at low cost as compared to when using chemical fertilizer. Best quality can be obtained with an application of 20 to 30 tons of manure per hectare (Chweya and Mnzava, 1997). According to Poole et al. (2022), fertilization program that provides 680.39kgN/ha/year from 3:1:2 ratio fertilizer source which is roughly comparable to 1.36kg N, 0.45kg P and 0.91kg K<sub>2</sub>0/305m or 4 gram of 19-6-12 per 15.24cm pot for 3 months is needed to produce excellent quality plant and it requires only 6 litres of water per day in a 1m<sup>2</sup> plot. Spider plant has a short vegetative cycle which results in short harvest period lasting for four to five weeks during summer while flowering period last for two to three months (Gonye et al., 2017). It is crucial to the livelihoods of rural populations as an important source of food to supplement diets and source of income to improve their standard of living. (Gonye et al., 2017).

It contains a lot of minerals like calcium, iron, magnesium and protein besides vitamins A and C. Rubaihayo (1997) and Labadarios and Steyn (2001) stated that spider plant is harvested and eaten at a younger stage of development when their leaves are tastier and nourishing. However, this crop can be sundried and preserved for consumption during the dry season when it is scarce as it is only plentiful in the rainy seasons. Apart from being a source of food, Spider plant can be used as an ingredient in the production of detergents, and it is a source of edible polyunsaturated oil which is extracted from seeds (Maumba, 1993) cited by (Gonye et al., 2017).

In most countries, the seeds and leaves are utilized in traditional medicines. The leaves contain lot of phenol compounds which include phenolic acid, flavonoids and condensed tannins, and other phytochemicals which can be employed to treat illnesses like cardiovascular diseases, cancer and asthma (Uusiku et al., 2010). Quercetin is the most abundant flavonoid in plant leaf tissue which has been revealed to have curative ability for treating breast cancer and the potential to preserve the reproductive system (Choi et al., 2021; Oh et al., 2010). Also, in some villages in other countries, leaves boiled in water are thought to cease mother's milk supply and consuming the vegetable is thought to help pregnant women experience less dizziness. Frequent intake of the leaves by expectant mothers will make childbirth easier by shortening their labour time and will assist them get back to their health following delivery (Mishra et al., 2011). Omondi et al. (2017) noted that *C. gynandra* can be useful for intercropping due to its insect repellent properties. Slow food foundation for biodiversity (2022) confirmed the latter statement, indicated that intercropping spider plant with cabbage reduced diamond back moth and thrips attack due to its carvacrol compound in the oil found in leaves which gives the plant its aroma and flavour.

#### 2.2.1 Environmental factors affecting the growth of Cleome gynandra L.

Crop growth and development is essential to research the plant behaviour under given set of environmental circumstances (Hassan et al., 2015). Yield is a complex character which is dependent on several agronomic features and is highly affected by many hereditary and environmental components (Emongor et al., 2017).

*C. gynandra* L. is a widely available species and spreads as a weed in common desolate lands and in agricultural lands. Despite its inherent nutritional quality and medicinal value, the plant is still unrecognised in many parts of the country while the necessary conditions for its best growth are still uncertain. However, (Mishra et al., 2011) noted that, it flourishes well up to about 1000 m above sea level in semi-arid, humid and sub-humid regions, and is suited to a wide range of soils, but flourishes freely around refuse heaps and soils abundant in organic manure. Other studies carried by Chweya and Mnzava (1997) alluded that the plant is suited to various surroundings, indicating that it grows successfully up to 2400 m above sea level, and it endure high and moderate temperatures but survives best between 18-25°C and require high light intensity for maximum growth. The authors further noted that this species is tolerant to numerous soil types which are deep, well drained with a pH range of 5.5 to 7 ranging from sandy loam to clay loam with greater amount of organic matter content and sufficient mineral reserves. Chweya et al. (1997) indicated that the plant is a C4 plant thus tolerant to drought conditions, this mechanism enables it to flourish in regions with brief periods of rainfall. In a review by Shilla et al. (2019), it is mentioned that C. gynandra is an uncultivated vegetable which is adaptable to numerous environmental conditions in the tropics and sub-tropics with the potential to thrive in drier and hotter conditions such as semi-arid and semi humid climates with a variety of soil types due to its advantageous C4 photosynthetic pathway. According to Osborne and Freckleton, (2009), this species does not grow well at temperatures less than 15°C and thrive less in very humid climates. According to Ifeanyichukwu and Afolayan, (2015) in a study conducted in South Africa on the impact of environmental variables and depth of planting on germination of seed in C. gynandra reported that best germination temperatures for the plant are 20-30°C at planting depth of 0.5cm and irrigated twice a week. The latter authors also indicated that the species also germinate best in dark conditions and can survive successfully under different climatic conditions despite seasonal temperature variations. Akbari-Gelvardi et al. (2021) evaluated the germination and emergence responses of Asian spider flower seeds (Cleome viscosa L.) to various environmental factors and find out that optimum temperatures for germination 15.7-17.32°C and 31.89-34.11°C, seeds germinated most effectively at neutral and alkaline pH level with optimal emergence at 1cm sowing depth. Furthermore, it is adapted to moderate drought conditions, however the species is said to be sensitive to high levels of soil salinity. However, Leafy vegetables such as *Cleome gynandra* production are limited by many factors such as soil fertility and rainfall patterns. All of these factors characterize the soils and rainfall pattern of Sub-Saharan African countries where spider plant is mostly consumed. Nevertheless Mutoro et al. (2012) indicated that this plant is well adapted to the Kenyan local environment and possess the resilience to biotic and abiotic stress traits. There are a variety of indigenous vegetables that have the potential to provide relish to rural households and income sources in Botswana. Since these AIVs grow under rainfed they can be produced cheaply. In response to this opportunity African counties have started the to introduce programs and policies to encourage small scale farmers to domesticate wild vegetables (Ndoro, 2007; Ngenoh et al., 2019). Past research mainly concentrated on conventional vegetables, despite its benefits spider plant remain underutilized and since critical information regarding its production techniques is still lacking. It is necessary to speed up development of optimal production packages for adoption in order to boost production and improve livelihoods of these small scale farmers in Botswana, therefore the study's objective was to evaluate spider plant productivity with bioslurry fertilization as alternative to chemical fertilizers.

# 2.3 Nutrient depletion

Soil nutrient depletion refers to all nutrient losses from a soil resulting from both natural and human-caused activities. It is a serious issue that is directly related to food insecurity in emerging and least developed nations because of the intensification of land usage for agricultural productivity without sufficiently utilizing outside inputs (Henao and Baanante, 1999). The ongoing lack of adequate nutrients to rebuild nutrient losses as well as degraded soils due to wind and water erosion are not only aggravating soil deterioration, but also jeopardizing agricultural resilience in these areas (Ayoub, 1999; Sheldrick et al., 2002) cited by (Tan et al., 2005). This is demonstrated by the long-term deterioration in crop yields in the presence of subpar input and unbalanced fertilization in most African regions, Latin America and Asia (FAO/UNDP/UNEP/World Bank, 1997) cited by (Tan et al., 2005). Soil nutrient depletion poses grievous threat to the global food security and sustainable agriculture. The nutrient budgeting strategy was utilized by Stoorvogel et al. (1993), Tan et al., (2005) and Smaling et al. (1993) to fully investigate the major issue of soil nutrient deprivation in most SSA countries, and to demonstrate the crucial role that nutritional equilibrium can play in evaluating expectations for food security and production. Large amounts of soil nutrients continue to be depleted due to food production in developing and least developing nations.

Insufficient replacement nutrient inputs, rapid soil degradation brought by improper land utilization and poor soil managerial techniques, over-cultivation and uneven fertilizing result in soil nutrient depletion and because of this ongoing nutrient depletion has a negative impact on socio-economic wellbeing, sustainability of soil resource, deterioration in environment quality, and low crop output produced by soil degradation (Osgood, 2001 and Lipper, 2001). Miller and Larson (1992) and Tan et al. (2005) reported that, utilization of replacement nutrients and over-cultivation affected about 45 Mha of soils in Africa. About 135 Mha of soils have reportedly been susceptible to nutrient exhaustion, with 97 percent of cases arising in underdeveloped and emerging nations.

#### 2.4 Nutrient replenishment

The consistent decline in soil fertility in Sub-Saharan Africa (SSA) poses a severe challenge in food security and agricultural output, defined as "a net decrease in available nutrients and organic matter in the soil" (Scherr, 1999), and brought on by farmers keeping on mining soil nutrients to improve their agricultural output. Several alternatives including fallows, crop rotation, fertilizers application or a combination of these are accessible to farmers to decrease net nutrient losses. Crop rotation or sequencing such as planting nitrogen-fixing legumes succeeding nitrogen-demanding cereals on a specific piece of land, enables a gradual consumption of macronutrients by crops. McIntire and Powell (1995) emphasize on how the

size of the pasture areas must be to generate enough manure to maintain soil fertility in the absence of mineral fertilizer, despite Williams et al. (1995) 's claim that, manure alone would not improve crop yields in semi-arid West African countries. However, in a study conducted by Lim et al. (2012) on growth and yield responses of four leafy vegetables to organic fertilizer, the results showed that optimum yields of vegetables were obtained from application of organic fertilizer. In agreement to the latter author, Kai and Tamaki, (2020) concluded that organic fertilizer enhanced growth parameters of Brassica napus L. compared to inorganic fertilization. To "kick-start" the process of soil replenishment, utilization of inorganic fertilizer must be expanded. Adding organic materials to soil increases the physical, chemical and biological qualities that will improve the accessibility and uptake of those nutrients by crops. According to Ould et al. (2010) incorporation of organic manure enhances the biological and physical qualities of plant and soil, affirmed to this statement (Noor et al., 2020) 's research findings showed that organic fertilizer greatly enhanced soil chemical health and bulk density (organic matter content, NO<sub>3</sub>-N, N, P, K, Zn, S, NH<sub>4</sub>-N, content). Similar findings were reported by Hashimi et al. (2019) when conducting a study on the impact of organic and inorganic fertilizer levels on spinach (Spinacia oleracea L). This is in conformity to (Sharma, 2017) results which showed that organic fertilizers boost soil microbial activity in the soil, organic matter content, cation and anion exchange ability and soil carbon content. Additionally, the author reported that this fertilizer improves the soil physical characteristics like soil water retention capacity, texture and resistant to erosion. Similar results by Kai and Tamaki, (2020) indicated that growing Brassica napus with organic fertilizer treatment increased total carbon, total nitrogen, and Carbon/Nitrogen ratio also improved soil bacterial biomass, enhancing nutrient (N and P) movement hence creating a rich soil ecosystem with active soil microorganisms. In a study by Ullah et al. (2008) on the influence of organic manures and inorganic fertilizers on the yield of brinjal and soil qualities in Bangladesh, the results revealed that organic manures positively

affected availability of essential plant nutrients (N, P, K) and increased soil organic matter compared to chemical fertilizers. Likewise, Hossain and Ryu, (2018) reported that the content of organic matter in the soil and available nitrogen improved when increasing organic matter doses, furthermore organic fertilizer supressed the number of heavy metals (Cd and Pb) content in the soil except Zn. However, Ullah et al. (2008) reported that soil pH was elevated with chemical utilization than organic fertilizer where as (Hossain and Ryu, 2018) came to differ, reporting that soil pH increased in organic fertilizer treated plots, which shows that organic fertilizers encourage the growth of bases in soil exchange complex.

## 2.5 Integrated nutrient management

This applies to a collection of techniques for managing soil health that incorporate the utilization of organic inputs, enhanced germplasm and fertilizer merged with understanding how to modify these techniques to distinct environments with the goal of optimizing the efficiency of agricultural use of the delivered nutrients and increasing crop yields (Dries Roobroeck et al., 2016). Agricultural agronomy and inorganic fertilizers are the main areas of concentration. The timing of inorganic nutrients inputs, formulation, rate and placement are areas targeted by the interventions on fertilizer use. In addition to intercropping with legumes or rotation and utilization of microorganisms that enhance plant growth, the second Integrated Soil Fertility Management (ISFM) access point concentrate on strategies for managing organic resources, such as compost, manure, crop residues among others (Dries Roobroeck et al., 2016). Numerous ISFM-based techniques have been examined and demonstrated a considerable positive effect on productivity, resilience, revenue, and/or anthropogenic gas emissions as aimed for in Climate-Smart Agriculture (CSA). Combined use of composted organic manures with chemical fertilizers could be more efficient, cost-efficient, and long lasting for both agriculture and the ecosystem. In a study carried by Dries Roobroeck et al. (2016), the maize productivity fell to 1 ton ha<sup>-1</sup> in trials where sole fertilizers were used, and the 0.26 and 2.4 ton ha<sup>-1</sup> that were registered when organic inputs and NPK fertilizers were integrated in comparison to the case where the same inputs were applied independently as opposed to when they were used separately in the Integrated Soil Fertility Management (ISFM) system (Dries Roobroeck et al., 2016). In addition, rotated cowpea crops in the ISFM system produced an average of 1.2ton ha<sup>-1</sup> compared to 0.7ton ha<sup>-1</sup> when organic inputs or fertilizers were utilized independently; results also authenticate that ISFM practice results in long term improvement crop productivity thereby enhancing farmers ability to support themselves.

Despite the significant merits of Integrated Soil Fertility Management for preserving the environment, household income and food security, farmers typically adopt these practices in a poor and incomplete manner, particularly in African smallholder systems. The most significant barriers to adoption are connected to the high financial costs of input and produce trade, little awareness and widespread skepticism regarding the advantages of soil fertility management, lack of organic wastes and competing with animals for residues (Alene et al., 2008; Rufino et al., 2011). Therefore, this research aims at raising awareness to the farming community in Botswana and provide knowledge about the beneficial use of bioslurry as a potential alternative fertilizer to chemical fertilizers that can be used to curtail high cost of chemical fertilizers and again reducing environmental impact of livestock waste. Also, the biogas produced energy that farmers can utilize it for cooking and lighting hence decrease fuel costs that are already a crisis.

# 2.6 Nitrogen Use Efficiency (NUE)

In developing countries with a high proportion of less fertile soils, it may be difficult to meet the nutritional needs for high yielding crops because many cultivated soils are deficient in the most critical nutrients needed to grow vigorous plants (Marschner, 2005). Efficiency of nitrogen application has been reported to be at or below 50 percent, causing major ecological impacts (Scheiner et al., 2002; Baligar et al., 2001). Thus, aiming for efficient nitrogen use is important because plants that are effective in nutrient uptake and utilization enhance the effectiveness of applied nutrients, limiting nutrient losses to ecosystems and reduce input costs (Baligar et al., 2001). Coulibaly et al. (2020) revealed that in an experiment with three fertilizer levels of urea at 0kg/ha, 43.5kg/ha and 65kg/ha, treatment level of 43.5kg/ha provided the maximum biomass yield (9.07 ton/ha) of *Amaranthus cruentus* as opposed to 65kg/ha level which produced low yield of 5.9 ton/ha. In another study conducted by Sage and Pearcy (1987) on nitrogen use efficiency of C3 and C4 plants, concluded that *Chinopodium album* outperformed *Amaranthus retroflexus* L. with greater nitrogen use efficiency at low applied N rates, however, *A. retroflexus* L. outyielded *C. album* at high applied N rates. In addition, they demonstrated that both species gathered comparable amounts of nitrogen (N) per plot when given equivalent amounts of N, but *C. album* accumulated more N due to its decreased dry matter output; consequently, *C. album* utilised the absorbed N more effectively than *A. retroflexus*.

#### 2.7 Role of macro nutrients and crop growth and yield

Primary macronutrients play a crucial part in enhancing crop yields and value. Nitrogen, phosphorus, and potassium (N, P, and K) the most essential elements that are needed in large quantities. They must be easily accessible through soil media or fertilizer. For successful production of agronomic crops, proper plant nutrition is essential as each individual macronutrient has its own unique character and is therefore involved in a variety of metabolic activities of plant life (Zewdie and Reta, 2021).

#### 2.7.1 Nitrogen

The most common mineral nutrient in plants is nitrogen, and it makes up 2-4 percent of plant dry matter. Although there is 79% of nitrogen in the air, plants can only utilise nitrogen in the form of ammonium (NH<sub>4</sub>) and nitrate (NO<sub>3</sub>-). Additionally, nitrogen is appraised as a necessary element of all enzymes and proteins, and it is employed in a number of metabolic processes that lead to energy conversion (Rajasekar et al., 2017). Therefore, adequate N accessibility in plants is needed because it is amongst the key fundamental factors of crop production thus required by the plant in biggest percentage. It is an essential component of chlorophyll, protein, nucleic acids and protoplasm. In addition, it gives plants a dark-green colour and improves the vegetative growth. Zewdie and Reta (2021) also mentioned that because plants contain 15% nitrogen by weight, nitrogen plays a significant role in agriculture by enhancing crop productivity. Nitrogen is essential for all plants' healthy growth and development as it improves the quality of food and yield (Leghari et al., 2016). It increases the quality of fodder and green vegetables and the level of protein in cereal grains. In agreement with the latter statement, Mauyo et al. (2008) reported that utilization of nitrogen fertilizer improved the quality of spider plant by increasing plant height, fresh weight, quantity of shoots and saleable leaves, also demonstrated that applying 80kg/ha of nitrogen fertilizer will generate the best vegetable quality and yields while extending the harvesting period. Similar results reported by Mavengahama (2013) indicated that application of nitrogen boosted fresh yield, leaf dry matter and whole plant dry matter. Ogweno and Ng'etich (2012) reported that, adding calcium ammonium nitrate as nitrogen sources resulted in improved plant height, quantity of shoots and usable leaves and shoot output recommending 150 kgN/ha from calcium ammonium nitrate for maximum yields of cleome gynandra. Same results were reported by Olaniyi et al. (2008) who revealed that, Amaranthus cultivars recorded an increase in leaf number, internodes and plant height with application of increased nitrogen rates from 0 to 60 kgN/ha. Similarly, Mutua et al. (2015) reported that applying 300 kg/ha of NPK fertilizer and deflowering gave maximum number of primary branches per plant, greatest leaf output and extended the harvesting period by eight weeks in their study about NPK fertilization and deflowering of C. gynandra. Generally speaking, adding N typically has a greater impact on plant development and a significant effect on the quality of a product, particularly through improvements in protein content and quality. The results of the study carried out by Nofal et al. (2021) on the impact of soil application of nano NPK rates on growth, head quality and yield of lettuce showed that when 50% of nano nitrogen was applied vegetative growth, yield and marketable yield of lettuce increased.

### 2.6.2 Phosphorus

Together with N and K, P is recognised as one of the three vital nutrients that plants need for growth and reproduction. It is essential for sugar metabolism, photosynthesis, energy transfer and storage, cell division and growth, and transfer of genetic information (Havlin and Halvorson, 1990). It also encourages healthy shoot and root growth, speeds ground cover for erosion protection, improves the quality of fruit, vegetable, and grain crops, and is needed for formation of seed. Adequate P boosts plant water use efficiency, improves the effectiveness of other nutrients like N. Ali et al. (2014) reported that phosphorus is crucial for plant growth, particularly in the premature phases of jointing and for increasing grain yield and yield components. In a study conducted by Nkaa et al. (2014), on the effect of phosphorus fertilizer on growth and yield of cowpea, the findings revealed that phosphorus significantly enhanced growth and yield characters (plant height, leaf number and number of branches) also, 40 kg/ha phosphorus application rate is advised for maximising yields. Similar findings from Aryal et al. (2021) indicated that phosphorus dose of 40 kg/ha had the highest effects on growth, development and yield attributing traits hence resulted in increased output and productivity. According to Turuko and Mohammed, (2014) application of 20 kg/ha phosphorus level greatly improved dry matter yield, yield components and all other growth metrics of common bean except plant height.

#### 2.6.3 Potassium

Potassium (K) is an essential plant macronutrient that is crucial in many physiological activities that are critical to plant nutrition, nutrient transport, growth and water uptake particularly under challenging circumstances (Jiang et al., 2018). As a result, it serves on different purposes in plant development and nutrition that affect both crop's quality and yield (Roy et al., 2006; Kow

and Nabwami, 2015). Zewdie and Reta, (2021) added to the assertion made by the later author by stating recent research found that applying K fertilizer significantly improved productivity of rice and wheat. Furthermore, it strengthens the product's resilience to different injuries sustained during delivery and storage, thus extending shelf life. In a study carried by Zaki et al. (2015) in Egypt on the effect of rates of potassium on broccoli cultivars, the findings showed that 40 kg/ha of K<sub>2</sub>O gave the highest crop's yield, vegetative growth and chemical contents. According to Inthichack et al. (2012), potassium concentration of 482.7 mg/L improved the output of celery, cabbage and lettuce when assessing the impact of potassium sources and rates of plant growth and yield for hydroponic cabbage, lettuce and celery. However, Mg concentration decreased with increasing P supply. The obtained results were in harmony with (Saud et al., 2013; El et al., 2022 ; Hossain et al., 2017) who reported that raising potassium fertilizer levels increased leaf quantity per plant, shoot and root fresh weight of plants, similarly El et al. (2022) found that addition of 866 kg/ha k<sub>2</sub>O with compost at rate of 25.98 ton/ha increased highest values of the majority of vegetative growth character measurements and output of lettuce. Nofal et al. (2021) reported that head quality of lettuce notably increased when 50% of nano potassium fertilizer was applied. These beneficial outcomes might result from improving the photosynthetic and the assimilation rates which might raise most vegetative metrics and head yield of lettuce (Saleh et al., 2010). On contrary Nemadodzi et al. (2017) in South Africa conducted a research study on the impact of NPK on physiology and biomass yield of baby spinach and concluded that potassium did not have any influence on leaf area index, chlorophyll content, stomatal conductance, and yield of spinach, nonetheless, growth was best maximized with NPK ratio of 45:45:60 kg/ha.
#### 2.8 Use of fertilizers in crop production

Insights of soil chemical makeup is critical due to its influence on accessibility of plant nutrients and these properties may be changed by adding lime or fertilizers (Hugh Savoy, n.d.) Furthermore plants require 18 elements which are vital plant nutrients for healthy growth and completion of life cycle, thus this called for development of soil amendments containing these nutrients and altering soil chemistry. Fertilizers, synthetic or natural are the additives that bloom plant productiveness and improvement, and they assist the soil nourish its fertility thereby enhancing productivity and production (Smith, 2022). Additionally, incorporating fertilizers into the soil ensures that the plant is getting right nutrients throughout its improvement. In conformity with the above statements, Fusire (2008) revealed that spider plant responds well to fertilizers, organic or inorganic and the findings by Gonye et al. (2017) revealed that doses of the organic or inorganic fertilizer applied in the study delivered adequate N which resulted into rapid increase in growth metrics. In a study conducted in Cameroon by Fonge et al. (2016) on effects of fertilizer rate on growth, yield and nutrient concentration of leafy vegetables, Amaranthus cruentus L. and Vernonia hymenolepis, the results revealed that A. cruentus and Vrnonia hymenolepis responded positively to fertilizer. The findings are consistent with (Ouda and Mahadeen, 2008) for Brassica oleracea L. which found that yield and nutrient content improved with increasing fertilizer rates. Hashimi et al. (2019) indicated that using chemical fertilizers, cow manure independently and in combination significantly increased soil nutrients levels, similarly Sharma (2017) reported an improvement in soil organic matter content, microbial activity, soil carbon and physical properties. They are made from various natural and synthetic substances, and they come in liquid, solid and gaseous form (anhydrous ammonia), they consist of at least one of the acknowledged plant nutrients, which are employed primarily for their plant nutrient content. These substances are intended for usage or claimed to have benefits in enhancing plant development or improving nutrient levels available to plants in the soils. According to Reetz (2016), the widespread use of commercial

mineral fertilizers is one of the key drivers in ensuring global food security in recent years and over 48% of more than 7 billion people alive today are alive due to improved agricultural output made possible by application of nitrogen (N) fertilizers. The focus of nutrient management is to provide sufficient supply of all essential nutrients for crops during the growing season and increase the biological, chemical and physical soil qualities. Additionally, they are required to replace nutrients lost during crop harvest and to add additional nutrients to improve crop yield (Sharma, 2017). If the availability of any nutrient is limiting at a specific period, there is a possibility of production loss. Moreover, Mani (2002), reported that lack of nitrogen causes slow and poor development, whereas excessive application of this nutrient cause low quality of leaves and delayed maturity. Likewise, Krasilnikov et al. (2022) also mentioned that improper application of inorganic fertilizers alters soil pH, increase pests and disease attack and cause acidification, all of which may lead to reduced soil organic matter and beneficial organisms, stunted plant growth and yield, and GHG's emissions. According to Reetz (2016), the farmer's financial returns have greatly improved due to fertilizer utilization in agricultural crop production. The issue, however, lies in managing fertilizers and soil in a sustainable way to consistently improve output of food and fibre crops through rational and efficient fertilizer use procedures.

## 2.8.1 Inorganic fertilizers

Benton, (2012) defined inorganic fertilizers as fertilizers which comprises of artificially manufactured elements that are prepared for use on plants. They contain all essential nutrients like phosphorus, nitrogen and potassium, and they provide plants with instant nutrients supply therefore found handy by most farmers (Ibrahim et al., 2014). Like organic fertilizers, inorganic fertilizers supply nutrients necessary to grow plants but unlike organic fertilizers they do not need to decay over time to provide plants with nutrients (Stewart et al., 2005). According to Benton, (2012) these fertilizers provide some benefits which include convenience,

affordability, and effectiveness in nourishing plants. However, there are drawbacks that may arise when using chemical-based fertilizers as they can burn or damage the whole plant (Rosen and Horgan, 2009), thus it is crucial to incorporate the accurate quantities of fertilizer into the soil. Sharma, (2017) indicated that improper use of inorganic fertilizers can result in nutritional imbalances which limit the absorption of some critical nutrients and increase acidity of soil which could be detrimental to nitrogen fixing bacteria leading to low yields. Inorganic fertilizers must be applied twice during a growing season for maximum plant growth, and this may depend on the crop type (Sartain, 2011), furthermore, constant utilization of these fertilizers degrade the soil structural and physical characteristics, lessen soil organic matter and increase erosion. Unutilized nutrients may travel into underground water and contaminate the water which is harmful to human beings and animals (Sharma, 2017).

One benefit of inorganic fertilizers is their quick act on plants, these salts abundant in nutrients liquefy easily and become instantly plant accessible. They are meant to give needed sustenance in the form of phosphorus, potassium and nitrogen. Inorganic fertilizer's quick supply of vital minerals and elements dismiss the possible issue of nutrient deficiency on plants (Stewart et al., 2005).

Inorganic fertilizers are made available in different convenient forms like dry grains, liquid concentrates and water-soluble powders, hence advantageous to use them over organic fertilizers such as manure. They come in tubular and spike forms made especially for indoor or outdoor container plants, as well as dry granules, liquid concentrates, and powders that dissolve in water.

The primary drawback of inorganic fertilizer is that they are more expensive than the organic fertilizers hence organic are more cost effective (Rainbow and Wilson, 2002). Inorganic fertilizers also have the problem of leaching so it is much more prevalent when inorganic

fertilizers are used because the nutrients are already present in their most fundamental components and hence can be washed away easily if plants roots are overirrigated (Mills and Jones, 1996). Also, inorganic compound contains compounds and salts which plants cannot absorb and are therefore left behind in the soil where they build up and even change the chemistry as time passes thus making the soil less suitable for upcoming plantings, lastly excessive use of inorganic fertilizers can turn out to be harmful to plants according to Stewart et al. (2005).

#### 2.8.2 Organic fertilizers

These are organic materials used as fertilizers derived from biological nature (Sharma, 2017). They are carbon-based compounds that add numerous secondary and important micronutrients hence increasing productivity, growth, and quality of plants (Chaney, 2012). They are derived from animal manure and vegetable matter (Bayani, 2011). According to Colting and Tagarino (2008) most fertilizers are extracted from minerals or produced industrially. These organic materials are naturally occurring as by product or end product of a naturally occurring process which include animal waste from meat processing, peat, manure and slurry (Singh, 2012). Peat, animal and plant wastes from agriculture, and sewage sludge respectively are the major organic fertilizers sources according to Bayani, (2011). Schrack, (2009) indicated that chicken manure which is made from chicken litter mixed with saw dust is an organic fertilizer that has shown to improve the state of soil for harvest than man-made fertilizers. Assefa and Tadesse, (2019) mentioned that organic fertilizers are cheap and can be transported easily in bulk hence accessible even to resource poor farmers. Also, these organic substances work as both soil conditioners and fertilizers. Affirming to the latter statement, Ibukunoluwa Moyin-Jesu (2015) indicated that chicken manure at 6 ton/ha significantly enhanced soil productivity, growth, and output of cabbage because of balanced nutrient composition and lower C/N ratio. Likewise, Kai and Tamaki (2020) reported that cultivation of *Brassica napus* L. with organic fertilizers treatment enhanced total carbon, total nitrogen, and increased soil bacteria biomass, producing a rich soil environment. The use of organic fertilizers to manage soil fertility has always been a fundamental principle to sustainable agriculture. Lazcano et al. (2013) and Ullah et al. (2008) stated that maintaining soil fertility is necessary for long term sustainable agriculture. The latter author continued to state that application of organic manures as components of sustainable agriculture has a good effect on soil chemical and physical qualities which eventually increases production. According to FAO (2012), as compared to inorganic fertilizers, these fertilizers have several of advantages which include guaranteeing that the food products generated are exempted from hazardous substances, aid in maintaining soil structure and enhancing soil nutrient holding capacity. Also, these fertilizers are quickly biodegradable and do not pollute the environment. Additionally, Sharma, (2017) mentioned that organic fertilizers supply nitrogen in utilizable form which help to promote better plant growth and, they prevent or suppress pest and disease attack by meeting plant nutritional needs and enhance plant tolerance.

Virtually every aspect of organic farming is informed by the health of the soil, organic fertilizers increase organic matter in the soil which increase the soil composition, increases air spaces and water holding capacity within the soil, lessen soil surface runoff and foster the development of a strong natural environment that is long-lasting and conducive to long term use (Fuller, 1994). According to Assefa and Tadesse, (2019), the decomposition of organic matter from the organic sources breaks down naturally providing essential nutrients and minerals to the soil increasing its fertility and quality of plants. Ouda and Mahadeen, (2008) reported that broccoli crops planted in soil modified with organic fertilizer had quick vegetative growth, head output with big head diameters. Same findings were reported by Shaheen et al. (2014), which revealed that organic fertilizer enhanced nutrient levels of soil N, P and K and spinach yield. The other advantage is that the organic fertilizers have a gradual release of nutrients therefore there is less chance of nutrient burn from over fertilization resulting in

stronger and stable plants than those growing at artificially accelerated rates, an in theory, have better nutritional content and taste (Fuller, 1994). Increased organic matter and reduced nutrients leaching work together resulting in components like nitrogen and phosphorus ending up in the plants roots rather than in local water ways hence preventing eutrophication which disrupts ecosystem and renders water retention for human use (Hue, 1994).

However, the rate at which nutrients are released from organic fertilizers can be affected by both availability of soil micro-organisms and air temperatures. Jaja and Barber (2017) added that release of organic matter is favoured by slow decomposition, temperature and moisture thus rapid decomposition results in more nutrients being released even when the plant does not need them. Inadequate decomposition can leave residual microbes in the organic waste and these pathogens can filtrate the water system on the food crops, posing risks to both human health and the environment (Fertilizer Europe, 2014).

#### 2.8.2.1 Bioslurry composition and Utilization

Amon et al. (2006) defined bioslurry as an end-product from anaerobic fermentation of organic materials during formation of flammable methane gas. When this product is well digested with the right amount of organic substrates, its composition consists of 93% of water while dry matter occupies the remaining 7% of which 2.5% is inorganic and 4.5% is organic matter (Warnars and Oppenoorth, 2014). pH of bioslurry lies above neutral range ( $\geq$ 7). Wirth et al. (2012) mentioned that bioslurry production goes through stages namely; (1) hydrolysis where bacteriodes, clostridum and bafidobacterium and obligate bacteria excrete hydrolytic enzymes like protease, cellulase, amylase, lipase, and others to act on polymers of carbohydrates, proteins and lipids which yield monomers like amino acids, fatty acids and sugars, (2) Acidogenesis phase, the simple monomers are transformed into simple compounds such as hydrogen and carbon dioxide. The third stage of acetogenesis involve acetogenic bacteria that transform the organic acids to acetic acid along with additional production of hydrogen, carbon dioxide and nitrates, then the last stage methanogenesis occurs when the methanogens microorganisms take part in converting the acetic acid and hydrogen from the acid fermentation into carbon dioxide and methane gas. Amongst different natural waste materials, cattle waste is the most utilized substrate in production of bioslurry and its anaerobic digestion process takes fifty days after which the slurry is disposed. Numerous variables, including water, breed and age of animals, feed rate, diet, type of waste (animal or human) and retention time may determine the composition and value of bioslurry (Warnars and Oppenoorth, 2014). Organic matter content in bioslurry is mainly influenced by the length of the digestion process as well as the type of the feedstock employed and the longer the retention time, the lower organic content bioslurry is produced (Apples et al., 2011; Grootboom, 2018).

Several studies illustrated that bioslurry has plentiful plants nutrients like nitrogen, phosphorus and potassium, and other elements such as magnesium, calcium, manganese, iron, zinc and copper which are vital for plant growth (Mdlambuzi et al., 2021). Biramo (2017) reported that bioslurry contains 1.6%, 1.55% and 1.0% of NPK respectively, likewise Fashaho, (2020) also reported a range of 1.4-1.8 N, 1.0-2.0 P, 0.8-1.2K and 25-40 percent of organic matter in a well digested slurry while Sharma et al. (2021) reported OC, N, P and K content of 41.6, 0.72, 0.59 and 0.9 percent respectively in dried slurry. Warnars and Oppenoorth, (2014) mentioned that a fully digested bioslurry is characterized by, dark brown or black color, an odourless smell which does not attract flies and contains tiny living creatures.

Bioslurry can be utilized in three forms while applying it to soil; (i) liquid form which can be applied directly in the field using a watering can or bucket or discharged through the irrigation canal, (ii) dried slurry which is dried first and applied to the soil in solid form and lastly (iii) composted bioslurry (Fashaho, 2020). Bioslurry nutrients are bioavailable, thus it can be utilized as a basal or top dressing, nevertheless if this fertilizer is applied to standing crops, it should be applied by diluting it with water in a ratio of 1:1.5-2.0 otherwise it will pose scorching effects on newly developing leaves because of greater level concentration of ammonia and phosphorus (Biramo, 2017), Warnars and Oppenoorth (2014) indicated that bioslurry is recommended to be applied at a rate of 10-20tons ha<sup>-1</sup> in areas that are irrigated and 5tons ha<sup>-1</sup> in dry farming for positive yield results. Furthermore, application of more than 25tons ha<sup>-1</sup> may not give any significant increase in yields. However, appropriate rate may be determined by the type of crop and soil (Warnars and Oppenoorth, 2014).

According to Devarenjan et al. (2019), a biodigester of 2 m<sup>3</sup> in volume can produce 50 kg of bioslurry daily with 1 m<sup>3</sup> containing about 0.16 to 1.05 kg of nitrogen which is equal to 0.35 to 2.5 kg of urea. On the same context Xu et al. (2021) reported that 800 m<sup>3</sup> volume of biogas plant emits 15 tons of bioslurry every day hence the need for storage for appropriate management of the daily volumes of bioslurry that is generated to prevent risks of environmental pollution since it is applied to soils on seasonal basis during crop production, thus proper storage is necessary to combat nitrogen leaching losses and nitrogen volatilization which may affect the quality of slurry. In agreement with (Xu et al., 2021), Bonten et al. (2014) reported that excess bioslurry can be kept in air tight or open lagoons, containers and tanks, alternatively he said bioslurry can be divided into liquid and solid fractions which can be kept dried and the author also continued to mention that it can be mixed with other organic materials making a slurry compost, however mixing bioslurry with inorganic materials cause nitrogen losses of 15 to 27% on average. Mamo, (2013) added by indicating that building bioslurry pits next to the biodigester plant as a storage source where bioslurry will be collected is important and the pits should be kept covered to prevent contamination. Karki, (2006) reported that composted slurry contains 0.75%, 0.65% and 1.05% NPK respectively.

#### 2.8.2.2 impact of Bioslurry on growth and yield indices

As it has been determined that nutrients in bioslurry are bio available, it is reasonable to anticipate that crop growth and output will be increased due to improved nutrient availability.

Islam (2006) indicated that utilization of bioslurry can increase crop revenue by 25%. Supporting studies by Ogbaji et al. (2018) reported that bioslurry vigorously increased growth in spinach on slight acid or alkaline soils irrespective of treatment over control thus concluded that soils treated with bioslurry was the most suitable for crop cultivation compared to soil treated with chemical fertilizers. In the same accord, Biramo, (2017) reported 18.4 percent and 28.4 percent yield increment for cabbage compared to farmyard manure application but a full dose of the prescribed inorganic fertilizer produced yield increment of 14 percent. Similar findings by Xu et al. (2005) showed that vegetables given organic fertilizer treatment grow more effectively and generate higher yields than those with inorganic fertilizer treatment. Likewise, Khan et al. (2004) indicated that the availability of some micronutrients such as zinc in bioslurry accounted for higher yields in crops. In a study comparing the impact of bioslurry and inorganic- fertilizer on growth and yield of kale, Haile and Ayalew (2018) reported that sole fertilization with 100 percent liquid bioslurry considerably increased leaf marketable weight (445.10g) and biomass (814.86g), additionally Warnars (2014) noted that nutrient delivery is vital in plant growth. According to Suthar (2009) and Shahabz (2011) plant height improved due to greater application rate of cow dung bioslurry than chemical fertilizer application. Shaheb et al. (2015) cited Shaheb and Nazrul (2011) for similar findings that cow dung slurry at 5ton ha<sup>-1</sup> combined with IPNS basis inorganic fertilizer generated the highest cabbage output. In conformity with the latter, Shahbaz et al. (2014) also reported an improvement in growth after treating okra with 200 percent cow waste bioslurry. Rahman et al. (2010) found that plant height is influenced by various factors including genetic makeup and soil fertility. Grootboom (2018) indicated that bioslurry can be a significant nutrient source

for crops since 100 to 150kg N/ha treatment of bioslurry enhanced dry matter yield of spinach, however combined application of chemical fertilizer and bioslurry boosted dry matter yield and nutrient uptake than sole application of bioslurry at the same Nitrogen dose. Similar findings by Jeptoo, (2013) reported higher plants growth and yield in plants treated with higher amounts of bio slurry than other treatments. On another parameter, Haile and Ayalew, (2018) found a high number of kale leaves after 100% bioslurry application over the control and other treatments. Attesting to the latter author's claims, Shakti (2006) conducted an observational field trial on the impact of cow and poultry waste bio slurry on brinjal, cabbage, and tomato and reported that bio slurry enhanced the yields of the crops as compared to those of recommended chemical fertilizer dose. However, Masarirambi et al. (2010) reported that inorganic fertilizers provide plants with enormous amounts of nutrients that are quickly released, improving the quantity of leaves of green vegetables as well as their quality. Bioslurry applied at 10t ha<sup>-1</sup> increased leaf area of spinach (Islam et al., 2016). In according with Mog, (2007) and Apahidean et al. (2012), fertilization with bioslurry enhanced cell elongation and division which led to an increase in leaf enlargement. Dumani et al. (2021) alluded with other findings and reported that ford hook giant variety of spinach fertilized with 200% cow dung bioslurry was higher-up for the majority of the assessed growth characteristics including plant height (7.63 cm), quantity of leaves (7.23) and leaf area (593.3 cm<sup>2</sup>), furthermore, the author reported high amount of macro nutrients compared to other treatments. Contrary Khanafi et al. (2018) reported that chemical fertilizer enhanced plant development due to fast delivery of nutrients directly to plants.

## 2.8.2.3 Influence of Bioslurry on soil physical and chemical properties

The organic matter usage in agricultural systems ought to be encouraged. It permits maintaining soil fertility, while enhancing the soil structure and mineral elements availability. In fact, accumulation of soil organic matter to ideal levels is a crucial component of any system for

organic production (Gaskell et al., 2000; Majekodunmi et al., 2021). Shaheb et al. (2015) reported insignificant changes on post-harvest chemical analysis of soil pH, organic matter (OM) and other nutrients after bioslurry utilization. Contrary to the latter author Jeptoo et al. (2012) indicated that implementation of bioslurry considerably enhanced soil major nutrients (Nitrogen, Phosphorus and potassium). Lolamo et al. (2023) reported that application of bioslurry and chemical fertilizer influenced the status of soil chemical properties, however, bioslurry performed better than chemical fertilizer and control by increasing soil pH from 5.6 to 7.4 while chemical fertilizer decreased pH. Further bioslurry amended soils increased organic carbon by 8.4% to 2.98% and other previous studies also showed significant increase in soil organic carbon after organic fertilizer application as opposed to chemical fertilizers (Jibril and Bekele, 2022; Tadesse et al., 2013). Similarly, study by Geremew (2017) revealed that application of full dose of dry bioslurry (14 ton/ha) increased organic carbon over control. The latter author also reported an increase in TN, available P, and CEC by 0.20%, 59.6 mg.kg<sup>-</sup> <sup>1</sup> and 29 cmol.kg<sup>-1</sup> respectively compared to 0.24%, 42.5 mg.kg<sup>-1</sup> and 23.6 cmol.kg<sup>-1</sup> of chemical fertilizer. This finding agrees with previous studies carried by (Tana and Woldesenbet, 2017; Mengistu et al., 2017) which demonstrated that fertilization with organic fertilizers raises soil cation exchange capacity while inorganic fertilizer lowers the cation exchange capacity. Also (77,78) stated that there is a direct correlation between CEC and OC as soils with low CEC tend to have lower OC level. High CEC and OC are important as they increase ability of the soil to buffer its fertility by protecting nutrients from leaking and improving their accessibility (Lolamo et al., 2023). Earlier research revealed that application of organic fertilizers increased organic matter content and available nutrients which in turn increased the cation exchange capacity and exchangeable bases of the soil (Assefa and Tadesse, 2019; Kome et al., 2021). Zhang et al. (2012) stated that bioslurry has been used as a soil conditioner to enhance soil fertility more especially in acid and nutrient-poor soils, additionally it has high

surface area thus its utilization to soils has a positive impact on a number of soil metrics like pH and nutrient retention, aggregate stability, porosity, water holding capacity and soil bulk density (Musse et al., 2020). Similarly, Kebede et al. (2022) reported improvement of potassium, soil pH, organic carbon, total nitrogen, organic matter and C:N ratio compared to chemical fertilizers, the author also reported an increase in soil physical properties and concentration of essential metals (Cu, Fe and Zn) with bioslurry application. Abubaker etal. (2015) reported that fertilization with pig bioslurry increased available mineral nitrogen in the soil compared to other treatments in a study they conducted on short-term effects of pig slurry and other biogas digestates usage on soil microorganisms' activity, similar findings were revealed by Rewe et al. (2022). Application of bioslurry slightly reduced bulk density by 17%  $(0.83 \text{ g/cm}^{-3})$  and increased porosity by 11% (70%) compared with application of chemical fertilizers in a study conducted by Lolamo et al. (2023) on the effects of bioslurry and chemical fertilizer application on soil properties and food safety of tomato. Apart from that application of bioslurry increased soil moisture content by 58.3% compared with chemical fertilizer 58.1% and control. This was in accordance with other studies which reported that soils amended with bioslurry, farmyard manure and other organic fertilizers increased porosity and decreased bulk density which are associated with increase in water holding capacity of the soil (Hartanto and Putri, 2013; Tana and Woldesenbet, 2017). According to Shahabz (2011) bioslurry application at 600 kg/ha gave highest total nitrogen uptake by shoot by 80 percent and phosphorus uptake by plants raised from 89-135 percent over control treatments in okra. On the other hand, Sasanya (2019) carried out a study on the impact of piggery and poultry slurry on the safe eating of Amaranthus hybridus and Conchorus olitorus and concluded that the vegetable's anions and heavy metals content were lower than the limits for safe consumption on bioslurry amended soils than chemical fertilizer amended soils thus, the two vegetable crops on bioslurry amended soils were safe to be eaten by humans over those planted on chemical fertilizer soils.

Radioactively active gases like carbon dioxide (CO<sub>2</sub>), nitrous oxide (N<sub>2</sub>O) and methane (CH<sub>4</sub>) often known as GHG's, have been emitted at a higher rate over the previous few decades. The greenhouse effect is brought on by these gases absorbing the emitted infrared light from the earth's surface. This alters regional and global changes in climatic parameters particularly temperature and precipitation. These changes in climate impact agriculture, human health, both terrestrial and aquatic ecological systems, water resources among others. A significant greenhouse gas, CO<sub>2</sub> account for about 60% of the entire greenhouse impact (IPCC, 2013). Soil is a significant source and sink of atmospheric CO<sub>2</sub>. For fending off the threats of global warming brought on by GHGs, reducing CO<sub>2</sub> by storing carbon in the soil is of prime importance. Application of crop residues and animal manure, crop diversification, biodiversity and reduced tillage are among the soil management practices that encourage increased soil organic carbon content by storing carbon in the soil.

The importance of CO<sub>2</sub> in global warming highlights the necessity to stop carbon leakages from the soil resource. Globally, the first metre of the soil profile has an abundance of organic carbon estimated to be around 1500 Gt (Jobbagy and Jackson, 2000; Guo and Gifford, 2002). This is significantly more than the 560 Gt of carbon (C) discovered in the biological pool (Lal, 2008) and double more than CO<sub>2</sub> in the atmosphere (IPCC, 2013). By storing this enormous amount of carbon, the soil is limiting CO<sub>2</sub> accumulation in the atmosphere which will curtail the issue of climate change. Decreasing soil organic carbon (SOC) loss is an effective technique to enhance food security because climate change can negatively impact agricultural production. Around 24 percent of global soils and 50 percent of its cultivated soils are already deteriorated (Batjes, 2013; IPCC,2014; World Bank, 2015), hence there is an opportunity for increased land productivity and absorption of atmospheric carbon in the soil. According to the IPCC (2014), it is possible for agricultural soils to sequester up to 1.2 billion tons of carbon annually. Agricultural land could store at least 10% of the present 8–10 Gt/year annual emissions (Hansen et al., 2013).

According to Stockman et al. (2013), carbon sequestration in soils can be a short-term climate mitigation strategy for lowering atmospheric CO<sub>2</sub> concentration. The process may be propelled naturally or human induced. The human induced sequestration guarantees that there is no net growth in the atmospheric C pool because the CO<sub>2</sub> sequestered comes from the atmosphere. Abdullahi et al. (2018) indicated that, generally there are two sorts of sequestration: non-living and living (abiotic and biotic). The abiotic approaches entail administering CO<sub>2</sub> into abandoned coal mines, deep oceans, geological strata, and oil wells, whereas the biotic component on the other hand, entails controlling higher plants and microbes to absorb additional atmospheric CO<sub>2</sub> and fixing this C un-stable soil pools. Carbon can also be stored in soil through humus build up onto the topmost layers (usually 0.5–1 m depth) of soil or human induced by changing land utilization or implementation of rightful management techniques in pastoral, agricultural, or forest environments (Lal, 2008).

The organic carbon content of the soils in the Sub-Saharan Africa is deficient due to high temperatures that encourage organic matter breakdown, sandy texture, and organic inputs (Lal, 2004). Nyamangara (2002) enriched the latter statement by indicating that on average in SSA, SOC content of soils is less than 1% even though productive soils typically need 25-30g C kg<sup>-1</sup> (Islam, 2006). Excessive reduction of SOC levels can degrade soil fertility, reduce biomass productivity and contribute to climate change (Zhang et al., 2021). Loss of SOC brought on by human disturbances are thought to have contributed 11-35cm<sup>3</sup>m<sup>-3</sup> globally to the increased atmospheric CO<sub>2</sub> concentration from 1850 to 2000 (Solomon et al., 2007). Soils of Botswana are generally sandy, have low SOC and poor in soil fertility. Consequently, there is a significant potential for carbon sequestration just like the rest of SSA.

There are tried-and-true methods and tactics that increase soil carbon stock in various terrestrial environments (Abdullahi et al., 2018). Most of these methods raise the SOC stock by biomass through photosynthesis. This builds up SOC both above ground by deposition of organic compounds and below ground by root growth and secretions. According to Post and Kwon (2000), By adding more organic matter, altering how easily it decomposes, burying it deep into the soil, and improving the physical stability of the soil aggregates or the synthesis of organo-mineral complexes, the organic carbon level of the soil can be raised. A study conducted by Shahzad et al. (2017) indicated that greatest increase in soil carbon was observed when the required nitrogen was applied as bioslurry as opposed to composted poultry manure and chemical fertilizers. The latter author further indicated that, compared to the control, adding the recommended rate of nitrogen to 8.4 tons of ha<sup>-1</sup> of bioslurry each year enhances soil carbon sequestration by 24.9 tons ha<sup>-1</sup> over 100 years, this sequesters 7.5 tons ha<sup>-1</sup> more carbon than applying the same quantity of nitrogen as poultry manure. In another study, Smith et al. (2014) reported that utilizing bioslurry as organic fertilizer capture less of the carbon in the soil from organic residue than burning on pyrolysis cook-stoves or composting. In Ethiopia, Mengistu et al. (2016) conducted a comparison study on biogas digesters and other organic residues and concluded that substitution of biomass fuels with biogas energy enabled the biogas users to cut GHG emissions roughly by 1.9 of CO<sub>2</sub> equivalents per digester annually. Additionally, decreased usage of chemical fertilizers helped lower GHG emission since bioslurry, the by product from biogas production was used as fertilizer.

In addition to lowering the amount of greenhouse gases (GHGs) in the atmosphere, soil carbon capture and storage supports initiatives aimed at increasing land (forest or agricultural land) profitability (Abdullahi et al., 2018). This is because all methods for storing carbon in soil also boost soil quality and agricultural productivity by improving the amount of organic matter in the soil. Organic matter improves the soil's ability to hold water, hold nutrients, and offer a favourable environment for soil microbes (Lal, 2004). Neba et al. (2022) indicated that soil carbon sequestration aids numerous soil related activities and services such as water holding capacity, structural stabilization retention and release of nutrients and, contribute significantly to the overall soil health, agricultural productivity and efforts to combat climate change.

Stomatal conductance determines the amount of CO<sub>2</sub> entering and water vapor existing plants leaves and so has an impact on photosynthetic rate and transpiration (Kusumi et al., 2012). Plants open and close their stomata to control CO<sub>2</sub> uptake and water evaporation in response to environmental and biochemical cues. According to Kusumi et al. (2012), increase in stomatal conductance in well-watered plants can increase in their CO<sub>2</sub> uptake and subsequently improve photosynthesis (Kusumi et al., 2012). An essential ingredient for plant photosynthesis is nitrogen from organic or inorganic materials because it modifies the distribution of nitrogen among the various photosynthetic components (Yamori et al., 2011). However, excessive nitrogen depositions break the soil nitrogen equilibrium and negatively affect photosynthesis (Wangfeng et al., 2002). Nitrogen deposition has increased dramatically globally because of climate change (Dore et al., 2005) and its effects on CO<sub>2</sub> diffusion conductance (both mesophyll and stomatal conductances, gm and gsc) in photosynthesis have drawn significant attention in the fields of physiological ecology, global change, plant physiology and other related fields. Numerous studies have been conducted on the relationship between gsc and soil nitrogen; generally, gsc rose in response to soil nitrogen increases (Li et al., 2004; Eller et al., 2017). Additionally, several recent articles noted that high N conditions reduced evapotranspiration, which constrained N uptake in almond trees (Kong et al., 2017; Sperling et al., 2019) and this appeared to be associated to the slight decrease in stomatal conductance and leaf N content under high nitrogen condition. Wang chunxue et al. (2022) indicated that organic fertilizers can alter the stomatal conductance of green jujube leaves and hence affect the photosynthetic pace and final yield. According to Ng'etich et al. (2012), in a study on the effects

of composted farmyard manure on growth and yield of spider plant in Kenya, farmyard manure application at 7.5 and 11.5 t ha<sup>-1</sup> increased leaf stomatal conductance and, leaf chlorophyll content by between 16 and 35 percent compared to the control, and increased yield parameters by between 36 and 57 percent. Furthermore, the amount of leaf chlorophyll increased which may be a result of the plants effective uptake and absorption of nitrogen from animal wastes, which serves as constituents of chlorophyll. This has reportedly been directly linked to photosynthetic potential and output of any plant (Biljana and Aca, 2009). Leaf photosynthesis and stomatal conductance are known to be highly correlated (Kusumi et al., 2012). Reduced stomatal conductance in control and greatest fertilizer rates are signs that there is a lack of or an excess of nitrogen in the soil, which inhibits leaf gaseous exchange and reduces photosynthesis, resulting in decreased yield of a particular crop (Ngétich et al., 2012). Furthermore, along with the stomatal conductance, the increased leaf chlorophyll content is one of the contributing factors to high photosynthetic rate in plants that have received the optimum fertilizer levels. In another study conducted by Zangani et al. (2021) nitrogen application decreased stomatal conductance in the early flowering phases whereas the stomatal conductance raised during late flowering phases at 100kg/ha N application, however the results showed that increase in nitrogen level also enhanced the amount of leaf chlorophyll.

## 2.8.2.4 Bioslurry compared to Farmyard manure and chemical fertilizers.

Bioslurry has demonstrated to be an excellent organic manure compared to compost and farmyard manure (Fashaho, 2020) and its structure and composition has fixed two-fold nitrogen content that differ from farmyard manure, and it constitutes readily available plant nutrients at high amounts than farmyard manure and compost (Warnars, 2013). Fashaho (2020) reported that bioslurry contains abundant nutrients whereas farmyard manure loses nutrients through leaching or volatilization due to exposure to sun. Different authors has reported different compositions of bioslurry and according to Mdlambuzi et al. (2021) bioslurry constitutes of

0.5-2.5N, 0.5-1.9P and 0.6-2.2K percent depending on the chemical composition of feed stock used, similarly Kumar et al. (2015) reported 1.60N, 1.55P and 1.0K percent which are on the same range with (Mdlambuzi et al., 2021) while NPK content of farmyard manure is 0.8%, 0.7%,0.7% respectively which is lower that of bioslurry as reported by the latter authors, and even less than NPK content of composted slurry which consist of 0.75, 0.65 and 1.05 percent N,P,K respectively (Amrit, 2006; Karki, 2006). Surendra et al. (2014) reported that animal farm manure contains 0.4-0.8%N, 0.6-0.8% P and 0.5-0.7%K. However, Bonten et al. (2014) suggested that if ammonia volatilization is avoided during anaerobic digestion, the NPK concentrations of bioslurry and farmyard manure may be comparable and according to Nguyen Thi Thu Ha (2015) bioslurry has more organic matter (30-50%) as compared to farmyard manure which has 18-20% organic matter and the latter has slightly acidic pH whereas its counterparts bioslurry contain an alkaline pH (> 7.5) which decreases acidity of soils and increases the ability of P absorption by plants. Also, the author indicated that bioslurry is weed seed free compared to farmyard manure as it contains many weed seeds, insect pupae and harmful bacteria to plants which may cause occurrence of harmful pathogens when applied to soil. Several studies have reported that bioslurry improved crops outputs, Warnars and Oppenoorth (2014) and Nguyen Nhu Ha (2015) reported experiences of farmers in Viet Nam, indicating that farmyard manure can increase crop yields from 10-20% while Warnars (2012) reported that bioslurry raised crop revenues by an average of 25%. As cited by (Shahabz, 2011), Singh (1995) noted that lower yield of crops was produced by 12.5 ton/ha of digested manure and came to conclusion that bioslurry was preferable to farm manure for increasing yields in okra, corn, pea and soybeans. Compared with farmyard manure co-application of bioslurry and recommended amount of chemical fertilizer produced higher yields, making bioslurry superior to farmyard manure for increasing yields. Even though bioslurry and farmyard manure have different nutrient composition, they bring similar effects of increasing yields of crops even though at different levels as they both contain all necessary nutrients for crops growth. Also, they possess high organic matter which when applied to the soil, they will enhance its chemical, biological and physical qualities, such as porosity, water holding capacity, stability, increasing number of beneficial microorganisms in the soil such as nitrogen fixation and phosphorus decomposition microorganisms thus equally effective in improving soil fertility. They increase plants' capacity to hold nutrients while limiting soil leaching and they are both cheap to access, reducing costs of chemical fertilizers (Warnars, 2012).

Chemical fertilizers increase soil nutrients and increase crop production just like bioslurry and farmyard manure, however the chemical fertilizers restore a small part of these soil nutrients and only mineral nutrients are given to the soil without adding any organic matter thus decreasing soil productivity (Warnars and Oppenoorth, 2014). Additionally, over utilization of chemical fertilizers result in nitrate and nitrite contamination in vegetables and similarly their continuous use produces crops that are easily targeted by insect attacks, intrusive weeds and microbial pathogens. Yu et al. (2010) reported that decomposition of organic material in bio slurry is a leisurely process which is good for nutrient absorption by plants as compared to chemical fertilizers. Over- fertilization of chemical fertilizers create negative environmental effects such as increase in greenhouse gas emissions due to high ammonia discharge in to the atmosphere and eutrophication as a result of phosphorus losses to drainage water also these mineral fertilizers are unable to afford them due tom their low economic capacity (Warnars and Oppenoorth, 2014). Therefore bio slurry can be an alternative option this affiliation of financial and environmental challenges associated with the utilization of chemical fertilizers.

#### 2.9 Limitations of Bioslurry as a fertilizer

Bioslurry possess various limitations such as bulkiness because of its high-water content (93%) which restrict its ability to fulfil the total nutrient demand in agriculture areas. There are possibilities of nitrogen loss through volatilization of ammonia, and this depends on contents and viscosity of the bio slurry, pH, total ammonical nitrogen in the slurry and the environmental temperature together with the direction and speed of the wind (Kumar et al., 2021). Bioslurry has a high pH (>7), and this high pH range promotes nitrogen loss through volatilization which impose complexity during handling, storage and field application (Rahaman et al., 2020). Bonten et al. (2014) mentioned risks of nitrogen and potassium leaching during storage when the proper covering is not provided thus high chance of ammonia leaking, therefore bioslurry can be dried or decomposed for convenient handling. The latter author also mentioned the low C/N ratio contents in anaerobic digestates caused by the transformation of carbon rich substrate into methane with susceptibility towards ammonia emissions increasing. Therefore, it is important to consider the C/N content while selecting substrates. Combined digestion with other diverse feed stocks has been reported to improve the process of anaerobic digestion (Goswami et al., 2016).



*Figure 2.1: Production of biogas and bioslurry through anaerobic digestion of pig manure for sustainable agriculture environment at Nnone farm, Ramaphatle, Kweneng District.* 

## CHAPTER 3

## **MATERIALS AND METHODS**

#### 3.1 Study site description

Field and greenhouse experiments were conducted at Botswana University of Agriculture and Natural Resources (BUAN), content farm located in Sebele (24°33'S, 25°54'E, 999 m ASL), Gaborone, Botswana. The research area experiences semi-arid climate characterized by an average summer temperature of 20.7 °C with an average annual rainfall of 526 mm (Jain et al., 2006). The research plot soils are Haplic Lixisols (Nachtergaele and De Wit, 1990) with loamy sand texture.

#### 3.1.1 Planting materials and treatments

Two spider plant accessions (*Cleome gynandra* L.) used in this study were Tot 89-26 and Rothwe. Tot 89-26 (originally from Kenya) was obtained from the Department of Crop Science, University of Namibia, while the other accession (Rothwe) was sourced from the National Plant Genetic Resource Centre of Botswana, Ministry of Agriculture, Botswana. The two accessions are native to Kenya and Botswana, respectively. These countries experience different climatic conditions that contribute to the differences in performance of accessions such as response to soil fertility conditions, growth and yield. Rothwe grows in Botswana where the climate is arid to semi-arid. This climate is characterised by extreme temperatures which range from 29.5°C to 35°C in summer and 19.8°C to 28.9°C in winter and an average rainfall ranging from 250mm in the southwest to 650 mm in the northeast (World Bank Group 2021; Statistics Botswana, 2003). Tot 89-26 grows in Kenya under various climates that include tropical, sub-tropical and temperate. Under these climates, summer temperatures range from less than 10 to 27°C at higher altitudes and 28 to 30°C at lower altitudes and receives an average annual rainfall of 250 mm in northwest and east areas to 2500 mm in western regions (Obiero and Onyando, 2013; World Bank Group, 2021; Parry et al., 2012).

The experiment composed of the two accessions and seven fertilizer treatments. The fertilizer treatments were different levels of bioslurry applied as nitrogen at 0 (control) 10, 20, and 30 tonha<sup>-1</sup>, nitrogen was applied as Urea (46% N) to achieve application rate of 0 (control), 60, 120 and 180 kg N ha<sup>-1</sup>. The experiments were conducted between March and June 2023.

## 3.1.2 Bioslurry sampling and analysis

Pig liquid bioslurry was collected from Nnone Farm Biogas Plant at Ramaphatle Village, Kweneng District located 44km southwest of Gaborone. The pigs were fed with commercial pig ration which was made up of maize grain, wheat, oilseed and oil cake and soybean products. The anaerobic digestion plant with a working volume of 20cm<sup>3</sup> was used to treat the pig bioslurry with a hydraulic retention time of 8 to 20 days with summer temperature that ranges from 28 to 35°C (Nekhubvi and Tinarwo, 2017). Five sub-samples of bioslurry were taken using 2 litre plastic bottles and mixed in a plastic container. From the mixture, a one litre volume of the combined sample of liquid bioslurry was taken for laboratory analysis. The sample was analysed for pH, electrical conductivity (EC, ds/m), organic carbon (OC, %), total nitrogen (TN, %) available phosphorus (P, mg/kg), and available potassium (K, mg/kg), at BUAN Soil Science Laboratory. Also, the density and C/N ratio were determined.

## **3.2** Experimental description

#### 3.2.1 Greenhouse experiment

A two-factor (fertilizer and accession) factorial experiment was laid out in a completely randomized design (CRD) at the Department of Crop and Soil Sciences, BUAN. Two spider plant accessions (Rothwe and Tot 89-26) were used in this study. Polythene planting bags (30 cm diameter and 20 cm tall) were filled with Haplic Lixisols collected from BUAN Garden at

Sebele. Each bag was filled with air dried 7kg soil which was thoroughly mixed and sieved through 2mm sieve. Three seeds of each accession were sown directly into the pots at approximately 2mm depth and the soil was watered to field capacity. After the seedlings had emerged and developed true leaves which occurred seven days after sowing, they were thinned to one seedling per pot. The distance between the replicates was 70cm apart and within the replicates, pots were separated by 50cm. There were 42 experimental units in the entire experiment.

Data on the following parameters was collected; days to 50% emergence and 50% flowering, leaf number, plant height, and leaf petiole length were recorded on weekly basis. The chlorophyll content, stomatal conductance, leaf fresh and dry mass were measured at two weeks interval while root mass, root dry mass, stover yield and nitrogen use efficiency were measured after final harvest of the plants. All other agronomic practices (watering, weeding and pest scavenging and control) were done accordingly.

#### 3.2.2 Field Experiment

A two-factor factorial experiment laid in a randomized complete block design (RCBD) was conducted at BUAN gardens, Sebele. The same treatments applied in the greenhouse were repeated in the field experiment. The experimental site measured 7.0 m  $\times$  22.5m (157.5 m<sup>2</sup>) and this included a 1 m wide perimeter buffer around the blocks and 0.5m between the plots. There were three blocks, each measuring 1.0 m  $\times$  22.5m. Within each block there were fourteen 1m square (1 m  $\times$ 1 m) plots. The seven factorial treatments of the two accessions and fertilizer levels were randomized within each block.

Seeds were sown into seedling trays on 22 March 2023 and seed emergence was at 85% for both accessions. Seven days after sowing, seedlings were transplanted into three rows per plot with 30 cm inter-row and 20 cm intra row plant spacing, totalling 12 plants per plot. Measurements were taken from three plants in the middle row while the two outside rows and the ends of the inside rows were boarder plants. The importance of the boarder plants in the study was to protect the inner plants from influences of other factors. Two weeks after transplanting, three plants in each inner row were selected at random for taking measurements. These plants were tagged for ease of identification during observation and measurement. The same measurements collected at the greenhouse were done in the field experiment. All other agronomic practices (watering, weeding and pests scavenging) were done accordingly.

At planting and transplanting, the liquid slurry was used as the nitrogen supply (N) and it was mixed into the soil following treatments described. The second split of bioslurry was applied three weeks after planting and transplanting. Inorganic fertilizer was applied as urea (46%) following treatments described and it was also applied in two splits, during planting and transplanting and three weeks after. Phosphorus applied as single super phosphate (SSP) and potassium were applied as per the soil test results and according to the recommendations for spider plant (Mutua, et al., 2015).

#### **3.3 DATA COLLECTION**

#### 3.3.1 Climate of the experimental site

Daily rainfall and the atmospheric temperature at the study area were recorded over the experimental period was sourced from Department of Meteorological Services station at Sir Seretse Khama Airport. The station is about 5 km, west of BUAN.

Table 3.1: Average monthly (January-June 2023) weather data recorded during the study period.

Month High temp °C		Low temp °C	Rainfall (mm)	Humidity (%)		
т	21.2	20.1	76.0	22		
January	31.3	20.1	/6.2	23		
February	30.8	19.7	72.4	22.5		
March	29.4	17.8	55.9	14		
April	27.1	13.9	27.9	2.5		
May	24.6	8.8	7.6	0		
June	22.1	5.6	4.6	0		

Source: weatherspark.com/h/m/148546/2023/1 accessed 10 May 2023

#### 3.3.3 Soil sampling and Analysis

Five subsamples were collected from a depth of 0-20cm at the experimental site and completely mixed to come up with a combined sample before and after the experiments. Soils samples collected for both experiments were taken from a fallow land (field experiment soil) which has not been under cultivation for a year and a cultivated land (greenhouse experiment soil) which is mostly used for students' practical lessons. Cowpea, maize, sunflower and sorghum were the main crops found to be grown in the area. Fertilizers containing N, P and K has been applied on this land over the years.

The following soil parameters were measured at the BUAN Soil Science Laboratory: soil pH (CaCl<sub>2</sub>), CEC, SOC, EC, soil particle size, moisture content, TN, available P and K were analysed. A glass electrode meter was used to measure soil pH (Sahlemedihn and Taye, 2000);wet digestion method by (Walkley and Black., 1934) was followed to determine OC; Available P was measured by Bray 1 extracting procedure and ammonium acetate extraction

solution (pH) was used to determine available K and CEC and measured using ICP-OES (Doll and Lucas, 1973; Musse et al., 2020). The amount of TN in soil samples were determined using micro Kjeldahl (1983) method. Hydrometer method (Sahlemedihn and Taye, 2000) was followed to determine soil particle size distribution.

#### 3.3.3.1 Gravimetric soil moisture content

Gravimetric soil moisture content was determined according to Sahlemedihn and Taye (2000).

$$W = \frac{Mw - Dw_s}{Dw_s} \times 100$$
 -------Equation 3.1

Where W = gravimetric soil moisture content (g/g)

 $Dw_s = dry weight of soil (g)$ 

Mw = Wet weight of soil (g)

#### 3.3.3.2 Soil Bulk density

A cylindrical core sampling technique was used to determine soil bulk density (Carter, 1990). Soil samples were oven dried at 105°C to obtain consistent weights (Black, 1965; Sahlemedihn and Taye 2000) calculated using formula:

**Db**  $(g/cm^{-3}) = Wd/Vt$  ------Equation 3.2 Where Db= bulk density  $(g/cm^{-3})$ 

Wd= weight of dry soil (g)

Vt= volume of bulk soil (cm<sup>-3</sup>)

## 3.3.3.3 Soil Total Porosity (%)

Soil total porosity was calculated using soil particle density and bulk density values. The soil particle density (Dp) was assumed = 2.65g/cm<sup>-3</sup> and was utilized for estimating overall soil porosity (Hillel, 1998; Sahlemedihn and Taye 2000) using the following formula:

 $P(\%) = (1 - (Db/Dp)) \times 100$  ------Equation 3.3 Where P = total porosity, Db= bulk density and, Dp = particle density of soil.

#### 3.3.3.4 Field capacity

Soil field capacity was determined by saturating undisturbed soil samples and let them freely drain for two days, while evaporation losses were prevented by covering the core with plastic (Dalgliesh and Foale, 1988). Then moisture content was calculated as described in chapter 3.3.3.1 above.

#### 3.3.3.5 Aggregate Stability

After field sampling, following two weeks of air-drying period, soil samples were run through a 4.75 mm sieve to exclude coarse materials such as plant residues and stones. The sample was thoroughly mixed before placing it in a stack of sieves (4.00, 2.00, 1.00, 0.5 and 0.25 mm). A mechanical shaker (Retsch GmbH and Co. KG 5657 HAAN 1, West Germany) was used to split the soil aggregates following dry sieving procedure by (Krivoshein et al., 2022). In this stack, the largest mesh size was at the top and closed recipient at the bottom. Then a 50 g sample was put onto the topmost of five successively laid out sieves and shaken for five minutes at a rate of 45 rotations per minute. There were five categories of soil aggregates 4.75-2.00; 2.00-1.00; 1.00-0.50; 0.50-0.25 and 0.25-0.00 mm, their mean diameter was calculated. Subsequently, the weight of five dry-stable aggregate (DSA) fractions were determined and expressed as a percentage (%).

**DSA (%)** = (Mi /weight of soil (g)) × 100------Equation 3.4  $MWD = \sum n \ i=1 \ Xi * Wi$  ------Equation 3.5 where DSA = dry-stable aggregate of each size fraction, Xi = mean diameter of each size fraction (mm), MWD = mean weight diameter of aggregates (mm), Mi = weight of each aggregate size fraction (g), and Wi = percentage of the total sample weight occurring in the corresponding size fraction.

#### 3.3.4 Soil carbon sequestration and carbon dioxide (CO<sub>2</sub>) emission determination

Calculating soil carbon sequestration in soil involves determining the amount of carbon stored in the soil over a specific period from a specific depth and area. Soil samples were taken between 0 and 20cm depth using a soil corer at the pre and post experiment to determine initial and final soil OC content as well as BD. Walkley and Black (1934) procedure was followed to determine soil organic carbon while BD measured using the core method (Hallel, 1998).

The OC content in a layer of soil at a particular period is: SOC  $(kg/m^2) = OC (\%) *SD (m)*BD$   $(kg/m^3)$ . Soil OC sequestered was calculated using the formula below (Yan et al. 2013; Zeng et al., 2021).

## 3.3.4.1 Soil Organic Carbon sequestered (SOC)

**SOC**  $(kg/m^2) = (OC_f - OC_0) X BD *SD$  ------Equation 3.6 Where:

 $OC_0$  = initial soil organic carbon (%)

 $OC_f = final soil organic carbon (\%)$ 

Db= soil bulk density  $(g/cm^{-3})$ 

SD= soil depth (cm)

#### 3.3.4.2 Soil carbon dioxide (CO<sub>2</sub>) emission

A method described by Rahman, (2013); Anderson (1982) and Zaman et al. (2021) was followed to determine soil carbon emissions. Carbon dioxide emission was measured four weeks after treatment application. A CO<sub>2</sub> trap was prepared by pipetting 20ml of sodium hydroxide (1.0N NaOH) into 50ml sampling bottles and placed on the surface of the soil in each plot then immediately covered the trap with a chamber (1L beakers of volume 91.61cm<sup>3</sup>). The chambers were covered with a heavy-duty foil to shield them from direct sunlight and pressed 2cm deep into the soil, delimiting a measuring soil volume of 183.2cm<sup>3</sup>. They were placed between plant rows and left for 24hours. The sampling bottles with the trap solution were then removed and immediately sealed tightly and brought to the laboratory for analysis. At the laboratory,  $BaCl_2$  and phenolphthalein solutions were used to titrate the trap solutions with 0.1 N HCl solution. The amount of  $CO_2$  that evolved from the soil was calculated utilizing the formula below.

# $CO_2 (g/m^2/day) = (B-V) * NE$ ------Equation 3.7

Where: B is volume in (ml) of acid needed to titrate NaOH in trap jars from control cylinders to the end point, V is volume (ml) of acid needed to titrate NaOH in the trap jars exposed to the soil atmosphere to the end point, N is the normality of the acid and E is the equivalent weight, to express the data in terms of  $CO_2$ ; E=22. The daily emission of carbon dioxide was expressed as mg CO2 per day per kg of soil.

#### **3.3.5 Plant Growth and Yield Parameters**

In the field, each plot had three rows and data was gathered from the plant row in the middle. Plants in middle row were randomly selected for collecting the following data.

#### 3.3.5.1 Seedling emergence (%)

It was obtained by deducting number of total seedlings emerged from total seeds planted.

## 3.3.5.2 Days to 50% flowering

Days to 50% blooming was determined as the amount of time needed for half of the plants to reach flowering.

## 3.3.5.3 Plant height

A measuring tape was used to measure the plant's height from the soil's surface to the top of its tallest shoot. Data was collected every 7 days after emergence and continued until final harvest.

#### 3.3.5.4 Total leaf number

The total leaf number was determined for every plant labelled for data gathering.

#### 3.3.5.5 Leaf petiole length

The leaf petiole measurement was taken from the beginning of the petiole to the leaf tip.

## 3.3.5.6 Leaves fresh weights $(g/m^2)$

The total mass of fresh leaves was determined immediately after harvesting every two weeks using an electronic balance.

#### 3.3.5.7 Root Count

Following reaping, roots from each plant were uprooted and cleansed using water from the tap, then number of roots counted and recorded.

## 3.3.5.8 Root dry weight/plant

After harvest, the stems of tagged plants were cut at the soil surface level, roots were uprooted and cleansed using tap water and dried for 48hours in an oven at 65°C. The weight of the oven dried roots was determined.

#### 3.3.5.9 Stover yield

All plants were harvested, weighed, and dried for 48hours in an oven at 65°C, the weight of oven dried stover was determined and recorded.

#### 3.3.5.10 Soil-plant analysis development (SPAD)

The level of chlorophyll content in leaves was determined with SPAD-502 meter (Nu-Tech International, India) fourteen days after sowing or transplanting and fortnightly until the end of the experiment.

## 3.3.5.11 Stomatal conductance

Leaf stomatal conductance was determined every after two weeks from three full grown sunny exposed leaves using a leaf porometer (Decagon device, Inc. Sc-1)

# 3.3.5.12 Nitrogen Use Efficiency (NUE)

NUE was determined using the formula by Dobermann 2007; Moll et al. 1982). This NUE formula determines measures agronomic NUE, which measures the amount of applied fertilizer that contributes to yield and it is as followed:

 $NUE = (Yield_f - Yield_0) / N$  fertilizer applied------Equation 3.8 Where: Yield<sub>f</sub> is yield in fertilized conditions kg ha<sup>-1</sup>

Yield<sub>0</sub> is yield in unfertilized conditions, kg ha<sup>-1</sup>

N fertilizer is amount of fertilizer applied kg ha<sup>-1</sup>

# 3.3.6 Plant nutritional analysis

Analysis of plants nutrients was performed using Association of the Analytical Chemists standard procedures (AOAC) (Hussain et al., 2011). Determination of nitrogen content was done by Kjeldahl method (Kjeldahl, 1983). Analysis of calcium, sodium, potassium, magnesium, iron, zinc, phosphorus and manganese was performed using ICP-OES (Hussain et al., 2009; Liberato et al., 2017) plant analysis procedures. After the experiment was over, each of these parameters was determined at BUAN Soil Science Laboratory.



Figure 3.1: Measurements of chlorophyll content (left) and stomatal conductance (right) on leaves of cleome gynandra L.

# 3.4 Statistical analysis

Data gathered from the experiment were subjected to analysis of variance (ANOVA) and least significant difference (LSD) at P=0.05 was used to separate variations between treatment means with the assistance of Statistix 10.0 programme (Analytical Software, Tallahassee, FL, USA).

#### **CHAPTER 4**

## RESULTS

The physical and chemical properties of soils from field and greenhouse studies (Table 4.1) and bioslurry (Table 4.2) were conducted. Textural classes of the soils in the two studies were sandy loam. Soil OC content, TN, available P, CEC, EC and pH were 0.22%, 0% 2.11 mg/kg, 4.56 cmol/kg, 0.06 dS/m and 5.19 for the field experiment. For the greenhouse experiment, soil OC content, TN, available P, CEC, EC and pH were 0.52%, 0%, 37.11 mg/kg, 3.76 cmol/kg, 0.18 dS /m and 7.07 (Table 4.1). Since N is sourced mainly from soil organic matter, which was low, it estimated that N was very low. The soil K was adequate, but P was adjusted to optimum level of 20 ppm as suggested by (Ravikumar and Somashelar, 2013; Singer and Munns, 2002). Also, lime was adjusted to raise the soil pH from 5.19 to 6.5 in the field (Rosen and Bierman, 2005; Goulding, 2016; Holland et al., 2018).

Bioslurry used in the study was alkaline (pH =8.0), EC= 0.04 dS/m, moisture content of 97.62% and OC (1.77%) with dry matter of 2.38%. Total nitrogen, phosphorus and potassium were 3.1%, 290.8 mg/kg and 5908.7 mg/kg, respectively (Table 4.2). This bioslurry was also odourless and brown in colour.

Experimental	Depth	pН	CEC	EC (dS/m)	OC (%)	TN (%)	Available	Available	Texture
site	(cm)	(CaCl <sub>2</sub> )	(cmol/kg)				P (mg/kg)	K (mg/kg)	
Field									Sandy
	0-20	5.19	4.56	0.06	0.22	0.002	2.11	77.18	loam
Greenhouse									Sandy
	0-20	7.07	3.76	0.18	0.52	0.002	37.11	209.10	loam

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Table 4.2: Chemical Composition of Pig liquid Bioslurry.

Organic	pН	EC	OC	C: N	TN	Р	К	Moisture	Dry matter	Density
material	(CaCl <sub>2</sub> )	(dS/m)	(%)	ratio	(%)	(mg/kg)	(mg/kg)	content (%)		$(g/cm^3)$
Liquid pig	8.0	0.04	1.77	5.5	3.11	290.83	5908.7	97.62	2.83	1.02
slurry										

#### 4.1. Effect of Bioslurry and Urea fertiliser on Soil Physiochemical Properties

# 4.1.1 Effect of Bioslurry and Urea fertiliser on Soil Chemical Properties after spider plant harvest. The findings from the greenhouse experiment indicated that soils amended with BS ranged between pH 6.99 and 7.65 while that of soil amended with CF ranged between pH 6.46 and 6.70. pH in the unfertilized soils was statistically the same (p>0.05) with pH from CF60, CF120 and CF180 soils. Figure 4.1a showed that inorganic N fertilizer decreased soil pH from the initial 7.07 while bioslurry increased soil pH. Combination of Rothwe and 30 ton/ha bioslurry resulted in the highest soil pH while CF120 and Tot89-26 resulted in the lowest pH (Figure 4.1b).

Soil analysis after harvest of spider plant demonstrated a substantial (p<0.05) variance in soil pH among treatments (Figure 4.1a). Application of bioslurry at 30 ton/ha significantly increased soil pH from pH 5.10 to 6.09 in the field experiment compared to 180 kgN/ha which did not significantly change the soil pH between CF (pH 5.39) and the control (pH 5.10). The control had the lowest soil pH. The results were not statistically different when bioslurry was applied at 20 ton/ha, 10 ton/ha and urea at 60 kgN/ha. The findings indicated that, as the inorganic N application dose rise soil pH decreased (CF60>CF120>CF180) while pH increased with an increase of organic N rates in the following range (BS10<BS20<BS30). Significant interactions existed between fertiliser treatments and accession (p<0.05) on soil pH among treatments (Figure 4.1c). Treatment combinations of BS30 and Tot 89-26 accession recorded the highest pH (6.12) while the lowest pH was obtained in untreated soils and Rothwe combination.




Figure 4.1: Effects of bioslurry (BS) in ton/ha and chemical fertiliser (CF) in kgN/ha on soil pH in greenhouse and field studies (a), accession and fertiliser combination effects on soil pH in greenhouse (b) and field(c). Results showed a highly significant effect of fertilizer treatments (P<0.05) on soil pH. Means that have same letter(s) do not differ significantly at p=0.05.

c)

Results in Figure 4.2a indicated that soil CEC was not significantly (p>0.05) altered by bioslurry and inorganic fertilizer use, together with their interactions with accession in both field and greenhouse studies. Application of fertilizer had a notable (p<0.05) impact on soil EC in the greenhouse study (Figure 4.2b). Results indicated a significant decline of soil EC in plots fertilized with 120 kgN/ha from the initial EC (0.18 dS/m). There was no significant difference between EC from CF120, CF60, BS10, BS20, BS30 and control while there was no change in soils fertilized with 180kgN/ha.



Figure 4.2: Effects of bioslurry (BS) in ton/ha and chemical fertiliser (CF) in kgN/ha on soil CEC (a) and EC (b) in greenhouse and field studies, means that have same letter(s) do not differ significantly at p=0.05.

Greenhouse study findings demonstrated that bioslurry and inorganic N fertilizer application rates substantially (p<0.05) reduced the final soil OC from the initial (0.52%) recorded on pre-experimental analysis results (Figure 4.3a). The OC concentrations in soils from the control pots significantly (p<0.05) decreased to 0.04% while BS10, CF180, CF60, CF120, BS20 and BS30 recorded 0.07, 0.08, 0.09, 0.10, 0.11 and 0.13% respectively. This changed the soil status from low to very low.

The final OC after harvest of spider plant in the field experiment was found to be significantly higher in BS30 (0.94%) and BS20 than the control and CF180 which recorded lower OC of 0.46 and 0.45% respectively (Figure 4.3a). BS10, CF60 and CF120 were in the range between

the high and low OC recorded in this experiment. The results showed that inorganic fertilizer decreased the soil OC in the range CF60>CF120>CF180 while liquid bioslurry increased soil OC in the range BS10<BS20<BS30.

The findings indicated a substantial (p<0.05) dissimilarity in the interaction of accession and fertilizer application among treatments. Figure 4.3b and c showed that soils where Tot 89-26 and Rothwe were planted under 30 ton/ha had the highest soil OC compared to when the accessions were planted under untreated soils (control) in both greenhouse and field experiments.







Figure 4.3: Effects of bioslurry (BS) in ton/ha and chemical fertiliser (CF) in kgN/ha on SOC in greenhouse and field studies (a), accession and fertiliser interaction effects on SOC in greenhouse (b) and field (c), means that have same letter (s) do not differ significantly at p=0.05.

Results in Table 4.3 below showed that bioslurry and inorganic fertilizer treatment application indicated a notable (p< 0.05) effect on soil TN and K concentration level in the greenhouse study. TN was substantially lower in the unfertilized soils than in bioslurry treated soils which had the highest soil TN. Inorganic fertilizer treatments CF120 and CF180 recorded statistically the same TN as bioslurry treatments. Generally, TN increased as treatment application rates increased. Soil potassium content was highest in BS20 and not significantly different from BS30, CF180 and control. However, the lowest K was obtained in CF120 which was statistically at par with CF60. The findings demonstrated a substantial relationship between accession and fertilizer treatments combination on TN and K (Figure 4.4a and b) while accession alone had no notable impact on soil nutrients. Application rate of 30 ton/ha and Tot 89-26 recorded the highest soil K and TN, (214.20 mg/kg and 0.13%) respectively and the lowest were found in 120 kgN/ha combined with Tot 89-26 and control with Tot 89-26. On the other hand, the results showed that, all treatments and their interactions did not demonstrate any substantial influence on soil P content in the greenhouse experiment (Figure 4.4c).

The application of bioslurry and inorganic fertiliser did not significantly (p>0.05) affect soil TN and K levels, however, a substantial (p<0.001) influence was noted on soil available P

content after harvest in the field experiment (Table 4.3). The highest level of P was observed in BS30 followed by BS20. The lowest P content was recorded in the control treatment which was not significantly different from inorganic fertilizer rates (CF60, CF120 and CF180) and lowest rate of bioslurry (BS10). Soil P levels rose with a rise of N rate of bioslurry application whereas there was no trend with inorganic fertilizer. Accession alone and its interaction with fertilizers were not markable (p>0.05) on soil P content (Figure 4.4d).

	FIELD			GREEN	HOUSE	
Treatment	TN (%)	K (mg/kg)	P (mg/kg)	TN (%)	K (mg/kg)	P (mg/kg)
0	0.02 <sup>a</sup>	40.35 <sup>a</sup>	1.61 °	0.03 °	155.36 <sup>abc</sup>	62.07 <sup>a</sup>
BS10	0.02 <sup>a</sup>	48.94 <sup>a</sup>	2.08 <sup>bc</sup>	0.08 <sup>abc</sup>	155.98 <sup>abc</sup>	79.12 <sup>a</sup>
BS20	0.02 <sup>a</sup>	58.00 ª	2.79 <sup>ab</sup>	0.12 <sup>a</sup>	194.12 <sup>a</sup>	127.33 <sup>a</sup>
BS30	0.03 <sup>a</sup>	49.39 <sup>a</sup>	3.15 <sup>a</sup>	0.11 <sup>a</sup>	168.03 <sup>ab</sup>	89.49 <sup>a</sup>
CF60	0.02 <sup>a</sup>	20.74 <sup>a</sup>	2.10 <sup>bc</sup>	0.05 <sup>bc</sup>	95.99 <sup>bc</sup>	84.35 <sup>a</sup>
CF120	0.03 <sup>a</sup>	16.62 <sup>a</sup>	2.05 <sup>bc</sup>	0.09 <sup>ab</sup>	79.29°	75.42 <sup>a</sup>
CF180	0.03 <sup>a</sup>	104.34 <sup>a</sup>	2.11 <sup>bc</sup>	0.11 <sup>ab</sup>	119.73 abc	102.44 <sup>a</sup>
Significance	NS	NS	***	*	*	NS
LSD	0.23	148.90	0.75	0.06	87.09	81.59
Mean	0.03	48.34	2.27	0.08	138.36	88.59

Table 4.3: Effect of Bioslurry and Inorganic fertilizer on soil TN, K and P

\*, \*\*, \*\*\*, NS. Significant at 0.05, 0.01, 0.0001 or not significant, respectively. Least Significant Difference (LSD) at p=0.05 was used to separate the means; means that have same letter(s) do not differ significantly from each other.



Figure 4.4: Interaction effects of accession and fertiliser on soil TN and K in greenhouse (a and b) and P content in greenhouse and field (c and d) respectively. Means that have same letter(s) do not differ significantly at p=0.05.

4.1.2. Effects of Bioslurry and Urea fertiliser on Soil Physical Properties after Spider plant harvest

Independent application of bioslurry and inorganic fertiliser (Table 4.4) had no substantial (p>0.05) influence on physical properties of soil (bulk density (BD), soil total porosity (DP), field capacity (FC) and soil aggregate stability (MWD) in the field study (Figure 4.5). Fertiliser and accession did not significantly interact (p>0.05) to influence soil physical properties. Control treatment in the field experiment recorded 1.20 g/cm<sup>3</sup>, 54.69%, 8.79% and 0.73 mm of bulk density, porosity, field capacity and structural aggregate stability respectively and the results were at par with treatment BS10. CF180 recorded high (1.31g/cm<sup>3</sup>) bulk density, low

porosity (50.45%), field capacity (8.76%) and structural aggregate stability (0.72mm) (Table 4.4).

In relative to the field study, the greenhouse study results indicated that CF180 recorded low bulk density (1.06 g/cm<sup>3</sup>), field capacity (21.07%), structural aggregate stability (0.78 mm) and high porosity (59.98%). Similar results were registered in BS30 (Table 4.4). Results showed that as fertilizer application rates increased, bulk density decreased while total porosity increased which resulted in increased field capacity of the soil (Table 4.4) and improved structural aggregate stability (Figure 4.5) in the greenhouse.

Table 4.4: Effect of Bioslurry and Inorganic fertilizer on soil bulk density (BD), porosity (DP) and field capacity (FC)

	FIELD			GREEN	HOUSE	
Treatment	BD	DP (%)	FC (%)	BD	DP (%)	FC (%)
	$(g/cm^3)$			(g/cm3)		
0	1.20	54.69	8.79	1.15	56.80	21.63
BS10	1.20	54.72	8.98	1.17	56.02	21.31
BS20	1.22	53.89	9.37	1.14	57.26	23.17
BS30	1.27	52.30	8.58	1.09	58.88	20.54
CF60	1.26	52.35	8.80	1.13	57.51	21.24
CF120	1.29	51.48	8.31	1.11	58.18	22.34
CF180	1.31	50.45	8.76	1.06	59.98	21.07
Significance	NS	NS	NS	NS	NS	NS
LSD	0.18	6.72	1.27	0.14	5.43	9.21
Mean	1.25	52.81	8.80	1.12	57.80	21.61

\*, \*\*, \*\*\*, NS. Significant at 0.05, 0.01, 0.0001 or not significant, respectively. Least Significant Difference (LSD) at p=0.05 was used to separate the means. The results showed a non-significant effect of fertilization on soil BD, DP, and FC.



Figure 4.5: Effects of bioslurry (BS) in ton/ha and chemical fertiliser (CF) in kgN/ha on soil structural aggregate stability (MWD) p>0.05. The means that have same letter(s) do not differ significantly at p=0.05.

### 4.2. Quantitative traits of spider plant as influenced by bioslurry and urea fertiliser.

The results in Table 4.5 below showed that leaf fresh weight, leaf dry weight and root number of spider plant were significantly affected (p<0.007, p<0.02 and p< 0001) by application of bioslurry and urea fertiliser in the greenhouse experiment. BS30 recorded the maximum leaf fresh (19.58 g/pot) and dry (3.35 g/pot) weight followed by CF120 which recorded 18.55 and 3.10g/pot of leaf fresh and dry weight. The least leaf fresh and dry weight were recorded in the control (10.39 and 1.90 g/pot) and BS10 (10.6 and 1.65 g/pot) respectively. Other treatments recorded leaf fresh and dry weight between 1.90 and 19.58 g/pot. Similarly, maximum root number was recorded at BS30 followed by BS20 and the latter treatments were not significantly different (p>0.05) from CF120, CF60 and control. However, CF180 recorded the lowest root number, that was statistically the same with BS10, CF60, CF120 and control. Stover yield showed no notable variations (p>0.05) between fertilizer treatments and accessions treatments. Root dry weight was not significantly (p>0.05) influenced by fertilizer treatment application.

Relative to the field experiment, bioslurry and inorganic fertilizer applications substantially (p<0.05) influenced leaf fresh and dry weight, and root weight of spider plants. Spider plant

grown in soils amended with 30 ton/ha liquid bioslurry recorded the highest leaf fresh (133.23 g/m<sup>2</sup>) and dry (36.89 g/m<sup>2</sup>) weight while lower leaf fresh (40.18 g/m<sup>2</sup>) and dry weight (15.38 g/m<sup>2</sup>) were recorded in the control plots. BS10 resulted in maximum root number (23.33/plant) compared with CF180 which recorded minimum root number (12.83/plant). No significant difference was shown between BS30, BS20, CF60, CF120 or BS10. These findings also revealed that stover dry weight and root dry weight were not influenced (p>0.05) by utilization of liquid bioslurry and inorganic fertilizer (Table 4.5).

Overall, there was a rise in leaf fresh and dry weight, stover dry weight and root dry weight as bioslurry application rates increased in both greenhouse and field experiments.

	GREEN	HOUSE				FIELD				
TREATMENT	LFWT	LDWT	STVDWT	RDWT	RNO	LFWT	LDWT	STVDWT	RDWT	RNO
0	10.39 <sup>b</sup>	1.90 <sup>ab</sup>	1.47 <sup>a</sup>	0.19 <sup>a</sup>	18.00 <sup>bc</sup>	40.18 <sup>b</sup>	15.38 <sup>b</sup>	30.62 <sup>a</sup>	0.57 <sup>a</sup>	13.33 <sup>b</sup>
BS10	10.60 <sup>b</sup>	1.65 <sup>b</sup>	1.50 <sup>a</sup>	0.18 <sup>a</sup>	17.00 °	69.63 <sup>ab</sup>	21.19 <sup>ab</sup>	46.70 <sup>a</sup>	1.17ª	23.33 <sup>a</sup>
BS20	16.01 <sup>ab</sup>	$2.97^{ab}$	2.28 <sup>a</sup>	0.29 <sup>a</sup>	27.17 <sup>ab</sup>	118.4 <sup>a</sup>	32.44 <sup>ab</sup>	48.18 <sup>a</sup>	0.90 <sup>a</sup>	$20.67^{ab}$
BS30	19.58 <sup>a</sup>	3.35 <sup>a</sup>	3.07 <sup>a</sup>	$0.37^{a}$	33.00 <sup>a</sup>	133.23 <sup>a</sup>	36.89 <sup>a</sup>	49.24 <sup>a</sup>	1.60 <sup>a</sup>	14.83 <sup>ab</sup>
CF60	16.12 <sup>ab</sup>	2.90 <sup>ab</sup>	2.72 <sup>a</sup>	0.29 <sup>a</sup>	21.33 <sup>bc</sup>	92.40 <sup>ab</sup>	27.90 <sup>ab</sup>	51.38 <sup>a</sup>	1.01 <sup>a</sup>	23.17 <sup>a</sup>
CF120	18.55 <sup>ab</sup>	3.10 <sup>ab</sup>	2.90 <sup>a</sup>	0.20 <sup>a</sup>	23.33 <sup>abc</sup>	69.54 <sup>ab</sup>	19.78 <sup>ab</sup>	57.95 <sup>a</sup>	1.09 <sup>a</sup>	18.33 <sup>ab</sup>
CF180	14.78 <sup>ab</sup>	2.31 <sup>ab</sup>	2.19 <sup>a</sup>	0.20 <sup>a</sup>	16.33 °	70.96 <sup>ab</sup>	21.07 <sup>ab</sup>	25.06 <sup>a</sup>	1.17 <sup>a</sup>	12.83 <sup>b</sup>
Significance	*	*	NS	NS	***	**	*	NS	NS	**
LSD	8.28	1.67	1.66	0.23	9.86	73.69	19.11	41.56	1.14	3.35
Mean	15.15	2.60	2.30	0.26	22.31	84.91	24.95	44.16	1.07	18.07

Table 4.5: Effect of bioslurry and inorganic fertiliser on leaf fresh weight, leaf dry weight, stover dry weight, root dry weight and root number.

\*, \*\*, \*\*\* and NS mean significantly different at p=0.05, 0.01, 0.0001 and not significantly different, respectively. Least Significant Difference (LSD) at P = 0.05 was used to separate the means; means that have same letter(s) within the same column do not differ significantly from one another. Bioslurry (BS), Chemical fertilizer (CF), leaf fresh weight (LFWT), leaf dry weight (LDWT), stover dry weight (STVDWT), root dry weight (RDWT), and root number (RNO).

## 4.2.1. Influence of Bioslurry and Urea fertiliser on plant height, leaf number and leaf with petiole length

Anova results revealed that the application of liquid bioslurry and inorganic fertilizer had no influence (p>0.05) on the height of plant and leaf number, but substantially (p $\leq$ 0.001) influenced leaf petiole length of *cleome gynandra* L. in the greenhouse. Accession interaction with fertiliser application had no markable effect (p>0.05) on the height of plant, leaf number and leaf petiole length.

Field experimental results showed that plant height, leaf number and leaf petiole length were influenced (p<0.05) by liquid bioslurry and fertilizer application. Highest plant height (27.04 cm), leaf number (23.39) and leaf with petiole length (9.43 cm) were recorded from BS20, CF60 and BS20 respectively. The lowest plant height (17.73 cm), leaf number (10.81) and leaf with petiole length (6.17 cm) were recorded from the control plots. There was no statistical difference (p>0.05) in plant height and leaf number between plants fertilized with inorganic fertilizer and bioslurry. Accession alone, significantly (p<0.05) influenced both leaf with petiole length and leaf number. Tot 89-26 produced high number of leaves (21.13) with long (9.14 cm) leaf with petiole length compared to Rothwe which produced low number of leaves (16.80) with short (6.57 cm) leaf with petiole length. The interaction of accession and fertilizer application had no significant effect (p>0.05) on plant height and leaf number but had a significant (p<0.05) effect of the petiole length. Application rate of BS20 on Tot 89-26 gave the highest leaf with petiole length (10.56 cm) while CF120 on Rothwe resulted on the lowest leaf with petiole length (4.71 cm). The results indicated that Tot 89-26 performs better than Rothwe accession in the field.

Dependent	Effect	GRENNHOUSE		FIELD	
variables					
		F-value	P-value	F-value	P-value
Plant height	Fert	0.70	0.06487	3.26	0.0042
	Accession	2.79	0.0960	0.00	0.9992
	Fert*Accession	0.81	0.5599	0.84	0.5402
Leaf number	Fert	1.48	0.1862	2.39	0.0290
	Accession	0.23	0.6337	3.98	0.0472
	Fert*Accession	0.40	0.8787	1.08	0.3743
Leaf petiole	Fert	3.68	0.0016	12.49	< 0.0000
length					
	Accession	287	0.0918	110.80	< 0.000
	Fert*Accession	1.96	0.0720	2.39	0.0320

*Table 4.6: Summary of ANOVA table for dependent variables: plant height, leaf number and leaf petiole length.* 

4.2.2. Number of days to 50% emergence and 50% flowering as affected by Bioslurry and Urea application.

Results showed no significant (p>0.05) effect of fertilisation on both number of days to 50% seedling emergence and flowering. The maximum days taken to 50% seedling emergence was found in treatment BS30 (3.33) which were statistically comparable to the other treatments. However, the low number of days taken to 50% seedling emergence were observed in BS20 (2.00) as shown in Table 4.7 for the greenhouse experiment. Data regarding number of days to 50% blooming (Table 4.7) indicated that the high number of days to blooming was recorded in BS10 (40.00) followed by BS20 (35.0) whereas minimum days to 50% blooming were recorded in treatment 0 (30.83).

For the field experiment, there was no data for days to 50% emergence as transplants were used therefore only the results for days to 50% blooming was presented. Maximum number of days (37.17) to 50% flowering were recorded in plots where spider plant was grown on soils treated

with 10 ton/ha of liquid bioslurry followed by 20 ton/ha and 60 kg N/ha. The minimum number of days (29.67) to 50% flowering were recorded in the control plots. Overall, none of the two fertilizers had a markable influence on number of days to bloom of spider plant. On the other hand, accession significantly ( $p\leq0.0001$ ) influenced the latter parameter (Figure 4.6). The results showed that Tot 89-26 outperformed Rothwe as it took more days to flower (39.24 and 36.10) while Rothwe took less days (29.05 and 28.20) in both experiments.

Treatment	Greenhouse	Greenhouse	Field		
	Days to 50%	Days to 50%	Days to 50%		
	Emergence	flowering	flowering		
0	2.17	30.83	29.67		
BS10ton/ha	2.83	40.00	37.17		
BS20ton/ha	2.00	34.33	32.67		
BS30ton/ha	3.33	33.00	31.50		
CF60kgN/ha	3.00	32.83	32.17		
CF120kgN/ha	3.00	35.00	29.83		
CF180kgN/ha	3.00	32.50	32.00		
Significance	NS	NS	NS		
LSD	1.62	9.30	8.22		
Mean	2.76	34.14	32.14		

Table 4.7: Effect of Bioslurry and Urea fertiliser on days to 50% seedling emergence and flowering.

\*, \*\*, \*\*\*, NS, means significantly different at p= 0.01, 0.05, 0.0001 or not significantly different, respectively. Least Significant Difference (LSD) at P = 0.05 was used to separate means. The results showed non-significance of bioslurry and inorganic fertilizer application. Key: Bioslurry = (BS), Chemical fertilizer =(CF).



Figure 4.6: Performance of spider plant accessions in response to days to 50% flowering showed highly significant difference (p<0.05). Means that have same letter(s) do not differ significantly at p=0.05.

## 4.2.3. Bioslurry and Urea fertiliser effects on the chlorophyll content and stomatal conductance of spider plant.

Chlorophyll content was influenced (p<0.05) by various levels of bioslurry and inorganic fertilizer in both greenhouse and field studies. Application of fertilizer at 120 kgN/ha in the greenhouse resulted in the highest SPAD units (112.15) followed by 110.75, 108.58, 104.79, 85.28, 77.99 and 60.18 SPAD units from BS20, BS30, CF180, CF60, BS10 and control, respectively (Figure 4.7a).

Leaves of plants subjected to application of bioslurry rate of 20 ton/ha had the highest chlorophyll content (109.18 SPAD units) but was not statistically indistinguishable (p>0.05) from the rest of the other treatments including the control which had the lowest chlorophyll concentration (86.23 SPAD units) in the field study (Figure 4.7a). The results in Figure 4.7b indicated that beyond the application rate of 120 kgN/ha inorganic fertilizer and 20 ton/ha of bioslurry, the chlorophyll content decreased in both greenhouse and field experiments. The influence of accession alone and its interactions with fertilizer application on chlorophyll content were not significantly different (p>0.05) in both experiments.



Figure 4.7: Effects of bioslurry (BS) in ton/ha and chemical fertiliser (CF) in kgN/ha on leaf chlorophyll content in greenhouse and field studies (a), results showed a significant difference of treatment application. The trend of bioslurry and inorganic fertilizer application rates (b). Means that have same letter(s) do not differ significantly at p=0.05.

Results of the study in Fig 4.8a revealed a notable (p<0.05) influence of bioslurry and inorganic fertilizer levels on leaf stomatal conductance. The highest stomatal conductance (600.69 mmol/m<sup>2</sup>s) was recorded at an application rate of 30 ton/ha followed by 20 ton/ha (562.26 mmol/m<sup>2</sup>s), 60 kgN/ha inorganic fertilizer at (464.37 mmol/m<sup>2</sup>s) and the least stomatal conductance was recorded for the control treatments (208.30 mmol/m<sup>2</sup>s) in the greenhouse.

The field experiment recorded the highest stomatal conductance (623.81mmol/m<sup>2</sup>s) from spider plant leaves subjected to 30 ton/ha of liquid bioslurry and the results were at par with BS10,BS20, CF60, CF120 and CF180 which respectively recorded 523.05, 590.47, 565.27, 467.95 and 525.63 mmol/m<sup>2</sup>s. Leaves of plants subjected to control plots had lower stomatal conductance (443.59 mmol/m<sup>2</sup>s) as shown in Figure 4.8a. In terms of stomatal conductance there were no statistical differences (p>0.05) between bioslurry and inorganic fertilizer application rates in all experiments. The interactions of accession with fertilizer application had no discernible (p>0.05) impact on stomatal conductance. However, Tot 89-26 accession performed better (605.77 mmol/m<sup>2</sup>s) than Rothwe (575.18 mmol/m<sup>2</sup>s) under the 30 ton/ha bioslurry rate in the field and Rothwe leaves had more stomatal conductance than Tot 89-26 in greenhouse experiment (Figure 8b and c).





Figure 4.8: Effect of Bioslurry (BS) in ton/ha and chemical fertiliser (CF) in kgN/ha on stomatal conductance (a), accession and fertiliser interaction effects on stomatal conductance in field and greenhouse studies (b and c). Results showed a highly significant difference of

fertilizer application (p < 0.05). Means that have same letter(s) do not differ significantly at p=0.05.

4.2.4. Effects of Bioslurry and Urea fertiliser on Nitrogen Use Efficiency of Spider plant Accessions.

Fertilizer application significantly (p<0.05) influenced the NUE of spider plant in the greenhouse experiment (Figure 4.9a). Spider plant efficiently used nitrogen when 60 kgN/ha of fertilizer was applied compared to the lowest NUE recorded for the control and the highest application of both BS (30 ton/ha) and CF (180 kgN/ha).

Just like in the greenhouse, results for the field experiment showed that, spider plant NUE was significantly (p<0.05) influenced by bioslurry and inorganic fertilizer application (Figure 4.9b). Spider plants were found to be more efficient in the use of nitrogen with low N input. Application rate of 60 kgN/ha resulted in significantly higher NUE (0.96%) and the lowest NUE (0.02 and 0.17%) was found in plants fertilized with 30 ton/ha of bioslurry and 180 kgN/ha respectively. Nitrogen use efficiency of spider plant dropped as the rate of N application increased in both experiments (Figures 4.9a and 4.9b). There was no discernible (p>0.05) relationship between accession and fertilizer application to influence NUE of spider plant. However, Tot 89-26 grown in both the greenhouse and field soils amended with 60 kgN/ha had the highest NUE (0.75 and 0.05%) compared Rothwe accession grown under the same level of fertiliser (59.68 and 4.73%) in both field and greenhouse experiments as indicated in Figure 4.9c and d below.



Figure 4.9: Effects of Bioslurry and chemical fertiliser on NUE in greenhouse (a) and field (b) experiments, and interaction effects of accession and fertiliser (c and d). Results indicated a highly significant variation (p<0.05) of treatment application. Means that have same letter(s) do not differ significantly at p=0.05.

### 4.3 CO<sub>2</sub> Emissions as affected by Bioslurry and Urea fertiliser application.

This study evaluated the effect of biogas slurry and inorganic nitrogen fertilizer on soil carbon dioxide emission. CO<sub>2</sub> fluxes were found to be high (4.88 g/m<sup>2</sup>/day) in the control plots followed by CF180 (3.25 g/m<sup>2</sup>/day). Nevertheless, there was no significant difference (p>0.05) between CO<sub>2</sub> emitted from CF180, BS10, CF120 and BS20. Plots amended with 30 ton/ha (BS30) of bioslurry recorded the lowest (0.19 g/m<sup>2</sup>/day) CO<sub>2</sub> fluxes which was not statistically different from BS20 and CF60 (Figure 4.10a). The results also indicated that as the rate of N

rise in inorganic fertilizer (CF60, CF120 and CF180 kgN/ha) the CO<sub>2</sub> fluxes released increased, but the emissions decrease with an increase of bioslurry rates.

In relative to the interaction analysis, results showed a non-significant (p>0.05) impact of fertiliser treatment and accession combination on CO<sub>2</sub> emissions (Figure 4.10 b). However, the results showed that where accession was planted without any fertiliser application, Rothwe released lower CO<sub>2</sub> into the atmosphere compared to Tot 89-26, same applied to when accessions were grown in soils amended with 30ton/ha.



Figures 4.10: Pie chart (a) showing the influence of Bioslurry (BS) in ton/ha and chemical fertiliser (CF) in kgN/ha and line graph(b) showing interaction effects of accession and fertiliser rates on  $CO_2$  emissions. Results showed a highly significant effect of treatments (\*\*\*) on  $CO_2$  emissions. Means that have same letter(s) do not differ significantly at p=0.05.

## 4.3.1. Effects of Bioslurry and Urea fertiliser on Soil Carbon Sequestration.

Soil OC sequestration was quantified based on the difference between the starting and end soil organic carbon contents. In the greenhouse study, SOC sequestration was significantly affected by bioslurry and urea fertilization. The results in Figure 4.11 indicated that no SOC was sequestered in the soil.

SOC sequestered ranged from 0.38 to 0.44 kg/m<sup>2</sup> in the field experiment and was discernibly influenced (p<0.05) by application of fertilizer (Figure 4.11). A higher carbon sequestration was observed in BS30 plots than in the unfertilized plots. The results indicated that soils amended with bioslurry sequestered more carbon than soil amended with inorganic fertilizer and the control.



Figure 4.11: Effects of Bioslurry (BS) in ton/ha and chemical fertiliser (CF) in kgN/ha on soil carbon sequestration in greenhouse and field. Results indicated a highly substantial impact of treatments (p<0.05) on Soil C sequestration. Means that have same letter(s) do not differ significantly at p=0.05.

#### 4.4 Effects of Bioslurry and Urea fertiliser on Plant mineral content

The mineral content of TN, crude protein, P, K, Zn, Mg, Mn, Ca, Cu, Fe and Na, was determined as described in chapter 3. The findings revealed that bioslurry and inorganic fertilizer had a substantial (p< 0.05) impact on TN and protein concentrations but did not significantly influence other mineral concentrations in the greenhouse grown spider plant (Table 4.8). Highest concentrations of TN and crude protein were found in spider plant grown in soils amended with 180 kgN/ha. This was statistically the same with other treatment applications except bioslurry application rate of 20 ton/ha which recorded the lowest concentrations. The mean mineral content recorded for TN, crude protein, Mg, Na, Mn, P, K

Zn, Ca, Cu and Fe was 0.35%, 2.22%, 38041, 52.34, 4014, 1210.9, 9134.4, 50.02, 22151, 22.85 and 840.39 mg/kg respectively (Table 4.8).

In the field study, bioslurry and inorganic fertilizer had no substantial (p>0.05) impact on mineral content of spider plant (Table 4.9). The mean mineral content was 3472, 27893, 3566.1, 137.52, 6622.8, 3730, 38.90, 0.27%, 1.70%, 11697 and 27.47 mg/kg for, Fe, Mg, Mn, Na, P, K, Zn, TN, crude protein, Ca and Cu. Plants from bioslurry treated soils resulted in higher TN, crude protein, Ca, Fe, Mg, Mn and P while plants from inorganic fertilizer treated soils recorded lower concentrations of the latter minerals even though they had high quantities of Zn, K, Cu and Na.

Treatment	TN	C.	Ca	Cu	Fe	K	Mg	Mn	Na	Р	Zn
		protein									
0	0.35 <sup>abc</sup>	2.16 <sup>abc</sup>	20074 <sup>a</sup>	23.21 <sup>a</sup>	834.6 <sup>a</sup>	29997 <sup>a</sup>	3521.8 <sup>a</sup>	66.86 <sup>a</sup>	1061.3 <sup>a</sup>	9886 <sup>a</sup>	80.95 <sup>a</sup>
BS10	0.38 <sup>abc</sup>	2.37 <sup>abc</sup>	24498 <sup>a</sup>	24.93 <sup>a</sup>	690.1 <sup>a</sup>	39337 <sup>a</sup>	4025.1ª	45.13 <sup>a</sup>	1380.0 <sup>a</sup>	10945 <sup>a</sup>	35.52 <sup>a</sup>
BS20	0.25 °	1.60 °	22692ª	21.41 <sup>a</sup>	833.3 <sup>a</sup>	37773 <sup>a</sup>	4036.2 <sup>a</sup>	42.81 <sup>a</sup>	1322.7 <sup>a</sup>	7611 <sup>a</sup>	40.56 <sup>a</sup>
BS30	0.33 <sup>abc</sup>	$2.04^{abc}$	21752ª	23.73 <sup>a</sup>	861.9 <sup>a</sup>	40947 <sup>a</sup>	4003.2 <sup>a</sup>	47.35 <sup>a</sup>	1305.0 <sup>a</sup>	9543 <sup>a</sup>	93.22 <sup>a</sup>
CF60	0.29 <sup>bc</sup>	1.84 <sup>abc</sup>	23598ª	24.13 <sup>a</sup>	690.5 <sup>a</sup>	36968ª	4306.0 <sup>a</sup>	53.59ª	1157.1ª	9276 <sup>a</sup>	49.77 <sup>a</sup>
CF120	0.43 <sup>ab</sup>	$2.66^{ab}$	22070 <sup>a</sup>	21.65 <sup>a</sup>	834.8 <sup>a</sup>	39325ª	4321.0 <sup>a</sup>	51.29 <sup>a</sup>	1057.2ª	9326 <sup>a</sup>	28.18 <sup>a</sup>
CF180	0.46 <sup>a</sup>	2.86 <sup>a</sup>	20375 <sup>a</sup>	20.85 <sup>a</sup>	1139.6ª	41940 <sup>a</sup>	3884.5 <sup>a</sup>	59.33ª	1193.1ª	7355 <sup>a</sup>	21.99 <sup>a</sup>
Significance	**	**	NS	NS	NS	NS	NS	NS	NS	NS	NS
LSD	0.16	0.98	9136	10.30	673.88	15459	1861.1	47.32	441.69	6036.6	153.6
Mean	0.35	2.22	22151	22.85	840.39	38041	4014	52.34	1210.9	9134.4	50.02

*Table 4.8: Effect of Bioslurry and Inorganic fertilizer on spider plant leave minerals (mg/kg) cultivated in the Greenhouse.* 

\*, \*\*, \*\*\*, NS. Significant at 0.05, 0.01, 0.0001 or not significant, respectively. Least Significant Difference (LSD) at p= 0.05 was used to separate

the means; means that have same letter(s) do not differ significantly from each other.

Treatment	TN	C.	Ca	Cu	Fe	K	Mg	Mn	Na	Р	Zn
		protein									
0	0.24 <sup>a</sup>	1.47 <sup>a</sup>	9442 <sup>a</sup>	22.95 <sup>a</sup>	3643.7 <sup>a</sup>	23283 <sup>a</sup>	3768.2 <sup>a</sup>	132.4 <sup>a</sup>	9870 <sup>a</sup>	3720.4 <sup>a</sup>	37.43 <sup>a</sup>
BS10	0.25 <sup>a</sup>	1.59 <sup>a</sup>	13290 <sup>a</sup>	19.25 <sup>a</sup>	3308.7 <sup>a</sup>	25232 <sup>a</sup>	4194.0 <sup>a</sup>	136.5 <sup>a</sup>	10052 <sup>a</sup>	4389.7 <sup>a</sup>	38.40 <sup>a</sup>
BS20	0.29 <sup>a</sup>	1.83 <sup>a</sup>	13577ª	20.25 <sup>a</sup>	3756.2ª	25117 <sup>a</sup>	3917.7 <sup>a</sup>	114.7 <sup>a</sup>	10157ª	3974.3 <sup>a</sup>	49.31ª
BS30	0.31 <sup>a</sup>	1.92 <sup>a</sup>	12945ª	23.11ª	$4009.7^{\mathrm{a}}$	25365 <sup>a</sup>	3554.8 <sup>a</sup>	161.0 <sup>a</sup>	2320 <sup>a</sup>	3579.3 <sup>a</sup>	34.57 <sup>a</sup>
CF60	0.31 <sup>a</sup>	1.94 <sup>a</sup>	11017 <sup>a</sup>	19.39 <sup>a</sup>	3517.5 <sup>a</sup>	21602 <sup>a</sup>	3307.8 <sup>a</sup>	131.99 <sup>a</sup>	1868 <sup>a</sup>	3462.5 <sup>a</sup>	29.43 <sup>a</sup>
CF120	0.28 <sup>a</sup>	1.75 <sup>a</sup>	11644 <sup>a</sup>	17.55 <sup>a</sup>	2550.9 <sup>a</sup>	20963 <sup>a</sup>	3172.3 <sup>a</sup>	113.1 <sup>a</sup>	1761 <sup>a</sup>	3268.2 <sup>a</sup>	9.21 <sup>a</sup>
CF180	0.22 <sup>a</sup>	1.38 <sup>a</sup>	9962 <sup>a</sup>	69.82 <sup>a</sup>	3517.3ª	53690 <sup>a</sup>	3048.2ª	143.0 <sup>a</sup>	10332 <sup>a</sup>	3715.7 <sup>a</sup>	73.93ª
Significance	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
LSD	0.13	0.84	7093.1	71.26	2526.9	52046.0	1580.2	47.69	20773	2189.7	99.80
Mean	0.27	1.695	11697	27.47	3472	27893	3566.1	137.52	6622.8	3730	38.90

Table 4.9: Effect of Bioslurry and Inorganic fertilizer on spider plant leave minerals (mg/kg) cultivated in the field.

\*, \*\*, \*\*\*, NS. Significant at 0.01, 0.05, 0.0001 or not significant, respectively. Least Significant Difference (LSD) at P = 0.05 was used to separate

means; means that have same letter(s) do not differ significantly from one another.

4.5 Assessment of optimum application rates of Bioslurry and urea fertiliser using different growth and yield components of spider plant



Figure 4.12: Bioslurry and Urea fertiliser application rates for optimum plant height (a and b), leaf fresh weight (c and d), leaf dry weight (e and f), chlorophyll content (g and h) and dry biomass yield (I and j) of spider plant in the field study at p=0.05.

Results of the study above (Figure 4.12) revealed that for optimum growth and yield of spider plant, bioslurry should be applied at 32.9, 59.8, 87.1, 23.4 and 30.5 ton/ha optimum plant height, leaf fresh weight, leaf dry weight, chlorophyll content and dry biomass yield respectively. Furthermore, when applying inorganic N fertiliser, 89.6, 107.3, 97.1, 88.2 and 89.9 kg/ha should be applied for the latter parameters. The results indicated no statistical difference between bioslurry and urea fertiliser rates.

#### **CHAPTER 5**

## DISCUSSION

#### 5.1. Soils of the study areas

The chemical and physical properties of the soils utilized in this study were analysed prior to planting. According to the results, the soil textural class was sandy loam for both the field and greenhouse experiments. The initial pH for greenhouse soil was neutral (7.07) with low TN, SOC, EC, CEC and high available P and adequate K. The initial soil pH for the field study was moderately acidic (5.19) with low TN, SOC, P, low EC, CEC, and adequate K. Low soil available P in the field might be due to the acidic soil pH which is reported to limit presence of P in the soil because of fixation by aluminium, iron, or calcium (Waugh et al., 1973; USDA, 2014) hence limiting plant growth. Soil organic matter is essential for increased crop yields because it is known to enhance soil structure, CEC and the capacity of soil to hold water resulting in good soil health (West and Post, 2002). Carter and Sterwart (1995) reported 2% SOC for optimal soil functions such as water infiltration capacity, structural stability, CEC and aggregate stability. Okalebo et al. (2002) recommended 3% concentration of SOC as the essential level for long term crop productivity in low input tropical soils. Soil nitrogen is also important in maintaining soil quality and crop production (Doran and Parkin, 1994). SOC and TN are directly correlated (Brevik et al., 2018). Lower SOC and lower Soil N availability indicate low soil fertility and pose an additional threat to crop yields. This is in line with the findings of the present study and suggesting that the soils are infertile hence spider plant grown in these soils will have stunted growth and poor yields. Efforts are needed to reimpose soil fertility and hence bioslurry and urea were added in the present study to enhance soil health for long term productivity

#### 5.2. Bioslurry nutrient composition analysis

It is crucial to quantify the nutritional composition of bioslurry prior to enable appropriate application. Results in Table 4.2 showed that bioslurry had 8.0 pH, 97.62% moisture content and 2.83% dry matter. These were like results of Warnars and Oppenoorth (2014) who reported 93% moisture content, 7% dry matter and pH  $\geq$ 7. The low organic carbon content, available P, and C:N ratio, high total N, K and EC were recorded in the current study. High pH and EC values in the slurry implies that it can buffer acidic soils as many plants perform optimally in neutral pH levels and depict the presence of soluble ions for plants root to uptake (Chen et al., 2017). This suggested that the bioslurry used in this study is an organic fertilizer and a potential nutrient pool of plant nutrients as their range was within what Jeptoo et al. (2013) and Fashaho (2020) reported. However, the findings of the current study for organic carbon, P and C: N differ and were less than those documented in other studies (Jeptoo et al., 2013; Fashaho, 2020) and this might be due to the type of waste used and the retention time of the bioslurry used. In line with the present study findings, Jeptoo et al. (2013) observed that bioslurry varies depending on type of feed. Similarly, Warnars and Oppenoorth (2014) reported that many variables which include water, type of waste, breed and age of animals, feed rate, diet, and retention time may determine the composition and value of bioslurry.

## 5.3. Quantitative traits of spider plant as influenced by bioslurry and urea fertiliser.

5.3.1 Number of days to 50% emergence and 50% flowering as affected by Bioslurry and Urea application.

Data recorded for days to 50% seedling emergence revealed that both bioslurry and inorganic fertilizer had no substantial influence on the number of days to emergence. The present results showed that plants in the control treatment took less days (2.17) to emerge and comparable to other treatments. This agrees with the results of a study conducted by Haile and Ayalew (2018) on kale (*Brassica oleracea* L.). High seedling emergence in the control treatments might be

attributed to the low levels of nitrogen which promoted seed germination by lowering the abscisic or gibberellins ratio (Song et al., 2016). Many studies have shown that low levels of nitrogen addition can have positive effect on seed germination (Yan et al., 2016, Tiansawart and Dalling, 2013). BS and CF fertilization increased nitrogen concentration in the soil which might have inhibited seed water absorption inhibiting seed germination resulting in low seedling emergence. Validating this statement, Frazen (2009) indicated that, nitrogen addition increases the salt content in the soil solution which influence the osmotic pressure causing water movement from seed to fertilizer pellet drying out the seed hence drying out the seed causing burn which lowers germination. In contrast to our study, Vaithiyanathan and Sundaramoorthy (2016) reported that administration of the suggested dosage of organic manure, inorganic fertilizer and biofertilizer raised the percentage of green gram seeds that germinated. In a different study conducted by Jilani et al. (2021) on rationalization of bioslurry and chemical fertilizer combination for pea growth and pod yield, less days of pea emergence (5.66) were observed on unfertilized plots while longer days (7.11) were recorded for emergence of pea in plots fertilized with 100:225:225 of NPK in both seasons. According to Liu, Zhang and Lal, (2016) addition of nitrogen can also increase the concentration of ferric iron and aluminium in soils through ion exchange processes that can be toxic to seed and seedlings inhibiting germination. Emergence of *cleome gynandra* L. seedlings in the present study could also be attributed to other factors that influence seed germination and emergence of plants like seed burial depth, soil pH, temperature, light, soil osmotic potential, and dormancy (Javaid et al., 2022; Akbari-Galvardi et al., 2021).

The number of days until 50% of the spider plant flowered was not significantly influenced by either fertilizer application rate. Moreover, no interaction was exhibited between fertilizer and accession. This insignificant effect from applied treatments might be the reason that N from urea and liquid bioslurry favour vegetative growth by its role in physiological and metabolic

function in the plant cells. Similarly, Ghimire et al. (2015) observed a delay in 50% flowering (51.5 days) after fertilizing with NPK at 75:60:50 compared to early flowering (48.67 days) recorded at 100% sole application of bioslurry in okra. Kafle (2010) also discovered shortened days to 50% flowering in green coronet cabbage treated with biogas slurry compost rather than chemical fertilizer. In a study carried by Hong et al. (2016) on chilli pepper, application of bioslurry at 1.4 kg/m<sup>2</sup> or 14 ton/ha recorded first flowering at 28 days compared to 30 days recorded at combined application of bioslurry and rice husk biochar at 50% rate. Early flowering may be explained by rapid increase of vegetative development and keeping enough stored food supplies for buds to differentiate into flower buds, while slow blooming caused by chemical fertiliser may result from the plant's longer vegetative phase being prolonged by the availability of inorganic nitrogen (Ghimire et al, 2015; Jilani et al., 2021). Our research's findings demonstrated that accession had a major influence on days to 50% blooming. Rothwe took less days (28.62) to flowering while Tot 89-26 took longer (37.67) days to flowering. This demonstrates that the accessions used in this study have a comparatively significant genotypic heterogeneity in days to blooming. This agrees with the findings of Ghimire et al (2015) who observed that Jaikisan-62 developed flowers earlier (48.1 days) than the Arka Anamika (50.3 days) variety of okra. According to Omondi (1990), the reproductive stage in vegetable crops hinders leaf formation hence, late blooming allows a genotype to maintain an extended vegetative phase throughout growing cycle. In this instance, farmers may find Tot 89-26 which blossomed in 37.67 days on average as a feasible alternative for cultivation, and for spider plant crops development to produce vegetables in Botswana.

## 5.3.2 Effects of Bioslurry and Urea fertilisation on Plant Growth Parameters

Plant growth and yield parameters are vital agronomic qualities that farmers need to focus on if they hope to reach the population's food needs and generate strong financial income therefore, utilization of fertilizers such as bioslurry to improve these qualities is crucial (Effa et al., 2019). Results in this study indicated that application of fertilizer increased the parameters related to plant development. The maximum plant height (27.04 cm) of spider plant in this study was obtained from application of bioslurry at 20 ton/ha in the field. However, plant height recorded from BS and CF was statistically the same. Bioslurry tended to slowly release nutrients, allowing efficient use of nutrients by plants which attributed to higher plant height in bioslurry fertilized plots. In accordance with this, Warnars and Oppenoorth (2014) reported that available plant nutrients from bioslurry are slowly released for plant uptake, while inorganic fertilizers provide plants with instant nutrients supply (Ibrahim et al., 2014). Bioslurry used in this study exhibited the higher levels of micronutrients such as N (3.11%) and this could explain the higher plant heights documented in this study. Nitrogen is more crucial than other elements for plants development. It affects the growth, yield, and quality of leafy vegetables by influencing cell division, expansion, and elongation, which results in large stems, leaves, and increased quality of crops (Onyango, 2002). The greater plant heights attained in this study could be related to bioslurry's quicker or more effective delivery of the necessary nitrogen, improved soil structure that increase capacity to hold water, thereby encouraging better plant growth (Jeptoo, 2013). Also, Stewart et al. (2005) stated that inorganic fertilizers dissolve easily in the soil and become instantly available to plants. The findings presented in this study are consistent with those documented by Suthar (2009) and Shahabz (2011) who reported a rise in plant height because of higher rate of bioslurry applications. Also, Haile and Ayalew (2018) reported taller plants 37.33cm with 100 kg/ha N application of bioslurry compared to 16.17cm of control treatment. Additionally, Suthar (2009) pointed out that the observed growth in plant height caused by greater dosages of organic fertilizer could possibly be the reason that they contain a wide variety of some critically important major and minor nutrients necessary for healthy plant growth. Similarly, Hailu et al. (2008) reported an increased height of carrots fertilized with organic phosphorus fertilizer (Orga) in comparison

to unfertilized plants, this was linked to phosphorus's capacity to promote early plant growth. Furthermore, Mutoro et al. (2019) reported a significant effect on plant height due to manure application on various accessions of spider plant while inorganic fertilizer application led to a non-substantial increase in the height of plants. The effect of fertilizer on plant height was also supported by Hasan and Solaiman (2012) and Biramo (2017) on cabbage and tomato respectively. The reduced plant height observed in the unfertilized soils might be caused by nitrogen exhaustion in the soil. This is affirmed by Ng' etich etal (2012) who reported that low N inhibits synthesis of proteins and molecules of chlorophyll which are necessary for proliferation of new cells and consequently slows down their growth. Although this study reported plant height less than 50cm, naturally growing *cleome gynandra* L. can reach a height of 150cm Chweya and Mnzava (1997). Jakse and Mihelie (1998) reported that the growth rate of spinach and cabbage was slow where chicken manure was applied compared to where inorganic fertilizer was applied. The stimulating effect of chicken manure on plant growth could be that this type of organic manure acts as soil amendment which improve soil structure parameter such as sandy soil's ability to hold water and enhanced nutrient availability in the rhizosphere which in turn boost plant growth (Ahmed, 2013). These results were consistent with those of Sowunmi and Oyedeji (2019) who found that 100kg N/ha produced taller spider plants. Moreover, a study by Mauyo et al. (2008) reported a considerable increase in the plant height and other growth indices of *Cleome gynandra* L. cultivated in Kenya with varying rates of inorganic fertilizer application. The notable rise in plant height is indicative of the beneficial impact of nutrients included in the fertilizer like N, P and K. These nutrients were most likely easily accessible and given in sufficient amounts to the plant and lead to better vegetative growth.

The leaf number was significantly impacted by the characteristics of the variety as opposed to application of different nitrogen rates (Mutoro et al., 2019). This statement is in line with the

results obtained in the present study which indicate that there was no interaction between accession and fertilization, but accession alone significantly (p<0.05) influenced the increase in leaf number of spider plant in the field experiment. The results in the present study showed that Tot 89-26 had higher (21.13) number of leaves than Rothwe which had the lowest (16.80) number of leaves. This difference in leaf number between the two accessions might be attributed to the different characteristics they possess and the location of growth. Fertilizer application significantly influenced plant leaf number in the field experiment. The lowest (10.81) leaf number was recorded from the unfertilized plots while the highest leaf number was recorded from plants fertilized with bioslurry and urea fertilizer and there was no substantial variation between both fertilizers. Brahma et al. (2002) observed that N boost leaf number per plant and bioslurry contains adequate amount of nitrogen. Similarly, Bachmann et al. (2011) reported bioslurry as a phosphorus and nitrogen source. The findings of this study are in line with the results reported by Haile and Ayalew (2018) who indicated that the highest leaf number (11) per plant was observed from sole application of 100 kg/ha N bioslurry while the control treatment recorded fewer leaf number. Sasanya and Ogedengbe (2019) also reported high number of leaves on plants with soils amended with piggery bioslurry than those produced on control soils. However, the results of Sasanya and Ogedengbe (2019) were contrary to our study as they reported low leaf number with NPK fertilization. High leaf number observed in our study could possibly be the result of better concentrations of essential nutrients in bioslurry, and faster release of nutrients for uptake from inorganic fertilizer. The reduced leaf count per plant in the control treatment may have been caused by the plants' inadequate nitrogen supply, which decreased plant productivity and, in turn, the number of leaves (Shangguan et al., 2000). Generally, the number of leaves is a function of the genetic makeup of the plant as well as environmental factors such as weather (Islam et al., 2010) and nutrient management, and this explains the lower leaf number in control that can be caused by lower nutrient content of the

growth media. Sowunmi and Oyedeji (2019) reported fewer leaves in the control plots where some of the plants in these plots experienced stunted growth as they were dependent on the native soil fertility which, from the result of chemical properties was lacking in certain macro nutrients.

The results indicated a substantial (p<0.05) impact of fertilizer application on the leaf petiole length. The findings are consistent to those of Esan et al. (2021) who observed the longest leaf petiole length after application of 36ton/ha of organic fertilizer. This rise in leaf petiole length by N application from bioslurry was linked to its role in stimulating cell division and elongation which in turn encouraged internode elongation (Raven et al., 1999; Mengel and Kirkby, 2001; Marschner, 2005; Eckert, 2011). However, Hasan and Solaiman (2012) reported the highest (20.7cm) leaf petiole length with urea applied at 330 kg/ha which was statistically comparable with treatment of poultry manure at 15 ton/ha.

#### 5.3.3 Effect of Bioslurry and Urea on Yield and yield components of Spider plant accessions

The yield parameters subsequently increased with increment of liquid bioslurry application rates. This might be ascribed to the availability of adequate plant available ammonium and nitrate together with other nutrients that were contained in bioslurry. This tended to result in high leaf quantity per plant through aided vegetative growth of spider plant. Also, nitrogen in bioslurry may have promoted vegetative development of spider plant which in turn caused photosynthesis to occur at a faster rate in the leaves increasing production (Rahman et al., 1985). These nutrients are necessary building blocks of numerous vital plant components like chlorophyll, protein, amino acids which improve vegetative growth (Haile and Ayalew, 2018). According to Brady and Weil (2002) the plant compounds subsequently encourage carbohydrate synthesis through photosynthesis leading to increased yields. Both organic and inorganic fertilizers constantly influence vegetative growth of cereals and fodder, therefore increased application of inorganic fertilizer and bioslurry optimized yield of spider plant in the

current study. This was comparable to the findings reported by Rahman et al. (2008). Better plant nutrient availability and favourable soil condition brought up by bioslurry application resulted in substantial vegetative growth which led to increased fresh and dry weight of spider plant. The results agree with the research documented by Mehdi et al. (2012) who reported that fertilizing with vermi-compost and municipal solid waste improved plant growth parameters such as plant height and leaf number through which it increased the leaf fresh weight. Production output of vegetables following the use of bioslurry have been documented by various researchers including okra (Shahabz, 2011), maize and cabbage (Karki, 2001; Zhou, 2009), and soybean (Haider et al., 2021). In consistency to the findings of this study, Kodzwa et al. (2023) indicated that organic fertilizers produced yields that are comparable to inorganic fertilizers, indicating that, for *Cleome gynandra*, poultry manure produced 24.38 ton ha<sup>-1</sup> of fresh leaf yield whereas 100:20:150 kg of N, P and K per ha respectively produced 20 ton ha<sup>-1</sup> of fresh leaf yield in their review on the optimization of African indigenous vegetable production in sub-Saharan Africa. In Egypt Abdelraouf (2016) concluded that raising the dose of N fertilizer application up to 224 kgN/ha improved fresh and dry leaf output of spinach. Also, frequent leaf harvesting may have enhanced partitioning of photosynthates to generate new shoots and subsequently leaf production (Frankow-Lindberg, 1997). This is line with the current fresh leaf mass results as harvesting was done fortnightly which might the other reason for high yields. However, Wang and Li (2004) reported that vegetable yields were not increased steadily as the dosage of applied N raised and excessive N fertilizer input inhibited plant growth which resulted in reduced yields. This corroborated with the findings of the present study, as there was a decrease in leaf dry and fresh weight of *cleome gynandra* L. as N inorganic fertilizer rates increased beyond 120 kgN/ha to 180 kgN/ha. A decline in production output at the elevated application rate may be caused by high nitrate levels in the soil brought on by excessive nitrogen fertilizer input (Ng'etich et al., 2012; Sanchez et al., 2004). In conformity

to our study, Eickhout et al. (2006) noted that nitrogen fertilization more than what crops need can also result in reduced yields because crops can only remove 40-60% of applied N from the soil and the rest can be lost through leaching, volatilization, denitrification, surface runoff and N<sub>2</sub>O emissions. In addition, Onyango (2002) and Wei et al. (2009) observed a reduction in leaf output at high N rates which could be a result of elevated levels of soluble N that raises the soil solution's osmotic potential, resulting in a decline in assimilation of water by the roots. On the contrary, the lowest leaf fresh weight from the control was due to potential loss of nutrients content in the soil. This was expected as initial soil analysis (Table 4.1) showed low nutrients level in the soil. Lack of nitrogen inhibits the synthesis of protein and other substances needed for development of new cells thus reducing leaf fresh weight (Kimani et al., 2012).

Biomass dry yield (root and stem) also increased as the concentration of bioslurry increased from 1.50 and 46.70 g/plant to 3.07 and 49.24 g/plant, but the increase reduced from 2.90 and 57.95 g/plant to 2.18 and 25.06 g/plant when inorganic fertilizer rates increased in the field and greenhouse trials. This increment was attributed to increased plant height, shoot growth, and leaf number per plant due to essential nutrients supplied by bioslurry which influenced the growth of spider plant. Khan et al. (2015) reported that bioslurry supplies essential nutrients and accelerates root growth, in addition it has traces of some important nutrients such as zinc, iron, copper and magnesium which are necessary for the growth and development of crops. Moreover, observation and documentation made by Souza et al. (2008) who showed that when organic fertilizer was applied to the soil certain metallic elements enhanced root growth that subsequently raised the dry biomass of kale crop, this supports the present study findings. In addition, Islam et al. (2010) concluded that using bioslurry has a substantial impact on dry matter and ash concentration level of fodder maize. According to Arisha et al. (2015) the organic manure's improving impact on dry weight could be attributed to the growing palisade tissues of leaf and this in turn increases the rate of photosynthetic assimilation and the process

of photosynthesis, which then increases vegetative growth and ultimately the plant's dry weight. Analysis of variance revealed that application of 30 ton/ha bioslurry produced significantly higher (33.0) and  $0.37 \text{g/m}^2$  root count and root dry mass compared to low (18.0) root count and  $0.19 \text{ g/m}^2$  root dry mass recorded in the control treatment. This improvement in root number and dry weight may be related to the elevated potassium levels in bioslurry available to plants which control various plant functions that normally promote plant biomass. This is supported by Haile and Ayalew's (2018) findings, which showed that high root mass and dry matter in Brassica oleracea crops was possibly caused by greater K fertilizer application. Potassium is essential for root development by enhancing uptake transfer from source to sink. Our findings are consistent with those of Mohammed et al. (2019) who found high root dry weight (9.97g and 9.07g) from chicken manure and compost respectively compared to 7.54g obtained from inorganic fertilizer and root volume of 49.1 and 43.6 compared to 40.6 of inorganic fertilizer on lettuce. A study carried by Sandrakirana and Arifin (2021) on soybean revealed that application of 1000 kg/ha of compost produced maximum 10.21g dry root weight. In accordance with the present results, Mohamed and Gabr (2002) reported high (7.0g) root dry weight compared to 5.2g of control for strawberry plant.

# 5.3.4 Effect of Bioslurry and Urea fertilisation on Leaf chlorophyll content of Spider plant accessions

Chlorophyll is a vital biological pigment used by the plants for light capturing which helps in the process of photosynthesis and it is used to measure the wellbeing of the plant (Nkcukankcuka et al., 2021). It has a significant impact in the metabolic activities subsequently economic yield of crop (Chatterjee, 2010). The of synthesis of chlorophyll requires several elements such as nitrogen, phosphorus, and other microelements from soils (Fredeen et al., 1990) therefore, the presence of these nutrients determines the plant chlorophyll content which affects the overall plant growth and photosynthetic capacity (Jeong et al., 2018). From the
findings of this study, it is evident that application of bioslurry and inorganic fertilizer led to subsequent increase in leaf chlorophyll content which might be attributed to efficient absorption of CO<sub>2</sub> and better photosynthesis process. Nitrogen is a vital part of the amino acids, molecule of chlorophyll, energy transfer compounds like ATP as well as other substances required for cell growth and development hence explaining the increase in chlorophyll content and other parameters (Mengel and Kirkby, 2001; Leghari et al., 2016). The current study results indicated that plant leaves had more chlorophyll content (109.18 and 112.15 SPAD units) after being fertilised with 20 ton/ha bioslurry and 120 kgN/ha inorganic fertilizer respectively than plants leaves under the control treatment which had the lowest chlorophyll concentration. Higher SPAD values in the results indicated higher N concentration and absolute chlorophyll in the leaf components which may have boosted the photosynthetic rate of spider plants and, as a result enhanced the overall plant growth leading to increased plant height and leaf yield. In a related study Sowunmi and Oyedeji (2019) reported values between 11.5 and 87.9 spad units in spider plant grown under different rates of inorganic and organic fertilizer. Similarly, Ng'etich et al. (2012) reported the highest 480.0 SPAD at the maximum application rate of 15 ton/ha farmyard manure compared to control treatment which recorded least (38.2 SPAD) leaf chlorophyll content. Complementary findings were reported on lettuce plants and African nightshade species respectively (Silva et al., 2017; Ondieki et al., 2011). In support to the results Liu et al. (2009) confirmed that biogas slurry substantially increased chlorophyll content of red fuji apple leaves at 3.319% higher than the control which might be due to increased chlorophyll density in leaf surface unit and/or increase in minerals needed in chlorophyll production (high magnesium density in slurry) such as magnesium as one of the main elements in chlorophyll structure (Shabanian Borujeni et al., 2005). Dalorima et al. (2018) reported beneficial impacts of vermicompost in enhancing chlorophyll content of watermelon, this suggests that vermicompost contain plentiful nutrients such as Mg, Cu, Fe, N, P and K that are

employed in the formation of chlorophyll, which is necessary for light synthesis and its then converted through photo-assimilation into chemical energy (Tanaka et al., 1998). Theunissen et al. (2010) and Ali (2014) also confirmed that adding 30% more vermicompost, raises total leaf chlorophyll content of marigold plant. Peyvast et al. (2007) postulated that the level of chlorophyll in plant leaves increases rapidly following application of organic fertilizer while Ciecko et al. (2004) indicated that the greater amount of NPK fertilizers is accompanied by higher total chlorophyll content in plant material. Moreover, Cerovic et al. (2012) reported that chlorophyll content is positively affected by fertilizer application especially nitrogen (Hokmalipour and Darbandi, 2011). This confirms the high chlorophyll content recorded from the bioslurry and inorganic fertilizer rates as they provided the plant with readily available form of nitrogen and other micronutrients needed for chlorophyll production. Nitrogen serves as an integral part of chlorophyll, which is said to be positively correlated to the photosynthetic capability and production of a specific plant (Biljana and Aca, 2009). Moreover, Muchecheti et al. (2016) reported that N concentration in leaves is closely correlated to chlorophyll content and may be used as a nutritional indicator. Ali et al. (2017) revealed that leaf chlorophyll is a key for leaf greenness, and it is often used to investigate leaf nutrient deficiencies and changes in chlorophyll. In line to the latter statement, the findings of this study indicated that, the control treatment recorded low chlorophyll content with pale green leaves which might be due to deficient N in the soil.

# 5.3.5 Effect of Bioslurry and Urea on leaf Stomatal conductance of Spider plant accessions Application of bioslurry at 30 ton/ha (BS30) in both experiments had the maximum (600.00

Application of biostury at 50 ton/na (BS50) in both experiments had the maximum (000.00 and 623.81 mmol/m<sup>2</sup>. s) stomatal conductance compared to (208.30 and 443.59 mmol/m<sup>-2</sup>. s<sup>-1</sup>) attained in the control. This maximum stomatal conductance might be due to the nutrients added through bioslurry application that are required to assist in evapotranspiration by plant. The results agreed with those of Dalorima et al. (2018) who reported that increased dosages of

vermicompost raised the watermelon leaf stomatal conductance. According to Khandaker et al. (2017), application of vermicompost resulted in increased stomatal conductivity of chilli. Contrary to the results Ng'etich et al. (2012) reported a decrease in leaf stomatal conductance in plants fertilized with 15 ton/ha farmyard manure while plants that were fertilized with 7.5 and 11.5 ton/ha recorded the highest stomatal conductance. The present findings corroborated with Elshaikh et al. (2018) who concluded that biochar increased leaf stomatal conductance and photosynthetic rate of okra plants. In affirmation, Ye et al. (2022) reported high (0.5014 mmol/m<sup>-2</sup>. s<sup>-1</sup>) stomatal conductance in plants fertilized with biogas bioslurry. This increased stomatal conductance from bioslurry and farmyard manure could be caused by their capacity to enhance hydrological and structural soil characteristics which is paired with increased root development and a rise in potassium assimilation which is necessary for controlling stomatal opening and shutting (Palm et al., 1997). However, the decrease in stomatal conductance in the control treatment and inorganic fertilizer levels could indicate that a deficit or too much N in the soil inhibits exchange of leaf gases which then limit the photosynthesis process resulting in lower yields of a given crop (Ng'etich et al., 2012). The regulation of leaf stomatal conductance is an important mechanism for plants as it is necessary for both CO<sub>2</sub> uptake and utilization during photosynthesis (Ng'etich et al., 2012) the end-product being an improved plant biomass and ultimate increase in yield (Dodd, 2003). Stomatal conductance increases the net photosynthetic rate by controlling CO<sub>2</sub> fixation in the leaf mesophyll tissue and is closely correlated with photosynthesis (Khandaker et al., 2013).

### 5.3.6 Effect of Bioslurry and Urea on Nitrogen Use Efficiency of Spider plant accessions.

Nitrogen is the most critical externally added input for any crop production system. Under use of N is associated with lower crop production while over-use leads to several soil and environmental related consequences (Yadav et al., 2017). In today's society, producing food with as little inorganic fertilizer as possible to lessen environmental impact is essential, hence

enhancing the use efficiency of nitrogen in cropping systems is necessary (Ranjan and Yadav, 2019). NUE offers valuable information on appropriate N fertilizer application, which is essential for sustaining productivity (Yadav et al., 2017). In the current study, Cleome gynandra L. utilized N more efficiently at low N application rates of inorganic fertilizer and bioslurry. Application rate of 60 kgN/ha to spider plant in the field and greenhouse resulted in considerably high NUE of 0.96% and 4.6% respectively. This implies that plants increased their physiological capacity to absorb N which is the limiting nutrient, in order to adjust to a lower nutritional status. Also, it is indicated that spider plant is not a heavy nutrient feeder (Chweya and Mnzava, 1997) and this could explain the reason why it flourishes well under various soil types and abandoned settlements. Our findings support those of Coulibaly et al. (2020) who found that high NUE of Amaranthus cruentus at low N rates of 43.5 kg/ha in contrast to 65 kg/ha of urea. NUE was maximum in reduced N doses and declined with higher N utilization dose in wheat (Sepaskhah and Hosseini, 2008; Faraj, 2011; Mehrabi and Sepaskhah, 2018). Similar results are reported in the current study. This decline observed in NUE with high N above 60 kg/ha was probably caused by increased N losses. Shahbaz et al. (2014) reported that maximum (22.61 g.g<sup>-1</sup>) NUE was observed where bioslurry was utilized at a dose of 600 kg/ha and minimum (5.66 g.g<sup>-1</sup>) was observed where 180:90:40 of N, P and K of inorganic fertiliser was applied. Contrasting results were found in the present study. This low NUE in bioslurry treatments in the present study might be due to losses of N due to gaseous loss of N to the atmosphere and runoff beyond the root zone (Mosier et al., 2001). In this study, Tot 89-26 had higher NUE compared to Rothwe when grown under the same applied N dose of 60 kg/ha. This could be the result of enhanced root morphological parameters including thickness, length, volume and surface area that have significant impact on a plant's capacity to take up soil nutrients (Baligar et al., 2001; Mamman et al., 2007).

#### 5.4. Effect Bioslurry and Urea on CO2 emissions

CO<sub>2</sub> is a significant greenhouse gas that makes up 60% of the total greenhouse impact and its rise in the atmospheric concentration is due to human agriculture practices (Rastogi et al., 2005). Atmospheric CO<sub>2</sub> concentration has been rising at the rate of  $3.2 \times 10^{15}$  g C year<sup>-1</sup> (IPCC, 1996). How quickly carbon is emitted is strongly controlled by the quantity and type of the organic material applied to the soil (Agehara and Warncke, 2005). It is important to comprehend how fertilizer application techniques affect dynamics of soil C, N and eventually GHGs emissions (Collier et al., 2014). From the current results it is indicated that application of bioslurry at 30 ton/ha emitted low CO<sub>2</sub> fluxes (0.19 g/m<sup>2</sup>/day) compared to control treatment (4.88 g/m<sup>2</sup>/day). However, there was no notable variation in the emissions between bioslurry and inorganic fertilizer rates. Comparable findings were reported by Ghu et al. (2013) in their report on the potential of small-scale biogas digesters to improve livelihoods and long-term sustainability of ecosystem services in sub-Saharan Africa. Contrary to the results, Sainju et al. (2012) indicated that applying N fertilizer increased CO<sub>2</sub> fluxes by 14% in contrast to where no N fertilizer was applied. In South Africa Mdlambuzi et al. (2021) reported that biogas slurry and chicken manure recorded elevated CO2 fluxes than chemical fertilizers in both seasons of 2016/2017 and 2017/2018, but the fluxes decreased in biogas slurry application rates later in the season. In agreement with the current results Fares et al. (2017) reported that application of organic fertilizer can assist in reducing CO<sub>2</sub> emission by sequestering carbon into the soil. Composted plant debris and animal manure often produce reduced GHGs emissions in comparison to green manures or synthetic fertilizers (Vallejo et al., 2006; Alluvione et al., 2010). On the other hand, Ray et al. (2020) found out that incorporation of organic amendments in the soil enhanced the amount of soil microbes which in turn stimulated CO<sub>2</sub> fluxes. Ramirez et al. (2010) and Abbas and Fares (2009) indicated that urea fertilization and organic amendments enhance CO<sub>2</sub> emissions in soils as result of greater supply of labile C and N which

enhance microbial activity. In agreement Taneva and Gozalez-Meler (2008) and Ni et al. (2010) observed that organic matter applied to the soil with high quantity of labile carbon enhance CO<sub>2</sub> emissions and thus limits the build-up of carbon in the soil. In affirmation Fares et al. (2017) highlighted that microorganisms are accountable for the breakdown of organic matter, which releases CO<sub>2</sub> fluxes particularly in organic amendments with low C/N ratio like bioslurry. On the contrary, bioslurry used in the present study exhibited low C:N ratio of 5.5 but there were low CO<sub>2</sub> emissions where it was applied, this might be due to the time taken to measure emissions after treatment application. The CO<sub>2</sub> emissions were measured four weeks after treatment application in the current study. In line with the current study findings, Czubaszek and Wysocka-Czubaszek (2018) revealed that CO<sub>2</sub> fluxes were affected when soil amendments were added to the soil thus higher CO<sub>2</sub> fluxes from bioslurry treatment transpired shortly after application (Mdlambuzi et al., 2021). Sänger et al. (2011) confirmed that following addition of biogas slurry CO<sub>2</sub> fluxes increased for a few weeks and then dropped 3-7 weeks after use. In addition, Rahman (2013) reported that the trend of CO<sub>2</sub> emissions from various organic substances declined after incubation period of three weeks with high and low emissions recorded from poultry manure and cow dung respectively in his study on carbon dioxide emission from soil. Reduced emissions of carbon in the present study might also be due to a decline in microbial activity as the growing season progresses. Studies have shown that root respiration affects CO<sub>2</sub> fluxes throughout the growth season (Pelster et al., 2017; Rochette et al., 2000). In line with the latter findings, our results indicated that where Rothwe accession was planted CO<sub>2</sub> fluxes were higher than in plots where Tot 89-26 was planted in combination with bioslurry at 20 ton/ha. This might be attributed to possible increase in root respiration in Rothwe hence high CO<sub>2</sub> emissions. Yi et al. (2007) reported that the addition of organic amendments to the soil resulted in a rise in root and carbon respiration. In addition, Fares et al. (2017) and Collins et al. (2011) reported that degradation of bioslurry and maize roots respiration assisted in the CO<sub>2</sub> fluxes. The discrepancy between the results of this study and those reported elsewhere in the literature (Sänger et al., 2011; Fares et al., 2017; Collins et al., 2011) may be related to the slurries properties and climatic conditions from both studies. The summer season in which the current investigation was carried out saw yearly minimum and maximum temperatures range from 24 to 29 °C and slurry having a pH of 8.0 and C/N ratio of 5.5 while the temperature conditions in the study by Sänger et al. (2011) were regulated at 13.5 and 23.5 °C with the slurry having a pH of 8.9 and a C/N ratio of 8.

#### 5.4.1 Effect Bioslurry and Urea on soil organic carbon Sequestration.

Soil CO<sub>2</sub> emissions lower soil organic pool which in turn affects soil fertility, productivity and soil structure therefore, it is crucial to reduce CO<sub>2</sub> emissions by sequestering carbon in the soil (Rahman, 2013). Soil organic carbon (SOC) serves as a key indication of soil fertility, and its storage into the soil is a key tactic to reduce greenhouse gases (Meng et al., 2014). Soil carbon sequestration has been promoted to combat changes in climate while improving soil fertility (Ma et al., 2023). Moreover, sequestering photosynthetically fixed organic carbon in soil has been suggested as a potentially low cost and highly scalable approach to CO<sub>2</sub> removal. Frey et al. (2014) has indicated that nitrogen input can progressively modify SOC stocks by changing how new organic carbon particles are imported into the soil and out of microbially degraded organic carbon. Prior research has indicated that organic carbon sequestration can vary in three various ways following N addition: rising, steady and decreasing (Sithole et al., 2019; Zhao et al., 2018). In line with the latter studies, findings from the present study showed an improvement in SOC sequestered in the order of bio slurry> chemical fertiliser > control. Comparable findings were reported by Yeasmin et al. (2014). This high carbon sequestration due to bioslurry application may be caused by high level of N in bioslurry which promoted plant growth and development of spider plant hence increasing SOC intake from plant remnants and soil root secretions. These root secretions aid in co-metabolic breakdown of the initial soil

organic carbon by supplying a readily accessible energy source (Wei et al., 2019). Similarly, Bhattacharyya et al. (2016) indicated that using animal waste and crop debris retention enhance carbon addition into the soil, thus reimposing SOC and contributing to SOC storage. According to Du et al. (2018) nitrogen enhance SOC input by encouraging plant growth and plant litter production, simultaneously nitrogen addition inhibits breakdown of SOC by lowering microbial activity and promote aggregates development (Ye et al., 2018). Soils applied with bioslurry showed improvement in soil field capacity, structural stability, total porosity and decreased soil bulk density and improved spider plant growth which might be due to hight SOC sequestered. Neba et al. (2022) affirms that soil carbon sequestration aids numerous soil related activities and services like the capacity to hold water, structural stabilisation, release of nutrients and contribute significantly to the overall soil health, agricultural productivity and efforts to combat climate change. However, there was no sequestration of soil OC in the greenhouse experiment. The reason for this is likely the decreased final SOC recorded in the study (Figure 4.11). Generally, these results indicated that more  $CO_2$  was emitted into the atmosphere instead of being stored in the soil and it is likely that amount of bioslurry and urea added were not enough for SOC decomposition due to low microbial level. It is still unclear how carbon input and sequestration relate to one another under various fertilisation conditions hence the intricate process of soil carbon sequestration can only be investigated in long-term fertilization trials, since it takes a while for soil C pools to balance (Xiang et al., 2022). This might possibly be linked to higher temperatures experienced in the greenhouse during the time when it had some technical faults in the process of the experiment which resulted in the accelerated soil respiration, mineralization and decomposition of bioslurry. In agreement with the findings of the current study, (Knorr et al., 2005; Zhou et al., 2012) reported that climate change, specifically global warming has been the primary factor driving the speeding up the losses of global soil C leading to low sequestration of SOC. In general, a rise in ambient

temperature enhanced mineralization and decay rates of SOC (Parihar et al., 2019): IPCC (2013) reported that changes in climate could affect the kinetics of carbon mineralization and as the environment warms, it works as positive feedback for climate change by speeding up the breakdown of SOC stock through increased microbial respiration and hence forth more SOC is released into the atmosphere (Davidson and Janssens, 2006). In line with the results, Kundu et al. (2007) indicated that soil organic carbon pool is an intricate process which is affected by various factors like fertilization, soil texture, conservation tillage, organic amendments, cropping sequences, climate conditions and human activities. According to Li et al. (2015) vulnerability of soils to elevated temperatures in tropical regions results in accelerated soil organic matter breakdown with more carbon let out to the atmosphere hence low sequestration. Previous studies have reported that soil warming accelerated SOC decomposition (Melillo et al.,2002; Fang et al., 2005) and the effect may have been caused by rising temperatures, which increased the speed of soil respiration and the overall quantity of mineralized carbon. At higher temperatures of 37°C, higher SOC decay rate was recorded under conventional tillage and control plots from reduced soil depth (7.5-15 and 15-30 cm), and they were more sensitive to temperature (Parihar et al., 2019). Similarly soil sampling for SOC analysis of the present study was collected from 0-20 cm depth.

### 5.5 Effect of Bioslurry and Urea fertiliser on Soil Physiochemical Properties

# 5.5.1 Effect of Bioslurry and Urea fertiliser on Soil Chemical Properties after spider plant harvest.

The adjustment of soil pH is key to plant nutrition as majority of the necessary elements such P is dependent on it and similarly Ca, Mg are also influenced by soil pH and affect plant development. The use of bioslurry and inorganic fertilizer influenced the status of soil pH and TN. Application of bioslurry increased pH while inorganic fertilizer decreased it. The high pH of bioslurry reduced the acidity of the experimental soil especially at the field and made it

favourable for the growth of spider plant. Daff (2013) asserted that spider plant requires a soil pH that ranges between 5.5 and 7.0 and Ghehsareh and Samadi (2012) reported that maximum uptake of nutrients is often done at the pH range of 5.5 to 6.5. The findings were consistent with the results from past research by Das and Dkhar (2012) who reported an increase in pH following bioslurry addition which could be attributed to the breakdown of organic substances that release basic cations ( $K^+$ ,  $Ca^{2+}$ ,  $Mg^{2+}$  and  $OH^-$ ) to the soil substituting acidic cations ( $H^+$ , Al<sup>3+</sup>, Fe<sup>3+</sup>), resulting in slight increase in pH. Kebede et al. (2022) and Opala et al. (2012) indicated that utilization of organic fertilizers increases soil pH and reduces exchangeable acidity while inorganic fertilizers influence soil pH. Similar previous studies (Kinaghi, 2016; FAO, 2013) reported similar findings that addition of bioslurry lowers the soil acidity and enhance the standard of agronomic soils by balancing acid conditions. Increase of pH upon application of bioslurry could be attributed to its alkaline nature necessary for survival of methanogenic bacteria and organic matter content (Khalid et al., 2011; Rajendran et al., 2012; Musse et al., 2020). Also, breakdown of plant materials to ammonia by soil microbes through mineralization increases pH since organic anions formed during decomposition consume protons from the soil (Haynes and Mokolobate, 2001; McCauley et al., 2017). The increase in pH found in this study has been reported in other studies. Thesis work of Mwanga (2016) reported an increase by 8.7% and 5.6% in two different sites in Tanzania on use of 6666.7 litres/ha of liquid bioslurry. Rewe et al. (2021) observed a rise in pH levels by 4% and 1% when 400 ml/hill bioslurry from dome and flexi biodigesters were used in maize crop respectively. On contrary Musse et al. (2020) observed a decline in pH by 0.7% when 20.6 and 41.2 cm<sup>3</sup>/ha liquid bioslurry was used.

The decrease in pH observed when inorganic fertilizer was used could be ascribed to nitrification or absorption of basic cations by plants (Rewe et al., 2021). The decrease of soil pH found in this study is like to one observed in the research work of Biramo et al. (2019)

where a decrease by 11.52% and 9.89% in two different tomato varieties were reported on using urea and triple superphosphate fertilizers respectively. Furthermore, he also reported a decline in soil pH by 12.07% and 12.99% in control treatment, in support to the current findings. Opala et al. (2012) noted a pH decrease by 7.82% and 12.94% at 16 weeks after incubation. This decrease in pH could be attributed to inhibition of soil ecological functions of inorganic fertilizer applications due to acidification (Xu et al., 2014; Zhou et al., 2016). Soil CEC was insignificantly affected (p>0.05) by effects of treatment utilization in both experiments. Contrary to our research findings, Odlare et al. (2008) noticed an improvement in CEC and pH in organic amendments soils. Furthermore, Alemneh (2011) observed a rise in CEC after using slurry compost and digested liquid slurry. An insignificant (p>0.05) decrease in soil EC was recorded in the field experiment while in the greenhouse, EC was substantially (p<0.05) affected by fertilizer application. Initial greenhouse soil EC was 0.18 dS/m which significantly decreased in CF180 and was statistically at par with CF60, BS10, BS20, BS30 and control. The findings were in confirmatory to the results reported by Rahman et al. (2008) that biogas slurry reduced soil EC because of its higher ability to exchange cations. Contrary Sarwar et al. (2003) and Niklasch and Joergensen (2001) reported that incorporation of various organic components to soil caused a rise in EC in both alkaline and acidic soils. Atiyeh et al. (2001) observed some improvement in EC with increasing rates of vermicompost with pig manure application. Angelova et al. (2013) reported an increasing trend in EC with the application of compost and vermicompost in a study they carried out on the effect of organic amendments on soil chemical properties.

Nitrogen is the most important element that plants take up from the soil and it contributes more than other nutritional components to growth and development of plants, it is essential for different biochemical and physiological processes (Crawford and Glass, 1998; Leghari et al., 2016). There is a general N deficiency in almost all agricultural soils and cropping systems globally, making the use of external N supply (N fertilizer) essential to produce the crops that meet the continually growing human population needs (Mohan et al., 2015). Soil total nitrogen was significantly increased (0.12%) by application of 20 ton/ha of bioslurry compared to the control (0.03%) but statistically at par with inorganic fertilizer rates in the greenhouse experiment. However, the increase in TN in the field was not significant. In conformity Hariadi et al., (2016) reported that bioslurry contains plentiful amounts of organic N and helps to directly supply of nitrogen to soils. Bioslurry used in this study contained 3.11% of total nitrogen which perhaps explains the rise in total TN after soil harvest from the initial (0.03 and 0.002%) before the experiment. In line with the current study results, Haque et al. (2015) indicated that applying bioslurry to the soil made more nitrogen accessible in the soil. These are consistent with the results of Ngala et al. (2020) who reported that bioslurry contains high nitrogen content which is easily metabolized and made plant accessible. The findings of the current study are corroborated by Lolamo et al. (2023) where bioslurry and CF caused an increase of total nitrogen by 15.38% and 8.3% in contrast to the control treatment respectively. In line with the current results, Musse et al. (2020) found increased TN values with increase in bioslurry as indicated by (Rewe et al., 2021) who observed the maximum increase in TN by 39% when 61.3m<sup>3</sup> of bioslurry was applied. Contrary Krishna (2001) reported that addition of slurry compost and liquid digested slurry to the soil did not alter soil TN. He further, reported that TN increased by 36.67% when using fixed dome bioslurry in maize.

Bioslurry and inorganic fertilizer applications to the soil statistically influenced soil available K as this nutrient increased with increased bioslurry application rates with the highest K obtained from BS20. However, soil available K from inorganic fertilizer applied soils was lower than the control soils, it dropped with rising rates of inorganic fertilizer. Higher K presence following bioslurry application could be related to high amount of K available in bioslurry used in the present research. Bioslurry analysis in the present research showed high

values of K (5908.7 mg/kg). Similarly, David (2015) attributed the increased K availability to the direct addition of K in the pig manure. In Nigeria, Ngala et al. (2020) observed that utilizing 7.5 and 10 ton/ha gave the highest soil K (2.91 cmol/kg) significantly higher than recommended K (1.54 cmol/kg) and control (0.49 cmol/kg) in maize. According to Kebede et al. (2022) application of 5ton/ha bioslurry recorded the highest (2.53 cmol/kg) K over the control and inorganic fertilizer. This indicated that bioslurry increases amount of K in the soil in contrast to inorganic fertilizers. Initial soil accessible P was 2.11 mg/kg and the status of P content in the soil improved significantly after crop harvest in plots amended with 30ton/ha of bioslurry in contrast to other treatments. The increase in soil available P could be because of the high amount of readily available P in the slurry which assisted in liberating more amount of P from soil like other organic manures. Similar findings were documented by Tolanur and Badanur (2003) in pigeon pea. He further indicated that the higher accessible P levels of the soil could be related to the liberation of organic acids during degradation which in turn assisted in letting out P. Hence, the utilization of bioslurry is anticipated to improve the availability of soil P and the assumption from these new findings could be that utilization of bioslurry had a significant contribution in enhancing phosphorus status in the soil more than using inorganic fertilizer. Mohanty et al. (2006) asserted that organic additions can improve the P recovery in the soil by boosting P movement in the soil. Similarly, Taye and Yifru (2010) implied that P availability increased due to the organic matter accumulation that forms complexes with amorphous ion in the soil hindering the binding and immobilisation of phosphates ions. This improvement in soil accessible P due to incorporation of bioslurry could be because decomposition of the fertiliser which solubilise insoluble organic P acids, thus leading to a substantial increase in soil accessible P (Sharma et al., 2013). Low soil accessible P levels in other treatments might be attributed to the sluggish pace of movement of the P element to the plants roots in the soil solution (Ngala et al., 2020; Brady and Weil, 2002). According to Brady

and Weil (2007) and Muindi (2019) P has low mobility and if not lost from the soil through erosion, plant uptake or adsorption, it can persist in the ground for a very long-time causing accumulation of P 'bank' (Nascimento et al., 2018).

In this study for field experiment, the highest and least soil organic carbon was recorded from soils supplied with bioslurry and control soils. This was similar to previous reports (Sarwar et al., 2003; Singh et al., 2001; Ali and Solomon, 2012) implying that bioslurry added greater organic carbon into the soil. In accordance, Franzluebbers (2005) observed a notable rise in SOC with application of poultry manure and Christensen (1996) reported a remarkable improvement in SOC when using organic manure than inorganic fertiliser. Similarly, the current study reported that bioslurry application increased SOC compared to chemical fertiliser. Shahzad et al. (2017) affirms that greatest rise in SOC was noted when required N was used as bioslurry as opposed to composted poultry manure and chemical fertiliser. Some studies suggest that chemical N fertiliser improves SOC over unfertilised soils (Buttle-Bayer et al., 2010). Similar results were reported in the current research findings. According to Lopez-Bellido et al. (2010) and Li and Zhang (2007) indicated that SOC either does not change or decrease with fertiliser application. This agrees with the findings of the research which indicated that SOC decreases with an elevation in N application rate. Though, the findings in the greenhouse experiment reported a decrease in SOC from initial soil SOC of 0.52%. This decrease in SOC could be caused by high temperatures experienced in the greenhouse which favoured the faster mineralisation of the organic matter hence low SOC in the soil. Even though there was minimal improvement in SOC, it made a significant contribution in lowering GHGs emissions, sustaining crop yields and soil health (Meena et al., 2020). In both varieties SOC improved under soils amended with bioslurry compared to chemical fertiliser.

# 5.5.2 Effect of Bioslurry and Urea fertiliser on Soil Physical Properties after spider plant harvest.

The use of inorganic fertilizer and bioslurry did not have any substantial (p>0.05) effect on soil bulk density, porosity and field capacity. This non-significant impact of treatment application on physical properties of soil after reaping spider plant could be due to a shorter experimentation period (May to June 2023). It takes a while for soil amendments to adequately affect soil structure attributes as organic matter did not have enough time to cause soil aggregation as expected. Even though a nonsignificant impact of fertilizer application was observed on soil physical properties, the results of the study indicated a slight decline in soil bulk density and a rise in porosity due to bioslurry application which is affirmed by the findings of Zhao et al. (2009) who observed a decline in soil crusting and bulk density due to organic amendments application. This decline in soil bulk density could be attributed to higher pore spaces, better aggregation, root growth, increased organic carbon and probably macropores in soil amendment in the field (Rashmi et al., 2022). This was consistent with the findings of the field experiment in this research which indicated that bioslurry treatments recorded low bulk density, high porosity and structural aggregate stability, compared to control and inorganic fertilizer. Similarly, Rashmi et al. (2022) confirmed that organic amendments improve soil structure, porosity, enhanced the capacity to hold water and reduced bulk density, a condition favourable for extensive root growth and development to extract plant nutrients. The findings agree with the research findings documented by Majajah and Dhyan (2001) and Khan et al. (2010) who observed that farmyard manure offers benefits in enhancing moisture content which results in a decline in bulk density and improved water diffusion rate. Lolamo et al. (2023) reported 17% reduction in bulk density and 11% rise in porosity compared with inorganic fertilizer. This decline in soil bulk density following utilization of slurry could be due to improvement in organic carbon content which in turn alters soil bulk density and porosity.

White (1997) reported that the value for bulk density was less than  $1g/cm^3$  for soils with high OC content, ranged between 1 to  $1.4 g/cm^3$  for well aggregated soil and was 1.4 to  $1.8 g/cm^3$  for sandy soils. Results from preceding research reported that addition of farmyard manure, bioslurry and other organic fertilizers into the soil improves porosity and lowers bulk density which is closely correlated with enhancing the ability of soil to hold water (Kebede et al., 2022; Khan et al., 2010).

#### 5.6 Effect of fertilisation on Nutrient Content of Spider plant

Numerous research investigations have documented that wild vegetables are an excellent source of micronutrients and occasionally exhibit high levels of nutritional properties in comparison to certain exotic varieties and thus these species have the capacity to eliminate human micronutrients deficits (Lewu and Mavengahama, 2010; Steyn et al., 2001; Ndlovu and Afolayan, 2008). Similarly, Moyo et al. (2018) reported that leaves of spider plant contain higher levels of protein, P, vitamin A and C, Mn, Mg, Fe, Zn, Ca, K, total phenolics and flavonoids than cabbage and Swiss chard. However, the plant's nutritional content is affected by environment, variety, maturity and production processes used (Manyoni et al., 2018; Ekpong, 2009). In agreement with past research, the present findings indicated that fertilizer and accession did not significantly interact on nutrient content in either of the two experiments. However, the fertiliser treatment had a substantial impact on the nutrient element composition of spider plant in the greenhouse study. In this case, only greenhouse study findings are being debated. The results revealed that plants treated with 180 kgN/ha had the highest leaf nitrogen content (0.46%) compared to the reduced N level (0.25%) obtained in 20 ton/ha. Comparable findings were confirmed by Herencia et al. (2011) in Swiss chard. This outstanding effect of urea fertiliser treatments regarding application of N to plant tissue compared to bioslurry treatments indicate that urea carries N which is easily accessible and quickly taken up by plants in the soil (Masarirambi et al., 2010). N content of plants is very significant since it directly

indicates plant protein level (Ahmad et al., 2014). In agreement, the results of the study revealed high (2.86%) crude protein content from pots fertilised with 180 kg/ha of urea compared 120ton/ha which recorded the lowest (1.60%) crude protein content. On contrary Seeiso (2014) reported an increase in crude protein where manure was applied in contrast to the control. Makinde et al. (2010) indicated that protein can be improved with any of the N fertiliser application. Furthermore, he proposed that the rise in protein content could possibly be explained by the reason that N is a vital component in protein buildup. High crude protein can be a result of nitrogen buildup in the juvenile tissues which also get soluble nitrogen carried from matured leaves (Salisbury and Ross, 1991). On contrary Kujeke et al. (2017) reported insignificant effect of fertiliser application on protein, calcium and iron in Zimbabwe. The elevated P was obtained in plants where 10 ton/ha of bioslurry was applied. In line with the present results, Warnars (2014) entrusted that P content improved on Swiss chard leaves after fertilization with bioslurry. According to Dumani et al. (2021) bioslurry like any other organic fertilisers enhance soil water holding capacity and physical structure which leads to deeper root growth and increased soil microbial activity that affects accessibility of micronutrients level in soils to plants. In their study with Swiss chard, Dumani et al (2021) reported high P level in leaves of Swiss chard fertilized with 20 litres/4.5m<sup>2</sup> of cow dung slurry. Warnars and Oppenorth (2014) reported that bioslurry includes substantial quantities of macro nutrients such as P that are easily accessible for plant absorption and improves soil physical and chemical properties of the soil which enhance nutrient uptake by plants. The present study results also indicated that high potassium content of leaves was recorded in inorganic fertiliser compared to bioslurry and control. The low efficacy of bioslurry could be explained by the sluggish release of nutrients from their slow mineralization rate, which is a characteristic of organic fertilisers. These findings are consistent with the results of Muhmood et al. (2014) who confirmed high micronutrient content levels in chilli and spinach leaves treated with NPK rates of 55:76:0 and

125:75:60 kg/ha respectively. Similarly, Dumani et al. (2021) reported high potassium content on plants fertilized with NPK 2:3:4 (30) while the lowest potassium was reported in plant fertilized with 40litres/4.m<sup>2</sup> of bioslurry. Plants fertilised with 10 ton/ha of bioslurry produced leaves with the highest calcium content in contrast to other treatments. Contrary, Ndololwana (2015) reported a significant high Ca level in Swiss chard leaves grown in unfertilized soils. Highest Fe, Mg and Mn content was found in plant leaves fertilised with inorganic fertiliser. In line with the results, Muhmood et al. (2014) showed high micronutrient content of Swiss chard leaves after fertilisation with inorganic fertiliser. Sowunmi (2015) reported that 100 kgN/ha improved the absorption of mineral nutrients like Ca, K, Fe, Na, Cu, Mn, Zn, Mg, while manure from goats only enhanced phosphorus absorption in plant, in a study she carried out on nutritional value and cultivation requirements of *Cleome gynandra* L. in South Africa. This suggests that elevated levels of micronutrients shown by urea is because of its faster release of nutrients to the soil. The low micronutrient content in bioslurry plots could be due to the gradual nutrient release to the soil by bioslurry like other organic fertilisers. Generally, this plant could be considered as possible contributor of essential nutrients needed for human consumption.

#### 5.7 Bioslurry and Urea application rates for optimum growth and yield of Spider plant

Though research on production and growing techniques of African leafy vegetables such as *C. gynandra* L. is scarce, there is a suggestion that such crops have little water requirement and thrive well in degraded soils (Aworh, 2015). However, there are claims that these ALVs can produce more even without the need to use production inputs such as fertilizers. According to Daff (2013), spider plant is acclimated to various soil types from clay loams to sandy and it prefers a pH range of 5.5 to 7.0. Schippers (2000) reported that utilization of fertilisers with high quantities of N enhanced leaf size and number. Similarly, Sowunmi and Oyedeji (2019) reported that application of 100 kgN/ha increased leaf number, plant height, stem girth and chlorophyll content of spider plant. Kimani et al. (2012) advised farmers to apply 80 kg/ha of

N for production of spider plant whereas Kujeke et al. (2017) recommended 30 ton/ha and 10 ton/ha of cattle and poultry manure respectively, for spider plant growth in Zimbabwe. Ng'etich et al. (2012) suggested that utilization of 150 kgN/ha from calcium ammonium nitrate is preferred in improving production hence approved for spider plant production in Kenya. Moreover, he recommended 11.5 ton/ha of farmyard manure for cultivation of spider plant by farmers. Mauyo et al. (2008) affirms that utilization of 80 kgN/ha improved growth and output parameters of spider plant. According to Warnars (2014) of 10 to 20 ton/ha of bioslurry resulted in substantial improvement in spider plant output in irrigated fields. In line with the findings from previous studies, our research reported results in the same range for bioslurry and urea fertiliser. Therefore, it is suggested that farmers should not use more than 20 ton/ha of bioslurry or 100 kgN/ha of urea fertiliser for maximum yields of spider plant, to save costs, improve soil physio chemical properties and conserve the environment in the study area.

# **CHAPTER 6**

# **CONCLUSION AND RECOMMENDATIONS**

#### 6.1 Conclusion

The results of this study showed that the impact of bioslurry and urea on the growth, development and yield of spider plant were not significantly different. Thus, bioslurry can be used as alternative source of nitrogen for spider plant cultivation. Bioslurry, significantly improved soil properties. There was no significant difference of NUE between bioslurry and urea fertilizers. NUE was high and low under low and high fertilization rates, respectively. This study determined that field fertilizer optimum rates are 20 ton/ha for bioslurry and 100 kgN/ha. Bioslurry as compared with urea fertilizer and control, improved carbon sequestration and reduced carbon dioxide emission from the soil. Besides enhancing soil health and crop production, the use bioslurry as soil amendment can positively contribute to sustainable agricultural production.

#### **6.2 Recommendations**

The study should be extended to other agroecological regions of Botswana and other crops grown under differing soil water regime. Also, the quality of produce must be evaluated to reveal the effect of different fertilizers on marketability of spider plant. Furthermore, this research needs to include on-farm trials as direct information dissemination and to initiate interest of the famers on this new wild and indigenous vegetable.

#### CHAPTER 7

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## Appendix 1



Figure 1a: Biogas plant at Nnone farm, Ramaphatle



Figure 1b: Bioslurry harvesting from the digester effluent tank.



Figure 1c: Land preparation and experimental layout at BUAN garden



Figure 1d: Soil CO<sub>2</sub> capture at the field



Figure 1e: Harvesting of spider plant in the greenhouse