HYT BIOFERTILIZERS AND BIOCHAR EFFECTS ON THE GROWTH, YIELD AND FRUIT QUALITY OF OKRA IN THE FOREST ECOLOGICAL ZONE OF GHANA.

BY

SASU GEORGE MENSAH

(10358693)

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DECLARATION

I hereby declare that except for references of other people's works which have been cited and duly acknowledged, this work is the result of my original research and that this thesis has neither in whole nor in part been presented for an award of a degree elsewhere.

SASU GEORGE MENSAH

(Student)

DATE----------

PROF. G.O. NKANSAH

(Supervisor)

DATE----------

DR. (MRS) C. AMOATEY

(Co-Supervisor)

DATE----------

ABSTRACT

Two different studies were conducted at the Forest and Horticultural Crops Research Centre FOHCREC), Kade- Okumaning, from May 2011 to April 2012. The main objectives were to investigate the biological effects of biofertilizers, inorganic fertilizer and biochar on the growth, yield and fruit quality of okra in the forest ecological zone of Ghana. High Yield Technology (HYT) biofertilizers, biochar and inorganic fertilizers at three levels each, 100%, 50% and 0% were considered. Two different experiments were set up on the field during the major and minor growing seasons. Twenty- seven treatments arranged in a randomized complete block design (RCBD) were replicated four times. Biochar was incorporated into the first 5cm of the soil to mix with the entire soil. HYT application was first done ten (10) days after emergence (DAE) and this was followed weekly, alternating the soil with foliar and inorganic fertilizer at the recommended rates. Data were collected on factors including vegetative growth characteristics, yield and fruit quality, soil chemical as well as microbial properties. Data was analyzed using analysis of variance (ANOVA). The results showed that application of HYT and biochar at 50% and 100% increased significantly the growth characteristics including plant height, stem diameter, fruit number, number of leaves, fresh and dry weight of shoots and roots, days to 50% flowering, days to harvesting, nutrient composition of plant foliage, nutrient uptake and reproductive characteristics such as days to flowering, fruit number and harvesting dates. HYT biofertilizers resulted in 62% increase in fresh fruit yield over inorganic fertilizers. A combination of HYT biofertilizers, biochar and inorganic fertilizers also yielded 55% better than inorganic fertilizer amended with biochar. Biochar also enhanced HYT efficiency by 8%.

DEDICATION

I solemnly dedicate this work to my mother Mad. Janet Korkor Abeka popularly known as Abena Korkor for her sacrifice towards my upbringing, education and this work. Sweet mother may the good Lord bless you and increase your years.

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LIST OF ABBREVIATIONS

PDB……Phosphorus dissolving bacteria

MVA……..Mycorrhiza vesicular arbuscular

NPK……..……Nitrogen phosphorus potassium

N……Nitrogen

P…… Phosphorus

K……... Potassium

O.M……....Organic matter

SOM……..Soil organic matter

OC……..…Organic carbon

DAG……..……Days after germination

DAS……..…Days after sowing

FYM……..……Farm yard manure

Cv …….Cultivar

FAO…….. Food and Agricultural Organization

FAOSTAT……FAO Statistics

HYT……..……High Yield Technology

SOC…….….Soil organic carbon

C……...……Carbon

CEC……..…Cation exchange capacity

O2.....Oxygen

OM……...Organic matter

EC…….Electrical conductivity/conductance

Cu…….…..Copper

Ca …….…….Calcium

Mg…….….Magnesium

C:N……...….Carbon nitrogen ratio

IAA…….…….. Indole acetic acid

NAA …… Naphthalene acetic acid

GA…….... Gibberellic acid

RDF……...Recommended dose of fertilizer

VAM…….…Vermicompost and manure

ADP……..Adenosine diphosphate

ATP……...…Adenosine triphosphate

SSA……..Sub-Saharan Africa

CHAPTER ONE

INTRODUCTION

Okra, (*Abelmoschus esculentus*, L. (Moench) belongs to the *malvacea* family. There are two cultivated types of okra, (*Abelmoschus esculentus*, L. (Moench) and West African okra, *(Abelmoschus caillei).*Okra plays an important role in the diet by supplying carbohydrate, protein, fat, minerals and vitamins that are usually deficient in the staple food. Okra is basically low in calories and dry matter constituents which when consumed in a meal with basic starchy food makes the food more palatable (Savello *et al*, 1982).

It is an important vegetable crop grown throughout the tropical and subtropical regions of Asia and Africa (Bisht and Bhat, 2006). Okra is believed to originate probably from South East Asia. It is popular in West Africa, Brazil, Phillipian, Thailand and India (ECHO, 2003). It is distributed also to other parts of the globe by the Portuguese (Sinnadurai, 1992).

India ranks first in the world with 35million tons (70% of the total world production) of okra produced from 0.35million hectares of land (FAOSTAT, 2010). The West and Central African regions account for more than 75% of okra produced in Africa, but the average production in the region is very marginal (2.5t/ha) compared to East (6.2t/ha) and North Africa (8.8t/ha)(FAOSTAT, 2006). In West Africa, Nigeria is the largest producer (1,039,000 t) with Ghana placing third (897,381t) after Cote d'lvoire.

In Ghana, okra is planted either as a sole crop or as an intercrop and it is grown extensively in terms of tonnage in Brong Ahafo, the three Northern regions, Volta, Greater Accra and Central regions, (NARP,1993). Okra is used for several purposes locally; the immature green pods and fresh leaves are used as pot herbs. Immature pods are steamed as baby okra, deep

fried, pickled or canned. The tender pods are used in stews or cut into slices and sun dried then grounded as powder and used in several dishes (Abdelmageed, 2010). The fruit also serve as soup thickeners (Schippers, 2000). The leaf buds and flowers are also edible. Okra seeds could be dried and used to prepare vegetable curds, (a nutritious material).

In Africa, okra is cultivated because of its high mucilage content which is used in thickening soup (Purseglove 1968, Wolfe *et al*., 1977). Fresh okra is high in vitamin A, B and C and in calcium (NARP, 1993). Significant levels of carbohydrates, potassium, magnesium and other vitamins are also present in okra (Norman, 1992, Adeboye and Oputa, 1996). Reports indicates that a good source of affordable vitamins, calcium, potassium and other minerals which are absent in the diet of most developing countries are supplied by okra (IBPGR, 1991). Essential and non- essential amino- acid that okra contains is comparable to that of soya bean. Okra therefore plays an important role in human diet. The green tender fruits of okra are highly nutritious containing 1107mg calcium and 8.9 mg of Iron for every 1000 g edible portion and fair amount of vitamins viz., A, B and C. It is also rich in protein and crude fiber, (Sona,Thamp and Indira, 2000). Recently, attention has been given to the use of okra seeds as sources of proteins (about 20% of dry matter) and vegetable oil (about 14 % of dry matter). Seeds contain mainly monounsaturated fatty acids (oleic) and palmitic acid (Martin and Rhodes, 1983) and have high lysine levels. The roasted seeds are used as a substitute for coffee. It is a potential export earner accounting for 60 percent of exported fresh vegetable (Sharma and Arora, 1993). Apart from its nutritive value, the stem and fruit sheath is used in the manufacture of paper as they contain of crude fibre.

The protein content of okra seeds varies between 15% - 26% and has edible oil content greater than 14% (NARP, 1993). For medical purposes, okra is very useful against genito-urinary disorders, sperma- torrhoea and chronic dysentery. It is reported that, okra is used in curing ulcers and relief from hemorrhoids (Adams, 1975).

A mucilaginous preparation from the pod can be used in plasma replacement or blood volume expander. The mucilage is also used to glace paper and also useful in confectionary (Markose and Peter, 1990). Almost all parts of the okra plant are consumed.

Continuous use of agricultural land over several years creates an imbalance in the store of nutrients available. Also, increase in cropping density and introduction of high yielding varieties have caused considerable drain of nitrogen and crops showed a positive response to the addition of nitrogen in the soil (Ali *et al*., 2004). Despite the nutritional value of okra, its optimum yields (2-3t^{-h}) and quality have not been attended in tropical countries partly because of continued declined in soil fertility

In Ghana, population pressure and shrinking level of land availability, declining soil productivity and issues around energy security and climate change have all had a significant effect on crop production and yield (Stoorvogel *et al*., 1993).

Serious depletion of soil fertility due to widening gap between nutrient removal and suppliers (Ramesh, 2008) has affected crop productivity. The use of inorganic fertilizers alone has not been helpful because it promotes and increases degradation of plant nutrients (Sharma and Mittra, 1991). The degradation accordingly is brought about by loss of organic matter which consequently results in soil acidity, nutrient imbalance and low crop yields.

Heavy application of chemical inputs, greatly deteriorate the environment and also decrease production (Nishio, 1996). Tropical soils after receiving chemical fertilizers tend to be unproductive due to lack of proper amendments of organic matter. With the growing environmental concerns the sole dependence on chemical input based agriculture is being replaced by integrated multi- approach involving conjunctive use of both organic and inorganic sources.

According to Ramesh, (2008), the use of organic manures particularly bio- fertilizers are the only option to improve the soil organic carbon for sustenance of soil quality and future productivity.

Bio- fertilizers are the formulation of living micro- organisms which are able to fix atmospheric nitrogen in the available form for plants either by living freely in the soil or being associated symbiotically with plants (Subba Roa *et al*., 1979). They are input containing micro- organisms which are capable of mobilizing nutritive elements from non-usable form through biological processes (Tient *et al*., 1979).

Bio-fertilizers are eco-friendly and supply the nutrient input of biological origin for plants. They are not only important for the reduction of quality chemical fertilizers but also for providing better yield in sustainable agriculture. Bio-fertilizers have been identified as alternatives to chemical fertilizers to increase soil fertility for crop production in sustainable farming (Amin, 1997). Phosphorous is a major growth limiting nutrient and unlike nitrogen, there is no atmospheric source that can be made biologically available (Ezawa *et al*., 2002). The role of phosphorous in increasing the yield and improving the quality of crops is well known. Soil micro- organisms play a dominant role in soil phosphorous dynamics and subsequent availability of phosphate to plants (Richardson, 2001). The benefit obtained from the use of organic materials as soil amendments have however not been fully utilized in Sub-Saharan Africa due to huge quantities required to satisfy the nutrient requirements of the crop as well as transportation and handling costs which constitute a major setback. They are thus rarely available in small subsistence farms in the required quantities (Nyathi & Campbell, 1995). Additionally it is capable of fixing nitrogen, mobilizing phosphate and micro-nutrients through the production of organic acids and lowering pH (Seber, 1993).

Climate change is now widely perceived as a serious threat to both human and the environment. It is important to recognize that the removal of excess $C0_2$ from the atmosphere will provide positive drive towards climate change mitigation supported by measures that will reduce or lower greenhouse gas emissions. A strategy that will ensure the removal of atmospheric $C0_2$ management is the use of bio char (Sombroda *et al*., 2003, Lehmann 2006).

Hamer *et al*., (2004) identified that some micro-organisms were able to live with Black Carbon (BC) as the sole Carbon(C) source and that Black Carbon in soils may promote the rate of decompositions of labile C compounds. Bio char is a charcoal substance produced from controlled, incomplete combustion of biomass in an oxygen- free environment. It creates virtually a permanent carbon sink in prime soils that have multiple environmental benefits when treated as soil amendments. Thus it can remove CO_2 in large quantities to combat climate change. Bio char gives the soil its black colour and improves soil structure, aggregation, water infiltration and retention and nutrient storage capacities (Lehmann *et al*., 2003). One significant feature of bio char is that it may increase stabilization of organic matter nutrient sources in the soil (Glaser *et al*., 2001) and reduce nutrient leaching losses (Lehmann *et al.,* 2003) and hence improve nutrient retention.

Biochar is important as a soil conditioner and also helps to spread and transform nutrients (Glaser *et al*., 2002; Lehmann *et al*., 2003). This therefore, makes it possible to modify N, P and S transformation in mineral soils. Besides, it has a high surface areas that is highly porous and so has the ability to increase soil water holding capacity, Cation Exchange Capacity (CEC) and surface sorption capacity when added to the soil (Glaser *et al*., 2002; 2004; Keech *et al*., 2005; Liang *et al*.,2006). It has been proven to have the ability to influence the population of soil microbes (Pietikainen *et al*.,2000) and also lower soil bulk density Gundale and Deluca, (2006). Bio char according to (Berglund *et al*., 2004), greatly increase net nitrification. It may also act as a habitat for soil micro- organisms (Pietikainen *et*

al.,2000) that are responsible for N, P and S transformation and has the capacity to support the presence of absorbed bacteria (Pietikainen *et al*., 2000; Rivera Utrilla *et al*., 2001). Bio char production is a perfect way of recycling forest and crop residues, mill residues, field crop residues and urban waste.

The use of bio char to meet the C and N requirements for crops would be inevitable in the years to come for sustainable improved agriculture since it generally retains nutrients in the soil for a long time (Sombreek *et al*., 1993; Lehmann and Rondon, 2005). It improves the soil physical and biological properties and efficient absorption of ammonium.

All plants require the same basic nutrient but plants differ in the way they respond to nutrient availability and use (Blackshaw *et al*., 2000). Many researchers have revealed the efficacy of bio char on increasing growth, yield and essential oils of okra and other crops in general. Modern agriculture have depended heavily on convectional fertilizers however the utilization of bio-fertilizers and bio char may be considered.

Essentially the use of chemical fertilizers alone or sole fertilizers may not be adequate enough to provide food needed to satisfy the needs of the populace. It is therefore expedient to properly combine the necessary soil amendment strategies to produce healthy food from an organic source with no environmental hazards. Soil nutrient and water resources need to be properly managed and conserved (Quansah, 1996).

To achieve this, several rates of chemical fertilizers, bio- fertilizers and bio chars were combined and investigated to ascertain the impact on crop yield and productivity.

1.1 JUSTIFICATION

One of the pragmatic ways to reduce soil pollution is the use of bio- fertilizers as substitutes for chemical fertilizers. The complementary use of organic manures and mineral fertilizers has been proven to be a sound soil fertility strategy in many countries of the World (Lombin

et al. 1991). It is envisaged that high and sustainable crop yield could be realized with judicious and balanced NPK fertilization combined with organic matter amendments (Palm *et al.,* 1997; Makinde *et al.,* 2001; Bayu *et al.,* 2006).

Keeping these in view, programmes on development of liquid bio- fertilizers and bio-fertilizer based integrated nutrient management packages for crop production especially vegetables need to be developed and used.

1.2 OBJECTIVES

1.2.1 MAIN OBJECTIVES

To assess the effect of High Yield Technology (HYT) bio- fertilizers and bio char on growth, yield and fruit quality of okra in the forest ecological zone of Ghana.

1.2.2 SPECIFIC OBJECTIVES

The objectives of the study were also:

- To determine the potential of HYT bio- fertilizer on the growth, and yield of okra.
- To determine the combined effect of HYT bio- fertilizers and bio char on the growth and yield of okra.
- To determine the effect of different rates of HYT and biochar on growth and yield of okra.

CHAPTER TWO

LITERATURE REVIEW

2.1The State of Agricultural Land in Africa

The magnitude of nutrient depletion in Africa's agricultural land is so great (Stoorvogel and Smaling, 1990; Smaling 1993; Smaling *et al*., 1997). An average of 660 kg N ha^{-1}, 75 kg P ha^{-1}, and 450 kg K ha^{-1} have been lost during the last 30 years from about 200 million hectares of cultivated land in 37 African countries, excluding South Africa (Stoorvogel and Smaling, 1990; Smaling 1993; Smaling *et al*., 1997). Africa is currently losing 4.4 million t N, 0.5 N million t P and 3 million t K every year from its cultivated land. These rates are several times higher than Africa's annual fertilizer consumption, excluding South Africa—0.8 million t N, 0.26 million t P, and 0.2 million t K (FAO, 1995). African soil nutrient balances are often negative due to a low level of fertilizer inputs, and soil nutrient depletion is a major reason for decreasing or stagnation of agricultural productivity (Sanchez *et al*., 1997).Traditional agriculture practices result in mining soils of plant nutrients through leaching, soil erosion and removal of crop residues leading to decreased fertility (Smaling *et al*., 1997).

Soil fertility decline includes the deterioration in the chemical, physical, and biological properties of the soil that affect plant nutrition, and is a result of specific processes (Lal and Singh, 1998) such as reduction of SOM and soil biological activity, adverse changes in soil nutrient resources and development of nutrient imbalances, and build-up of toxicities and acidification through incorrect fertilizer use, etc. In recent decades, unsustainable land cultivation practices (e.g. inadequate replacement of soil nutrients taken up by crops) have led to accelerated depletion of the natural soil base available for food production (Hossner

and Juo, 1999). Soil productivity maintenance remains a major environmental issue in countries of Sub-Saharan Africa (SSA) (Oyetunji *et al*., 2001).

As populations grew, fertility gradually depleted with crop-harvest removals, leaching, and soil erosion. Farmers were unable to sufficiently compensate these losses by returning nutrients to the soil via crop residues, manures, and mineral fertilizers (Shepherd and Soule, 1998) thus increasing pressures on agricultural land resulted in much higher nutrient outflows and the subsequent breakdown of many traditional soil-fertility maintenance strategies, such as fallowing land, intercropping cereals with legume crops, mixed crop-livestock farming. Notwithstanding, such strategies have not been replaced by an effective fertilizer supply and distribution system (Sanders *et al*., 1996). Traditional African coping strategies have not been capable of adjusting quickly enough to rapid population growth combined with decreasing farm size, soil fertility, and fuel-wood availability (Cleaver and Schreiber, 1994).

2.2 BIOCHAR

2.2.1 What is Biochar?

Biochar is a carbon-rich product obtained when biomass is heated in a closed container with limited air with the intent of being applied to soil to improve soil productivity, carbon storage or remediation and infiltration of soil water (Lehmann and Joseph, 2009). Besides it is able to increase productivity and soil organic carbon concentrations though there seems to be no improvement of nutrient availability (Kimetu *et al*., 2008). Biochar and charcoal essentially have similar characteristic, they can therefore remain in the soil for a long time and again have conditioning effect (Glaser *et al*., 2002).

Several researches have indicated that the addition of biochar to the soil may increase soil organic carbon and improve the supply of nutrient to plants and thereby enhance plant growth

and soil chemical, physical and biological properties (Glaser *et al.*,2002, Lehmann *et al.*,2003., Rondon *et al*., 2007).

2.2.2 Biochar composition

A lot of research on wildfire occurrences and the development of Anthrosols (Terra Preta Soils) in the Amazon shows that charcoal can remain in the soil for several hundred to thousand years (Agee, 1996., Lehmann and Rondon, 2006). Besides, it can rapidly increase the recalcitrant soil C fraction of the soil. The C in biochar is held in aromatic form which cannot decompose easily when applied as soil amendment (Amonette and Joseph, 2009) making it a tool to sequester C.

Biochar composition according to (Downie *et al*, 2009), varies by the type of material and condition of pyrolysis. The C content of biochar normally ranges between 172gkg^{-1}and *90*5kg-1. Nitrogen content is in the range from 1.8gkg^{-1} to 56.4gkg^{-1}, a total phosphorous (p) from 2.7gkg^{-1} to 480gkg^{-1} and total potassium (k) from 1.0gkg^{-1} (Lehmann *et al*., 2003, Lima & Marshall (2005) Chan *et al.*, 2007). There are however varying characteristics of oxygen (O), Hydrogen (H), N, Sulphur, P. base cations and heavy metals (Goldberg 1985, Preston and Schmidt, (2006). Biochar produced freshly consist of a crystalline phase with graphene layers and amorphous phase of aromatic structures (Lehmann *et al*., 2005; Cohen-Ofri *et al*., (2007). The outer surface contains various O and H functional groups and the graphene sheets may contain O groups and free radicals (Bourke *et al*., 2007).

Biochar has a wide range of pH values ranging from between 4 and 12 depending on the feedstock and operating conditions (Lehmann, 2007). Biochar produced at low pyrolysis temperatures (< 400^{o}C) gives acidic biochar. Conversely biochar produced at higher temperatures becomes alkaline due to reaction of water, O^2 and various soil agents, surfaces

oxidation occurs when incorporated to the soil (Lehmann, 2007, Cheng *et al*., 2006,). The cation exchange capacity (CEC) of fresh biochar is very low but increases with time as it ages in the presence of water (Cheng *et al*., 2006; Liang *et al*., 2006, Cheng *et al*., 2008).

2. 2 .3 Effect of biochar application on soil physical, chemical and microbial properties

Mineral fertilized fields show yield decreases, reduced nutrient cycling and reduced nutrient-use efficiency of applied fertilizer associated with a loss of SOC (Zech *et al*., 1990, Goldammer 1993, Silva-Forsberg and Fearnside 1995, Hölscher *et al*,. 1997).

Biochar addition to soils has a multitude of potential agricultural benefits. These include liming of acid soils, addition of basic cations and micronutrients, improving water holding capacity, and a gradual release of nutrients to the growing plant (Glaser *et al*., 2002; Laird *etal*., 2010; Sohi *et al*., 2010; Van Zwieten *et al*,. 2010). Leached sandy soils typically have low soil pH values, poor buffering capacities, low cation exchange capacity (CEC), with values ranging from 2-8 cmolc kg^{-1}, and can have Al toxicity (Novak *et al*., 2009). The addition of biochar to these highly leached, infertile soils gives an almost immediate increase in the availability of some basic cations (Glaser *et al*., 2002; Liang *et al*,. 2006), as well as a significant improvement in crop yields, particularly where nutrient resources are in short supply (Lehmann and Rondon 2006). Over time, these additions continue to promote soil nutrient availability by giving rise to greater stabilization of organic matter and a subsequent reduction in the release of nutrients from organic matter (Glaser *et al*., 2001; Lehmann and Rondon 2006).

Biochar is becoming a popular alternative to organic amendments that are being applied to soils to increase and sustain soil productivity (Lehmann and Joseph 2009). This is attributed to the large amounts of highly porous black carbon found in biochar. The carboxylate groups found in black carbon provide CEC, increase the O/C ratio, and are the primary source of

biochar's high nutrient retention ability (Glaser *et al*., 2001). In addition, biochar may aid in maintaining or increasing nutrient cycling and the stable pools of soil organic carbon (Gaskin *et al*., 2008). Despite biochar being able to improve and sustain soil fertility, fresh biochar shows moderately low cation retention properties relative to aged biochar (Lehmann 2007).

Biochar has the potential to increase nutrient availability for plants (Lehmann *et al*., 2003). Nutrient availability can be affected by increasing cation exchange capacity, altering soil pH, or direct nutrient contributions from biochar. One potential mechanism for enhanced nutrient retention and supply following biochar amendment is increasing (CEC) by up to 50% as compared to unamended soils (Lehmann 2003 and Liang 2006).

Biochar can act as a soil conditioner enhancing plant growth by supplying and, more importantly, retaining nutrients and by providing other services such as improving soil physical and biological properties (Glaser *et al*., 2002; Lehmann *et al*., 2003; Lehmann & Rondon 2005).

Soil microbial biomass and activities increase with biochar additions (Steiner *et al*., 2008 Kolb *et al*., 2009). Biochar provides a microbial refuge due to its porous nature (Peitikainen *et al*., 2000). The size of the microbial community can be linked to nitrogen mineralisation within the soil (McElligott, 2011). As large portion of crop nitrogen is derived from biological processes, changes in microbial processes derived from biochar addition to soils is enhanced. The addition of biochar to soil via microbial habitat provision (Peitikainen *et al*., 2000) induce an increased microbial biomass, nitrogen mineralization also increases, due to the increased microbial biomass and its intimate link to enzyme production (McElligott, 2011).

2.2.4 Plant growth effects with biochar additions

Biochar can be used as a soil amendment to improve soil quality and crop productivity in a variety of soils (Blackwell *et al*., 2009). In a pot experiment, Lehmann *et al*., (2003) found biochar to increase rice biomass by 17% and cowpea by 43% when applied at rates of 68t C ha^{-1} to 135t C ha^{-1}. This growth was attributed to direct nutrient additions from biochar of P, K and Copper (Cu). Iswaran *et al*., (1980) reported a 51% increase in biomass in soybean crops with biochar additions of 0.5t ha-^{1} and Hoshi (2001) found a 20% increase in volume and 40% increase in height of tea trees with biochar additions. Chidumayo (1994) reported better seed germination (30% enhancement), shoot heights (24%) and biomass production (13%) among seven native woody plants on soils under charcoal kilns compared to the undisturbed Zambian Alfisols and Ultisols. Positive plant growth and nutrient content responses to biochar are commonly observed in association with fertilizer application, while neutral or even negative plant growth responses have been observed succeeding biochar only amendments. Much greater yields in plant growth are observed with fertilizer additions plus biochar, as opposed to fertilizer additions alone (Gundale and DeLuca 2007; Yamato *et al*., 2006; Asai *et al*., 2009; Blackwell *et al*., 2009).

2.3 Bio fertilizers HYTa/b

Microorganisms such as bacteria, fungi and actenomyces play a principle role in N fixation and P availability in soil which may increase the uptake of N and P through plant roots. These microorganisms could be grown under laboratory conditions and then applied to seeds, roots or directly to the soil. The aim of using N-biofertilizers is to increase soil content of free living bacteria such as*; Azotobacter* sp., *Azospirillum* sp., *Klebsiella* sp. and others which are expected to increase N- fixation in the soil. Of course symbiotic bacteria of genus Rhizobium is also considered as a good way of N- fixation in legume crops.(Barakat and Gabr. (1998).

Biofertilizers produce the growth stimulating substances viz., auxin, gibberellins and cytokinins which contribute towards vigorous growth of the plant (Singh *et al*., 2003). Phosphate solubilizing bacteria plays a fundamental role in correcting the solubility problem in many soils by transforming this insoluble part again to soluble. Several soil bacteria, particularly those belonging to the genera Pseudomonas and Bacillus, and fungi belonging to the genera *Penicillium* and *Aspergillu*s possess the ability to bring insoluble phosphates in soil into soluble forms by secreting organic acids such as formic, acetic, propionic and succinic acids. These acids lower the pH and bring about the dissolution of bond forms of phosphate and render them available for growing plants (El-Hadad *et al*., 1986).

The response of plants to biofertilizers depends greatly on;

The genus and species of the used microorganism responsible for N fixation such as; *Azotobacter, Azospirillum* ,*Klebsiella* and its species . The microorganism responsible for P-availability such as P-dissolving bacteria (*Bacillus* sp.) and fungi such *Mycorrhiza* use single or mixed genus of the microorganisms for inoculation to exhibit considerable effect on N or P uptake.

The inoculation method used for adding bacteria or fungi; are seeds inoculation, treating roots of seedlings and transplants or inoculating soil directly.

The rate of organic and chemical fertilizers applied to soils or plants treated with biofertilizers especially NPK levels (Rao, 1993)

HYT[a] is 100% organic and safe blend of naturally occurring, non-pathogenic soil-based microbial complex and enzymes which restores and increase microbial activity in the soil, increase fertilizer efficiency and fix atmospheric. It contains twenty one different strains of microbes with *Azobacter vinelandii* and *Clostridium pasteurianum* as primary species and

100% organic and non-pathogenic immobilized cells which fixes N from the atmosphere, mineralize N from organic matter and solubilize N from NPK fertilizers. Mineralization, solubilization and desorption of P, K and other essential nutrients from organic matter or NPK fertilizers is accomplished by the microbes. The activity of the microbes breaks through the soil pan thereby opening new nutrient soil and providing better root formation. The softening of the soil leads to better water and nutrient retention. The increased microbial activities normalize the soil pH and increase the organic matter content of the soil via their demise (Agrinos, 2011).

HYT^{b} is an organic free L-amino acids and mineral nutrient source which is plant and microbial bio-stimulant and stress relief (Agrinos 2011). It is constituted of 12% L-free amino acids of L-triptophan, L-aspartic acid, L-serine, L-histodine, L-glycine, L-thereonine, L-threonine, L-alanine, L-proline, L-tryosine, L-arginine, L-valine, L-methionine, L-isoleucine, L-phenylalanine. 6% ultra-soluble minerals i.e. Ca y Mg and 82% transport(Agrinos 2011). Jain and Patriquin (1985) found that bacteria of the genera, *Azotobacter* and *Azospirillum*, could produce more than 30 mg of indole acetic acid (IAA) solutions of enzymatic complex, lactic acid, polysaccharides, polypeptides and carbohydrates. HYT^{b} increases stress resistance, increases and supports photosynthesis, pollination and fruit set, stimulates vitamin formation and increases sugar content (Agrinos 2011 cited by Adu, 2012)

The role of PSB as a bio fertilizer is unique in making the fixed soil phosphorus available to plants. PSB produce plant growth regulating substances, which promote root growth. Different mechanisms of mineral phosphate solubilization included synthesis of organic acids by phosphate solubilizing bacteria, CO^{2} and $H^{2}S$ production and chelation of other acids (Gaur, 1990). The amount of organic acids liberated by these microorganisms is said to be roughly about five per cent of the carbohydrate consumed (El-Hadad *et al.*, 1986). It is also

shown that solubilization of mineral phosphate by bacteria is the result of acidification by the direct oxidation of glucose or other aldose sugars (Goldstein, 1995).

2.3.1Effect of bio fertilizers on plant growth and development

Okon, (1985) reported that it is possible to increase plant height, leaf size and early flowering by use of *Azospirillum*. Sattar and Gaur (1987) reported that P-solubilizers improved the plant growth and development by the production of plant growth hormones like indole acetic acid (IAA), gibberellic acid (GA) and cytokinins. The increase in growth characters like plant height in French marigold by *Azospirillum* inoculation was observed and this might be due to the added nitrogen to crop through associative symbiosis and increased production of growth hormones like NAA, GA and cytokinins (Balasubramanium, 1989). In China aster, there was an increase in vegetative growth due to use of biofertilizers (VAM and phosphobacteria) (Prabhatkumar *et al*., 2003). This was related to nutrient uptake and biosynthesis of plant growth regulators, thereby stimulating the growth and development process of the plant. Naik and Patil (2004) isolated different plant growth promoting rhizobacteria such as Rhizobium, *Azospirillum* and Phosphate solubilizers. The efficient isolates having beneficial traits such as N_2 fixation, phosphate solubilization, production of IAA, GA and bicontrol activity were used to study the effect individually and in combination with each other by inoculating to soybean. The results indicated that the combined application of three or more beneficial organisms exerted more favourable effect on growth and productivity of soybean than dual or single inoculations (Naik and Patil, 2004).

In roses, the early flowering due to inoculation with *Azospirillum* was observed (Preethi *et al.* 1999). This was due to induced cytokinin synthesis and rapid assimilation of photosynthates resulting in early transformation of the axillary bulb from vegetative to reproductive phase. Prabhatkumar *et al*., (2003) obtained increase in vegetative growth of China aster by use of

biofertilizers viz., VAM+PSB which might be related in simulating nutrient uptake and biosynthesis of plant growth regulators, thereby improving the growth and development process of the plant.

Naik and Hosamani (2003) attributed the increased yield of green chili (*Capsicum annuum* L. cv. ByadagiDabbi) by *Azospirillum* inoculation to improved vegetative growth and maximum number of fruits per plant fruit and fruit parameters. Jeevansab (2000) noticed that Azospirillum+ RDF produced significantly higher number of fruits fruit length and girth and fruit yield compared to 50 per cent RDF (8.5, 7.3, 5.3 and 690.2 g, respectively) in capsicum. Wange & Kale (2004) reported 74% yield increase of *brinjal* (eggplant) over the recommended rate of N fertilizer when inoculation with mixture of *Azotobacter* + *Azospirillum* and followed by application of 75 kg N per ha. The results further revealed that reducing N rate to 50 kg while using these biofertilizers did not help in achieving yields at par with recommended 100 kg N per ha. Thus, only 25 per cent N saving through the use of biofertilizers can be achieved with increase in yield over recommended 100 kg N per ha (Wange and Kale, 2004).

2.4 Inorganic fertilizers

Major plant nutrients including nitrogen, phosphorus, potassium, magnesium and calcium play important roles in the growth and development of crops including cell division, cell elongation and formation of chlorophyll molecules (Mohammed *et al*., 2008, Adekayode and Olojugba, 2010). Studies on the effects of major plant nutrients on growth of plants have received major attention in different part of the world.

Nitrogen is an essential element and an important determinant in growth and development of crop plants. It plays an important role in cholophyll, protein, nucleic acid, hormone and vitamin synthesis and also helps in cell division and cell elongation. Several research works

have reported increase in green pod yield of okra with application of N from 56 to 150kg/ha (Hooda *et al*., 1980; Mani & Ramanathan, 1980; Majanbu *et al*., 1985, Singh, 1995).

Phosphorus is a key constituent of adenosine tryphosphate (ATP) and plays significant role in energy transformation in plants and also in various physiological processes (Shivasankeb *et al.*, 1982). It helps in nutrient uptake by promoting root growth and thereby ensuring a good pod yield through the increase in total dry matter (Roa ; 1982, 1995). However, Phosphorus deficiency results in poor root development, poor pod setting and subsequently reduces yield (Jam *et al*.,1990).

Potassium increases crop yield and improves fruit quality. It is one of the major nutrients essential for plant growth and development. Plant accumulates large quantities of Potassium, which constitutes between 2% and 10% of plant dry weight (Tisdale *et al*., 1993).

Calcium is an essential plant nutrient required for structural roles in the cell wall and membranes, as a counter cation for inorganic and organic anions in the vacuole, and as an intracellular messenger in the cytosol (Marschner, 1995).

2.4.1Effect of inorganic fertilizers on plant growth and development

According to Prabhakar *et al*. (1987) maximum plant height in chilli pepper was noticed with N application rate of 90 kg ha-1 while the crop did not respond to P application in respect of plant height. Dharmatti *et al*. (1992) found that increased level of nitrogen, phosphorus and potassium resulted in an increased number of branches per plant in bell pepper as a result of better availability of soil nutrients especially NPK which has enhancing effect on the vegetative growth of plants by increasing cell division and elongation. However, according to Srinivas (1983) application of nitrogen and phosphorus did not show any significant effect on plant height and number of branches per plant in chillies. Sharma (1995) in a study for the

determination of optimum doses of nitrogen, phosphorus, potassium and magnesium fertilization in tomato found that increase in the levels of nitrogen application showed increase in plant height and number of branches per plant. This was as a result of the involvement of Mg in chlorophyll formation which aided cell division and expansion leading to the formation of new cells. An experiment conducted by Balaraj (1999) showed significantly higher plant height and number of branches per plant with the application of 150:75:75 kg NPK per ha compared to 100:50:50 and 125:62.5:62.5 kg NPK per ha in chilli pepper. The author explained the significant height and branch number based on climatic condition prevailing at the time of the study and also sufficient nutrient supply to the plants in the form of the fertilizer.

The time taken for plants to flower differs from plant to plant and also depends on the nutrient store supply to the plants through the growth stages of the plant. The effect of NPK on the days taken to flowering has been studied over time by some researchers. Gnanakumari & Satyanarayana (1971) found that NPK fertilizer application hastened the initiation of flowering in egg plant. Plants that received 234 kg ha^{-1} each of N, P and K flowered earlier compared to the control. The researchers attributed this to early growth by fertilizer leading to early flowering. Gill *et al*. (1974) found that plant receiving high NPK dose produced flower buds earlier than in plants with low NPK in sweet pepper. This resulted from sufficient nutrient supply to the crops resulting in high vegetative material production leading to promotion of flower bud formation hence early flowering of the plants. In studying N application rates in chilli pepper, Dod *et al*. (1989) reported delayed flowering with increased N levels. The authors attributed the delay in flowering to increased N levels which enhanced plant vegetative growth at the expense of reproduction development of the plants.

Yield and yield components of vegetables as affected by NPK fertilizer have been studied throughout the world. Increases in yield have been attributed to plant nutrition, climatic

conditions, soil chemical and physical conditions as well as genotype by different authors (Adu, 2012). An experiment conducted by Adeboye and Oloyede (2007) to evaluate the effects of single super phosphate on the fruit yield of two landraces of *Trichosanthes cucumerina* L. showed that, P levels at 90kg P_2O_5 ha^{-1}gave significantly higher number of fruits compared to other P levels. The authors' explained this by the release of adequate nutrients to the plants due to the favourable soil pH. According to the authors, nutrient supply was good leading to effective building of photosynthetic structures which caused the yield increase. Similarly, Liu *et al*. (2008) designed a field experiment to study the effect of N and K fertilizers to optimize yield of vegetable crops. The findings by the authors indicated that, application of N and K fertilizer significantly increased the yield of kidney bean when 750kg N ha^{-1} and 300kg K_20 $ha^{-1}$was applied. Subsequent yields of tomato grown after the kidney bean gave significant yield response to the application of N and K. The yield increase of tomato after kidney bean was attributed to release sufficient nutrients to the plants. The findings of Liu *et al.* (2008) are similar to the findings of Adeboye and Oloyede (2007).

2.5 Effect of integration of bio fertilizer, inorganic fertilizer and biochar on:

2.5.1Plant vegetative growth

In a field trial with banana cv. Poovan, Jeeva (1987) reported that inoculation of Azospirillum along with nitrogenous fertilizers increased plant height, girth, total number of leaves, leaf area, sucker production, length of bunch, number of hands, fingers and total soluble solids. Nanthakumar and Veeraragavathatham (2000) stated that the results clearly indicated that combining organic fertilizer, namely 12.5t ha^{-1} of farmyard manure and 2kg each of *Azospirillum* and phosphobacteria, with inorganic fertilizers at 75% of the recommended dose of N and P and 100% of K (namely 75 kg N, 37.5 kg P and 22.5 kg K/ha) favourably influenced the growth parameters in *brinjal* cv. Palur-1. Binisha *et al*. (2002) revealed that

the treatment combination of NPK along with *Azospirillum* was more effective in improving vegetative and floral characters of *Dendrobium* than NPK alone. Wange and Kale (2004) reported that the results revealed significant improvement in vegetative characters such as plant height, number of leaves per plant in *brinjal* recorded higher values over the recommended rate of N-fertilizer due to inoculation with mixture of *Azotobcte*r + *Azospirillum* and followed by application of 75kg ha N .

2.5.2 Flowering status and maturity of plants

Seetha, (1999) obtained an early flowering in Gerbera plants when inoculated with *Azospirillum* and VAM in addition to 50% nitrogen and phosphorus dose.

Anburani and Manivannan (2002) reported that FYM at 25t ha along with 100% NPK + biofertilizers (*Azospirillum*+ phosphate solubilizing bacteria) recorded the lowest number of days to first flowering and days to 50% flowering as well as the most.

Okon (1985) and El-Hadad *et al*., (1986) reported early flowering by use of Azospirillum in tomato and rose respectively. This could be due to induced cytokinin synthesis and rapid assimilation of photosynthates resulting in early transformation of the axillary bulb from vegetative to reproductive phase. (Murti and Upreti, 1995). However, Jeevansab (2000) reported that *Azospirillum* + RDF (150:75:50) took more number of days to 50% flowering compared to RDF alone in capsicum. This was attributed to enhanced plant vegetative growth at the expense of reproductive growth and development of the plants.

2.5.3 Fruit yield and Yield components

Integrated nutrient management on commercial vegetables studied by Patil (1995) revealed that the combination of RDF (100:75:100 NPK kg/ha) + 50% recommended dose of vermicompost (2.5 t/ha) recorded significantly higher number of tomato fruits per plant and

average fruit weight over absolute control, RDF, FYM and vermicompost alone but was on par with combined application of organic and inorganic fertilizers. Singh *et al*. (1997) registered higher fruit yield per plant in chili with the application of vermicompost at 10 t/ha, whereas Patil (1995) observed that inclusion of vermicompost along with 100% RDF + FYM resulted in additional dry chili yield of 1.68g/ha . Balasubramani *et al*. (1997) reported that the seed and soil treatment of *Azospirillum* with 75 percent recommended dose of nitrogen per hectare recorded the highest yield of bhendi 17.5 t/ha whereas control registered 9.6 t/ha only at Madurai, Tamil Nadu.

Furthermore, the authors observed that soil and seed treatment of *Azospirillum* alone recorded the highest number of bhendi fruits per plant (26.4) followed by the *Azospirillum* seed treatment and soil application of nitrogen 30 kg/ha with 25.5 fruits, whereas control recorded only 17.3 fruits per plant.

The effect of biofertilizers and inorganic fertilizers on crossandra cv. Dindigul local was examined by Narasimha and Haripriya (2001). Number of spikes per plant, spike length, number of flowers per spike and flower yield per plant showed better results when 100 percent NPK (75:50:125 kg/ha) + *Azospirillum* and phosphobacteria each at 2 kg/ha was used. The increased flower yield might be due to the indirect effect of more number of branches as stimulated and developed by the influence of inorganic fertilizers along with biofertilizers.

2.5.4 Role of inorganic fertilizer, bio fertilizers and biochar in chemical composition of plant foliage

Radwan, (1983) studied the effect of tomato seeds inoculation with PDB. He showed that, P-content of tomato plants was increased. The same result was found by (Abd-El- Moneim *et al*., 1988 , Hewedy 1999; and Tantawy , 2000) all working on tomato inoculated with PDB .

Mohandas (1987) mentioned that, inoculating tomato plants with *Azotobacter vinelandii*resulted a significant increase in N-content of plants as compared with the control. Also, Gomaa (1989) showed that, inoculating tomato seeds with *Azotobacter chroococcum* and *Azospirillum brasilense* resulted in an increase in N and P content of plant leaves.

El- Shanshoury *et al*., (1989) indicated that, N and K content of tomato plants were increased by using *Azotobacter chroococcum*. Similarly, Monib *et al*. (1990) and Sorial *et al*. (1992) found that, N and P content of tomato plants were increased with seeds inoculation by *Azospirillum brasilense* and *Azotobacter chroococcum*.

Gomaa (1995) using a mixture of six genera ; Azospirillum + *Azotobacter* + Bacillus + Candida + Klebsiella + Pseudomonas , exerted tremendous increase in nitrogen content of tomato plants by 192%, as compared with untreated control , under the same conditions of N – free nutrient fertilization. Gomaa, (1989) also on tomato found that seed inoculation with a mixture of *Azospirillum brasilense, Azotobacter chroococcum* and phosphate dissolving bacteria resulted a significant increase in N and P contents over *Azospirillum brasilense* or *Azotobacter chroococcum* alone.

Barakat and Gabr, (1998) inoculated tomato plants with non- symbiotic N-fixing bacteria of the genera; *Azotobacter sp. ,Azospirillum sp*. and *Klebsiella sp*. alone (single biofertilizers) or together (mixed bio fertilizer) or in combination with four N fertilizer levels ; 0, 50,100 and 150 Kg/ N. Results showed a significant increase in N content and leaf chlorophyll of plants with increasing N applied rate up to 100 kg/ N or inoculation either with the single or mixed biofertilizers.

Tantawy in 2000 recorded an increase in nitrogen content of tomato plants due to inoculation with Rhizobacterin compared with unioculation under all nitrogen levels ; 0 , 50 or 100 kg/ N. Poi (1998) found that inoculating the soil with *Azospirillum* and *Pseudomonas* amended

the soil characters and increased the available N and P in the soil and its uptake by chili and tomato plants than untreated ones. Furthermore, Ouda (2000) found that a mixture of 1 kg Phosphorin + 1 kg Microbin + 1 kg Rhizobacterin per fed increased NPK content of tomato plant foliage and gave similar influence to 75% of recommended mineral NPK fertilizers. Tantawy (2000) found that, the maximum values of nitogen and phosphorus were more distinct via using Microbin + Phosphorin treatment, which came in the first rank, followed by the treatments of Microbin or Phosphorin alone and Rhizobacterin + Phosphorin. However, potassium content of tomato plants was not affected by using any bio fertilizer treatments.

CHAPTER THREE

MATERIALS AND METHODS

3.1 Experimental site

The experiment was carried out at the Forest and Horticultural Crops Research Centre (FOHCREC) Kade, located at latitude 6009and 6006N and longitude 0055and 0049W and 135.9 m above sea level. The centre is located in the semi-deciduous forest agro-ecological zone of Ghana in the Kwaebibrem district of the Eastern Region, 175km NE of Accra.

3.1.1 Soil characteristics

The soils of the research area developed from rocks of the Birimian system (middle Pre-Cambrian) (Adu, 1992) which constitute mainly of argillaceous sediments metamorphosed into phylite. These soils are well drained and belong to the forest Ochrosol Great Soil Group of the Ghanaian soil classification system (Bramner, 1962; Owusu-Benoah *et al*, 2000) and are generally accommodated as Acrisols in the FAO-Nesco Revised Legend (FAO, 1998) and as Utisols in Soil Taxonomy (Soil Survey Staff, 1998). The soil moisture regime is udic and

soil temperature regime is isohyperthermic (Van Wambeke, 1982; Owusu-Benoah *et al*., 2000).

3.1.2 Climate.

The climate of the area is humid tropical, with an average annual temperature of about 28^{0C}, Maximum temperatures is in March and minimum in August. The rainfall pattern is bi-modal with a 30-year average of 1433mm with peaks in June and a brief dry spell in August. The dry season is from December to March. The highest relative humidity of 75% to 80% was

recorded during the major rainy season whilst the lowest of 65% to 75% were recorded in the minor rainy season.

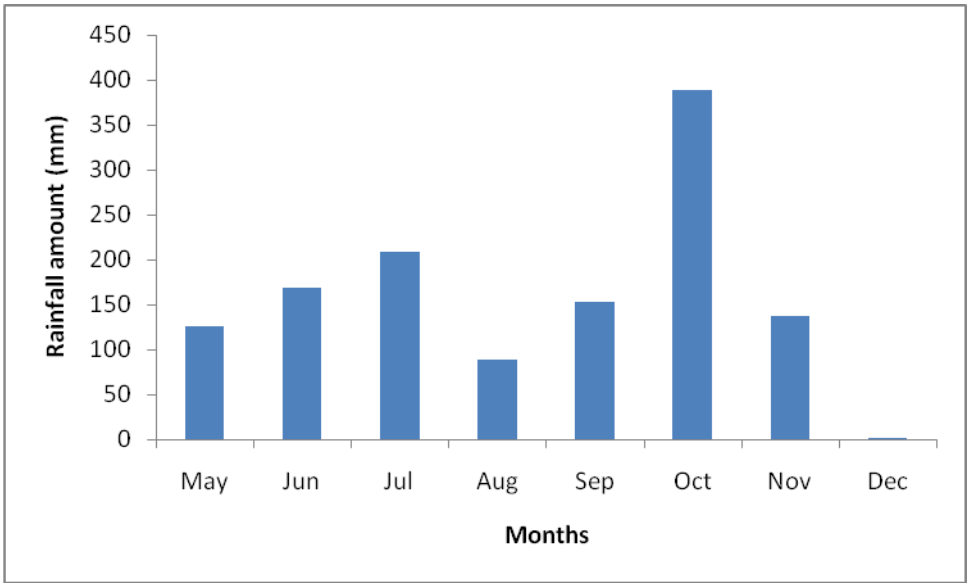

Figure 1: Rainfall distribution at the experimental site during the experimental period 2011/2012.

3.2 Experimental Material

Thiram treated seeds of okra cv Clemson Spineless was used for the study. Seeds were obtained from Agrimat Agrochemicals Co. Ltd, Madina, Accra. HYT biofertilizers were provided by Agrinos Company Ltd while inorganic fertilizers used were acquired from the Forest and Horticultural Crops Research Centre. Biochar was prepared at the centre by pyrolising rice husk.

3.3 Experimental details

Two types of experiments, field (both major and minor planting seasons) were conducted. The major planting season began in April, 2011 and ended in July, 2011 and the minor season commenced in August, 2011 and ended in November, 2011.

3.4 Experiment I; Field experiment (major and minor season planting)

3.4.1 Previous crop on site (major season)

The site selected for the project was selected from a site which had been allowed to lie uncultivated for three years after a previous harvest of maize.

The weeds growing on the research sites were predominantly *Chromoleana odorata* interspersed with *Cyperus rotundus, Imperata cylindrica, Panicum maximum*, and *Ageratum conyzoides.*

3.4.2 Previous crop on site (minor season)

This site was selected from an old citrus farm which had been planted to avocado. Weeds growing on this research site were mainly *Chromoleana odorata* and some underground twines.

3.4.3 Experimental layout and treatments

The experiments were laid out in a randomized complete block design (RCBD) comprising

twenty-seven (27) treatments and (4) four replications. There were three (3) levels of the different amendments at 100%, 50% and 0% in combinations (Table 1). Each experimental plot was $8m^2$ (8m x 1m) in major season and (8m x 2m) in the minor season with 0.3m between plots and 0.6m pathway. The planting distance adopted at both sites was intra row 0.5m and inter row 0.6The structural representation of the combination is thus seen in Figure 2.

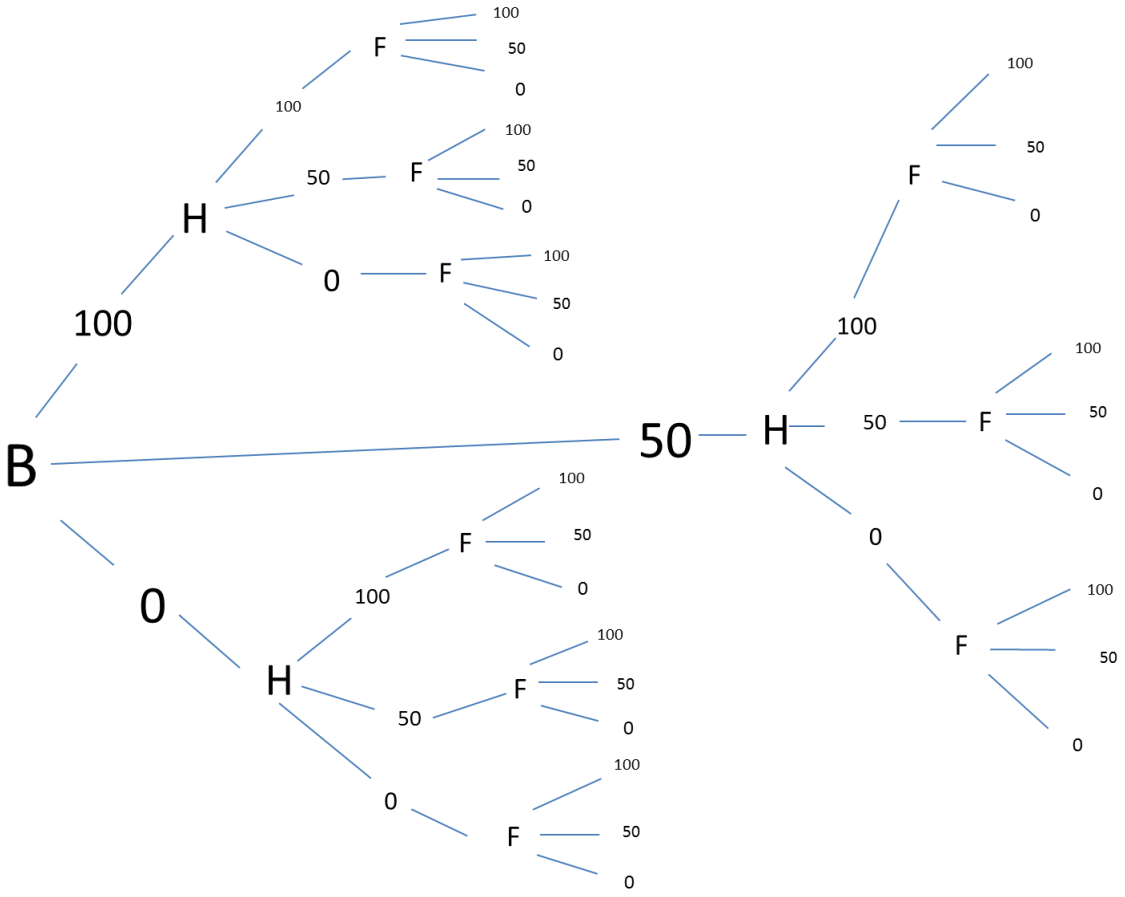

Figure 2: Structure of Treatment Combinations.

Table 1: Treatment combinations

Treatment	Treatment explanation
T1:F0B0H0	Control
T2:F0B50H0	3.5t biochar
T3:F0B100H0	7t biochar
T4:F50B100H0	50% fertilizer+7t biochar
T5:F50B50H0	50%fertilizer+ 3.5t biochar
T6:F50B0H0	50% fertilizer
T7:F100B100H0	100% fertilizer+ 7t biochar
T8:F100B50H0	100%fertilizer + 3.5t biochar
T9:F100B0H0	100% fertilizer
T10:F100B100H100	100% fertilizer+ 7t biochar+ 100% HYT
T11:F50B50H50	50% fertilizer +3.5t biochar+50% HYT
T12:F100B0H100	100%fertilizer+ 100% HYT
T13:F100B0H50	100% fertilizer + 50% HYT
T14:F50B0H100	50% fertilizer + 100% HYT
T15:F50B0H50	50% fertilizer + 50% HYT
T16:F100B100H50	100%fertilizer+7t biochar+ 100%HYT
T17:F100B50H100	100%fertilizer+3.5t biochar + 100% HYT
T18:F100B50H50	100%fertilizer+3.5t biochar +50% HYT
T19:F50B100H100	50%fertilizer+ 7t biochar +50%HYT
T20:F50B100H50	50%fertilizer+7t biochar+ 50%HYT
T21:F50B50H100	50%fertilizer+ 3.5t biochar+100%HYT
T22:F0B100H100	7t biochar +100% HYT
T23:F0B0H50	50% HYT
T24:F0B0H100	100% HYT
T25:F0B100H50	7t biochar +50%HYT
T26:F0B50H100	3.5t biochar +100% HYT
T27:F0B50H50	3.5t biochar +50% HYT

3.4 Plot Size and planting Distance

Refer to 3.4.3, each experimental plot was 8m x 1m in major raining season and 8m x 2m in the minor raining season with 0.3m between plots and 0.6m pathway. The planting distance adopted was intra-row 0.5m and inter-row 0.6m.

3.4.4 Cultural practices

3.4.4. 1 Germination test

Prior to land preparation, seed germination test was carried out to ensure their viability. Fifty seeds were placed in a petri- dish containing moist filter tissue covered and kept in the dark

room to ensure maximum seed germination since bright conditions may inhibit other seeds from germinating. This process was repeated three times and the mean germination percentage determined by the formula:

$$Mean\ germination = \frac{Number\ of\ total\ seeds\ germination}{Total\ number\ of\ seeds\ sown}$$

3.4.4.2 Biochar preparation and application

An empty metal drum was perforated on the sides (about 5mm) and an opening measuring 30cm in diameter was created on top of the container to facilitate the arrangement of fuel wood for burning. The bottom of the drum was completely removed. Fire was set in the container to facilitate charring. In the course of burning, rice husk was heaped on the sides of the drum container that had the fuel wood burning. Heat passed through the perforated holes and charred the rice husk into carbonated rice husk. After the pyrolises it was watered to cool down. It was then air dried and weighed into sacks. Biochar was spread and mixed thoroughly with the soil for the study.

3.4.4.3 Land preparation

The land was deep ploughed and brought to a fine tilth by repeated harrowing on 11 May 2011 and 12 August 2011 in the major and minor seasons respectively. The plots were prepared as per the specifications and biochar was applied on 22 May 2011 and 23 August 2011 at 7.0t/ha (100%), 3.5t/ha (50%) and 0t/ha (0%) for major and minor seasons respectively. The application was done by incorporating biochar to the top10cm of the soil with the aid of a hoe. The first ground application of HYT was applied on the 26 May and 1 September respectively after the 72 hours of activation.

3.4.4.4 HYT activation.

In activating HYT for use, 2L of HYT[b] representing 100%, and 1L representing 50% was added each to 100L of clean, non- chlorinated portable water or ground water with a neutral pH and electrical conductivity above 40ms filled into three big rubber drums each containing different levels, and stirred or agitated thoroughly to mix. The solution was allowed to stand for between 10- 15 minutes respectively before HYT[a] was added. The solution was stirred and stored 72hrs to activate the enzymes and microbes. Seventy-two hours (72hrs) thereon, the colour and smell of the activated solution changed or altered and became "sweeter" and less pungent in smell. A light film then formed on the surface of activated solution with bubbles or foam. Thereafter, the same volume of HYT[b] was added and agitated to awaken and reactivate the enzymes in accordance with the protocol stipulated by Agrinos (2011).

3.4.4.5 HYT Application

The solution prepared was used for ground application at 0.9L per experimental treatment and applied very close to the root zone this was repeated every 14 days(Table 2).

Table 2: Application regime

Time/WAG	1	2	3	4	5	6	7	8	9	10	11
Application	soil	Foliar	Soil	foliar	soil	foliar	soil	foliar	soil	foliar	soil

WAG: weeks after germination

At two leaves stage, a foliar application was done. 100L of water was mixed or agitated to ensure proper mix with 1L of HYT[b] solution and applied at 450ml per experimental

treatment. Foliar application was repeated every 14 days (Table 2). Knapsack sprayer was used to for the application.

3.4.4.6 Planting

Seven days after the ground application of HYT, seeds were sown on 30th May, 2011 in the major season and 7th September, 2011 in the minor season. At planting three seeds were placed in each hole created with a dibber at a depth of 5cm and at a spacing of 0.5m x 0.6m. These were thinned to one plant per stand at 2 weeks after germination (WAG).

3.4.4.7 Weed control

In order to keep the soil porous and also free from weeds, hand weeding and hoeing were done as and when necessary. Weeding was done manually with cutlass and hoe. Weeds were controlled until harvesting was completed at least weeds were controlled every 14days.

3.4.4.8 Irrigation

Supplementary irrigation was done as and when it was necessary. Watering can and rubber hose were used. Water was sourced from a bore hole that was dug nearby.

3.4.4.9 Fertilizer application

Three WAG (Weeks after germination) N.P.K 15:15:15 fertilizers were applied by ring application at the following rates specified: Each plant received a treatment of 8.6g per plant at 100%, 4.3g per plant at 50%.and 0g per plant at 0%. At flowering, ammonium sulphate at the rates specified was applied by ring application. Each received a treatment of 4.3g per plant at 100%, 2.1g per plant at 50% and 0g per plant at 0%. After the first harvest, potassium nitrate was applied by ring application with each treatment receiving a dosage of 8.6g per plant at 100%, 4.3g per plant at 50% and 0g per plant at 0%.

3.4.4.10 Plant protection

To control pest and diseases, the recommended fungicide and insecticide like Plan-D was applied at the vegetative stage. The necessary plant protection measures were taken up as per the recommended package. Flea beetle (*Podagrica* spp) the only pest that was noticed was controlled with PLAN- D and HYT[a] combinations.

3.4.4.11 Harvesting

The crop was harvested at 55 days after sowing (DAS) with a sharp knife. Harvesting was done at three days intervals.

3.5 Experimental data collection

Five plants from each treatment in each replication were randomly selected and tagged for record taking on growth, yield and yield components.

3.5.1 Plant height

Plant height was recorded from the base of the stem at soil level to the terminal bud of the plant of a fully opened leaf on the main shoot and the mean height was expressed in cm. Plant height was recorded from 15, 30 and 45 DAG (Days after germination).

3.5.2 Number of leaves

Number of leaves on each tagged plant was counted manually at 15, 30 and 45 DAG.

3.5.3 Stem diameter

The diameter of the stem 5cm from the ground level was measured with Vernier calipers at 15, 30 and 45 DAS.

3.5.4 Reproductive parameters

3.5.4.1 Days taken to first flowering

Daily observations were critically made to find out when tagged plants in each treatment flowered. The day the first plant flowered was considered as number of days to first flower production in each replicate. The average gave days to first flowering.

3.5.4.2 Days to 50% flowering

Daily observations were made on the five randomly tagged plants for flowering. The days on which 50% of plants per replicate flowered were recorded and considered as 50% flowering. The number of days taken from the date of sowing to flowering was expressed in number of days taken for 50% flowering.

3.5.5 Yield parameters

3.5.5.1 Number of fruits per plant

Fruits numbers and fruits weights were recorded each time harvesting was done. Five plants per treatments per replicate were used. The mean of the five plants was used to determine the number of fruits and fruit weights (g) per plant

$$\text{Number of fruits per plant} = \frac{\sum \text{Number of fruits from tagged plants per treatment}}{\text{Number of tagged plants per treatment}}$$

3.5.5.2Fruit Yield per plant (g)

The number of immature fruits were recorded and multiplied by the average weight of fruits per treatment to give the yield per plant in grams. Thus number of fruits per treatment by the average weight in a treatment.

$$\text{Fruit yield per plant (g)} = \frac{\text{Fruit yield per treatment (tagged)}}{\text{Number of tagged plants per treatment}}$$

3.5.5.3Fruit Yield per hectare (t)

Tender green fruits were harvested every three days. The number per plant and fruit weight was determined and the yield per plant calculated to determine the total yield per hectare as

$$\text{Fruit yield per hectare (t)} = \frac{\text{Area of a hectare}}{\text{Area of net plot}} \text{xYield per net plot}$$

3.5.5.4 Average length of fruit

Green tender and marketable fruits that were picked from tagged plants periodically. These were measured from the pedicel to the tip of the fruit and the average worked out and expressed in centimetres. Measurement was done with a flexible tape measure.

$$\text{Mean fruit length (cm)} = \frac{\sum \text{Lengths of selected fruits}}{\text{Number of fruits selected}}$$

3.5.5.5 Average fruit diameter

Fruits form harvested samples per treatment were used for measuring the diameter of the fruit. The diameter was measured at the centre of the fruit with Vernier calipers. The diameter was expressed in centimetres.

3.5.5.6 Fruit weight

The fruits harvested for market were weighed as and when they were harvested. The fruits were weighed with an electronic balance and values recorded for analysis.

3.6 Soil, biochar and plant nutrient analyses

Prior to the laboratory analyses, soil and biochar samples were air dried after sieving using a 2mm mesh. Plant samples were oven dried at 70^{0C} for 72 hours milled and sieved for the analyses of chemical properties.

3.6.1 Soil and biochar chemical properties analyses

Core soil samples were collected randomly from the 0-15cm depth on the site using a soil auger. Soil was then mixed thoroughly and the bulk density was taken to the laboratory, air-dried and sieved to pass through a 2mm screen for chemical analysis. The soil pH (1:1 soil/water) and biochar pH (1: 2.5 biochar/water) were determined using a glass calomel electrode system (Crockford and Nowell, 1956). The soil N was determined by the microkjedahl method (AOAC, 1970). Available soil P was extracted by the Bray P1 extractant, measured by the Murphy blue colouration and determined on a spectronic 20 at 882 Um (Murphy and Riley, 1962). Soil K, Ca, and Mg were extracted with a 1M NH4OAC, pH 7 solution, then K analysed with a flame photometer while Mg and Ca were determined with an atomic absorption spectrophotometer (Jackson, 1973).

Total nitrogen in sample was calculated as shown:

$$\%N = \frac{\text{molar mass of N x titre value x volume of extract x100}}{\text{weight of sample x 1000 x volume of aliquot}}$$

Available P in samples was calculated as shown

$$\text{P (ppm)} = \frac{\text{meter reading x volume of extract}}{\text{weight of sample x volume of aliquot}}$$

The concentration of potassium in the soil or biochar sample expressed in percentage was calculated as follows:

$$K\left(\frac{me}{100g}\right) = \frac{\text{meter reading}}{\text{atomic weight of K}}$$

The concentration of calcium and magnesium in the soil or biochar sample expressed in percentage was calculated as follows:

$$\text{Ca and Mg}\left(\frac{me}{100g}\right) = \frac{\text{AAS meter reading}}{\text{atomic weight of Cation}}$$

Soil particle size determination was done using the Bouyoucos Hydrometer Method (Bouyocous, 1962). The particle size distribution was determined using the formula:

$$(\text{clay} + \text{silt})\% = \frac{5 \text{ minutes hydrometer reading}}{\text{mass of soil(g)}} \text{x100} \rightarrow \quad (\alpha)$$

$$(\text{clay})\% = \frac{5 \text{ hours hydrometer reading}}{\text{mass of soil(g)}} \text{x100} \rightarrow \quad (\beta)$$

$$(\text{sand})\% = \frac{\text{oven dry mass(g)of particles retained on 45}\mu\text{m sieve}}{\text{mass of soil}} \text{x100}$$

(silt)%= (α) – (β)

3.6.2 Soil microbial analysis

A screw cap bottle of about 250 ml volume was washed, covered with aluminium foil and sterilized in an autoclave at 121^{oC} for 15 minutes. A long rope was tied around the neck of the bottle. The cap of the bottle was opened aseptically and lowered down into the well to a depth of about 1m making no air to escape. The bottle was raised out of the well and carefully replaced and labelled before, placing in an ice chest loaded with ice packs and transported to

the laboratory for incubation immediately. 9mls of ¼ strength phosphate saline buffer were added to 1ml of well water sample for 1 in 10 dilutions.

The pour plate method was used in which 1ml aliquot of the well water sample were transferred aseptically with a micro pipette into a sterile petri dish. 10 ml of the sterile plate count agar (PCA) was added when palm hot (45^{oC}), mixed and allowed to set. It was then incubated at 35^{oC} for18-24 hours. The microbial growth on the media was counted using the Start Scientific Colony Counter.

3.6.3 Plant analyses

The Kjeldahl digestion procedure as described by Okalebo *et al*. (2002) was used in determining N, P, K, Mg and Ca in the plants. A 0.1 g of milled and sieved plant samples were weighed into cleaned dry 125ml pyrex conical flask. Five milliliters H_2SO_4 was added and left to stand for about 1 hour. The flask and its contents were heated on a hot plate in a fume chamber and few drops of H_2O_2, added (3- 4 drops at a times) to avoid vigorous reaction of the content until the solution turned colourless. The solution was cooled and transferred into 100mls volumetric flask. The content was topped to the mark using distilled water and used to determine N, P, K, Ca and Mg. Total N in plant samples was determined using the microkjedal method of distillation and titration as described for soil and biochar above. Available P was determined following colour development using the Bray P1 extractant, measured by the Murphy blue colouration (Murphy and Riley, 1962) and determined on a Spectrophotometer (model Perkin Elmer Lamda 45). Exchangeable K in samples was read by aspirating directly into Jenway flame photometer (PFP7). Calcium and Mg in the extract was determined using the Atomic Absorption Spectrometer (AAS). All parameters determined were expressed in percentages using the equations already shown.

3.7 Statistical Analysis

The data collected were analyzed using analysis of variance (ANOVA) at p=0.05 (GenStat, version 11). Significant differences among treatment means were separated using least significant difference (LSD) test 5%.

CHAPTER FOUR

RESULTS

4.1. Initial soil chemical and physical properties from the experimental site

Results of the initial soil analysis for the experimental site are presented in Table 3. The soil at the experimental site was strongly acidic (pH 4.2), high in N and low in exchangeable K and Mg. Exchangeable Ca was marginal at the site. The soil was silty sandy loam texture and moderate in microbial population.

Table1: Initial soil chemical microbial and physical properties from the experimental site

pH	EC	Total N	Available P	Exchangeable cations			Microbial load	Sand	Silt	Clay
				K	Ca	Mg	CFU/100ml			
(1:1H20)	mS	%	ppm	me100g^{-1}				100%		
4.52	0.50	0.23	6.24	0.71	5.36	1.63	1.9x104	36	53	11

4.2 Chemical properties of biochar used in the experiment

The results indicated that biochar used for the experiment was alkaline (pH, 8.24) and contained relatively low values of K, Ca, Mg, N and P (Table 4).

Table 3: Chemical properties of biochar used in the experiment

pH	EC	Total N	Water Holding Capacity	Organic matter	Available P	Exchangeable cations		
						K	Ca	Mg
1: 2.5 H2O	mS		%		µg g^{-1}		cmol+ kg^{-1}	
7.91	4.35	1.4	75	5.67	95.5	1.6	2.2	0.35

4.3 Vegetative growth parameters of okra in the major and minor raining season (Field Experiment)

Plant height, stem diameter, number of leaves and leaf chlorophyll content were measured. The results are presented in tables (Tables 5 to8).

4.3.1 Effect of HYT bio fertilizers and biochar on plant height of okra.

The results showed that in the major season, at 15DAG okra plant height on treated soils were significantly higher than all the control (9.25cm) except for 7t biochar alone (10.75cm) and 3.5t biochar alone (10.25cm). Plant height at 30 DAG and 45DAG were all significantly higher than the controls of (23.50) and (35.50) respectively.

During the minor season, HYT biofertilizers and biochar significantly resulted in taller plant heights in all treatments at 15, 30 and 45DAG (Table 5). However, plant height at 45DAG resulted in significant increased compared to those at 15 and 30DAG

Table 4: Effect of HYT bio fertilizers, biochar and inorganic fertilizers on the height of okra

Treatment combinations	Major rainy season			Minor rainy season		
	15DA G/cm	30DA G/cm	45DA G/cm	15DA G/cm	30DA G/cm	45DA G/cm
Control	9.25	23.50	35.50	10.75	25.25	47.65
100% HYT	13.50	35.00	54.82	14.25	36.25	61.10
50% HYT	13.75	33.25	51.05	14.50	35.25	64.50
7t biochar	10.75	33.75	60.60	17.25	38.50	58.07
7t biochar +100% HYT	16.75	47.80	59.12	20.75	51.50	75.62
7t biochar +50%HYT	17.50	51.00	61.05	15.75	50.75	73.80
3.5t biochar	10.25	34.50	59.57	13.00	34.00	58.30
3.5t biochar +100% HYT	14.00	45.63	56.80	15.00	45.00	68.72
3.5t biochar +50% HYT	14.00	50.80	60.50	18.00	44.25	66.50
100% fertilizer	13.00	34.75	49.25	16.50	44.00	67.75
100%fertilizer+ 100% HYT	15.50	35.50	46.50	15.00	41.50	60.75
100% fertilizer + 50% HYT	14.25	40.75	51.05	16.00	46.50	69.35
100% fertilizer+ 7t biochar	19.75	36.55	48.27	14.75	41.25	63.25
100% fertilizer+7t biochar+ 100%HYT	15.50	49.25	62.97	19.25	48.75	68.75
100% fertilizer+7t biochar+ 50%HYT	18.88	44.75	55.37	18.50	47.75	74.00
100% fertilizer + 3.5t biochar	13.00	37.50	47.50	18.75	51.50	73.00
100%fertilizer+3.5t biochar + 100% HYT	15.75	42.25	51.75	18.50	50.25	80.50
100% fertilizer+3.5t biochar +50% HYT	17.25	43.50	54.25	19.25	48.00	73.50
50% fertilizer	14.50	37.25	48.45	15.50	40.00	71.25
50% fertilizer + 100% HYT	17.00	41.23	51.75	18.25	44.25	65.25
50% fertilizer + 50% HYT	19.65	50.75	62.05	17.00	46.75	69.75
50% fertilizer+7t biochar	14.50	44.00	54.80	17.25	43.50	77.75
50% fertilizer+ 7t biochar + 100% HYT	19.00	55.25	65.37	21.62	63.50	90.05
50% fertilizer+ 7t biochar + 50% HYT	18.75	61.62	73.75	18.88	50.00	72.37
50% fertilizer +3.5t biochar	14.75	36.50	47.50	15.50	44.50	78.00
50% fertilizer+ 3.5t biochar + 100% HYT	14.00	44.25	56.05	20.75	49.00	73.97
50% fertilizer+ 3.5t biochar + 50% HYT	15.50	36.62	47.75	14.87	41.25	64.00
$LSD_{0.05}$	3.998	9.032	9.263	2.557	7.170	9.106

Table 5: HYT bio fertlizers, biochar and inorganic fertilizers on chlorophyll content (mgchl/100gFW) of okra.

Treatment	Major rainy season			Minor rainyseason		
	Chlorophyll content ((mgchl/100gFW)					
	15DAG	30DAG	45DAG	15DAG	30DAG	45DAG
Control	15.70	18.80	18.60	10.38	15.12	15.58
100% HYT	36.75	31.43	29.50	18.35	28.37	28.83
50% HYT	33.95	39.50	30.18	16.25	26.00	26.68
7t biochar	20.15	30.05	30.55	28.48	28.57	29.18
7t biochar +100% HYT	29.70	30.55	25.57	16.68	26.80	26.58
7t biochar +50%HYT	27.70	34.35	26.95	26.40	25.10	25.40
3.5t biochar	21.70	27.18	27.95	24.88	25.60	26.10
3.5t biochar +100% HYT	28.00	33.75	33.00	26.38	26.47	27.83
3.5t biochar +50% HYT	20.40	31.15	39.90	23.73	26.57	28.05
100% fertilizer	45.88	42.10	43.88	23.20	27.57	29.08
100% fertilizer+ 100% HYT	44.23	44.18	44.98	26.25	26.97	27.60
100% fertilizer + 50% HYT	48.85	47.98	46.95	25.50	26.02	27.35
100% fertilizer+ 7t biochar	27.80	34.53	32.62	23.45	25.42	26.43
100% fertilizer+7t biochar+ 100%HYT	46.63	49.30	43.60	24.43	26.32	25.20
100% fertilizer+7t biochar+ 50%HYT	26.85	30.78	27.70	27.70	27.72	27.83
100% fertilizer + 3.5t biochar	43.83	41.80	36.00	26.48	25.97	26.75
100% fertilizer+3.5t biochar + 100% HYT	30.58	36.28	29.78	19.80	21.62	23.15
100% fertilizer+3.5t biochar +50% HYT	32.25	39.55	36.35	19.53	25.27	25.40
50% fertilizer	35.15	33.88	36.00	29.10	31.22	28.58
50% fertilizer + 100% HYT	38.03	33.78	33.60	34.65	31.60	32.08
50% fertilizer + 50% HYT	33.33	37.68	35.73	32.33	23.95	28.58
50% fertilizer+7t biochar	31.88	34.95	35.58	28.63	28.07	27.88
50%fertilizer+ 7t biochar + 100% HYT	34.75	32.60	36.88	19.33	19.82	22.00
50% fertilizer+ 7t biochar + 50% HYT	30.15	27.40	31.40	18.20	23.77	24.00
50% fertilizer +3.5t biochar	34.20	35.20	35.83	26.70	27.25	27.00
50% fertilizer+ 3.5t biochar + 100% HYT	34.78	32.55	33.73	35.35	36.55	30.15
50% fertilizer+ 3.5t biochar + 50% HYT	35.75	33.90	36.08	23.70	25.20	25.38
$LSD_{0.05}$	3.688	6.439	5.037	2.229	5.250	6.107

4.3.2 Effects of HYT bio fertilizers and biochar on chlorophyll content.

During the major season, the application of biochar, HYT and inorganic fertilizers significantly improved chlorophyll content resulting in greener leaves in all treatments observed at 15, 30 and 45 DAG (Table 6)

For data collected in the minor season, HYT, inorganic fertilizers and biochar significantly improved the chlorophyll content of the leaves at 15, 30 and DAG (Table 6). However, treatments imposed with 50% fertilizer + 100% HYT gave greener chlorophyll content than all treatments at 15, 30 and 45DAS with 34.65; 31.60 and 32.08 respectively.

4.3.3 Effects of HYT bio fertilizers and biochar on number of leaves

The application of HYT and biochar increased leaf number statistically at 15 DAG with the exception of soil amendment combinations of 7t biochar(3) 7t biochar +50%HYT (3), 3.5t biochar(4), 3.5t biochar +100% HYT(4),3.5t biochar + 50% HYT (3), 100% fertilizer (4), 100% fertilizer +3.5t biochar (4) and 50% fertilizer (4) from the control. In the minor season at 15DAG only 3.5t biochar + 100% HYT(10) and 50% fertilizer +7t biochar +100% HYT(11) recorded significantly more leaves than the control (Table 7)

At 30DAG in the major season, all treatment combinations except 100% fertilizer + 7t biochar (7), 50% fertilizer + 7t biochar + 100% HYT (20), produced lesser number of leaves than the control (12). In the minor season, 100% fertilizer +100% HYT (15), 100% fertilizer +7t biochar +50% HYT(16) 50% fertilizer + 100% HYT (16), 50% fertilizer + 7t biochar + 100% HYT (20), 50% fertilizer +7t biochar +50%HYT (16) and 50% fertilizer +3.5t biochar + 100% HYT (19) produced more number of leaves than the control (9).

At 45DAG only 100% fertilizer +7t biochar +50% HYT (22), 50% fertilizer +7t biochar +100% HYT (25), 50% fertilizer +7t biochar +50% HYT (26), 50% fertilizer +3.5t biochar +100% HYT (22) and , 50% fertilizer +3.5t biochar +50% HYT (20) produced significantly more leaves than the control (16) the rest were not significantly different than the control. In the minor season, all treatment combinations produced significantly more number of leaves over the control (12) (Table 7).

Plant leaves at 35 and 45DAG significantly increased as compared to 15DAG in both minor and major seasons.

4.3.4 HYT inorganic fertilizers and biochar on plant diameter

Table 8 indicates that all treatments had higher stem girths at 15, 30, and 45 DAG with the exception of plant treatments with 50% or 100% inorganic fertilizers. A similar pattern was observed for stem girth of plants at 15 DAG in the minor season but at 30DAG and 45 DAG, treatments had higher stem girth compared to the control (Table 8).

Table 6: Effect of HYT bio fertilizers and biochar on number of leaves of okra

Treatment	Major rainy season			Minor rainy season		
	Leaf number/ plant 15DAG	Leaf number/ plant30DAG	Leaf number/ plant 45DAG	Leaf number/ plant 15DAG	Leaf number/ plant 30DAG	Leaf number/ plant 45DAG
Control	2.75	11.75	15.50	7.00	9.75	12.00
100% HYT	5.75	10.50	17.75	5.00	11.00	18.50
50% HYT	5.75	10.50	17.00	6.00	11.00	19.00
7t biochar	2.75	11.50	17.75	5.00	10.75	19.25
7t biochar +100% HYT	4.25	11.75	16.75	9.00	14.50	21.00
7t biochar +50%HYT	3.25	11.75	15.50	8.00	19.75	23.00
3.5t biochar	3.75	10.75	16.00	6.00	10.75	18.25
3.5t biochar +100% HYT	3.75	13.00	16.00	10.00	11.25	18.50
3.5t biochar +50% HYT	3.25	11.00	14.75	8.00	15.75	20.25
100% fertilizer	3.75	11.75	16.00	8.75	11.75	18.75
100% fertilizer+ 100% HYT	5.25	12.75	17.50	6.00	11.25	19.50
100% fertilizer + 50% HYT	5.50	13.50	18.25	7.00	15.25	19.50
100% fertilizer+ 7t biochar	5.50	14.50	18.75	7.75	14.75	18.00
100% fertilizer+7t biochar+ 100%HYT	5.50	13.50	18.00	7.75	11.75	18.25
100% fertilizer+7t biochar+ 50%HYT	7.50	17.25	22.00	8.25	16.00	20.50
100% fertilizer + 3.5t biochar	4.00	12.50	17.00	8.75	12.75	19.50
100% fertilizer+3.5t biochar + 100% HYT	5.50	14.00	18.25	7.25	13.50	18.00
100% fertilizer+3.5t biochar +50% HYT	6.25	15.50	19.75	9.00	14.25	18.25
50% fertilizer	3.75	11.50	15.75	8.25	11.50	20.50
50% fertilizer + 100% HYT	5.00	14.25	18.75	9.00	16.25	19.25
50% fertilizer + 50% HYT	5.50	15.75	20.00	8.50	13.75	19.00
50% fertilizer+7t biochar	4.25	14.25	18.25	6.75	12.50	18.50
50% fertilizer+ 7t biochar + 100% HYT	7.25	20.25	24.75	10.50	19.50	24.50
50% fertilizer+ 7t biochar + 50% HYT	9.00	22.75	26.25	8.75	16.25	20.25
50% fertilizer +3.5t biochar	5.75	14.50	18.25	6.50	13.00	18.75
50% fertilizer+ 3.5t biochar + 100% HYT	6.00	17.75	22.25	8.75	18.50	21.75
50% fertilizer+ 3.5t biochar + 50% HYT	5.25	15.50	20.25	7.00	12.00	18.25
$LSD_{0.05}$	2.430	4.486	4.655	2.237	4.647	4.771

Table 7: HYT inorganic fertilizers and biochar on plant girth (cm)

Treatment	Major rainy season			Minor rainy season		
	Stem Girth 15 DAG	Stem Girth 30 DAG	Stem Girth 45 DAG	Stem Girth 15 DAG	Stem Girth 30 DAG	Stem Girth 45 DAG
Control	1.38	3.63	4.1	1.57	2.67	3.77
100% HYT	1.46	3.46	4.71	1.57	3.14	4.47
50% HYT	1.54	3.22	4	1.59	3.42	4.39
7t biochar	1.49	3.36	5.25	1.04	3.22	4.08
7t biochar +100% HYT	1.74	3.85	5.42	1.76	3.69	4.71
7t biochar +50%HYT	1.66	4.01	5.5	1.66	3.3	4.4
3.5t biochar	1.56	3.61	4.26	1.65	3.54	4.39
3.5t biochar +100% HYT	2.3	3.77	5.5	1.57	2.99	4
3.5t biochar +50% HYT	2.15	3.93	5.65	1.65	2.95	3.92
100% fertilizer	1.32	2.55	4.16	1.47	3.46	4.79
100% fertilizer+ 100% HYT	2.91	3.91	4.47	1.79	3.3	4.32
100% fertilizer + 50% HYT	2.61	3.18	4.79	1.64	3.73	4.67
100% fertilizer+ 7t biochar	2.46	3.14	4.83	1.61	3.54	4.4
100% fertilizer+7t biochar+ 100%HYT	2.77	3.63	4.16	1.99	3.77	5.47
100% fertilizer+7t biochar+ 50%HYT	2.5	3.3	4.71	1.78	4.09	5.91
100% fertilizer + 3.5t biochar	2.2	2.75	4.08	1.73	3.26	4.32
100% fertilizer+3.5t biochar + 100% HYT	2.36	3.22	5.26	1.57	3.65	4.93
100% fertilizer+3.5t biochar +50% HYT	1.99	3.46	5.02	1.99	3.93	5.18
50% fertilizer	1.24	2.91	4.4	1.22	3.38	4.47
50% fertilizer + 100% HYT	1.68	3.06	4.63	1.65	3.73	4.95
50% fertilizer + 50% HYT	1.41	3.85	5.57	1.65	3.46	4.63
50% fertilizer+7t biochar	1.54	3.49	4.98	1.57	4.01	4.95
50% fertilizer+ 7t biochar + 100% HYT	2.51	4.83	6.83	1.82	4.28	5.69
50% fertilizer+ 7t biochar + 50% HYT	1.81	3.69	6.79	1.89	3.69	5.79
50% fertilizer +3.5t biochar	1.57	3.96	4.49	1.61	3.61	4.63
50% fertilizer+ 3.5t biochar + 100% HYT	3.16	4.01	5.65	2.29	3.46	5.63
50% fertilizer+ 3.5t biochar + 50% HYT	2.5	3.95	5.47	1.73	3.14	5.79
$LSD_{0.05}$	0.439	0.568	0.699	0.339	0.568	0.658

Table 8: Effect of HYT bio fertilizers and biochar on fresh weight in (cm) of okra

Treatment	Major rainy season			Minor rainy season		
	Root fresh weight (g)	Stem fresh weight (g)	Leaf fresh of weight (g)	Root fresh weight (g)	Stem fresh weight (g)	Leaf fresh weight (g)
Control	13.33	48.97	44.20	11.75	50.89	37.40
100% HYT	13.73	71.80	64.80	16.52	86.73	69.87
50% HYT	13.10	80.30	68.70	14.87	79.53	62.77
7t biochar	18.90	87.35	72.05	17.17	109.48	86.30
7t biochar +100% HYT	20.15	130.65	119.97	23.92	162.23	153.75
7t biochar +50%HYT	17.28	97.32	90.88	26.10	239.40	84.92
3.5t biochar	15.65	77.00	69.60	12.80	111.98	70.82
3.5t biochar +100% HYT	19.23	127.50	113.80	15.60	114.90	89.80
3.5t biochar +50% HYT	17.62	118.27	108.50	16.45	126.65	50.52
100% fertilizer	19.33	121.97	117.30	21.00	124.72	96.02
100% fertilizer+ 100% HYT	16.40	93.05	74.73	18.50	99.03	103.30
100% fertilizer + 50% HYT	15.43	87.30	69.45	12.20	71.95	54.70
100% fertilizer +7t biochar	14.23	94.92	82.38	11.02	75.85	63.05
100% fertilizer+7t biochar+ 100%HYT	9.85	67.07	58.98	8.52	71.45	59.35
100% fertilizer+7t biochar+ 50%HYT	15.18	108.30	94.00	30.55	181.12	121.60
100% fertilizer + 3.5t biochar	13.45	170.10	73.18	10.65	128.55	67.32
100% fertilizer+3.5t biochar + 100% HYT	13.05	71.87	64.10	22.45	89.40	54.70
100% fertilizer+3.5t biochar +50% HYT	21.33	138.72	106.00	28.85	85.50	67.62
50% fertilizer	17.05	97.15	67.93	17.97	94.15	62.02
50% fertilizer + 100% HYT	15.93	78.82	131.78	12.85	103.40	111.97
50% fertilizer + 50% HYT	17.40	86.95	72.80	17.77	87.23	71.27
50% fertilizer+7t biochar	17.90	83.85	58.98	17.65	98.58	68.38
50% fertilizer+ 7t biochar + 100% HYT	13.75	83.22	95.80	11.27	104.10	94.85
50% fertilizer+ 7t biochar + 50% HYT	19.25	109.35	92.08	25.55	111.25	84.00
50% fertilizer +3.5t biochar	17.03	67.20	57.80	11.67	68.73	56.70
50% fertilizer+ 3.5t biochar + 100% HYT	30.48	270.60	228.60	33.15	119.65	112.47
50% fertilizer+ 3.5t biochar + 50% HYT	14.55	61.40	49.80	14.52	61.20	58.90
$LSD_{0.05}$	6.152	45.063	33.144	8.232	34.383	19.915

4.3.5 Effect of HYT bio fertilizers and biochar on plant biomass (roots, leaves, stem)

Fresh root weight differences among treatments were significant for only treatments with 100% HYT and biochar combinations or HYT and biochar alone. 50% fertilizer +3.5tbiochar+100%HYT giving the most outstanding performance of (30.48g) beside, combinations of 100% fertilizer +3.5tbiochar+50% HYT (21.33g) and 50% fertilizer + 3.5t

biochar+100% HYT(30.47g). The rest were significantly lighter in weight than the control (13.33g) (Table 9). For instance performance at 100% and 50% fertilizer, biochar and HYT alone did not show any better result than the control.

For the minor season, fresh root weight soil amendment combinations that were significantly heavier than the control (11.75g) were those treated with maximum levels of biochar in combination with HYT,50% fertilizer 3.5t biochar +100%HYT (33.15g) and 100% fertilizer alone(21.00g) and maximum levels of fertilizer and biochar +50% HYT(30.53g) or in combination with 3.5tbiochar,maximum levels of HYT in combinations with 3.5tbiochar,100% HYT in combination with 50% fertilizer +7t biochar +50% HYT (25.55g). All other treatment combinations aside these were significantly lighter in weight than the control (11.75g) (Table 9).

Differences in fresh stem weight in the major season revealed that soil amendment combinations with biochar and HYT at maximum levels and fertilizer alone in combination with 50% fertilizer + 3.5t biochar +100% HYT (270.60g), 100% fertilizer + 3.5t biochar (170.10g), 100% fertilizer + 3.5t biochar + 50% HYT (138.72g), 100% fertilizer alone (121.92g), 50% fertilizer + 7t biochar +50% HYT (109.35g) and 100% fertilizer + 7t biochar + 50% HYT (108.30g) gave values that were significantly heavier than the control (48.97). (Table 9).

For the minor season, except for sole soil amendments of 100%HYT(79.53g),100% fertilizer+50% HYT (71.95g),100% fertilizer +7t biochar (75.85g),100% fertilizer +3.5t biochar+50% HYT (61.20g), all other soil amendments combination gave significantly heavier stem weights than the control (50.89),the heaviest occurring at 100% fertilizer + 7t biochar + 50% HYT (181.12g)(Table 9). 7t biochar and 50% HYT significantly gave the highest fresh stem weight.

For fresh leaf weight in the major season, indicated in Table 9, except for maximum levels of biocharandHYTcombinations,50%fertilizer+3.5tbiochar+100%(228.60g), 50% fertilizer +100%HYT(131.78g)100%fertilizer+3.5tbiochar+50%HYT(106.00g),50%fertilizer+7tbioch ar+100%HYT (95.80g), 50% fertilizer+7tbiochar+50%HYT (92.08g) that resulted in heavier leaf weight statistically , all other soil amendment combinations gave statistically lighter leaf weights than the control (44.20g). 7t biochar + 50% HYT (239.40g) significantly gave the highest fresh stem weight.

In the minor season, apart from soil amendment combinations of 3.5t biochar + 50% HYT (50.52g), 100% fertilizer + 50% HYT (54.70g), 100% fertilizer + 3.5t biochar + 100% HYT (54.70g), 50% fertilizer + 3.5t biochar (56.70g) and 50% fertilizer + 3.5t biochar +50% HYT (58.90g) all treatment combinations significantly produced heavier fresh leaf weights than the control with the following treatments giving the most significant weights, 100% fertilizer + 7t biochar + 50% HYT (121.60g), 50% fertilizer +3.5t biochar + 100% HYT (112.47g), 50% fertilizer + 100% HYT (111.97g) and 100% fertilizer + 100% HYT (103.30g). (Table 9). However 7t biochar + 50% HYT treatment combination produced the heaviest fresh leaf weight 153.75g.

Table 9: Effect of HYT bio fertilizers and biochar on days to flowering of okra

Treatment	Major rainy season		Minor rainy season	
	Days to 1st flowering	Days to 50% flowering	Days to 1st flowering	Days to 50% flowering
Control	48.50	50.75	43.50	46.75
100% HYT	54.75	56.75	53.25	54.00
50% HYT	55.50	58.00	51.50	53.00
7t biochar	51.50	53.00	53.00	54.25
7t biochar +100% HYT	45.25	48.25	46.75	50.00
7t biochar +50%HYT	46.00	50.25	46.50	48.50
3.5t biochar	55.00	57.00	52.25	54.00
3.5t biochar +100% HYT	44.00	47.25	45.25	46.50
3.5t biochar +50% HYT	44.25	47.50	46.50	48.50
100% fertilizer	51.75	56.00	46.25	49.25
100%fertilizer+ 100% HYT	55.00	57.00	50.75	51.75
100% fertilizer + 50% HYT	54.00	55.25	48.75	51.50
100% fertilizer+ 7t biochar	50.25	55.00	49.75	52.25
100%fertilizer+7t biochar+ 100%HYT	53.50	55.00	48.50	51.00
100%fertilizer+7t biochar+ 50%HYT	51.00	54.75	50.50	52.50
100%fertilizer + 3.5t biochar	51.75	55.50	49.75	51.75
100%fertilizer+3.5t biochar + 100% HYT	53.50	54.75	45.25	47.75
100%fertilizer+3.5t biochar +50% HYT	52.25	56.00	45.50	47.00
50%fertilizer	51.00	54.25	43.50	47.00
50% fertilizer + 100% HYT	53.25	54.25	50.00	52.25
50% fertilizer + 50% HYT	53.25	54.00	50.50	51.75
50% fertilizer+7t biochar	50.50	52.25	42.75	46.00
50%fertilizer+ 7t biochar + 100% HYT	53.25	56.50	49.50	51.25
50%fertilizer+ 7t biochar + 50% HYT	53.50	54.25	52.25	53.75
50% fertilizer +3.5t biochar	49.50	52.75	41.25	45.50
50%fertilizer+ 3.5t biochar + 100% HYT	49.25	51.00	48.25	49.75
50%fertilizer+ 3.5t biochar + 50% HYT	55.25	55.75	49.25	51.25
$LSD_{0.05}$	3.101	3.372	2.567	2.057

4.4.0 Days to first flowering and days to 50% flowering

Table 10 shows that there were no significant differences in days to flowering in the following treatments 7t biochar(53days),7t biochar +100% HYT(48days),7t biochar + 50% HYT(50days),3.5t biochar +50%HYT(47days), 50% fertilizer +3.5t biochar +100% HYT(51days) compared to the control in the major season. In the minor season, the data indicate that days to 50% among 7t biochar + 50% HYT (48), 3.5t biochar + 100% HYT (46days), 3.5t biochar + 50% HYT (48days) 100% fertilizer + 3.5t biochar + 100% HYT (48days), 100% fertilizer + 3.5t biochar + 50% HYT (47days) and 50% fertilizer (47days)

were not significantly different from the control (47days). The other treatments took significantly longer days to flower. (Table 10). Table 10 again shows that, days to 50%flowering was longer in the major season compared to the minor season

4.5.1Effect of HYT bio fertilizers and biochar on yield and yield characteristics of okra

Plants treated with 3.5t biochar+ 100% HYT, 7t biochar + 50% HYT (23) and 7t biochar + 100% HYT (22) recorded the highest number of fruits. (Table11).

The results also show that in the minor season 100% HYT (21), 7t biochar + 100% HYT (24), 7t biochar + 50% HYT (21), 3.5t biochar + 100%HYT (23), 3.5t biochar + 50%HYT (22) significantly had the highest fruit number compared to the control (10) (Table 11).

Yield per plant (g) in the major season experiment as shown in Table 11, indicates that 7tbiochar + 50% HYT (253.50g), 7t biochar +100% HYT (247.45g) and 3.5t biochar + 100 HYT (247.53g) significantly had the highest fruit weight per plant compared to the control (88.8g).

In the minor season 7t biochar +100% (248.00g), 3.5t biochar + 100% HYT, (247.55g) and 7t biochar + 50% HYT (224.65g), 7t biochar + 50% HYT (224.18g) significantly recorded the highest yield compared to the control (92.47g) (Table 11).

Significant differences were observed in fruit number and yield per plant between the control and the other treatments in both the major and minor seasons. In the major season the highest yield were recorded for the following soil amendments combinations, 3.5t biochar + 100% HYT (8.25t/ha), 7t biochar + 100% HYT (8.23t/ha), 7t biochar + 50% HYT (7.88t/ha) 50% HYT alone (7.53t/ha). All results in the minor season revealed that soil amendment combinations gave significant higher yields than the control (3.07t/ha). The best results were obtained with soil amendment combinations of 7t biochar + 100% HYT (8.25t/ha) 3.5t

biochar +100% HYT(7.67t/ha) and 3.5t biochar +50% HYT(7.45t/ha). Treatment combination that gave the least yield statistically was only 3.5t biochar (4.40t/h) Table 11.

Table 10: Effect of HYT bio fertilizers and biochar on number of fruits and yield of okra

Treatment	Major rainy season			Minor rainy season		
	Number of fruits	Yield/ plant(g)	Yield/ hectare (g)	Number of fruits	Yield/ plant (g)	Yield/ hectare (t)
Control	9	89	3	10	92	3
100% HYT	21	221	7	22	218	7
50% HYT	21	225	8	21	217	7
7t biochar	18	181	6	14	136	5
7t biochar +100% HYT	23	247	8	24	248	8
7t biochar +50%HYT	23	254	8	22	224	7
3.5t biochar	13	131	4	13	132	4
3.5t biochar +100% HYT	23.	248	8	23	230	8
3.5t biochar +50% HYT	21	225	8	22	224	7
100% fertilizer	17	166	6	17	158	5
100%fertilizer+ 100% HYT	21.	214	7	19	192	6
100% fertilizer + 50% HYT	19	167	6	17	164	5
100% fertilizer+ 7t biochar	15	144	5	17	162	5
100%fertilizer+7t biochar+ 100%HYT	20	220	7	20	212	7
100%fertilizer+7t biochar+ 50%HYT	20	217	7	20	208	7
100%fertilizer + 3.5t biochar	15	142	5	17	162	5
100%fertilizer+3.5t biochar + 100% HYT	19	197	7	20	200	7
100%fertilizer+3.5t biochar +50% HYT	19	206	7	19	196	7
50%fertilizer	18	172	6	15	147	5
50% fertilizer + 100% HYT	19	185	6	19	188	6
50% fertilizer + 50% HYT	18	195	6	16	170	6
50% fertilizer+7t biochar	18	178	6	15	148	5
50%fertilizer+ 7t biochar + 100% HYT	20	210	7	21	211	7
50%fertilizer+ 7t biochar + 50% HYT	20	208	7	19	202	7
50% fertilizer +3.5t biochar	18	189	6	16	155	5
50%fertilizer+ 3.5t biochar + 100% HYT	20	218	7	21	197	7
50%fertilizer+ 3.5t biochar + 50% HYT	20	216	7	19	193	6
$LSD_{0.05}$	1.25	15.08	0.56	1.61	18.60	0.62

4.5.2 Effects of HYT bio fertilizers and biochar on fruit weight, fruit length and fruit diameter.

The results in Table 12 indicate that fruit weight in all the soil amendment combinations treatment was higher than the control (6.45g). Plant treated with 50% fertilizer + 3.5t biochar + 50% HYT (11.00g), 50% fertilizer + 100% HYT (10.95g), 50% fertilizer + 3.5t biochar +

100% HYT (10.83g), 50% fertilizer + 7t biochar +100% HYT (10.88g), 100% fertilizer +3.5t biochar + 100% HYT (11.18g) had the highest fruit weights during the major rainy season. (Table 12)

During the minor season, 100% fertilizer + 7t biochar (11.00g) 100% fertilizer + 3.5t biochar (10.30), 50% fertilizer + 7t biochar +100% HYT (10.48g), 100% fertilizer + 3.5t biochar + 100% HYT (10.63g), 100% fertilizer + 3.5t biochar + 50% HYT (10.43g) significantly recorded the highest fruit weight than the control (6.45g).

Fruit length in treated plots was significantly longer than the control in both major and minor reasons. In the major season, soil amendment combinations at 100% biochar and HYT gave longer fruits. Also soil amendment combinations of 50% fertilizer + 7t biochar + 100% HYT (8.20cm) at both major and minor seasons had longer fruits length than the controls. (Table12).

Differences in fruit diameter were also recorded between 50% fertilizer + 3.5t biochar +100% HYT and 50% fertilizer + 7t biochar + 100% HYT in the major and minor seasons. Fruit diameter was 9.10cm and 8.92cm compared to 8.63cm and 9.22cm in the major and minor seasons for the former and latter respectively (Table 12). Fruits diameter as shown in the Table were also significant in both major and minor seasons. The combination of biochar and HYT at various levels resulted in bigger diameter in both major and minor seasons.

Table 11: Effect of HYT bio fertilizers and biochar on yield characteristics of okra

Treatment	Major rainy season			Minor rainy season		
	Fruit weight (g)	Fruit length (cm)	Fruit girth (cm)	fruit weight (g)	Fruit length (cm)	Fruit girth (cm)
Control	6.45	6.45	6.73	6.63	6.85	6.25
100% HYT	9.88	7.65	8.20	9.98	7.40	7.95
50% HYT	9.90	7.68	7.70	9.58	7.30	7.52
7t biochar	9.78	7.60	7.70	9.65	7.72	8.07
7t biochar +100% HYT	10.05	8.38	8.43	10.23	8.42	8.80
7t biochar +50%HYT	9.60	8.20	8.45	10.15	8.10	8.52
3.5t biochar	9.78	8.18	8.25	9.58	7.80	8.62
3.5t biochar +100% HYT	9.70	8.15	8.15	9.65	8.02	8.60
3.5t biochar +50% HYT	10.10	7.68	7.25	9.18	7.82	8.52
100% fertilizer	10.70	7.53	7.98	10.53	8.25	7.62
100%fertilizer+ 100% HYT	10.23	7.50	8.13	9.85	7.62	7.55
100% fertilizer + 50% HYT	10.40	7.78	7.73	10.28	7.07	7.70
100% fertilizer+ 7t biochar	10.65	7.40	7.93	11.00	8.55	7.67
100%fertilizer+7t biochar+ 100%HYT	10.65	7.38	7.98	9.98	7.15	7.72
100%fertilizer+7t biochar+ 50%HYT	10.70	7.58	7.70	9.75	7.32	7.55
100%fertilizer + 3.5t biochar	10.68	7.45	8.08	10.30	7.20	7.22
100%fertilizer+3.5t biochar + 100% HYT	11.18	7.78	8.30	10.63	7.77	8.60
100%fertilizer+3.5t biochar +50% HYT	10.90	7.68	7.83	10.43	8.05	8.42
50%fertilizer	10.65	7.95	8.03	9.68	7.85	7.25
50% fertilizer + 100% HYT	10.95	7.80	8.15	10.20	7.55	7.35
50% fertilizer + 50% HYT	10.78	7.98	7.65	10.00	7.27	6.50
50% fertilizer+7t biochar	10.05	7.90	8.20	9.73	7.97	8.12
50%fertilizer+ 7t biochar + 100% HYT	10.88	8.20	8.63	10.48	8.20	9.22
50%fertilizer+ 7t biochar + 50% HYT	10.78	7.75	7.95	10.15	8.15	8.52
50% fertilizer +3.5t biochar	9.88	7.73	8.20	9.88	7.25	7.80
50%fertilizer+ 3.5t biochar + 100% HYT	10.83	7.85	9.10	10.40	8.15	8.92
50%fertilizer+ 3.5t biochar + 50% HYT	11.00	7.43	7.98	10.23	7.35	8.25
$LSD_{0.05}$	0.663	0.823	0.323	0.492	0.531	0.179

Table 12: Effect of HYT bio fertilizers and biochar on soil chemical properties

Treatment	N (%)	P (ppm)	K (me $100g^{-1}$)	Ca (me $100g^{-1}$)	Mg (me $100g^{-1}$)	pH	E.C. (mS)
Control	0.18	94.95	0.45	1.13	1.41	5.10	0.43
3.5t biochar	0.35	116.98	0.80	0.94	1.36	5.18	0.78
7t biochar	0.30	116.9	0.78	0.83	1.15	4.94	1.20
50% fertilizer+7t biochar	0.35	86.33	0.62	0.63	1.38	5.30	0.13
50%fertilizer+ 3.5t biochar	0.31	32.95	0.63	0.75	1.06	4.88	1.00
50% fertilizer	0.59	85.10	0.62	1.12	1.26	5.56	0.43
100% fertilizer+ 7t biochar	0.35	176.58	0.32	0.61	1.06	5.12	1.70
100%fertilizer + 3.5t biochar	0.55	171.03	0.98	0.77	0.89	5.16	1.00
100% fertilizer	0.49	125.48	0.83	1.67	0.94	5.06	1.70
100% fertilizer+ 7t biochar+ 100% HYT	4.32	115.03	0.82	0.75	1.51	4.94	1.98
50% fertilizer +3.5t biochar+50% HYT	0.27	66.30	0.82	0.74	1.51	5.02	2.00
100%fertilize+ 100% HYT	0.47	94.78	0.67	1.35	0.84	5.66	0.30
100% fertilizer + 50% HYT	0.36	70.40	0.77	0.58	0.81	5.15	1.13
50% fertilizer + 100% HYT	0.38	84.98	0.96	1.13	0.94	5.18	1.65
50% fertilizer + 50% HYT	0.34	115.9	0.95	0.74	1.09	5.10	2.00
100%fertilizer+7t biochar+ 100%HYT	0.37	75.88	0.62	3.43	1.38	5.32	0.20
100%fertilizer+3.5t biochar + 100% HYT	0.46	174.1	0.96	1.14	1.02	5.05	1.65
100%fertilizer+3.5t biochar +50% HYT	0.39	152.2	1.08	1.19	1.06	4.98	1.93
50%fertilizer+ 7t biochar +50%HYT	0.40	155.5	0.95	0.93	0.76	4.99	1.13
50%fertilizer+7t biochar+ 50%HYT	0.45	75.8	0.52	3.00	1.56	5.45	0.20
50%fertilizer+ 3.5t biochar+100%HYT	0.39	85.6	0.83	1.48	0.90	4.78	1.18
7t biochar +100% HYT	2.84	32.1	0.26	0.96	1.24	5.73	0.20
50% HYT	0.43	30.6	0.30	2.62	1.00	5.54	0.20
100% HYT	0.38	28.9	0.30	1.20	1.55	5.88	0.10
7t biochar +50%HYT	0.29	30.5	0.23	1.26	1.56	5.58	0.10
3.5t biochar +100% HYT	0.38	3.49	0.23	0.95	1.33	5.99	0.10
3.5t biochar +50% HYT	0.52	2.55	0.40	3.00	1.18	5.67	0.10
L.S.D (0.05)	0.07	6.75	0.09	0.10	0.08	0.19	0.17

4.5.3 Effect of HYT bio fertilizers and biochar on soil chemical properties

NITROGEN

Soil amendment combination for nitrogen studied had higher values than the control (0.18) with 7tbiochart+100%HYT (2.84) proving to be the most outstanding.

PHOSPHORUS

Soil phosphorus under different soil amendments combinations gave higher values in 100% fertilizer +7t biochar(176.58);100% fertilizer +3.5t biochar +100%HYT (174.09), 50%

fertilizer +7t biochar +50% HYT (155.50);100%fertilizer +7t biochar +50%HYT (152.23), 100% fertilizer (125.48) and soil biochar amendments at 3.5t biochar(116.98) and 7t biochar (116.93) and were statistically significantly higher than the control(94.95) The least was recorded at 3.5t biochar +100%HYT (3.49) (Table 13).

POTASSIUM

All soil amendments combinations except for 50% fertilizer + 7t biochar +50% HYT (0.52); 7t biochar +100 HYT (0.26); 50% HYT (0.30); 100% HYT (0.30); 7t biochar +50% HYT (0.23); 3.5t biochar + 100% HYT (0.23); 3.5t biochar + 50% HYT (0.40)and 100% fertilizer +7tbiochar (0.32) were statistically significantly higher than the control(0.45).The combination that gave the highest effect was recorded at 100%fertilizer +3.5tbiochar+50%HYT (1.08). (Table 13).

CALCIUM

In Table 13, soil calcium measured indicated that 100% fertilizer (1.67); 100% fertilizer + 100% HYT (1.35); 50% fertilizer + 7t biochar + 100% HYT (1.48) gave statistically significantly higher values than the control (1.13),50% fertilizer +7t biochar +50%HYT (3.00); 3.5t biochar + 50% HYT (3.00)and 50% HYT (2.62) gave the most significant values whilst the rest of the treatments had significantly lower calcium than control (1.13).

MAGNESIUM

Magnesium conditions studied under different soil amendment combinations gave statistically lower values than the control(1.4) except,100% fertilizer + 7t biochar 100% HYT (1.51),50%fertilizer +3.5 biochar +50% HYT (1.51),50% fertilizer +7t biochar + 50% HYT (1.56), 100% HYT (1.55) and 7tbiochar + 50% HYT (1.56) that showed statistically significantly higher values than the control, as is shown in Table 13..

SOIL pH

Table 13 also shows that all soil amendments combinations analyzed were acidic from the pH measurement. However 100%biochar and HYT combination or HYT alone,50% fertilizer + 7tbiochar(5.30),50% fertilizer (5.56); 100% fertilizer + 100% HYT(5.66); 100% fertilizer + 7t biochar + 100% HYT(5.32); 50% fertilizer +7t bioiclar+50% HYT (5.45) 7t biochar + 100% HYT (5.73) 50% HYT (5.54) 100% HYT (5.88) 7t biochar + 50% HYT (5.58); 3.5t biochar + 100% HYT (5.99) and 3.5t biochar + 50% HYT (5.67)were all less acidic and had values higher than the control of (5.10). The rest of the soil amendment combinations were highly acidic and had significantly lower pH than the control(5.10) (Table 13).

ELECTRICAL CONDUCTIVITY (EC)

EC content of the various soil amendments levels showed that 50% fertilizer +7tbiochar and 50% fertilizer alone(0.20),100%fertilizer +100% HYT (0.30), 1100% fertilizer, biochar and HYT(0.20), 50%fertilizer +7t biochar+50%HYT(0.20) together with 100% biochar and HYT were all not statistically different from the control (0.43) (Table 13). EC (0.10) was recorded to be the least while all other soil amendment combinations were statistically significantly higher than the control (0.43) the most significant soil amendment combination recorded for EC was 50% fertilizer +3.5t biochar+50%HYT (2.00) and 50% fertilizer +50% HYT (2.00) (Table 13).

Table 13: Effect of HYT biofertilizers, biochar and inorganic fertilizer application on soil microbial population at pre and post-harvest stages of study

Treatment	Pre harvest stage 10^3/ CFU/100ml	Post-harvest stage 10^3/ CFU/100ml
Control	1.9	2.0
3.5t biochar	3.7	6.3
7t biochar	2.8	5.7
50% fertilizer+7t biochar	5.8	5.2
50%fertilizer+ 3.5t biochar	7.6	8.8
50% fertilizer	5.6	6.1
100% fertilizer+ 7t biochar	8.7	10.5
100%fertilizer + 3.5t biochar	3.8	7.0
100% fertilizer	2.6	5.5
100% fertilizer+ 7t biochar+ 100% HYT	4.8	5.5
50% fertilize +3.5t biochar+50% HYT	4.7	5.4
100%fertilize+ 100% HYT	6.2	16.7
100% fertilizer + 50% HYT	5.7	4.8
50% fertilizer + 100% HYT	7.5	17.8
50% fertilizer + 50% HYT	4.8	11.0
100%fertilizer+7t biochar+ 100%HYT	3.8	5.9
100%fertilizer+3.5t biochar + 100% HYT	5.2	17.8
100%fertilizer+3.5t biochar +50% HYT	5.1	19.5
50%fertilizer+ 7t biochar +50%HYT	4.5	7.0
50%fertilizer+7t biochar+ 50%HYT	5.1	7.0
50%fertilizer+ 3.5t biochar+100%HYT	4.5	6.6
7t biochar +100% HYT	3.7	99.1
50% HYT	5.0	5.2
100% HYT	5.7	17.8
7t biochar +50%HYT	5.2	11.5
3.5t biochar +100% HYT	4.8	19.5
3.5t biochar +50% HYT	5.2	18.8
L.S.D (0.05)	1.10	1.89

4.5.4 Effect of HYT biofertilizers, biochar and inorganic fertilizer application on soil microbial population at pre and post- harvest stages

Pre harvest stage

With the exception of 7t biochar (2.8) and 100% fertilizer (2.6) all soil amendment combinations significantly recorded higher microbial count than the control (1.9). (Table 14).

The best microbial results were obtained from 100% fertilizer +7t bioclar (8.7); 50% fertilizer + 3.5t biochar (7.8); 50% fertilizer + 100% HYT (7.5) and 100% fertilizer +100% HYT (6.2).

Post-harvest stage

Microbial count during the post-harvest stage at all the soil amendments combinations were significantly more than control (2.0).The soil amendments combination that gave the maximum microbial population count was 7t biochar + 100% HYT (99.1). (Table 14). The combination of biochar and HYT or HYT alone greatly enhanced microbial development than fertilizer and biochar combined or biochar or fertilizer alone.

CHAPTER FIVE

DISCUSSION

5.1 Effect of HYT biofertilizers and biochar application on soil chemical and microbial properties

The increase in the pH of the soil after the application of biochar could be attributed to the high pH level of the biochar and carbonate concentration which had a liming effect on the soil. Similar observations were reported by (Glaser *et al*., 2002; and Van Zweiten *et al*., (2007). The high concentration of potassium and relatively high concentrations of calcium and magnesium in the biochar amended fields could also be due to low exchangeable acidity. This may be explained using results of (Cochrane and Sanchez 1980, Fisher and Binkely 2000), who reported that precipitation of Al as hydroxyl releases cations in the soil. The increased available P content and total N of the soil with the application of bio fertilizer and inorganic fertilizers could be attributed to release of P from complexes of Al and Fe under increasing soil pH, the higher sorption affinity of biochar for organic and inorganic compounds and higher nutrient retention ability of biochar as explained by (Kleineidam *et al*., 2002) in similar studies supported and that the availability of biochar increased the availability of all major cations, as reported by (Glaser *et al*., 2002 and Lehmann *et al*., 2003).The pH of the experimental plot at the start of the experiment was 4.52 compared to a range of 4.78- 5.99 obtained at 90 days after application of the biochar.

Though there was not much difference in the number of microbes in the soil at the initial stages, however, the microbial population increased due to the application of biochar which is

consistent with findings of (Kolb *et al*., 2009) and repeated application of HYT biofertilizers supported by work done on rice by Subashini *et al*., (2007). The increase in microbial load due to application of biochar could be attributed to creation of microclimate that encouraged microbial colonization. Biochar served as refuge by protecting microbes from predation and desiccation while the organic matter adsorbed to biochar provided C energy and mineral nutrient requirements due to its porous nature. These observations agree with reports by (Warnock *et al*., 2007; Saito and Muramoto 2002). The addition of biochar to soil via microbial habitat provision (Peitikainen *et al*., 2000) induce an increased microbial biomass, nitrogen mineralization also increases, due to the increased microbial biomass and its intimate link to enzyme production and enhanced fertility of the soil, thus conferring reports by (Zama *et al*., 1999).

5.2 Effect ofHYT biofertilizers and biochar on plant nutrient composition

Plant nutrient analysis indicated an antagonistic association among P and N, K, Mg and Ca. Increased application of biochar, biofertilizers and inorganic fertilizer increased plant nutrient composition compared to the control. Findings of the study agrees with those of Abd -El – Moneim *et al*.,(1988), Hewedy (1999) and Tantawy, (2000) all working on tomato. Mohandas (1987), Gomaa (1989) Barakat and Gabr (1998), Ouda (2000) reported that applying bio fertilizer or inorganic fertilizer increased the N, P and K content of plants

5.3 Effect of HYT biofertilizers and biochar, application on plant growth

Significant increase in plant height as a consequence of biochar addition could have resulted from improved pH, EC and soil fertility leading to better nutrient absorption as reported by (Lehmann *et al*., 2003; Liang *et al*., 2006; and Solomon *et al*., 2007). Similar results were reported by Hoshi (2001) in tea trees where HYT biofertilizers were found to increase plant growth with the application of plant growth substances like indole acetic acid (IAA), gibberellic acid (GA) and cytokinins. Also they increased uptake of nutrients in the plants

leading to enhanced chlorophyll content and carbohydrate synthesis that led to the increase cell division and enlargement of the cell size which might have helped in increased plant height, stem girth and number of leaves. This significant increase in vegetative growth is in agreement with (Balasubramanian, 1989; Anburani and Manivannan, 2002; Prabhu *et al.*, 2003 and Wange and Kale, 2004). The increase in plant vegetative growth as a result of application of inorganic fertilizers alone or in combination with biochar could be attributed to increased uptake of nutrients in the plants leading to enhanced cell division and cell formation and hence in the height, stem girth and number of leaves increase. The outcome agrees with work done by Balaraj, (1999) in pepper, and Hoshi, (2001) in tea. The increase in height, stem girth and number of leaves obtained by the application of integrated nutrient management of biochar, bio fertilizer and inorganic fertilizers could be due to better nutrition associative symbiosis increased production of growth hormones like NAA, GA and cytokinins and improved nutrient availability and uptake through the sorptive capacity of biochar as indicted by studies conducted by (Major *et al*., 2009). Similar results were also noticed by Paramaguru and Natarajan (1993) in pepper, Ranganathan and Raniperumal (1995) in pepper, Deka *et al*., (1996) in chili, (Nanthakumar and Veeraragavathatham (2000), Anburani & Manivannan, (2002) and Wange and Kale (2004) all in egg plant.

The bigger canopy diameter observed in the biochar, bio fertilizer and inorganic fertilizers treated plots could be attributed to increased uptake of nutrients by the plants due to increased soil pH leading to enhanced carbohydrate synthesis which might have resulted in increased cell division and enlargement and therefore increased in plant canopy. The results herein agree with the findings of work done eggplant by (Prabhu *et al*., 2003;Wange and Kale, 2004).

Leaf chlorophyll content was highest in plants treated with biochar, bio fertilizer and inorganic fertilizers. This could be due to the relatively low uptake of K in treated plants

compared to the control. The low K uptake promoted the uptake of Mg by the plants, hence increasing the formation of chlorophyll molecules resulting in high chlorophyll content in plants. The result here is in agreement with the findings of Sharma (1995) who reported that, leaf chlorophyll content was higher when optimum doses of NPK was applied to the plants leading to the uptake of Mg which is the central molecule of chlorophyll. Also these findings agree with that of Sutpal and Saimbhi (2003) who reported N and P supply increased the uptake of Mg leading to the formation of more photosynthetic structures including chlorophyll.

Significant increase in growth could have resulted from improved pH, EC and soil fertility as credited in the studies of (Lehmann *et al*., 2003; Liang *et al*., 2006; Solomon *et al*., 2007). Increase in dry matter as a result of nutrient application is attributed to balanced nutrient uptake by plants which results in enhanced cell division and enlargement leading to shoot growth and development. It could be due to the production of plant growth substances like indole acetic acid (IAA), gibberellic acid (GA) and cytokinins and improving the availability and acquisition of nutrients which promoted the vegetative growth of treated plants. Gaur and Alagawadi (1987) and Fallik and Okon (1996) reported similar results with *Azospirillum brasilense* and phosphate solubilizing *Pseudomonas striata* or *Bacillus polymyxa* on rice and maize respectively. Iswaran *et al*., (1980) reported a 51% increase in biomass in soybean crops with biochar additions of 0.5t ha^{-1} while Lehmann *et al*., (2003) found biochar to increase rice biomass.

5.4 Effect of HYT biofertilizers, biochar and inorganic fertilizer application on yield attributes of okra

The increase in yield as a result from biochar addition could be due to the beneficial effects of biochar had on soil chemical and microbial properties which positively affected the uptake of available nutrients in the soil by the plants leading to the development of adequate

photosynthetic structures which increased the synthesis of carbohydrates and subsequent accumulation in the fruits leading to the high yields. Yield increase had been reported with biochar additions applied together with inorganic or organic fertilizer treatments by (Glaser *et al*.,(2002); Lehmann *et al*., (2002) Van Zwieten *et al*., (2007). Though integration of HYT biofertilizers with inorganic fertilizers gave higher yield than inorganic fertilizer alone or in addition to biochar, they were significantly lower than HYT applied alone or with biochar. This phenomenon could be attributed to excessive vegetative growth at the expense of reproductive growth hence reduction in yield complementing the findings of (Shahi *et al*., 2002).

Higher number of fruits has also been attributed to large plant canopy size and higher number of fruiting branches due to nutrient application which provided more space for flowering and subsequent higher number of fruits and or growth promoting substances like IAA and GA. These could enhance pollen germination and tube growth, which ultimately increase the fruit set. The higher fruit set may also be due to higher percentage of productive flowers. Balasubramanium *et al*., (1998), Anburani and Manivannan, (2002).Premsekhar and Rajashree (2009), reported similar results in tomato.

CHAPTER SIX

CONCLUSION AND RECOMMENDATION

The application of HYT and the use of biochar as soil amendment significantly promoted growth and increased yield in okra. The study also suggest that biochar additions to the soil increased the soil N, P, K, Ca and Mg, soil pH and soil microbial population, plant height and fresh shoot weight of okra. It also played a major role in lowering the EC of the soil to enhance better nutrient availability and absorption as well as serving as a liming agent which increased the pH from 4.2 to 5.99 which is ideal for of okra production, thus therefore increasing the yield of okra in all biochar treatment combinations. It also enhanced the efficiency of inorganic and HYT biofertilizers in the study. Biochar enhanced the efficiency of HYT by 7.73%.

HYT biofertilizers resulted in 62.28% increase in yield compared to inorganic fertilizers. A combination of HYT biofertilizers, biochar and inorganic fertilizers yielded 54.75% compared to inorganic fertilizer amended with biochar treatments. From the study, the following soil amendment combinations gave the highest yield, 7t biochar + 100% HYT (8.23); 7t biochar + 100% HYT (8.25), 3.5t biochar + 100% HYT (8.25); and (7.67), 3.5t biochar+ 50% HYT (7.50) and (7.45) respectively for major and minor seasons. It is therefore recommended that biochar at 7t ha^{-1} or 3.5t ha^{-1} + 50% and 100% HYT should be used or adopted to give better results for farmers. This recommendation corroborates with the fact that inorganic fertilizers are becoming too expensive to procure by small- scale farmers and also poses environmental hazards while rice husk which is abundant in rice--producing areas

could be obtained cheaply and exploited. Besides organic fertilizers like HYT biofertilizers have some secondary beneficial effect on the soil properties and hence environmentally friendly.

It can therefore be concluded that 7t biochar in combination with 100% and 50% HYT gave the best result and could prove cost effective when the application protocol is properly followed.

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