

# **Characterization, Conversion and Evaluation of Selected Lignocellulosic Biomass**

*by*

**ANUSHMA S.**

**(2012-11-111)**

**THESIS**

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**COLLEGE OF AGRICULTURE**

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**KERALA, INDIA**

**2014**

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I, hereby declare that this thesis entitled “**Characterization, Conversion and Evaluation of Selected Lignocellulosic Biomass**” is a bonafide record of research work done by me during the course of research and the thesis has not previously formed the basis for the award to me of any degree, diploma, associateship, fellowship or other similar title, of any other university or society.

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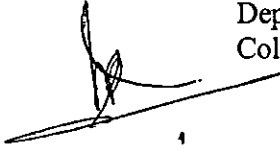
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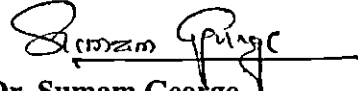
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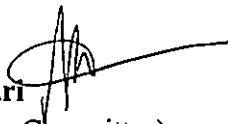
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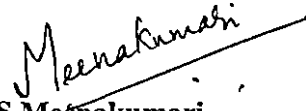
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## LIST OF ABBREVIATIONS

|                    |                                     |
|--------------------|-------------------------------------|
| %                  | Per cent                            |
| @                  | At the rate of                      |
| $\alpha$           | Alpha                               |
| $\mu\text{g}$      | Microgram                           |
| \$                 | Dollar                              |
| AAS                | Atomic Absorption Spectrophotometry |
| ADF                | Acid Detergent Fibre                |
| ADL                | Acid Detergent Lignin               |
| BAF                | Bio Accumulation Factor             |
| BCF                | Bio Concentration Factor            |
| $\beta$            | Beta                                |
| $^{\circ}\text{C}$ | Degrees Celsius                     |
| C                  | Carbon                              |
| CD                 | Critical difference                 |
| CEC                | Cation Exchange Capacity            |
| cm                 | Centimetre                          |
| CRD                | Completely Randomized Design        |
| DAP                | Di Ammonium Phosphate               |
| dS                 | Desi Siemen                         |
| EC                 | Electrical Conductivity             |
| FAO                | Food and Agriculture Organization   |
| <i>et al.</i>      | And others                          |
| Fig.               | Figure                              |

|                     |  |
|---------------------|--|
| FYM                 | Farm Yard Manure                               |
| g                   | Gram   |
| hr                  | Hour   |
| ha <sup>-1</sup>    | Per hectare                                    |
| <i>i.e.</i>         | That is  |
| IUCN                | International Union for Conservation of Nature |
| K                   | Potassium                                      |
| KAU                 | Kerala Agricultural University                 |
| kg                  | Kilogram                                       |
| M                   | Molar  |
| m                   | Metre  |
| Mg m <sup>-3</sup>  | Megagram per metre cube                        |
| mg                  | Milligram                                      |
| ml                  | Millilitre                                     |
| mg kg <sup>-1</sup> | Milligram per kilogram (ppm)                   |
| MOP                 | Muriate of Potash                              |
| MUB                 | Modified Universal Buffer                      |
| MSW                 | Municipal Solid Waste                          |
| N                   | Nitrogen                                       |
| NARP                | National Agriculture Research Project          |
| NDF                 | Neutral Detergent Fibre                        |
| nm                  | Nanometre                                      |
| P                   | Phosphorous                                    |
| plant <sup>-1</sup> | Per plant                                      |
| PNPP                | Para Nitro Phenol Phosphate                    |

|             |                                      |
|-------------|--------------------------------------|
| POP         | Package of Practices                 |
| ppm         | Parts per million                    |
| S           | Sulphur                              |
| SOM         | Soil Organic Matter                  |
| s           | Seconds                              |
| t           | Tonnes                               |
| TPF         | Triphenyl Formazone                  |
| TTC         | Triphenyl tetrazolium chloride       |
| UNEP        | United Nations Environment Programme |
| USA         | United States of America             |
| WHO         | World Health Organization            |
| <i>viz.</i> | Namely                               |

# *Introduction*

## 1. INTRODUCTION

The most widely available and also the most wasted energy source are a wide variety of agricultural wastes. Most of the farm wastes including weed plants are of biological origin and thus are easily decomposable by microorganisms and can be exploited for the production of biofuels and manures. Apart from the direct wastes from farm, weed plants can also be included in farm wastes. Wetland plants like water cabbage (*Limnocharis flava* L. Buchenau) and water hyacinth (*Eichhornia crassipes* (C. Martius) Solms-Laub) have now emerged as very devastating weed plants in the wet land ecosystem especially in Kerala. The first goal of any waste management system is to maximize the economic benefits from the waste resources and maintain acceptable environmental standards. The spread of invasive alien plant species is neither easy to manage nor easy to reverse, threatening not only biodiversity but also to economic development and human wellbeing (UNEP, 2012). These troublesome weeds can be effectively utilized for production of quality manure.

Water cabbage (*L. flava*) is considered as a major weed in many countries. This perennial aquatic plant colonizes shallow wetlands and margins of deeper waterways. It is an emergent aquatic weed in the Limnocharitaceae family, which has invaded in the flood plains of Kuttanad wetland system and other low lying areas of Kerala. Water cabbage has become a serious weed in rice fields, irrigation canals and wetlands in South-East Asia (Waterhouse, 2003). Clumps of the weed provide congenial breeding sites for disease-vectors, including mosquitoes, which encourage the spread of diseases such as Japan fever and dengue fever on human (Abhilash, 2004) and other pest and diseases of crop plants.

Another major wet land weed water hyacinth (*E. crassipes*), a native of Amazon Basin in South America, has emerged as a major weed in more than fifty countries in the tropical and subtropical regions of the world with profuse and permanent impacts (Patel, 2012). Water hyacinth has been identified by the

International Union for Conservation of Nature (IUCN) as one of the hundred most aggressive invasive species and recognized as one of the top ten worst weeds in the world. Efficient in utilizing aquatic nutrients and solar energy for profuse biomass production, water hyacinth can cause extensive environmental, social and economic problems. They cause substantial economic losses to the tune of total \$120 billion annually in US (Kettunen *et al.*, 2009). It usually found in lakes, estuaries, wetlands, marshes, ponds, dams, slow flowing rivers, streams and waterways in the lower latitudes where growth is stimulated by the inflow of nutrient rich water from urban and agricultural runoff, deforestation, products of industrial waste and insufficient wastewater treatment (Ndimele *et al.*, 2011). Decomposed water hyacinth can be used as a green manure or as compost to improve the poor quality of soils.

In Kerala, coir pith is another important agricultural waste generated from coir fibre extraction industries. On an average  $3 \times 10^{11}$  metric ton with an annual biosynthetic rate of  $2 \times 10^{10}$  metric ton are produced in the planet and constitutes second most abundant group of biopolymer in biosphere. High content of lignin (30 per cent) and cellulose (26 per cent) in coir pith causes very slow decomposition. Degradation of coir pith can be effectively done with suitable species of Basidiomycetes fungi (*P. sajor-caju*) and in combination with nitrogen fixing bacteria. Coir waste after biodegradation can effectively used as manure for increasing the yield of crops.

Banana is one of the important fruit crops grown almost in every state of India (7.1 lakh ha). Apart from fruit, it generates huge quantity of biomass as waste in the form of pseudostem, leaves, suckers etc. Of these, on an average about 60 to 80 tons  $\text{ha}^{-1}$  is pseudostem alone. Presently the disposal of pseudostem is being carried out in routine ways *i.e.* dumping on field bunds, burning etc. causing environmental problems. A baseline survey conducted in Gujarat during 2008-09 revealed that 33 per cent of the growers are either composting or chopping and incorporating the pseudostems into soil while rest of the farmers are disposing it on field bunds .



The total amount of crop residues generated in India is estimated as  $350 \times 10^6 \text{ kg yr}^{-1}$ . Management of this voluminous residue is a major challenge and the farmers generally get rid of this waste by burning it in the field itself. The burning results in huge losses of N (up to 80 per cent), P (25 per cent), K (21 per cent) and S (4- 60 per cent) besides polluting the air, thereby depriving the soils of its organic matter. Therefore there is a need to explore some eco-friendly, low cost, easily adoptable residue management strategies that can replenish the soil of its nutrients. Agriculture waste recycling can bring tremendous benefits to agriculture and land management in long run. In addition, there are the benefits of a cleaner environment, a healthier habitat and an intelligent use of all available recyclable resources without condemning them as wastes.

Composting is by definition the solid-phase biological decomposition of organic residues that occurs in aerobic condition by exploiting substrate self heating as a consequence of microbial oxidative reactions. This process leads to the production of compost, a humus-like, dark, crumbly material that can be used as fertilizer to reintegrate organic matter in agricultural soils. In nature's laboratory, there are a number of organisms (micro and macro) that have the ability to convert organic waste into valuable resources containing plant nutrients and organic matter, which are critical for maintaining soil productivity.

In this context the present study is envisaged with the following objectives.

- Characterization of the lignocellulosic biomass from selected plant sources
- Assessment of various microbial and enzymatic sources for degrading the lignocellulosic biomass into compost.
- Evaluation of manurial value of the resultant composts.

# *Review of Literature*

## 2. REVIEW OF LITERATURE

India with its varied agro climatic zones is amenable to grow a wide variety of food crops as well as horticultural crops. These crops form a significant part of the total agricultural produce in the country. From the agriculture sector, a large quantity of waste is produced as crop residues, post harvest loss, weed plants etc. Management of this voluminous residue is a major challenge. Hence the present study was designed with the objective of managing wastes by characterization of lignocellulosic biomass. In this chapter, an attempt was made to review the previous related works under various subheads.

### 2.1. COMPOSTING

Composting is a thermogenic, solid state fermentation process, carried out by a succession of microbial populations beginning with mesophilic bacteria, actinomycetes and fungi followed by thermophiles and ending again with mesophiles (Johri *et al.*, 1999). Composting process creates stable, soil enriching humus and concentrates the N, P, K, Ca and Mg contents (Eneji *et al.*, 2001).

Many alternatives for the disposal of the organic wastes have been proposed, composting being one of the most attractive on account of its low environmental impact and cost as well as its capacity for generating a product valuable for increasing soil fertility (Pascual *et al.*, 2002) or as a growing medium for horticultural purposes (Bustamante *et al.*, 2008). The main advantages of using compost as growing media are their high content of organic matter and nutrients, their pathogen free nature and suppressive effect against phytopathogens (Entry *et al.*, 2005). The addition of mature compost to soil favours plant development and improves soil quality, as well as having a suppressive effect on many diseases caused by soil borne plant pathogens (Cotxarrera *et al.*, 2002; Erhart *et al.*, 1999).

Composts are good organic manures which help to maintain and enhance the fertility and productivity of agricultural soils, there by promoting

sustainability. Composting is the transformation of raw organic materials into biologically stable, humic substances suitable for a variety of soils and plant uses (Brown and Subler, 2007).

Essentially, composting is controlled decomposition; the natural breakdown process that occurs when organic residue comes in contact with the soil. It is an ancient technology and there are roman and biblical references to composting and numerous accounts of composting practices in subsequent millennia (Ros *et al.*, 2006).

Aerobic composting involves a process of biological decomposition and stabilization of organic substrates under conditions that allow multiplication and activity of thermophilic microorganisms as a result of biologically produced heat, to produce a final product that is stable, free of pathogens, pests and plant seeds, useful in agriculture and forestry as manure (Balasundaran, 1999; Saravanan *et al.*, 2003). High temperature within waste heap undergoing composting has been considered as consequence of microbial activity, whereby heat is liberated through respiration of microbes and built up within the pile (Tiquia and Tam, 2000).

## 2.2. SUBSTRATES

### 2.2.1. Water Cabbage

Water cabbage (*L. flava*) is considered to be a major weed in many countries. This perennial aquatic plant colonises shallow wetlands and margins of deeper waterways. It can quickly grow to dominate native aquatic plants and affects the ecology of stream. It hinders agricultural production by infesting irrigation channels, drainage ditches and rice paddies. These rice paddies are rendered useless and are often abandoned as a result.

Water cabbage (*L. flava*) is an emergent plant in rice cultivated areas that may become noxious over the growing areas (Karim *et al.*, 2004). In west Java and Thailand, it is commonly cultivated as a food crop in fertile soil and harvested after 2-3 months before being marketed (Maisuthisakul *et al.*, 2008).

Study by Thien (2005), Amalina (2006) and Nihla (2006) showed that the amount of Fe uptake was higher compared to Mn in the *L. flava* plant tissues. The  $\text{Fe}^{2+}$  was a micronutrient for the plants required in higher concentration than  $\text{Mn}^{2+}$  (Kamal *et al.*, 2004).

Saupi *et al.* (2009) reported a relatively high moisture content of 79.34 percent when compared to ash (0.79 per cent), crude fat (1.22 per cent), crude fibre (3.81 per cent) and total carbohydrate (14.56 per cent) in *L. flava*.

Abhilash *et al.* (2009) investigated the potential of *L. flava* grown for phytofiltration of Cd in polluted water with low concentrations of Cd in a hydroponic experiment. They spiked 45 days old seedlings of *L. flava* with different concentrations of Cd (0.5, 1, 2 and 4  $\text{mg l}^{-1}$ ) and found that the Cd content was highest in the roots followed by leaves and peduncle. This suggested that *L. flava* was a suitable species for phytofiltration of low concentrations of Cd in water.

The presence of nutrients ( $\text{NH}_3\text{-N}$  and  $\text{PO}_4\text{-P}$ ) and heavy metals (Fe and Mn) in the *L. flava*'s tissues have proved the ability of this plant to uptake pollutants, where the highest accumulation was obtained in the root tissue for both nutrients and heavy metals. It has been successfully proved to play an important role in expediting the treatment process through various mechanisms such as phytoextraction, phytoaccumulation and rhizofiltration (Kamarudzaman *et al.*, 2011).

*L. flava* is a potential plant to remediate water polluted by Pb. The bioaccumulation factor (BAF) is more than one with the translocation factor less than one indicated that it is tolerant to Pb metals. The highest Pb concentration in the plant was in the roots, then followed leaves, and stem. The concentration of Pb in leaves tissues of *L. flava*, exposed to 10  $\text{mg l}^{-1}$  Pb was 1.90  $\text{mg kg}^{-1}$  which is lower than the criteria given by FAO and WHO (5  $\text{mg kg}^{-1}$ ) as reported by Rachmadiarti *et al.* (2012).

According to Anning *et al.* (2013) high rate of hyper accumulation of heavy metals by *L. flava*, was reported. Additionally, the species tolerated phytotoxic levels of Fe, Hg, and Pb, as no signs of wilting were found during the study.

### 2.2.2. Coir Pith

Coir pith has been widely utilized in agriculture due to its physical properties such as high porosity and water holding capacity rendering it suitable for employing as planting materials (Ravindranath, 1991).

Coirpith is known as organic waste composed of lignocellulosic fibrous material, separated from the husk of the coconut fruit. It is abundantly available as an agricultural waste from the local coir industry. In India, 7.5 million tonnes of coirpith are produced every year. Disposal of accumulated coirpith is a major problem as coirpith gets decomposed very slowly in the natural environment because of its chemical and structural complexity (Ramalingam *et al.*, 2004).

Fuangworawong *et al.* (2008) found that several months were necessary in order to obtain matured coir pith compost even though aeration was provided. Naturally, coirpith degradation is very slow because of the chemical and structural complexity of its lignin–cellulose complex (Ramalingam *et al.*, 2004).

Tripetchkul *et al.* (2012) reported that initial moisture content of coir pith as 6.35 per cent, pH 5.68, ash content 47.11 per cent, organic matter 52.89 per cent, total C 29.38 per cent, total N 0.44 per cent and C:N ratio of 66.13.

Abbiramy and Ross (2012) reported that lignin, cellulose, organic carbon and NPK content of coir pith were 37 per cent, 35 percent, 28.30 per cent, 0.68 per cent, 0.27 percent, 0.04 per cent respectively. Lignin content was decreased from 37 per cent to 18.3 per cent in coir pith treated with earthworms while cellulose content decreased from 35 per cent to 2.76 per cent.

Muthurayar and Dhanarajann (2013) reported that raw coir pith recorded P, K, cellulose and lignin contents of 0.02, 0.30, 35.7 and 54.3 per cent respectively. The pH was found to be acidic (5.4) and C: N ratio was found to be high (162:1).

Prabha *et al.* (2013) reported that the percent of N in raw coir pith was about 0.34 per cent which was very low and as composting proceeds the percentage of N also increased.

Reghuvaran and Ravindranath (2014) reported that the lignin content of the raw coir pith was reduced from 32 per cent to 17 per cent when treated with the consortium of *P. sajor-caju*, *Azotobacter vinelandii* and *Azospirillum brasilense*. They also reported that the maximum carbohydrate, protein and chlorophyll and plant growth (leaf number, shoot length and root length), were achieved in plants grown in soil and composted coir pith.

Kadalli *et al.* (2000) observed highest N and P contents in coir pith, composted with *P. sajor-caju*, cow dung, garden weeds, sun-hemp, rock phosphate and micronutrients. After 12 weeks of composting period there was a considerable increase in the N, P, and K contents. They also reported that the N content of the composts ranged from 1.02 to 1.38 per cent and the organic carbon content ranged from 30.24 to 43.76 per cent and C:N ratio has been drastically reduced to 21.91 due to increase in N content and loss of carbon as CO<sub>2</sub>.

It is also reported that when the coir pith was treated with the mushroom *P. sajor-caju*, the nutrient content of coir pith such as N, P and K showed the variation and it got increased (Crawford, 1978).

### **2.2.3. Water Hyacinth**

Use of water hyacinth as a feedstock for compost has been reported as early as 1925 (Gopal, 1987). It was made using earth, cow manure, and wood ash between layers of fresh water hyacinth which was then covered with earth.

The water hyacinth, (*E. crassipes*) is considered to be one of the most invasive plant species worldwide (Gopal, 1987). It is a native of the Amazon River, most likely from Brazil (Penfound and Earle, 1948). It became a nationwide aquatic weed problem during the last century after its introduction to the United States in 1884 at the Centennial Exposition in New Orleans (Gopal, 1987).

Carina and Cecilia (2007) reported that water hyacinths can be rich in N, up to 3.2 per cent of dry matter and have a C/N ratio around 15 and it can be used as a substrate for compost or biogas production.

It has effectively resisted all attempts of eradicating it by chemical, biological, mechanical or hybrid means (Rai, 2009; Chauhan and Joshi, 2010).

Montoya (2010) reported that large scale composting is an effective means of managing water hyacinth by rendering the seeds and other propagules non-viable. Water hyacinth has been used in phytoremediation because it has exceptionally high affinity and accumulation capacity for several metals (Malik, 2007; Chunkao *et al.*, 2012). The accumulation of metals in the water hyacinth occurs by intracellular component through energy dependent process and passive adsorption onto the body surface. The body of water hyacinth contains many polyfunctional metal binding sites for both cationic and anionic metal complexes (Mahamadi, 2011).

The mobility of trace metals, their bioavailability and related eco-toxicity to plants depend strongly on their specific chemical forms or ways of binding rather than total metal concentration (Fuentes *et al.*, 2006; Gupta and Sinha, 2007). In order to screen the suitability of a material, such as water hyacinth for land application, not only depends on their total content in a matrix, but also their bioavailability and their capacity for remobilization (Peruzzi *et al.*, 2011).

The bioavailability of metals in soil is a self - motivated process that depends on explicit combinations of chemical, biological and environmental



parameters. These include soil properties such as pH, organic matter content, redox potential, cation exchange capacity, sulphate, carbonate, hydroxide, soil texture and clay content (Prabpai *et al.*, 2009; Guala *et al.*, 2010).

The pH and organic matter content are the major critical factors for heavy metal accumulation by plants (Li *et al.*, 2010). Gupta *et al.* (2007) analyzed the physico-chemical characteristics of fresh water hyacinth plant and the results showed that the fresh plant contained moisture of 92.8 per cent, ash content 417 g kg<sup>-1</sup>, pH 8.1, total organic carbon 338 g kg<sup>-1</sup>, total N 9.5 g kg<sup>-1</sup>, C:N ratio 36:1, total P 9.7 g kg<sup>-1</sup>, K 5.4 g kg<sup>-1</sup>, total Fe 1640 mg kg<sup>-1</sup>, total Cu 312 mg kg<sup>-1</sup>, total Cd 1.36 mg kg<sup>-1</sup>, total Cr 41.18 mg kg<sup>-1</sup>, total Pb 67 mg kg<sup>-1</sup> and total Zn 640 mg kg<sup>-1</sup>.

According to Nyananyo *et al.* (2007) water hyacinth besides being a nuisance in public water bodies had high protein and total organic matter content, which made a potential raw material for the production of animal feed and low cost alternative source of organic fertilizers.

Kafle *et al.* (2009) found that the compost obtained from water hyacinth had acceptable composition of 1.78 per cent N, 0.93 per cent P, 0.75 per cent K, pH 8.4 and could be used in agricultural land for crop production.

Elserafy *et al.* (2012) reported that water hyacinth seems to be a good source of organic carbon and may be used as a compost to meet the great demand for organic manure.

Singh and Kalamdhad (2013) reported that the total concentration of metals like Zn, Cu, Mn, Fe, Ni, Pb, Cd and Cr are increasing during the period of composting. The order of total metal content in the composted water hyacinth was Fe > Mn > Pb > Ni > Zn > Cu > Cr > Cd. These heavy metals were concentrated during the composting process, due to weight loss in the course of composting following organic matter decomposition, release of CO<sub>2</sub> and water and mineralization processes (Zorpas *et al.*, 2000).

Studies on crops grown with water hyacinth compost have produced some interesting results. In Sudan, increase in yield has been reported in carrots, red beans, and onions, but the same study also reported a decrease in the yield of okra (Philipp *et al.*, 1983; Gopal, 1987). The losses in yields were attributed to the relatively high KCl content of water hyacinth compost (Gopal, 1987).

The pot containing soil amended with water hyacinth compost was reported to achieve significantly better height, larger number of leaves, more favourable shoot- root ratio, and greater biomass per unit time and larger length of inflorescence in *Crossandra undulaefolia*. In terms of root length, quicker on set of flowering and harvest index the treated plants performed better than the controls (Gajalakshmi and Abbasi, 2002).

The water hyacinth can be used on the land either as surface mulch (Woomer *et al.*, 2000) or as compost. Mulching field crops with water hyacinth was found to increase the production of lady's finger (67 per cent), potato (14 per cent) and tomato (90 per cent) as compared to control (no mulching) (Sannigrahi *et al.*, 2002).

#### **2.2.4 Banana Pseudostem**

The main residual wastes of the banana crop are leaves and pseudostems, both containing high levels of lignocelluloses (Reddy, 2001). These lignocelluloses materials are efficient substrates for white-rot fungi, which produces lignolytic and cellulolytic enzymes that have numerous applications in industrial processes for food, drug, textile and dye use (Robinson *et al.*, 2001; Pointing, 2001).

According to Reddy *et al.* (2003) production of lignolytic and cellulolytic enzymes on pseudostems of banana waste by *P. sajor-caju* caused 52 per cent reduction of lignin occurred in banana leaves while 23 per cent reduction in pseudostem when compared to cellulose contents (4 per cent reduction in banana leaves; 1.5 per cent in pseudostem). He also reported that the banana waste can be utilized for the production of lignolytic and cellulolytic enzymes by solid

substrate fermentation using two *Pleurotus* species (*P. ostreatus* and *P. sajor-caju*).

In general, banana pseudostem is an abundant natural resource in subtropical and tropical regions and has potential for providing profitable products such as feed (Ullola *et al.*, 2004), and manures (Ultra *et al.*, 2005) which call for practical techniques and processes to exploit this natural resources.

Banana pseudostem can be recycled to be used as bio fertilizer (Phirke *et al.*, 2001). It contains good amount of cellulose and starch and can be used as cattle feed (Katongole *et al.*, 2008). Outer covering of pseudostem is mostly cellulosic material while core or pith is rich in polysaccharides and other trace elements, but lower in lignin content (Cordeiro *et al.*, 2004).

A study conducted by Shah *et al.* (2005) revealed that banana waste can be used as an alternative substrate to other agricultural/agro-industrial waste, wheat bran/straw, sawdust and bagasse, which are already in use for the production of ligno and cellulolytic enzyme production using two lignocellulolytic fungi (*Phylosticta spp.* MPS-001 and *Aspergillus spp.* MPS-002). Laccase activity was high in relation to cellulase and xylanase during the initial stage of degradation. Laccase activities of *P.ostreatus* and *P. sajor-caju* began to decrease before 20<sup>th</sup> day on leaf and pseudostem biomass. Growth of *P. ostreatus* and *P. sajor-caju* on banana waste by utilizing the lignin contents leading to their greater percentage reduction (Ahmad, 2010).

In a study conducted by Lekshmi (2011), banana pseudostem was utilized as a substrate for the production of enriched composts and observed significant influence on yield and yield attributes of the crops as well as improvement in the soil physico-chemical and biological properties.

### 2.3. COMPONENTS OF BIOMASS

Lignocellulose is the major component of biomass, comprising around half of the plant matter produced by photosynthesis (also called photomass) and representing the most abundant renewable organic resource in soil. It

consists of three types of polymers *viz.* cellulose, hemicellulose and lignin that are strongly intermeshed and chemically bonded by non-covalent forces and by covalent cross-linkages (Perez *et al.*, 2002). Cellulosic biomass, sometimes called lingocellulosic biomass, is a heterogeneous complex of carbohydrate polymers and lignin, a complex polymer of phenyl propanoid units (Wright, 1988). Only a small amount of the cellulose, hemicellulose and lignin produced as by-products in agriculture or forestry is used, the rest being considered waste.

The enzymatic machinery for degrading cellulose, hemicellulose and lignin is possessed only by microorganisms (Perez *et al.*, 2002). Many microorganisms are capable of degrading and utilizing cellulose and hemicelluloses as carbon and energy sources. However, a much smaller group of filamentous fungi has evolved with the ability to break down lignin, the most recalcitrant component of plant cell walls. These are known as white-rot fungi, which possess the unique ability of efficiently degrading lignin to CO<sub>2</sub> (Blanchette, 1995). Other lignocellulose degrading fungi are brown-rot fungi that rapidly depolymerize cellulosic materials while only modifying lignin (Eriksson *et al.*, 1990).

There are many genera of actinomycetes and eubacteria which can degrade extracted lignin (Buswell and Odier, 1987). Many bacterial strains, especially actinomycetes, can solubilize and modify the lignin structure extensively, but their ability to mineralize lignin is limited (Buswell and Odier, 1987; Ball *et al.*, 1989; Eriksson *et al.*, 1990; Godden *et al.*, 1992).

### 2.2.1. Cellulose

Cellulose, an insoluble polymer consisting of  $\beta$ -(1-4) linked glucose residues has been the subject of intense research for more than a century, and new insights into a better understanding of its molecular architecture continue to emerge (Somerville *et al.*, 2004). It is well known that native cellulose molecules are found in fibril form, and its molecular architecture has a high degree of

individuality, depending on its source (cell wall layer or plant type) (Ding and Himmel, 2006).

The visually dominant structural features of cellulose in higher plants are cellulose microfibrils with diameter of 2-10 nm, cross-linked by other cell wall components such as xyloglucans (Somerville *et al.*, 2004). Microfibrils are unbranched fibrils composed of approximately 30-36 glucan chains aggregated laterally by means of hydrogen bonding and van der Waals forces to produce crystalline structures (Ding and Himmel, 2006).

Krassing (1993) has shown that a higher degree of fibrillar aggregation produces a more compact fibre structure, with fewer, smaller interstices resulting in a smaller internal accessible surface area. An important feature of the highly ordered regions is that the cellulose chains are packed so tightly that even small molecules such as water cannot penetrate these highly organized structural entities. The limited accessibility to these regions leads to alteration of their reactivity to swelling and reactive agents such as cellulases. With this type of structure, it is apparent that only the cellulose molecules situated on the surface of these aggregations would be susceptible to the degrading actions of enzymes.

Coughlan (1985) coined the term 'amorphogenesis' to suggest a possible mechanism by which the dispersion, swelling or delamination of cellulosic substrate occurred, resulting in a reduction in the degree of fibrillar aggregation and/or crystallinity, and the creation of a larger accessible surface by increasing the reactive internal surface.

If cellulose hydrolysis only occurs on the surface of the cellulose aggregations, the available surface area is a potential determinant of the maximum rate of hydrolysis that can be achieved. It has been proposed that the tightly packed cellulose regions are a major factor in contributing to the resistance of cellulose to degradation, by limiting the accessibility to cellulases (Wood *et al.*, 1989).

Once the cellulose network is accessible to the enzymes, the synergistic action of endo and exo-glucanases promotes the fragmentation of accessible molecules to soluble cello-oligosaccharides (cellulosic molecules with a degree of polymerization of less than 6 units) which are quickly hydrolyzed, mostly to cellobiose (Mansfield *et al.*, 1999). In most commercial cellulase systems, an extraneous source of  $\beta$ -glucosidase is usually added to completely hydrolyze the cellobiose to glucose enhancing the overall reaction by minimizing end product inhibition.

There are several microorganisms especially fungi were reported as the efficient cellulose degrading microorganisms with the ability to produce significant quantity of cellulase enzymes. *T. reesei* is one of the most widely used species of filamentous fungi for the production of cellulolytic enzymes (Peterson and Nevalainen, 2012)

Sufficient  $\beta$ -glucosidase activity is needed for the complete and efficient cellulose hydrolysis (Reczey *et al.*, 1998; Lynd *et al.*, 2002). The  $\beta$ -glucosidases are widely produced by different genera and species of the fungal kingdom including Ascomycetes and Basidiomycetes, where especially the ascomycete genus *Aspergillus* has been widely studied for  $\beta$ -glucosidase production. *A. niger* has been setting the standard in commercial  $\beta$ -glucosidase production (Dekker, 1986), but within the last few years more research papers have been published on efficient  $\beta$ -glucosidases from other *Aspergillus* species and *Penicillium* genus (Sorensen *et al.*, 2012; Liu *et al.*, 2012).

### 2.3.2. Hemicellulose

Hemicelluloses, the second major plant constituents and sources of energy and nutrients for soil microflora, are water-soluble polysaccharides and consist of hexoses, pentoses, and uronic acids. Xylan is the most abundant component of hemicellulose contributing over 70 per cent of its structure (Reczey *et al.*, 1998).

Hemicellulose hydrogen-bonds to cellulose microfibrils, thus forming a network that provides the structural backbone to plant cell wall. All types of

cellulose microfibrils are composed of linearly linked (1,4)-D glucopyranose units and differ only by the degree of polymerization (Ho *et al.*, 1998). The remaining polysaccharides are known collectively as hemicelluloses and exhibit species related composition. These amorphous, complex heteropolymers characterized by a branched molecular structure exhibit a lower degree of polymerization than cellulose and lower in molecular weight. Cellulose and hemicellulose are potential sources of fermentable sugars (Hinman *et al.*, 1989; Taherzadeh *et al.*, 1999; Sreenath and Jeffries, 2000).

Xylanases can hydrolyze  $\beta$ -1, 4 linkages in xylan, the most abundant component of hemicellulose and produce oligomers which can be further hydrolyzed into xylose by  $\beta$ -xylosidase. The enzymes such as  $\beta$ -mannanases, arabino furanosidases or  $\alpha$ -L-arabinanases can hydrolyse mannan-based or arabino furanosyl-based hemicelluloses. The xylose, arabinose, galactose and mannose are further converted to organic acids, alcohols, CO<sub>2</sub> and H<sub>2</sub>O. Uronic acids are broken down to pentoses and CO<sub>2</sub> (Reczey *et al.*, 1998).

### 2.2.3. Lignin

The amounts of the carbohydrate polymers and lignin depend on the type of material. Wyman (1996) compiled the compositions of lignocelluloses from different hardwoods, softwoods, and agricultural residues. The hardwoods such as white birch, aspen, red maple, eucalyptus, populus, and oak contain 39-54 per cent cellulose, 14-37 per cent hemicellulose, and 17-30 per cent lignin. The corresponding values for softwoods (*eg*: pines and firs) are 41-50 per cent cellulose, 11-27 per cent hemicelluloses and 20- 30 per cent lignin. The composition of different agricultural residues varies widely. For instance, rice straw consists of 32-47 per cent cellulose, 19-27 per cent hemicellulose, and 5- 24 per cent lignin.

In biomass, polysaccharides are 'protected' in the cell walls by lignin. Lignin provides plant tissue and individual fibres with compressive strength and

stiffens the cell wall of the fibres to protect the carbohydrates from chemical and physical damage (Saheb and Jog, 1999).

Lignins act as binding agents for the cellulose and hemicellulose fibres through a variety of linkages involving ether and C-C bonds of aromatic rings and propyl side chains (Agarwal, 2006).

The presence of lignin in some cell walls imparts further strength, and provides resistance against pests and diseases. The presence of lignin in the cell wall, however, impedes enzymatic hydrolysis of the carbohydrates. The chemical characterization of *Ampelocissus cavicaulis* fibre by Agu *et al.* (2012) revealed 38.66 per cent cellulose, 33.21 per cent lignin and 1.47 per cent acid soluble lignin. He also reported the fibre contains 2.88 per cent ash and 5.60 per cent moisture. The hemicellulose content was found to be 25.90 per cent and the cellulose to lignin ratio was 1.16.

Natural biomass decay is carried out by microbial communities in which polysaccharides are consumed by microbes as a food source; lignin is transferred from plants to soil as carbon storage. As the structure of lignin is complex and variable, only a few microorganisms, including bacteria such as *Streptomyces* spp. and *Nocardia* spp. and basidiomycetes (brown-rot and white-rot fungi, respectively), are able to degrade lignin relying on the oxidative action of unspecific and extracellular enzymes, such as lignin peroxidase, manganese peroxidase, and laccase (Ding *et al.*, 2012)

Dry plants in general comprise 40-50 per cent cellulose, 15-25 per cent hemicelluloses, 5-10 per cent other components (ash, minerals, etc.), and 20-25 per cent lignin (Faik, 2013). Lignin is the second most abundant biopolymer besides cellulose, consisting primarily of three units: guaiacyl (G), sinapyl (S), and p-hydroxyphenyl (H) units linked by aryl ether or C-C bonds. It is an irregular, high molecular weight polymer formed by enzyme-initiated, free-radical polymerization of coniferyl alcohol (in hardwoods), coniferyl plus sinapyl



alcohols (in softwoods), or coumaryl alcohol plus both above mentioned alcohols (in grasses).

The degradation of lignocellulosic material is affected by the existence of aerobic mesophilic and thermophilic bacteria which are capable of producing lignocelluloses degrading enzymes (Kala *et al.*, 2009; Nakasaki *et al.*, 2009). Zainudin *et al.* (2014) reported that during the composting of oilpalm empty bunch and palm oil mill effluent anaerobic sludge, the initial composition of cellulose, hemicelluloses, and lignin were about 52 per cent, 26 per cent, and 19 per cent respectively, and the temperature was around 34°C. The initial amount of cellulose at first day was higher than the amount of cellulose for the rest of the days during the composting process. They also reported that the reduction of cellulose and hemicellulose components within the 20 days of composting was due to the presence of thermophilic bacterial groups capable of degrading lignocellulosic material.

#### 2.4. COMPOSTING OF FARM WASTES

Composting is a spontaneous, biological decomposition process of organic materials in a predominantly aerobic environment. During the process of composting bacteria, fungi and other microorganisms, including micro arthropods, break down organic materials to stable, usable organic substances called compost. It is a useful way of transforming organic waste into valuable organic matter for use as an organic amendment for soils (Gajdos, 1992).

The bioconversion process is gradually emerging as a natural, promising, environment friendly and potential microbial process to degrade environmental contaminants (Colwell, 1994). It is seen as an environmentally acceptable method of waste management which uses naturally occurring microorganisms, to convert biodegradable organic matter into humus like product. The process destroys pathogens, converts N from unstable ammonia to stable organic forms, reduces the volume of waste and improves the nature of the waste (Georgacakis *et al.*, 1996; Sequi, 1996).

Samarta and Patro (1996) established that organic farming was the backbone of sustainable agriculture, improved the soil health and the crops grown in rich organic manure, resist pest and disease attack. It was an ecologically sound and sustainable way of growing more food.

Lekshmi (2011) developed an organic nutrition schedule for improving the soil health and productivity of chilly (*Capiscicum annum* L.). According to her, soil physico-chemical and biological properties were improved by the application of various composts prepared from farmwastes viz. banana pseudostem and dried leaves. Various natural additives viz. rockdust, biological agents (*Trichoderma* spp., consortium of effective microorganisms, vermiwash, panchagavya etc.) were used for the enrichment which increased the productivity and fruit quality was also found to be superior to the conventional methods.

According to Keener *et al.*(2000), the composting process composed of three steps (i) an initial mesophilic phase which lasted for 1-3 days, where mesophilic bacteria and fungi degraded simple compounds such as sugars, amino acids, proteins etc. and led to an increase in temperature (ii) thermophilic phase, where thermophilic microorganisms degraded fats, cellulose, hemicelluloses and some lignin, during this phase the maximum degradation of the organic matter occurred together with the destruction of pathogens (iii) cooling phase, characterized by decreased temperature due to the reduction of the microbial activity associated with the depletion of degradable organic substrates, the composting mass was recolonised by mesophilic microorganisms which were able to degrade the remaining sugars, cellulose and hemicellulose.

The volume of the finished compost is 50 per cent or less of the volume of raw material (Rynk *et al.*, 1992). This makes composting an effective means of waste control (Stoffella and Kahn, 2001).

#### 2.4.1. Role of Compost in Soil Health

The use of organic manures as amendments to improve soil organic matter level and long term soil fertility and productivity is gaining importance. The benefits of composted organic wastes to soil structure, fertility as well as plant growth have been increasingly emphasized (Esse *et al.*, 2001).

Preventing the loss of top soil by directly adding compost to the soil surface also makes compost an effective means of erosion control (Stoffella and Kahn, 2001). Compost has also been used in revegetation projects and mine reclamation as a topsoil and soil amendment for disturbed landscapes (Rynk *et al.*, 1992). Currently, the highest demand for compost lies in the horticultural industry where it is primarily used in landscaping and in the greenhouse (Stoffella and Kahn, 2001).

In addition, compost is utilized by horticulturalists in vegetable, fruit, ornamental, nursery, and turf crop production systems. Compost has also been found to reduce plant diseases, help with weed control, and increase the accessibility of nutrients by plants (Stoffella and Kahn 2001). In Texas, the potential industry opportunity is very high for compost, as it used by the Texas Department of Transportation for erosion control on highway right-of-ways and during construction of highways (Pearson 2003).

Application of organic waste with high organic matter content has been reported to improve soil fertility (Tejeda *et al.*, 2009). Application of organic manure and farm yard manure improved the soil physico-chemical properties such as pH, soil moisture, organic carbon and nutrient status of the soil (Verma *et al.*, 2009). According to Singh and Rao (2009) organic manures are nature's best mulches and soil amendments which improve soil structure, aeration and also increase the water holding capacity of the soil. Application of water hyacinth compost as an organic source enhanced the uptake of N by groundnut, maize and barley (Rabie *et al.*, 1995).

#### 2.4.2. Role of Bio Inoculants in Composting

Fungi are known agents of decomposition of organic matter especially cellulosic substrates. Being efficient consumers of carbon, fungi build up much higher biomass than other microorganisms. The most commonly observed species of cellulolytic fungi in composting materials are *Aspergillus*, *Penicillium*, *Rhizopus*, *Fusarium*, *Chaetomium*, *Trichoderma*, *Alternaria*, and *Cladosporium* (Ashraf *et al.*, 2007). Some of the species of *Paecilomyces* and *Sporotrichum* have also been named as efficient degraders of lignocellulosic waste (Kapoor *et al.*, 1978). Fungal species were found to be numerous during both mesophilic and thermophilic phases of composting. Their importance along with actinomycetes has been reported in composting, especially during the late curing stage (Finstein and Morris, 1975).

Actinomycetes have an important role in carbon cycle because they are well adapted to the penetration and degradation of organics such as lignocelluloses. They appear during the thermophilic phase as well as the cooling and maturation phase of composting and can occasionally become so numerous that they are visible as a white film on the surface of the compost. The genera of the thermophilic actinomycetes isolated from compost include *Nocardia*, *Streptomyces*, *Thermoactinomyces*, and *Micromonospora* (Strom, 1985a.). The group, thermomonosporas is of particular interest because they have the ability to produce thermostable cellulolytic enzymes (Ball and McCarthy, 1989). The thermophilic filamentous bacterium *Thermobifida fusca* (formerly *Thermomonospora fusca*) is a major cellulose degrader in soil (Irwin *et al.*, 1993).

Fungi with cellulolytic enzymes and/or wood-degrading capability were *Cladosporium*, *Fusarium*, *Geotrichum*, *Myrothecium*, and *Trichoderma* (Deuteromycetes); *Aspergillus*, *Bulgaria*, *Chaetomium*, *Helotium*, *Paecilomyces* and *Penicillium* (Ascomycetes); *Coriolus*, *Phanerochaete*, *Poria*, *Schizophyllum* and *Serpula* (Basidiomycetes) (Carlile and Watkinson, 1997). The conversion of cellulosic mass into fermentable sugars through bio catalyst cellulose derived from cellulolytic organisms has been suggested as a feasible process and offers

potential to reduce the use of fossil fuels and also reduce environmental pollution (Dale, 1999; Lynd *et al.*, 1999).

Yau and Murphy (1998) reported that the biodegradation of coir waste (coco peat) was enhanced by the addition of N fertilizer and inoculation with soft-rot fungus, *Chaetomium globosum* and resulted in the reduction of hemicelluloses. The active component mediating the biodegradation and conversion processes during composting was the resident microbial community, among which fungi played a very important role. Therefore, optimization of compost quality was directly linked to the composition and succession of microbial communities in the composting process (Peters *et al.*, 2000; Taiwo and Oso, 2004).

Microbial degradation of cellulosic materials is the result of synergistic action of enzymes such as endo- $\beta$ -1,4- glucanase, exo- $\beta$ -1,4-glucanase and  $\beta$ -glucosidase, all of which attack  $\beta$ -1,4-glycosidic bonds. Endo- $\beta$ -1,4- glucanase and exo- $\beta$ -1,4-glucanase both act upon cellulose to produce cellobiose. Endo- $\beta$ -1,4- glucanase cleaves randomly  $\beta$ -glycosidic bonds in  $\beta$ -1,4-glucan chains to produce free chain ends and exo- $\beta$ -1,4-glucanases acts at chain ends by removing cellobiose units from the free chain. On the other hand,  $\beta$ -glucosidase hydrolyses cellobiose to glucose, reducing the inhibition effect of cellobiose on endo-glucanase and exo-cellobiohydrolase (Fernandez *et al.*, 2002; Brienzo *et al.*, 2008).

#### 2.4.2.1. *Trichoderma reesei*

Today, 87 per cent of energy used in the world comes from non-renewable sources like natural gas, oil, and coal (Merino and Cherry, 2007). Although biofuel production is now being pushed in order to decrease the requirement for fossil fuels, the raw materials therefore originate from commodities and land also needed for food. In this respect, production of the so-called second generation biofuels from agricultural waste products by the aid of cellulases and hemicellulases produced for example by *T. reesei* and fermentation of the resulting oligosaccharides by yeast provides an alternative strategy. However, for an economically competitive process an increase in efficiency of more than 40-

fold would be necessary, which is a formidable challenge for research with *Trichoderma*. Cellulolytic enzymes hydrolyze cellulose and are produced by a wide range of bacteria and fungi, *T. reesei* which is one of the best known cellulolytic organisms (Mach and Zeilinger, 2003).

Microbial inoculation and certain additives in either soil or soil-less media might offer considerable benefit to growers (Carlile and Wilson, 1991). The benefits of microbial inoculation in compost for various field crops was demonstrated by Espiritu (2011) and reported that the tallest plants were produced by the application of compost prepared with the combined inoculants of *Azotobacter* sp. and *T. harzianaum*. In the same study, the combined use of inoculants of *Azotobacter* sp. and *T. harzianaum* added into the coconut coir dust-chicken manure compost supplied into the test plants (mung bean-*Vigna radiata* and pechay- *Brassica napus*) have significantly promoted growth with improved degradation of compost, enhancing N fixation and supply of nutrients.

Xi *et al.* (2002) studied the effects of complex microorganisms (*B. casei*, *Lactobacillus buchneri*, *Candida rugopelliculosa*, *Trichoderma* and White rot fungi) in composting process of the municipal solid waste and sludge and the experimental results showed that the complex microorganisms were effective in composting organic matter, speeding up the composting process and changing into humus.

Muthurayar and Dhanarajan (2013) studied the effect of *Trichoderma* on composting of coir pith and found that the lowest C:N ratio as 21.8, 22.8 per cent cellulose, 10.3 per cent lignin and highest nutrient content viz. 1.28 per cent N, 0.47 per cent P, 1.2 per cent K when compared with other inocula.

The addition of specific microorganisms (biological control agents) to compost is recommended to enhance disease suppression (Segall, 1995; Trillas *et al.*, 2006; Siddiqui *et al.*, 2008). *T. harzianum* is among the biological control agents most widely used against soil borne pathogens (Lorito *et al.*, 2010).

The genus *Trichoderma*, filamentous ascomycetes are widely used in industrial applications because of high secretory capacity and inducible promoting characteristics. *T. reesei* was selected as the best cellulase producing strain which have the capacity to secrete large amounts of cellulolytic enzymes (cellulases and hemicellulases). Microbial cellulases have industrial application in the conversion of cellulose, a major component of plant biomass, into glucose (Mach and Zeilinger, 2003).

Sangeeth and Padmaja (2006) investigated the efficacy of *Phanerochaete chrysosporium* and *T. viride* for composting bagasse. They reported that the inoculated (*P. chrysosporium* and *T. viride*) bagasse showed a drastic reduction in organic carbon content from 41.13 to 21.13 per cent (raw bagasse) within 60 days of decomposition.

As a potent cellulase producer, research with *T. reesei* is nowadays particularly focused on improvement of efficiency of the enzyme cocktail produced in order to decrease overall costs of production of bioethanol from cellulosic waste material (Kumar *et al.*, 2008).

Ajnavi (2008) recycled agricultural waste comprising of garden waste (grass cutting) and leaf litter by different fungi (*A. niger* FS1 and *T. reesei* MTCC-164) and using cow dung and Di Ammonium Phosphate - 0.1 per cent as activators and reported that organic carbon was decreased from 28.6 to 14.5 per cent and cellulose from 534.4 to 115.1 ppm, with concomitant increase in available N content from 74.6 to 356.5 ppm over 90 days of incubation. In garden waste, cellulose was decreased by 77 per cent and in leaf litter (comprising mainly with *Bambusa vulgaris* leaves) by 70 per cent when the biomass was treated with fungal consortia with the addition of 0.1 per cent DAP as an activator.

Parveen and Padmaja (2009) observed a drastic reduction in cellulose content in *T. viride* inoculated spent mushroom substrate from 19.24 to 8.78 per cent within 60 days of decomposition.

#### 2.4.2.2. *Pleurotus sajor - caju*

Singh and Sharma (2003) accelerated the process of composting a mixture of municipal solid waste (MSW) and horticultural waste by inoculating different microflora viz. *P.sajor-caju* (fungus), *T. harzianum* (fungus) and *A. chroococcum* (bacteria) in different combinations and reported that the combination of *P.sajor-caju*, *T.harzianum* and *A.chroococcum* produced the best quality compost. The growth of the crop was also enhanced significantly with the combination of *P. sajor-caju*, *T. harzianum* and *A. chroococcum* over other treatments.

Theradimani *et al.*(1991) and Nallathambi and Marimuthu (1993) highlighted the efficacy of *Pleurotus* spp. in reducing organic carbon content of plant residues like paddy straw, cotton and sorghum stalks, saw dust, paper waste and bagasse. They found that among the different species of *Pleurotus* (*P. platypus*, *P. florida*, *P. sajor-caju*, *P. citrinopileatus*) and *Volvariella volvacea*, *P. platypus* registered maximum reduction of organic carbon content of the substrates after 15 day of incubation.

Yasotha and Vijayalakshmi (2000) during their study on efficient utilization of lignolytic fungi for biodegradation of sugarcane bagasse observed a maximum loss of organic carbon (56 per cent) in *P. sajor-caju* and *T. harzianum* inoculated treatment. Suri and Pramnik (1995) observed that *P. sajor-caju* increased total N in rice husk during decomposition.

Marimuthu *et al.* (1991) observed that *Pleurotus* spp. have the capacity to detoxify phenolic compounds present in raw plant residues, by complexing them to non toxic melanins. Saroja *et al.* (1999) found that the total phenolic content of sugarcane bagasse was decreased from 58.40 to 45.28 mg g<sup>-1</sup> during 30 days of spawing with *Pleurotus* spp.

Muthurayar and Dhanarajan (2013) conducted a study regarding the influence of bio inoculants viz. *P. sajor-caju* and *T. viridae* on the decomposition of coir pith. They claimed the highest P content of 0.47 per cent and K content of 1.2 per cent and the least cellulose and lignin contents of 22.8 per cent and 10.03



per cent respectively were recorded under coir pith + cow dung +vegetable market waste + poultry waste + mixed microbial culture (*T. viridae* + *P. sajar-caju*).

*P. sajar-caju*, can reduce the amount of lignin in the coir pith considerably thus converting the waste pith into a useful product in an eco-friendly manner. The product obtained after bio-degradation can be used as manure (Reghuvaran and Ravindranath, 2013).

#### **2.4.2.3. Composting Inoculum and Commercial Enzyme Cocktails**

Xi et al. (2005) used a method to improve the composting efficiency by seeding with microbial consortia (A- a blend of *Bacillus azotofixams*, *Bacillus megaterium* and *Bacillus mucilaginosus*, B- a blend of effective cellulolytic strains, viz. *T. koningii*, *Streptomyces cellulosa*, and white-rot fungi; and C, a mixture of A and B).

Gaind et al. (2009) composted poultry droppings, neem cake, castor cake, jatropha cake and grass clippings in the presence of a fungal consortium which included *Aspergillus awamori*, *Aspergillus nidulans*, *T. viride* and *Phanerochaete chrysosporium*. Evaluation of compost maturity showed that mixture of wheat straw, poultry dropping and jatropha cake had the lowest C:N ratio of 10:1, the highest humic acid fraction of 3.15 per cent, the lowest dehydrogenase activity and a germination index exceeding 80 per cent in 60 days of decomposition. Inoculated and grass clipping amended wheat straw-poultry dropping mixture resulted compost with highest humus content of 11.8 per cent and C: N ratio of 14:1, humic acid fraction of 2.84 per cent and germination index of 59.66 per cent. They also reported that fungal consortium was effective in improving the humus content of all the composted mixtures.

Parveen and Padmaja (2009) carried out an experiment to assess the degrading efficiency of cellulolytic fungi (*T. viride*, *T.koningii* and *T. harzianum*) on the biodegradation of spent mushroom substrate.

According to Gautam *et al.* (2010), composting was a time consuming process and to reduce the composting period, efficient decomposing micro organisms were needed. Among various isolates, three isolates viz. *Pseudomonas sp.*, *T. viride* and other *Trichoderma sp.* were found most efficient for composting and a microbial consortium was developed using these isolates. They also reported the temperatures ranging from 40-60 °C and pH ranging from 6.0 to 7.0 was found as favourable for cellulases and pectinases enzymes production.

Chaturvedi *et al.* (2010) conducted an experiment to analyse and evaluate the effect of microbial bio-augmentation on composting of jatropha cake in terms of changes in physico-chemical and hydrolytic enzymatic parameters. The microbial inoculants of fungal strains namely, *A. awamori*, *A. nidulans*, *T. viride*, and *P. chrysosporium* amended with beneficial soil microorganisms like *P. striata* and *Azotobacter chroococcum* produced bio-augmented compost with most desirable phosphorus availability at the time of maturity while hydrolytic enzymes cellulase and xylanase displayed a significant increase after 60 days, indicating their contribution in the process of rapid decomposition.

## 2.5. QUALITY OF COMPOSTS

According to Saravanan *et al.* (2003) the three most important factors for making good compost are the chemical makeup of the raw ingredients or feedstocks (quality and quantity of carbon and minerals, pH), the physical size and shape of the feedstock and the porosity of the pile, and the population of organisms involved in the composting process (macrofauna, mesofauna, and microorganisms including bacteria actinomycetes and fungi).

Compost “happens” either aerobically or anaerobically when organic materials are mixed and piled together. Aerobic composting is the most efficient form of decomposition and produces finished compost in the shortest time (Adediran *et al.*, 2003).

Various chemical, biological and physical changes are reported during the process of composting. Initial temperature of 28-30 °C was recorded at the start

of composting and highest temperature was observed at 14 days of composting which rose up to 46<sup>0</sup>C under water hyacinth and then declined gradually. In the case of organic carbon the initial content was varied from 40 to 48 per cent (Goyal *et al.*, 2005).

### 2.5.1. Nutrient Status of the Compost

The decrease in total N content at early stages of decomposition was due to losses of N in the form of ammonia which in turn depends upon the type of material and C:N ratio. The composting of materials with low C:N ratio result in more N losses than in wastes with high C:N ratio ( Monedero *et al.*, 2001).

N is an important nutrient for composting process since the quantity of N determines the microbial population growth. During composting process, microorganisms oxidize organic matter and release essential minerals for plants such as N, P, and S and therefore, the amount of N increases at the end of the process. Nallathambi and Marimuthu (1993) and Theradimani and Marimuthu (1993) were opined that the increase in N content of agrowaste during decomposition may be due to lignolytic activity of *Aspergillus* spp., *Trichoderma* spp. and *Pleurotus* spp. Yaghmaelan *et al.* (2005) reported that the composting of poultry waste, leaves and garbage from MSW increased the N content from 0.62 to 1.03 per cent.

Rao (2007) found that the N content of *P. sajor-caju* inoculated municipal solid waste showed an increase from 0.78 to 1.29 per cent on composting of municipal solid waste and agricultural waste. The apparent increase in total nutrient content in compost was not only due to enrichment but also due to the reduction in weight because of decomposition. Parveen and Padmaja (2009) recorded a maximum increase in N content from 1.1 to 2.7 per cent in *T. viride* inoculated spent mushroom substrate over 60 days of degradation.

As the decomposition progressed due to losses of C mainly as carbon dioxide, the C content of the compostable material decreased with time and N

content per unit material increased, which resulted in the decrease of C: N ratio as reported by Goyal *et al.* (2005).

### 2.5.2. Enzyme Status and Plant Growth Promoters in Composts

Enzyme activities have been used as suitable indicators for evaluation of the degree of alteration of soils in both natural and agro ecosystems. Soil microbial properties have a strong correlation with enzyme activities and soil health. Gianfreda and Ruggiero (2006) observed a typical increase in enzyme activity shortly after the addition of organic amendments to the soil.

Gilani and Bahmanyar (2008) observed a positive correlation between soil enzyme activity and organic matter content of the soil, and with the water soluble soil organic C. Tejada *et al.* (2006) found an increase of urease,  $\beta$ -glucosidase, alkaline phosphatase and arylsulfatase activities after the application of diverse organic wastes such as cotton gin compost, beet vines composted with crushed cotton gin compost and poultry manure to the soil.

Various hydrolytic enzymes are believed to control the rate at which various substrates are degraded. Enzymes are the main mediators of various degradative processes (Tiquia, 2002). Important enzymes involved in the composting process include cellulases, hemicellulases, proteases, lipases, phosphatases and arylsulphatases. High levels of protease, lipase and cellulase activities have been detected throughout the active phase of composting (Herrman and Shann, 1993; Queda *et al.*, 2002; Mondini *et al.*, 2004).

Different hydrolytic enzymes are released by microorganisms, which are involved in the depolymerization of different constituents of organic wastes (Kandeler *et al.*, 1999; Marx *et al.*, 2001). The activity of dehydrogenase is considered an indicator of the oxidative metabolism in soils and thus of the microbial activity, because it is exclusively intracellular and theoretically, can function only within viable cells. Srinivas and Saroja (2002) reported that the addition of organic manures as farm yard manure at 10 t ha<sup>-1</sup> caused significant differences in dehydrogenase activity in submerged vertisol planted with rice.

Additions of organic amendments *viz.* composts stimulated microbial production of enzymes such as dehydrogenase and phosphatase which enhanced organic matter decomposition and organic P mineralization in the soil (Garcia-Gil *et al.*, 2000; Takeda *et al.*, 2009).

Goyal *et al.* (2005) claimed that the cellulase activity increased during decomposition and was maximum at 30 days and declined thereafter ( at 60 and 90 days). The initial xylanase activity increased with increase in composting period up to 60 days and declined later on. The activities of both cellulase and xylanase showed that cellulose and hemicellulose are actively degraded during first 60 days of composting. Protease activity was also increased up to 60 days and declined further.

Urease catalyse the hydrolysis of urea to CO<sub>2</sub> and NH<sub>3</sub>, which is of specific interest because urea is an important N fertilizer. Urease is released from living and disintegrated microbial cells and in the soil it can exist as an extracellular enzyme absorbed on clay particles or encapsulated in humic complexes (Yang *et al.*, 2006).

Bhattacharyya *et al.* (2005) and Krishnamurthy *et al.* (2011) reported an increased urease activity on addition of organic manures over mineral N application and absolute control.

According to Krishnakumar *et al.* (2005) phosphatases catalyses the hydrolysis of both organic phosphate esters and anhydrides of phosphoric acid into inorganic P. Krishnamurthy *et al.* (2011) reported higher phosphatase activity in organic manure amended rice soil.

### **2.5.3. Maturity of the Compost**

A number of criteria and parameters have been proposed for testing compost maturity. Physical characteristics such as colour, odour and temperature give a general idea of the decomposition stage reached, but give little information as regards the degree of maturation (Gomez-Brandon *et al.*, 2008).

The most important factors affecting the successful application of compost for agricultural purposes are its degree of stability and maturity (Wu *et al.*, 2000). The terms stability and maturity are both commonly used to define the degree of decomposition of organic matter during the composting process even if they are conceptually different. Compost maturity refers to the degree of decomposition of phytotoxic organic substances produced during the active composting stage. Application of unstable or immature compost may inhibit seed germination, reduce plant growth and damage crops by competing for oxygen or causing phytotoxicity to plants due to insufficient biodegradation of organic matter (Brewer and Sullivan, 2003 and Cooperband *et al.*, 2003).

A large variety of techniques have been reported for the determination of compost stability. Chemical parameters such as pH, electrical conductivity (EC), cation exchange capacity (CEC), dissolved organic carbon, C: N ratio and  $\text{NH}_4^+$  to  $\text{NO}_3^-$  have been applied as indicators of stability (Wang *et al.*, 2004).

Mondini *et al.* (2006) reported that microbial biomass could be used as a stability parameter in lignocellulosic waste composts because it clearly reflects the transformation of organic matter during the composting process. Respiration ( $\text{CO}_2$  evolution rate and/or  $\text{O}_2$  uptake rate) is a general measure of microbial activity, and it has been widely used to evaluate the stability of compost (Gomez *et al.*, 2006). The ATP content and enzyme activities were useful indicators of compost stability (Tiquia *et al.*, 2002 and Bitzer *et al.*, 2006).

Biological methods involving seed germination tests and plant growth bioassays have been used to evaluate the maturity of compost (Cooperband *et al.*, 2003).

The principal requirement of compost for it to be safely used in soil is its degree of stability or maturity, which implies stable organic matter content and the absence of phytotoxic compounds and plant or animal pathogens. Maturity is associated with plant-growth potential and phytotoxicity (Iannotti *et al.*, 1993) whereas stability is often related with the compost's microbial activity.

Application of undecomposed wastes or non-stabilized compost to land may lead to immobilization of plant nutrients and cause phytotoxicity (Butler *et al.*, 2001; Fuchs, 2002; Cambardella *et al.*, 2003). For this, chemical methods are widely used, including measurement of the C/N ratio in the solid phase and in water extract, inorganic N, the cation exchange capacity, as well as the degree of organic matter humification. The supply of carbon relative to N (C:N ratio) determines whether net mineralization or immobilization of N will occur. Mineralization is conversion of organic N to mineral forms (*i.e.* ammonium and nitrate); immobilization is incorporation of N into microbial biomass (Golueke, 1991).

The ease with which compounds degrade generally follows the order carbohydrates > hemicellulose > cellulose = chitin > lignin. Fruit and vegetable wastes are easily degraded because they contain mostly sugars and starches. In contrast, leaves, stems, nutshells, bark, and tree limbs and branches decompose more slowly because they contain cellulose, hemicellulose, and lignin (Kumar *et al.*, 2010).

Hue and Liu (1995) suggested the water soluble organic-C/total organic-N ratio as a suitable parameter for assessing compost maturity.

Finstein and Miller (1985) defined compost maturity in terms of nitrification. When the  $\text{NH}_3^+$  concentration decreases and  $\text{NO}_3^-$  appears in the composting material it can be considered ready to be used as compost.

According to Riddech *et al.* (2002) development of mesophilic and thermophilic microorganisms during composting are related to the mesophilic and thermophilic stages of the composting system.

Microbial succession plays a key role in composting process and appearance of some microorganisms reflect the quality of maturing compost (Ryckeboer *et al.*, 2003).

Soil enzymes derived primarily from soil microbes are important due to the fact that they participate in elemental cycling and decomposition of organic residues and are considered fundamentally good indicators for soil quality (Venkatesan and Senthurpandian, 2006).

Dehydrogenase is one such enzyme indicative of overall biological and microbial activity of soils (Quilchano and Maranon, 2002) because it is associated with living cells and microbial oxidoreduction process (Alef and Nannipieri, 1995; Stepniewska *et al.*, 2007) which are important for organic matter degradation and transformation. Dehydrogenase activity in soils is very sensitive to various natural and anthropogenic factors like soil aggregation, soil aeration status (Brzezinska *et al.*, 2001), organic content (Gajananda, 2007), vegetation (Bastida *et al.*, 2006), agricultural management (Truu *et al.*, 2008), addition of pesticides (Stepniewska *et al.*, 2007), insecticides (Singh and Kumar, 2008) and heavy metal combined pollution (Gao *et al.*, 2010).

## 2.6. IMPACT OF COMPOST ON SOIL

### 2.6.1. Yield and Yield Attributes

Oworu *et al.* (2010) reported that the application of 12 t ha<sup>-1</sup> compost resulted the tallest plant, highest number of leaves and wider leaf area, but there was no significant difference among the different rates of composts applied in terms of other growth parameters measured. Thus, it appears that the application of compost enhanced leaf production in grain amaranth.

Pradeepkumar *et al.* (2011) claimed that Bhindi plants receiving 100 per cent of manure through sludge compost showed earliness in flowering and significantly increased the number of fruits per plant and yield in bhindi.

Organic fertilizer had more balanced nutrient supply, create better soil structure thereby enhancing root growth while the slow release of the nutrients would contribute to the residual pool of organic N and P in the soil though organic fertilizer being low in nutrient content would make larger quantity to be needed to provide enough nutrients for crop growth and the long term or heavy application



may result in salt and heavy metal accumulation which may adversely affect plant growth (Ramesh *et al.*, 2009).

Adekayode and Ogunkoya (2011) studied the effect of organic and inorganic fertilizers on the yield and yield attributes of amaranthus and observed significant increase in the growth, yield and vitamin C content of amaranth. The yield and vitamin C content of amaranth correlated with the soil organic matter content.

Finstein and Miller (1985) reported that the significantly higher amaranth yield of 8.33 t ha<sup>-1</sup> obtained in NPK plot compared to the organic compost plot could be attributed to the immediate release and availability of nutrients while the higher residual nutrient build-up in the organic compost treated plot which reflected in the higher yield in the subsequent years.

Akparobi (2009) has reported that the highest manure level of 35 t ha<sup>-1</sup> attained the highest plant height of 123.27 cm than no manure (80.20 cm). Adeyemi *et al.* (1987) observed the adequacy of manure decreased the number of days from planting to first harvesting, and it increased the plant height of grain amaranthus.

Tindall (1975) reported that amaranthus require soil with high organic content, and adequate mineral nutrients favoured the production of higher plant height in amaranthus. Kipkosgel *et al.* (2002) reported that the addition of various rates of organic fertilizers significantly improved vegetative growth and increased leaf yields of amaranth.

According to Ainika *et al.* (2012) all the growth and yield parameters of amaranths were significantly increased in response to the application of farmyard manure at the highest rate of 10 t ha<sup>-1</sup>. This may be attributed to the fact that farmyard manure like other organic matter contains essential plant nutrients though in lower concentrations compared with inorganic fertilizer.

### 2.6.2. Enzymes

Enzyme activities are widely used as an index of soil fertility since they are involved in the biological transformations of native and foreign compounds in soils (Tate, 2000). Microbial activity is of great importance for biological and biochemical soil processes because it directly influences the transformation of nutrients. It is also qualitatively and quantitatively associated with the presence of extracellular hydrolytic enzymes which are important in the process of decomposition and mineralization of organic matter (Kiss *et al.*, 1975; Nakas *et al.*, 1987; Martens *et al.*, 1992; Elliott *et al.*, 1993). The most important general indicators of soil microbial activity are microbial biomass C and soil respiration, while specific indicators are related to the activity of extracellular hydrolytic enzymes such as phosphatase and glucosidase, that are involved in nutrient cycling (Gil-Sotres *et al.*, 2005).

To evaluate biological and biochemical soil properties and the glucosidase activity has been considered because of their relationship to the soil C cycle and the sensitivity of these indicators to detect changes resulting from agricultural management practices (Nannipieri *et al.*, 1990; Dick and Tabatabai, 1993; Gil-Sotres *et al.*, 2005; Lagomarsino *et al.*, 2009). The activity of phosphomonoesterase enzymes, such as acid and alkaline phosphatases, has been widely studied because of its importance in organic P mineralization, releasing orthophosphates that are readily assimilated by plants and soil microorganisms (Sylvia *et al.*, 1999).

Microbial communities in the soil are enhanced and stimulated by the addition of organic waste, especially due to the presence of readily available nutrients and C compounds. In general, organic waste has high levels of macronutrients and secondary nutrients such as N, P, K, Ca (Ros *et al.*, 2003) and micronutrients such as B, Zn and Mn. Since the application of organic waste can change biological and biochemical indicators, studies are needed to measure the effect of this practice on soil (Martens, 2000; Tejada *et al.*, 2009).

The enzyme activities largely reflected the diversity of the microbial population and in turn reflected the composting process. The three enzymes *viz.* cellulase, dehydrogenase, phosphatase were recognized as very important enzymes involved in the mineralization of nutrients. Characterizing and quantifying the enzymatic activities during composting reflected the dynamics of the composting process in terms of decomposition of organic matter and N transformations and provide information about the maturity of composted products (Tiquia, 2002). In addition, on the basis of the well demonstrated relationship between enzymatic activity, quantity and quality of organic organic matter, compost stability which was defined as the degree of decomposition of the readily bio-degradable organic matter can also be assessed (Garica *et al.*, 1993).

The enzyme urease is responsible for breakdown of urea into CO<sub>2</sub> and NH<sub>3</sub> during the composting process. Pallab-De *et al.* (1990) reported that there was a significant positive correlation between the organic C and urease activity. Frankkenberger and Dick (1982) also reported the correlation of urease activity with organic C and total N content of the soil.

Phosphatase plays an important role in transforming the organic phosphorus into the available form of phosphorus (Pallab-De *et al.*, 1990).

Dehydrogenase, the intracellular enzyme involved in microbial oxidoreductase metabolism and found to be higher in compost than soil (Garcia *et al.*, 2000). The activity of these enzymes basically depends on the metabolic state of the microbes and widely used to measure metabolic activities, which in turn is correlated with total microbial activity.

Sangeetha *et al.* (2012) studied the activity of enzymes during the composting of silk worm litter using different inoculants *viz.* *Cellulomonas cellulans*, *P. putida*, *A. terreus*, *P. chryosporium*, *B. subtilis* and microbial consortium of the afore said organisms. The maximum urease activity (266.4 µg N g<sup>-1</sup>soil) and phosphatase activity (34.3 µg PNPP g<sup>-1</sup>soil) were reported with the application of compost from the silkworm litter pupal waste and microbial

consortium. The highest dehydrogenase activity of  $37.7 \mu\text{g TPF g}^{-1} 24 \text{ h}^{-1}$  was recorded with the compost from silkworm litter pupal waste and *C. cellulans*. Dehydrogenase activity increased during the mesophilic period of composting and reached maximum during the thermophilic period and subsequently declined to a constant value. Cellulase activity was highest ( $346.4 \mu\text{g reducing sugars g}^{-1}$ ) with the application of compost from silkworm litter and microbial consortium.

Pedrazzini and McKee (1984) reported that increased dehydrogenase activity in soil was largely due to available nutrients and higher amounts of organic C.

### 2.6.3. Biological Status- Microflora

During composting microorganisms transform organic matter into  $\text{CO}_2$ , biomass, thermo energy (heat) and humus like end-product. Microorganisms require a carbon source, macronutrients such as NPK, and certain trace elements for their growth. Carbon serves primarily as an energy source for the microorganisms, while a small fraction of the carbon is incorporated into their cells (Zimmermann and Frey, 2002.).

Composting is a dynamic process carried out by a rapid succession of mixed microbial populations. The main groups of microorganism involved are bacteria, including actinomycetes, and fungi (Golueke, 1991). Although the total number of microorganisms does not significantly change during composting, the microbial diversity can vary during the different phases of composting (Atkinson *et al.*, 1996a).

Manure application is known to stimulate and improve soil structure, fungal and bacterial population and thus biological activity (Chaoui *et al.*, 2003).

The precise nature of succession and the number of microorganisms at each composting phase is dependent on the substrate and on the preceding microorganisms in the succession (Crawford, 1983). At the beginning of composting mesophilic bacteria predominate, but after the temperature increases

to over 40°C, thermophilic bacteria take over and thermophilic fungi also appear in the compost. When the temperature exceeds 60°C, microbial activity decreases dramatically, but after the compost has cooled mesophilic bacteria and actinomycetes again dominate (McKinley and Vestal, 1985; Strom, 1985a).

Composting is an aerobic process in general, but anaerobic microenvironments may develop. Atkinson *et al.* (1996b) estimated that almost 1 per cent of all the bacteria found in municipal solid waste compost was anaerobic. All the anaerobic bacteria found were highly cellulolytic and thus may play a significant role in the degradation of macromolecules. The majority of the mesophilic anaerobic bacteria were facultative, while under thermophilic conditions more obligate anaerobic bacteria were found (Atkinson *et al.*, 1996b).

Bacteria are typically unicellular with a size ranging from 0.5 to 3.0 µm. Because of their small size, bacteria have a very high surface/volume ratio which allows rapid transfer of soluble substrates into the cell. As a result, bacteria are usually far more dominant than larger microorganisms such as fungi. A wide range of bacteria have been isolated from different compost environments, including species of *Pseudomonas*, *Klebsiella* and *Bacillus* (Nakasaki *et al.*, 1985; Strom, 1985a,b). Typical bacteria of the thermophilic phase are species of *Bacillus* (*eg.* *B. subtilis*, *B. licheniformis* and *B. circulans*).

Strom (1985b.) reported that as much as 87 per cent of the randomly selected bacterial colonies during the thermophilic phase of composting belong to the genus *Bacillus*. Many thermophilic species of *Thermus* have been isolated from compost at temperatures as high as 65°C and even 82°C (Beffa *et al.*, 1996).

Actinomycetes are bacteria which form multicellular filaments, thus they resemble fungi. They appear during the thermophilic phase as well as the cooling and maturation phase of composting, and can occasionally become so numerous that they are visible on the surface of the compost. Thermophilic actinomycetes have been isolated from a wide range of natural substrates (*eg.* from desert sand and compost) (Cross, 1968).

The genera of the thermophilic actinomycetes isolated from compost include *Nocardia*, *Streptomyces*, *Thermoactinomyces* and *Micromonospora* (Waksman *et al.*, 1939; Strom, 1985a).

Actinomycetes are able to degrade some cellulose, and solubilize lignin, and they tolerate higher temperatures and pH than fungi. Thus, actinomycetes are important agents of lignocellulose degradation during peak heating, although their ability to degrade cellulose and lignin is not as high as that of fungi (Crawford, 1983; Godden *et al.*, 1992).

Fungi are the dominant group of microorganisms found highest in terms of biomass in soil literary. A moderately high level of N is needed for fungal growth although some fungi, mainly wood-rotting fungi, grow at low N levels. Indeed, a low nutrient N level is often a prerequisite for lignin degradation (Eriksson *et al.*, 1990; Dix and Webster, 1995). Low nutrient N is a rate-limiting factor for the degradation of cellulose (Dix and Webster, 1995).

Most fungi prefer an acidic environment but tolerate a wide range of pH, with the exception of the Basidiomycotina which do not grow well above pH 7.5. *Coprinus* species are the only Basidiomycotina which prefer an alkaline environment (Dix and Webster, 1995). The majority of fungi are mesophiles which grow between 5°C and 37°C, with an optimum temperature of 25-30°C (Dix and Webster, 1995). However, in the compost environment the elevated temperature means that the small group of thermophilic fungi is an important biodegradation agent.

Cooney and Emerson (1964) defined thermophilic fungi as fungi with a maximum growth temperature of 50°C or higher and a minimum growth temperature of 20°C or higher. Thermo tolerant species have a maximum growth temperature of about 50°C and a minimum well below 20°C.

The ligninolytic capacity of all thermophilic fungi is not known. However, most of them are known to degrade wood or other lignocellulose,

cellulose or hemicelluloses (Sharma, 1989; Kuhad *et al.*, 1997). The ability of fungi to hydrolyse hemicelluloses is probably more common than cellulose hydrolyzation (Dix and Webster, 1995). The most effective lignin degraders are Basidiomycotina, but according to Mouchacca (1997) opined that all Basidiomycotina are mesophilic. However, a few Basidiomycotina grow well at elevated temperatures. *P. chrysosporium* is a white-rot fungus with an optimum temperature of 36-40°C and maximum temperature 46-49°C (Mouchacca, 1997). *Ganoderma colossus* is another white-rot fungus which is still capable of growing at 45°C and has an optimum temperature of 40°C (Adaskaveg *et al.*, 1995).

In the genus *Coprinus* there are some species that have an optimum temperature of above 40°C (Crisan, 1973). Some of the wood-rotting *Coprinus* species are brown-rot fungi which modify rather than degrade lignin (Rayner and Boddy, 1988).

It is known that plants and plant products (organic amendments, crop residues, green manures) can dramatically affect soil microbial communities, and are primary drivers of soil microbial dynamics (Garbeva *et al.*, 2004; O'Donnell *et al.*, 2001; Van-Elsas *et al.*, 2002), and thus may be important components in establishing and maintaining soil health.

The results of the study conducted by Larkin (2008) indicate that, in general, manures containing biological amendments *viz.* *T. virens* and *T. harzianum* strain can effectively deliver microorganisms to natural soil, resulting in a wide variety of effects on soil microbial communities depending on the particular types, numbers, and formulations of organisms added. The amendment *T. virens* did not contain bacteria, yet significantly, higher bacterial populations were observed under this treatment than other treatments, indicating some stimulation of bacterial activity with the addition of this amendment.

Plant effects on soil microbial communities, as well as different positive effects among plant species, are presumed to result primarily from the release of

qualitatively and quantitatively different nutrients and organic compounds through roots and the breakdown of plant organic matter and residues (Curl and Truelove, 1986; Grayston *et al.*, 1998).

#### 2.6.4. Heavy Metal Content in Composts

The presence of heavy metals in final compost raises serious concern about the adverse environmental impact, as a result of excessive compost application to agricultural lands. Heavy metal uptake by plants and consecutive accumulation in human tissues and biomagnification through the food chain causes both human health and environment concern (Wong and Selvam, 2006).

The mobility of trace metals, their bioavailability and related eco-toxicity to plants depend strongly on their specific chemical forms or ways of binding rather than total metal concentration (Fuentes *et al.*, 2006; Gupta and Sinha, 2007). In order to screen the suitability of a material, such as water hyacinth for land application, not only depends on their total content of heavy metal in a matrix, but also their bioavailability and their capacity for remobilization (Peruzzi *et al.*, 2011). Water soluble fraction of metal may be readily leachable and bioavailable during thermophilic stage due to decomposition of organic matter, and reduced in final compost due to changes in other oxidizing and anionic conditions in the medium (Ahmed *et al.*, 2007).

In several field trials in compost application lead to a decrease of the extractable fraction of Cd and Zn and to a lower uptake by plants. This was explained by enhanced adsorption capacity (surface activity) introduced with compost (Petruzzeli and Pezzarossa, 2003).

Castaldi *et al.* (2006) reported that water soluble Pb and Cd were about 0.89 and 0.005 mg kg<sup>-1</sup> dry matter respectively in the mature compost of municipal solid waste. Sims and Kline (1991) reported that water extractable Ni was about 7.2 per cent of total Ni during sewage sludge composting. The order of water soluble metal concentration in the composted water hyacinth was Fe > Mn > Cu > Zn > Cr.



During the composting process, heavy metals distribution influenced the process of organic matter mineralization or metal solubilisation by the decrease of pH, metal bio sorption by the microbial biomass or metal complex formation with the newly formed humic substances which made the metals insoluble and less easily extractable (Garcia *et al.*, 1995; Cai *et al.*, 2007).

Soil organic matter (SOM) has probably the greatest capacity and strength of bonding with most trace metals of any soil component. As a consequence there are often statistically significant correlations between solubility of trace metals such as Cu, Hg and Cd, and Soil organic matter content. The strongest relations for total Cd and SOM ( $\text{g kg}^{-1}$  soil), carbonyl and sulph-hydryl groups are the key to the bonding of metals (Castaldi *et al.*, 2006). The bonding strength of these functional groups varies considerably.

Large metal additions force bonding onto the predominant groups (carboxyl). In this case Cu is often found to have weak bonding strength. Generally the metals that bond most strongly to soil organic matter tend also to be the most rapidly adsorbed. Most metals *viz.*  $\text{Pb}^{3+}$ ,  $\text{Cd}^{2+}$ ,  $\text{Cu}^{2+}$  and  $\text{Fe}^{3+}$  when complexed with soil organic matter have low lability. In contrast dissolved humic and fulvic acid-metal complexes of metals such as  $\text{Cu}^{2+}$ ,  $\text{Ni}^{2+}$  and  $\text{Co}^{2+}$  appear to be largely labile. Lability is particularly sensitive to pH and metal/organic ratio, decreasing as pH is raised and as the metal/organic ratio is decreased (Petruzzelli and Pezzarossa, 2003; Leita *et al.*, 2003).

The concentration of heavy metals in compost is generally higher than the normal concentration in soil, so the greater possibility of metal accumulation when the compost repeatedly applied (Zhang *et al.*, 2006). Over ten years trial period, the most abundant metals in the upper most soil horizon were Cu, Zn, and Pb. Cd was the least plentiful, corresponding to the mean metal concentrations in the municipal solid waste compost applied (Businelli *et al.*, 2009). This is supported by the findings of Bergkvist *et al.* (2003) who reported, that during a period of 41 years, 92 per cent of applied Cd was recovered in the topsoil in

sludge treatment, indicating measurable losses by both downward movement and crop uptake.

Six year consecutive applications of a swine compost resulted significantly higher concentrations of Cu and Zn at 10-20 cm depth of the compost amended soil, relative to the control, with an increase from 102.8 to 127.4 mg kg<sup>-1</sup> for Cu and from 111.9 to 165.7 mg kg<sup>-1</sup> for Zn (Zhao *et al.*, 2006). On the other hand, Bartl *et al.* (2002) found that 32 t ha<sup>-1</sup> of biowaste compost did not influence the total contents of Cd, Mn, Mo or Ni in soil, in 5 years. The total soil contents of Zn and Pb were significantly higher in soils with compost treatment than in the unfertilized soils. From experiments longer than 10 years, it is possible to suggest that sludge and composted sludge showed a high accumulation of Zn, Cu and Cr probably due to the notably higher concentration of these metals in the raw materials (Saviozzi *et al.*, 1999; Kunito *et al.*, 2001).

Activity of soil enzymes *viz.* dehydrogenase, urease and  $\beta$ -D-glucosidase were also found to be adversely affected by the metals derived from the addition of sewage sludge (Kunito *et al.*, 2001). According to Tittarelli *et al.* (2007) the main factors that affect the environmental behaviour of heavy metals are: (i) cation exchange capacity, as an index of the soil capacity to adsorb and hold metal cations; (ii) humic substances, that can interact with heavy metals, forming complexes with different solubility, and consequently mobility, and (iii) the water and thermic regime of the soil, which affect the organic matter decomposition. It can also be generally assumed that extractability and uptake of heavy metals decline as the soil pH becomes more alkaline, especially after repeated compost application. By contrast, Takeda *et al.* (2005) opined that low pH in the soil was caused by more than 60 years of rice straw compost applications, and might have enhanced the concentration of metals in the water-soluble fraction.

Ten years after municipal solid waste compost application, a part of the heavy metals were further re-mobilized in the soil profile, also leading to a decrease in the percentage distribution of organically-bound heavy metals with

time. According to Businelli *et al.* (2009), metal mobilization was primarily influenced by organic matter dynamics. On the contrary, Sukkariyah *et al.* (2005) reported that in aerobically digested sewage sludge-amended soils, the extractability of the heavy metals steadily declined over 17 years, despite a significant decline in organic matter concentration in the amended soils.

In general, heavy metal uptake by crops increases in leafy plants and it is higher in cereal leaves than in grains. Lettuce, for example, has higher assimilative capacity for heavy metals *viz.* Zn and Cd uptake than other non-leafy crops (Sukkariyah *et al.*, 2005). Mantovi *et al.* (2005) reported that biosolid applications significantly increased the content of Zn and Cu in wheat grain and of only Cu in both sugarbeet roots and maize grain. Cd is one of the most significant potential contaminants of food supplies on arable lands and may limit sewage sludge suitability for soil amendment, because of organic material contained a large amount of metal (Singh and Pandeya, 1998). Being relatively soluble in soils, it is readily taken up by crops and it is quite toxic to humans (Miller and Miller, 2000). This behaviour is particularly evident at low soil pH. Cd solubility in equilibrium extracts of Ca (NO<sub>3</sub>)<sub>2</sub> was increased, during the 41<sup>st</sup> - year trial, by a factor of 20 in the sludge treatment compared with the control (Bergkvist *et al.*, 2003).

# *Materials and Methods*

### 3. MATERIALS AND METHODS

The present study entitled “Characterization, conversion and evaluation of selected lignocellulosic biomass” was carried out in the Department of Soil Science and Agricultural Chemistry at College of Agriculture, Vellayani during August 2013 to February 2014. The study is envisaged for the characterization of the lignocellulosic biomass from selected plant sources, assessment of various microbial and enzymatic sources for degrading the lignocellulosic biomass into compost and evaluation of the resultant composts for assessing manurial value. The investigation pertaining to the study consists of three parts

- i. Collection and proximate analysis of samples for characterization
- ii. Identification of the best degrading source for biomass degradation and conversion into compost.
- iii. Evaluation of the resultant composts through a pot culture experiment using amarathus (variety Arun) as the test crop. The materials and the methods adopted for the study are briefly discussed in this chapter.

#### 3.1. DETAILS OF THE EXPERIMENTAL SITE

##### 3.1.1. Location

Composting of different substrates and the pot culture experiment were done at the experimental yard attached to the Department of Soil Science and Agricultural Chemistry, College of Agriculture, Vellayani, Thiruvananthapuram.

##### 3.1.2. Soil

The soil for pot culture experiment was collected from the Instructional Farm, College of Agriculture, Vellayani. Soil belongs to the Vellayani series (Loamy Kaolinitic Isohyperthermic Rhodic Kandistult). Soil was collected in bulk and was mixed with sand and cowdung powder in the ratio 1:1:1. Each pot was filled with 7 kg of this soil. Important physico-chemical and biological parameters of the soil were analysed as per standard procedures presented in Table 1.

Table1. Physical, chemical and biological properties of initial soil

| Sl No                 | Soil properties   | Content  |                  |
|-----------------------|---|--|------------------|
| 1.                    | Bulk density( $\text{Mg m}^{-3}$ )  | 1.53   |                  |
| 2.                    | Particle density ( $\text{Mg m}^{-3}$ )   | 2.28   |                  |
| 3.                    | Water holding capacity (%)  | 25.54  |                  |
| 4.                    | pH  | 6.01   |                  |
| 5.                    | EC( $\text{dSm}^{-1}$ )   | 0.64   |                  |
| 6.                    | Available N ( $\text{kg ha}^{-1}$ )   | 224.84   |                  |
| 7.                    | Available P ( $\text{kg ha}^{-1}$ )   | 89.18  |                  |
| 8.                    | Available K ( $\text{kg ha}^{-1}$ )   | 248.6  |                  |
| 9.                    | Organic carbon (%)  | 0.983  |                  |
| 10.                   | Available micronutrients<br>( $\text{mg kg}^{-1}$ )   | Fe   | 15.63            |
| 12.                   |   | Mn   | 15.04            |
| 13.                   |   | Cu   | 0.732            |
| 14.                   |   | Zn   | 1.968            |
| 15.                   |   | B  | 0.271            |
| 16.                   | Heavy metals<br>( $\mu\text{g kg}^{-1}$ )   | Pb   | 58.63            |
|                       |   | Cd   | 0.593            |
|                       |   | Sn   | N.D              |
|                       |   | Ni   | 82               |
| Biological properties |   |  |                  |
| 17.                   | Urease activity ( $\mu\text{g urea hydrolysed g}^{-1} \text{ soil hr}^{-1}$ )                                   | 79.67  |                  |
| 18.                   | Phosphatase activity ( $\mu\text{g p-nitrophenol released g}^{-1} \text{ soil hr}^{-1}$ )                       | 12.46  |                  |
| 19.                   | Dehydrogenase activity ( $\mu\text{g TPF hydrolysed g}^{-1} \text{ soil 24 hrs}^{-1}$ )                         | 52.48  |                  |
| 20.                   | Arylsulphatase ( $\mu\text{M p- nitro phenol released g}^{-1} \text{ soil hr}^{-1}$ )                           | 0.0125   |                  |
| 21.                   | $\beta$ -D glucosidase ( $\mu\text{g p-nitrophenyl } \beta\text{- D Glucosidase g}^{-1} \text{ soil hr}^{-1}$ ) | $2.1 \times 10^{-4}$                               |                  |
| 22.                   | Soil respiratory activity( $\mu\text{g of CO}_2 \text{ evolved g}^{-1} \text{ of soil hr}^{-1}$ )               | 2.8  |                  |
| 23.                   | Microflora.   | Bacteria ( $\text{cfu g}^{-1} \text{ soil}$ )      | $19 \times 10^6$ |
|                       |   | Fungi ( $\text{cfu g}^{-1} \text{ soil}$ )         | $2 \times 10^4$  |
|                       |   | Actinomycetes ( $\text{cfu g}^{-1} \text{ soil}$ ) | Nil              |

N.D - Not Detectable

### 3.1.3. Weather

The experiment was conducted during the period of December 2013 to February 2014. The weekly averages of the weather parameters *viz.* maximum and minimum temperature, relative humidity and rainfall received during the cropping period were collected from the Meteorological Observatory attached to NARP, Southern Region, at College of Agriculture, Vellayani and are presented as Appendix 1 and graphically presented in Figure 1.

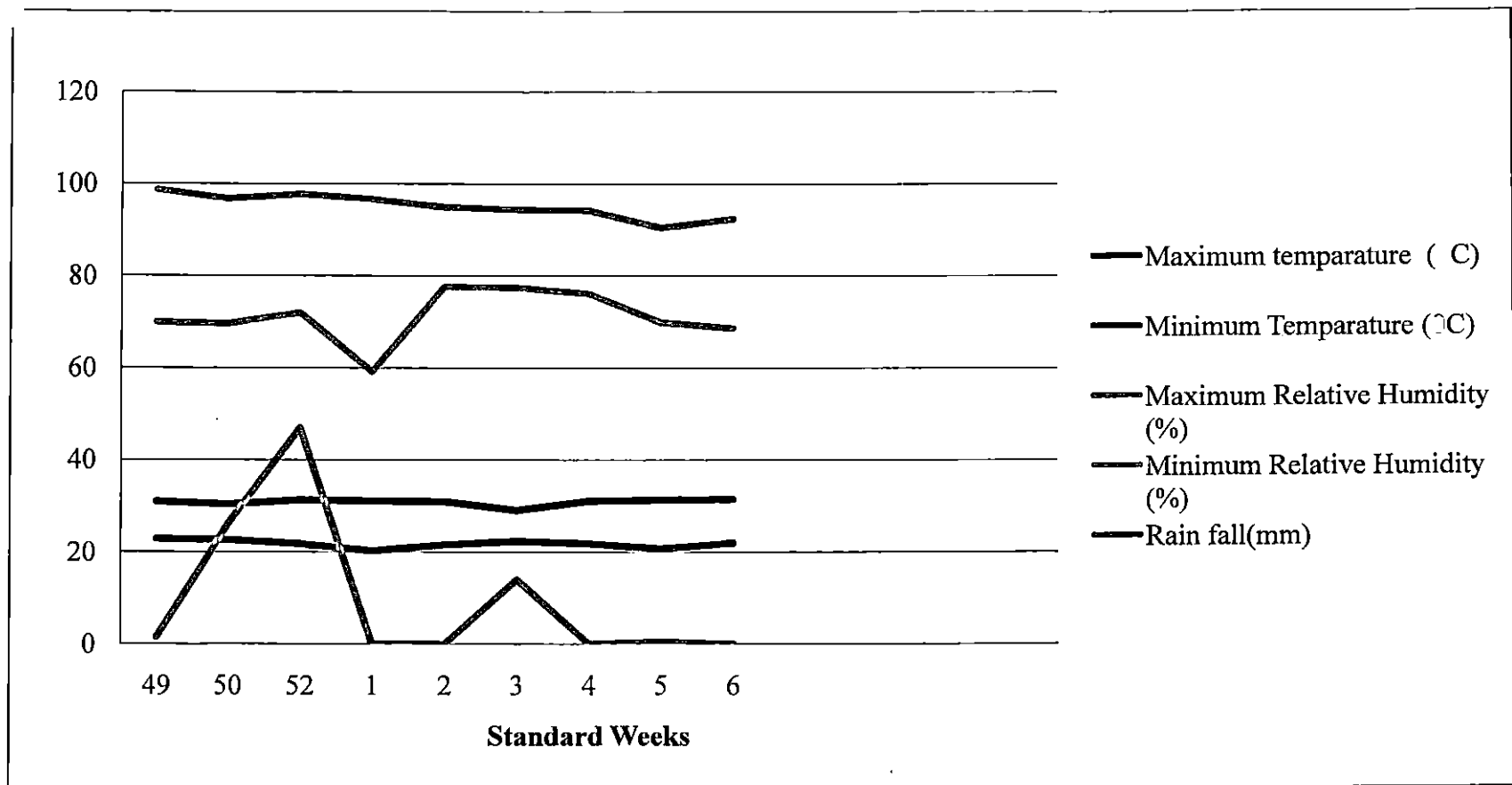


Figure 1. Weather parameters during the cropping period ( December 2013- February 2014)



## 3.2. EXPERIMENTAL MATERIALS

### 3.2.1. Collection and Proximate Analysis of Samples for Characterization (Experiment I).

This involved collection of representative samples such as water cabbage (*Limnocharis flava*), water hyacinth (*Eichhornia crassipes*), farm wastes (dried leaves and pseudostem of banana) and coir pith from different locations. Water cabbage (*Limnocharis flava*) was collected from the rice field of the College of Agriculture, Vellayani, coir pith was collected from Muttakkad, water hyacinth from Thiruvallam, and farm waste from Instructional farm, College of Agriculture, Vellayani. These plant materials were then subjected to chemical characterization for identifying the constitutional makeup.

Design: Completely Randomized Design (CRD)

Treatments: 4

Replication: 5

The substrates used in the study are

- S<sub>1</sub> Water cabbage
- S<sub>2</sub> Coir pith
- S<sub>3</sub> Water hyacinth
- S<sub>4</sub> Farm waste

### 3.2.2. Identification of Best Degrading Source for Biomass Degradation and Conversion into Compost (Experiment II)

By use of various inoculants, the substrates *viz.* water hyacinth, water cabbage, farm waste and coir pith were converted to compost. Good quality compost from each category of agro waste used for composting was identified and evaluated further for manurial value.

#### 3.2.2.1. Method of Composting:

The method followed was aerobic heap method with 100 kg of biomass was mixed with 10 kg of cowdung. After the completion of thermophilic stage, 1 kg of the following inoculants were added to the substrates.

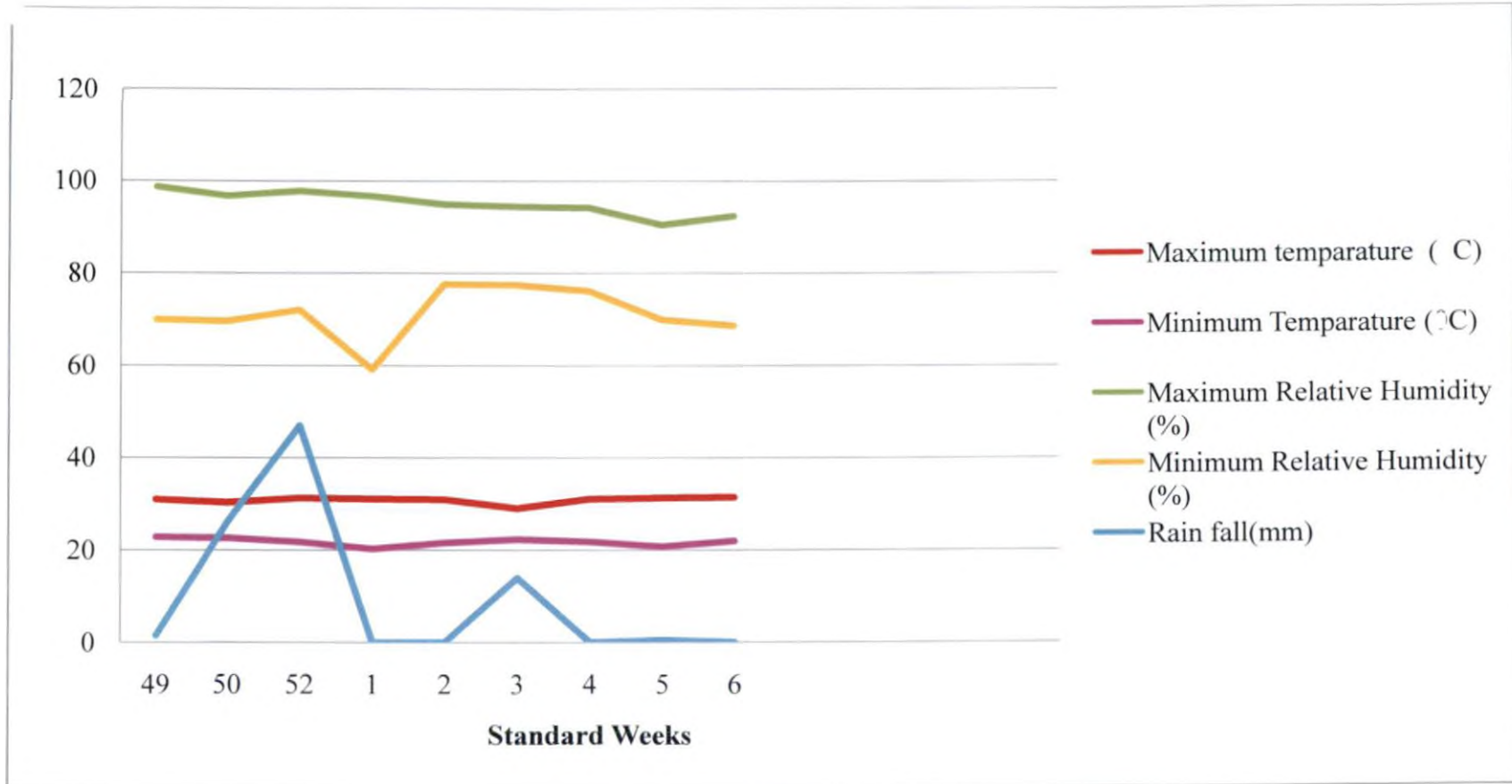


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Plate 1. A general view of the initial stage of the compost heap



Plate 2. A general view of the initial stage of the compost heap

The inoculants used in the study were:

I<sub>1</sub> - *Trichoderma reesei*

I<sub>2</sub> - *Pleurotus sajor-caju*

I<sub>3</sub> - Composting Inoculum developed by the Dept. of Agricultural Microbiology,  
College of Agriculture, Vellayani

I<sub>4</sub> - Commercial enzyme cocktail (Cellulase / pectinase and laccase)

Design : Completely Randomized Design (CRD)

Treatments: 16 (4x4 Factorial)

S<sub>1</sub> - Water cabbage

I<sub>1</sub> - *T. reesei*

S<sub>2</sub> - Coir pith

I<sub>2</sub> - *P. sajor-caju*

S<sub>3</sub> - Water hyacinth

I<sub>3</sub> - Composting Inoculum

S<sub>4</sub> - Farm waste

I<sub>4</sub> - Commercial enzyme cocktail

Replication: 3

Details of the composts are presented in Appendix II

### **3.2.3. Evaluation of the Resultant Composts through Pot Culture (Experiment III)**

The resultant compost from the previous stage was then evaluated for its performance as manure in pot culture experiment.

#### **3.2.3.1. Details of the Pot Culture Experiment**

Design: Completely Randomized Design (CRD)

Treatments: 19

Replications: 3

Crop : Amaranthus

Variety: Arun

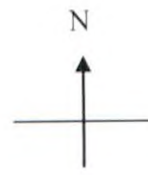
##### **3.2.3.1.1. Experimental Site Preparation**

The experimental site was cleared, levelled and weeds were removed.

##### **3.2.3.1.2. Preparation of Potting Mixture**

Soil required for preparing the potting mixture was collected from Instructional farm Block No 4. Soil, sand and cow dung were mixed in the ratio 1:1:1 and 7 kg potting mixture was filled in fifty seven pots.





| R <sub>1</sub>  | R <sub>2</sub>  | R <sub>3</sub>  |
|-----------------|-----------------|-----------------|
| T <sub>10</sub> | T <sub>6</sub>  | T <sub>12</sub> |
| T <sub>16</sub> | T <sub>12</sub> | T <sub>14</sub> |
| T <sub>5</sub>  | T <sub>11</sub> | T <sub>10</sub> |
| T <sub>9</sub>  | T <sub>16</sub> | T <sub>8</sub>  |
| T <sub>18</sub> | T <sub>5</sub>  | T <sub>4</sub>  |
| T <sub>4</sub>  | T <sub>4</sub>  | T <sub>7</sub>  |
| T <sub>13</sub> | T <sub>19</sub> | T <sub>9</sub>  |
| T <sub>2</sub>  | T <sub>14</sub> | T <sub>1</sub>  |
| T <sub>19</sub> | T <sub>8</sub>  | T <sub>2</sub>  |
| T <sub>7</sub>  | T <sub>15</sub> | T <sub>13</sub> |
| T <sub>17</sub> | T <sub>7</sub>  | T <sub>11</sub> |
| T <sub>11</sub> | T <sub>3</sub>  | T <sub>6</sub>  |
| T <sub>15</sub> | T <sub>1</sub>  | T <sub>18</sub> |
| T <sub>3</sub>  | T <sub>17</sub> | T <sub>3</sub>  |
| T <sub>14</sub> | T <sub>9</sub>  | T <sub>16</sub> |
| T <sub>1</sub>  | T <sub>13</sub> | T <sub>17</sub> |
| T <sub>8</sub>  | T <sub>10</sub> | T <sub>15</sub> |
| T <sub>6</sub>  | T <sub>2</sub>  | T <sub>19</sub> |
| T <sub>12</sub> | T <sub>18</sub> | T <sub>5</sub>  |

Fig. 2. Layout of the experimental field

### **3.2.3.1.3. Planting Materials**

Seeds of the amaranthus variety "Arun " was obtained from College of Agriculture, Vellayani. It is the most popular red amaranthus variety suitable for Kerala having duration of 45 days.

### **3.2.3.1.4. Sowing**

A few seeds were dibbled in each pot in order to get even, healthy and vigorous seedlings. Seed germination was noted on the fourth day after sowing.

### **3.2.3.1.5. After Sowing**

Uniform germination was observed. Thinning was done two weeks after to retain two vigorous plants per pot. Regular weeding was done throughout the cropping period. Regular irrigation was provided as and when required.

### **3.2.3.1.6. Treatment Application**

After two weeks of sowing, nineteen treatments including POP recommendation, vermicompost, prepared sixteen compost combinations (Experiment I) and one absolute control were imposed to the pots according to the technical programme. Details of the treatments are presented in Table 2.

### **3.2.3.1.7. Manures and Fertilizers**

Nutrient recommendation for amaranthus is 100 N: 50 P<sub>2</sub>O<sub>5</sub>: 50 K<sub>2</sub>O kg ha<sup>-1</sup>. (KAU POP, 2011). 100 per cent N requirement was complimented by the addition of respective composts. The P and K requirement of the crop were supplemented by the inorganic fertilizers in addition to the composts. The rock phosphate (20 per cent P<sub>2</sub>O<sub>5</sub>) and MOP (60 per cent K<sub>2</sub>O) were used as sources of P and K respectively. For the pots with KAU POP (2011) recommendation as treatment (T<sub>1</sub>) Urea (46 per cent) alone was given as N source. Vermicompost given as one treatment and one was kept as absolute control without any treatments.



Table 2. Treatment Details

| Treatments      | Details   |
|-----------------|---|
| T <sub>1</sub>  | N, P, K as per POP – 1.36 g N+ 1.56 g P+ 0.52 g K |
| T <sub>2</sub>  | 100 % N as vermicompost - 15 g                    |
| T <sub>3</sub>  | 100 % N as compost ( C <sub>1</sub> ) - 14.3 g    |
| T <sub>4</sub>  | 100 % N as compost ( C <sub>2</sub> ) -10.73 g    |
| T <sub>5</sub>  | 100 % N as compost ( C <sub>3</sub> ) - 7.97 g    |
| T <sub>6</sub>  | 100 % N as compost ( C <sub>4</sub> ) -11.87 g    |
| T <sub>7</sub>  | 100 % N as compost ( C <sub>5</sub> ) - 26.1 g    |
| T <sub>8</sub>  | 100 % N as compost ( C <sub>6</sub> ) - 23.25 g   |
| T <sub>9</sub>  | 100 % N as compost ( C <sub>7</sub> ) - 27.9 g    |
| T <sub>10</sub> | 100 % N as compost ( C <sub>8</sub> ) - 26.57 g   |
| T <sub>11</sub> | 100 % N as compost ( C <sub>9</sub> ) - 16.9 g    |
| T <sub>12</sub> | 100 % N as compost ( C <sub>10</sub> ) -15.1 g    |
| T <sub>13</sub> | 100 % N as compost ( C <sub>11</sub> ) - 10.2 g   |
| T <sub>14</sub> | 100 % N as compost ( C <sub>12</sub> ) - 10.73 g  |
| T <sub>15</sub> | 100 % N as compost ( C <sub>13</sub> ) - 21.5 g   |
| T <sub>16</sub> | 100 % N as compost ( C <sub>14</sub> ) - 18.6 g   |
| T <sub>17</sub> | 100 % N as compost ( C <sub>15</sub> ) - 7.97 g   |
| T <sub>18</sub> | 100 % N as compost ( C <sub>16</sub> ) - 16.91 g  |
| T <sub>19</sub> | Absolute control                                  |

\* The phosphorus and potassium requirement of the crop were supplemented by the inorganic fertilizers in addition to the composts added.

Details of the pot culture experiments were detailed in the appendix II





Plate 3. A general view of the pot culture experiment

### **3.2.3.1.8. Plant protection**

At initial stages leaf caterpillar attack was observed. As a remedy turmeric powder, *Capsicum frutescens* and garlic were ground, mixed, strained and sprayed. *Rhizoctonia* leaf spot attack was also noticed.

### **3.2.3.1.9. Harvesting**

After 45 days of sowing, the plants were cut off at the base and biometric observations were taken immediately.

## **3.3. OBSERVATIONS**

### **3.3.1. Biometric Observations**

#### **3.3.1.1. Yield**

The fresh weight of each plant was recorded immediately after the harvest and expressed in grams.

#### **3.3.1.2. Yield Attributes**

##### **3.3.1.2.1. Plant Height**

Plant height was measured from the base of the plant to the terminal leaf at the time of harvest and expressed in cm.

##### **3.3.1.2.2. No of Branches Plant<sup>-1</sup>**

Numbers of branches were noted at the time of harvest.

##### **3.3.1.2.3. No of leaves Branch<sup>-1</sup>**

Numbers of leaves branches<sup>-1</sup> were recorded at the time of harvest.

##### **3.3.1.2. 4. Girth of the Stem**

Girth of the stem was measured and recorded at the time of harvest and expressed in cm.

## **3.4. CHEMICAL ANALYSIS**

### **3.4.1. Analysis of Plant**

#### **3.4.1.1. Cellulose Content**

Cellulose content of the various substrates were determined by treating the

acetolysed plant sample with sulphuric acid and the hydrolysed glucose was treated with anthrone reagent (Hodge and Hofreiter, 1962).

0.5 g of the plant sample was taken, 3 ml of acetic: nitric reagent (150 ml of 80 per cent acetic acid + 15 ml of concentrated  $\text{HNO}_3$ ) was added and mixed using a vortex mixer and placed in a water bath at 100 °C for 30 minutes. This mixture was cooled, centrifuged for 20 minutes and the supernatant was discarded. The residue was washed with water, and then 10 ml of 67 per cent  $\text{H}_2\text{SO}_4$  was added and left for 1 hr. 1 ml of this solution was diluted to 100 ml. 1ml of the diluted solution was taken in a test tube and 10 ml of anthrone (200mg anthrone/100ml conc.  $\text{H}_2\text{SO}_4$  prepared fresh and chilled before use) reagent added and mixed well. The tubes were heated in a boiling water bath for 10 minutes, cooled and measured the absorbance at 630 nm. A blank was prepared with anthrone reagent and water. Standard curve was prepared by taking 0.4 , 0.8, 1.2, 1.6, and 2 ml of standard cellulose solution (corresponding to 40-200  $\mu\text{g}$  of cellulose), equalized the volume and anthrone reagent was added and developed the colour as above. Standard cellulose solution was prepared by dissolving 100 mg of cellulose in 10 ml of 67 per cent  $\text{H}_2\text{SO}_4$  and kept for 1hr. A working standard was prepared by diluting the 1 ml of standard cellulose solution to 100 ml (100 $\mu\text{g}/\text{ml}$ ). The results were expressed in terms of per cent cellulose.

#### ***3.4.1.2. Hemicellulose Content***

Hemicellulose content of the various substrates were analysed gravimetrically by treating with neutral detergent solution . Here the samples were refluxed with neutral detergent solution to remove the water solubles and materials other than fibrous component. The left out material was weighed after filtration and expressed as neutral detergent fibre (NDF) (Thimmaiah, 2004).

One gram of the powdered sample was taken in a refluxing flask and 10 ml of cold neutral detergent solution was added (neutral detergent solution - dissolved 18.61 g of disodium ethylene diamine tetra acetate and 6.81 g of sodium borate decahydrate dissolved in 200 ml of water by heating. To this, added 100

ml of solution containing 30 g of sodium lauryl sulphate and 10 ml of 2-ethoxy ethanol. Then added 100 ml of a solution containing 4.5 g of disodium hydrogen phosphate. The pH was adjusted to 7.0 and made up the volume to one litre). 2ml of decahydronaphthalene and 0.5 g sodium sulphite was added to the sample and boiled and refluxed for 60 minutes. The sample mixture was filtered through sintered glass crucible (G-2) by suction and washed with hot water. The residue was washed twice with acetone, transferred to a crucible and dried at 100°C for 8 h. The crucible was cooled in a desiccator and weighed.

Hemicellulose = Neutral detergent fibre (NDF) - acid detergent fibre (ADF).

Acid detergent fibre (ADF) was found from lignin estimation.

#### **3.4.1.3. Lignin Content**

Lignin content of the various substrates was analysed gravimetrically by treating with acid detergent solution. The sample material refluxing in acid detergent solution removed the water solubles and materials other than fibrous component. The left out material was weighed after filtration and treated with 72 per cent H<sub>2</sub>SO<sub>4</sub>, filtered, dried and ashed. The loss of weight on ignition was expressed as the acid detergent lignin (ADL) (Thimmaiah, 2004).

##### **3.4.1.3.1. Acid Detergent Fibre (ADF)**

One g of powdered sample was placed in a round bottom flask and added 100 ml of acid detergent solution prepared by dissolving 20 g of cetyl trimethyl ammonium bromide in one litre of 1N H<sub>2</sub>SO<sub>4</sub>). Sample was heated to boil in 10 minutes. The heating was reduced to avoid foaming as boiling begins. It was then refluxed for 1 hr after onset of boiling. The container was removed, swirled and filtered the contents through a pre-weighed sintered glass crucible (G-2) by suction and washed with hot water twice. The residues were washed with acetone and the lumps were broken. Acetone washing was repeated until the filtrate was colourless. The residues were dried at 100°C overnight, weighed after cooling in a desiccator. Expressed ADF content in percentage *i.e.*  $W/S \times 100$  where W was the weight of the fibre and S was the weight of the sample.

#### 3.4.1.3.2. Determination of Acid Detergent Lignin (ADL)

ADF was transferred to a 100 ml beaker with 50 ml of 72 per cent H<sub>2</sub>SO<sub>4</sub>. 1 g asbestos was added and allowed to stand for 3 hours with intermittent stirring with a glass rod. The acid was diluted with distilled water and filtered with preweighed Whatman No.1 filter paper (wet the filter paper in hot water, dried in oven at 102 °C for 2 hours. It was then cooled in a desiccator and weighed in a cover dish). Glass rod and residue were washed several times to get rid of the acid. The filter paper with residue was dried at 100 °C and weighed after cooling in a desiccators and then transferred the to a preweighed silica crucible and ashed the filter paper with the content in a muffle furnace at 550°C for about 3h. The crucible was cooled in a desiccator and weighed. The ash content was calculated. For blank 1g asbestos was taken, 72 per cent H<sub>2</sub>SO<sub>4</sub> was added and followed the above steps (If the weight loss of asbestos blank on ashing is below 0.002 g/g, the determination of blank was discontinued).

Acid detergent lignin (%) =

$$\frac{\text{Washed fibre (Test - asbestos blank)} - \text{Ash (Test - asbestos blank)} \times 100}{\text{weight of sample}}$$

#### 3.4.1.4. Protein

Analysis was carried out by Kjeldahl's method, which evaluated the total N content of the sample after it has been digested in sulphuric acid with digestion mixture (K<sub>2</sub>SO<sub>4</sub> + CuSO<sub>4</sub> in 1:5 ratio). Crude protein was found out by multiplying the nitrogen content of the substrates with a factor of 6.25.

#### 3.4.1.5. C: N Ratio

Carbon contents of the plant substrates were found out by loss on ignition method. Plant sample in a preweighed silica crucible was weighed and placed in a muffle furnace and ignited for 30 minutes at bright red heat. Then cooled in a desiccator and weighed. The loss in weight represented the loss on ignition of organic matter. This value was multiplied with the factor 0.58 represents the

carbon content of the sample. Nitrogen contents of the substrates were found by kjeldahl method .

Table 3. Standard analytical methods followed in the analysis of plants.

| Sl. No                       | Properties                       | Method  | Reference               |
|------------------------------|----------------------------------|---|-------------------------|
| <b>Elemental composition</b> |                                  |   |                         |
| 1                            | Nitrogen                         | Micro Kjeldahl method   | Jackson (1973)          |
| 2                            | Phosphorus                       | Nitric-perchloric acid digestion (9:4) and spectrophotometry using vanadomolybdophosphoric yellow colour method | Jackson (1973)          |
| 3                            | Potassium                        | Nitric - perchloric acid (9:4) digestion and flame photometry   | Jackson (1973)          |
| 4.                           | Iron, manganese, zinc and copper | Nitric- perchloric acid (9:4) digestion and AAS   | Jackson (1973)          |
| 5.                           | Hot water extractable B Content  | Nitric- perchloric acid (9:4) digestion and Turbidimetry  | Chesnin and Yien (1950) |
| 6.                           | Heavy metals (Pb,Cd, Ni,Sn)      | Nitric- perchloric acid (9:4) Polarographic method in ion tracer equipment                                      | Pinta (1966)            |

### 3.4.2. Analysis of Compost

Table 4. Standard analytical methods followed in the analysis of composts

| Sl. No | Properties       | Method  | Reference      |
|--------|------------------|---|----------------|
| 1.     | Moisture content | Gravimetric method  | Jackson (1973) |
| 2.     | pH               | pH meter  | Jackson (1973) |
| 3.     | EC               | Conductivity meter  | Jackson (1973) |
| 4.     | Ash content      | Loss by ignition  | Jackson (1973) |
| 5.     | Organic matter   | Loss by ignition  | Jackson (1973) |
| 6.     | Nitrogen         | Micro Kjeldahl method   | Jackson (1973) |
| 7.     | Phosphorous      | Nitric-perchloric acid digestion (9:4) and spectrophotometry using vanadomolybdophosphoric yellow colour method | Jackson (1973) |

|     |                                    |   |                             |
|-----|------------------------------------|---|-----------------------------|
| 8.  | Potassium                          | Nitric-perchloric acid digestion (9:4) and flame photometer                             | Jackson (1973)              |
| 9.  | Dehydrogenase activity             | Using 3% Triphenyl Tetrazolium Chloride and spectrophotometry                           | Casida <i>et al.</i> (1964) |
| 10. | Heavy metal content (Pb,Cd,Ni, Sn) | Nitric-perchloric acid digestion (9:4) and Polarography method in ion tracer equipment. | Pinta (1966)                |

#### 3.4.2.1. Cellulase Activity

Cellulase activity was estimated as per the method suggested by Pancholy and Rice (1973). Five gram of air dried compost was taken in a 100 ml Erlen Meyer flask. Ten ml of acetate buffer and 1 per cent carboxy methyl cellulose were added. Flasks were incubated for 24 hrs at 37°C and left undisturbed. After the incubation, 50 ml of the filtrate was taken and 4 ml of anthrone reagent was added. The intensity of the green colour developed was read in a spectrophotometer at 620 nm. Glucose was used as standard at different concentrations for the preparation of standard calibration graph. The results were then expressed as the amount of glucose hydrolysed  $\text{g}^{-1}$  of soil  $24 \text{ hrs}^{-1}$  in ppm.

#### 3.4.3. Analysis of Soil

##### 3.4.3.1. Collection of Soil Sample

Soils were collected by the method of destructive sampling of the plants. Plants were uprooted and the soils were collected in polythene bags. These soils were stored in deep freezers to ensure the viability of microorganisms. Soil for chemical analysis were collected, dried in shade, powdered with a wooden mallet, sieved through a 2 mm sieve and stored in polythene containers.

### 3.4.3.2. Soil Characteristics

Table 5. Standard analytical methods followed in the analysis of soil

| Sl. No                                   | Properties                              | Method   | Reference                       |
|--|---|--|---------------------------------|
| <b><i>Physical Properties</i></b>        |   |  |                                 |
| 1.                                       | Bulk density                            | Core method  | Black <i>et al.</i> (1965)      |
| 2.                                       | Particle density                        | Pycnometer method  | Black <i>et al.</i> (1965)      |
| 3.                                       | Water holding capacity                  | Core method  | Gupta and Dakshinamurthy (1980) |
| <b><i>Chemical Properties</i></b>        |   |  |                                 |
| 4.                                       | pH                                      | pH meter (1:5 soil water ratio)                                    | Jackson (1973)                  |
| 5.                                       | EC                                      | Conductivity meter (1:5 soil water ratio)                          | Jackson (1973)                  |
| 6.                                       | Organic carbon                          | Walkley and Black rapid titration method                           | Walkley and Black (1934)        |
| 7.                                       | Available N                             | Alkaline potassium permanganate method                             | Subbiah and Asija (1956)        |
| 8.                                       | Available P                             | Bray No.1 extraction and Spectrophotometry                         | Jackson (1973)                  |
| 9.                                       | Available K                             | Neutral Normal NH <sub>4</sub> OAC extraction and Flame photometry | Jackson (1973)                  |
| 10.                                      | 0.5 N HCl extractable Fe, Zn, Cu and Mn | Atomic Absorption Spectrophotometry                                | O'Connor (1988)                 |
| 11.                                      | Hotwater extractable B Content          | Turbidimetry   | Chesnin and Yien (1950)         |
| <b><i>Microbiological Properties</i></b> |   |  |                                 |
| 12.                                      | Bacterial population                    | Serial dilution and plate count method                             | Timonin, 1940                   |
| 13.                                      | Fungal population                       | Serial dilution and plate count method                             | Timonin, 1940                   |
| 14.                                      | Actinomycetes population                | Serial dilution and plate count method                             | Timonin, 1940                   |



#### **3.4.3.3. Urease Activity**

The urease activity was determined by following the method described by Broadbent *et al.* (1964).

Twenty gram soil was weighed into an Erlenmayer flask, to which 4 ml of urea substrate solution was added. Enough water was added to each flask to maintain a tension of 1/3 bar and incubated for 24 hours at 30<sup>0</sup>C. Then the flasks were removed and CaSO<sub>4</sub> solution was added to make up the volume to 100 ml. About 15 ml of the supernatant was taken and colour was developed by adding 10 ml of p-dimethyl amino benzaldehyde which was then read in a Spectrophotometer at a wavelength of 420 nm. Standards were also prepared by using urea solutions of known concentrations. The results were expressed in terms of urea hydrolysed g<sup>-1</sup> of soil hr<sup>-1</sup> in ppm.

#### **3.4.3.4. Phosphatase Activity**

The phosphatase activity was determined by following a procedure described by Eivazi and Tabatabai (1977).

To 1 g soil in a 50 ml Erlen Meyer flask, 0.2 ml toluene, 4 ml modified universal buffer (pH-6.5) and 1ml p-nitrophenyl phosphate solution were added and incubated at 23<sup>0</sup>C for one hour. After incubation, 0.5 ml CaCl<sub>2</sub> (1ml) and 0.05M NaOH (1ml) were added. The contents were swirled and filtered through Whatman No.2 filter paper and the intensity of yellow colour developed was read in a spectrophotometer at a wavelength of 420 nm. One percent of p-nitrophenyl phosphate was used for the preparation of standards. The results were expressed in terms of p-nitrophenol hydrolysed g<sup>-1</sup> of soil hr<sup>-1</sup> in micrograms.

#### **3.4.3.5. Arylsulphatase Activity**

Aryl sulphatase activity in soil was determined using modifications of the aryl sulphatase assay of Tabatabai and Bremner (1970) suggested by Pettit *et al.* (1977), Sarathchandra and Perrot (1981) and Tabatabai (1994).

One gram soil was pre-incubated for 1 hr with 0.2 ml toluene to inhibit enzyme activity from enzyme proliferation and de novo enzyme synthesis. 4 ml



Plate 4. A view of the soil phosphatase enzyme activity study



Plate 5. A view of the soil dehydrogenase enzyme activity study

of 0.5 M sodium acetate buffer (pH-5.8) and 1 ml 0.05 M p-nitrophenyl sulphate were added and the mixture was incubated for 1 hr at 37°C. The reaction was terminated by cooling to 0 °C in an ice bath. And the samples were centrifuged at 11000 rpm for 10 minutes to collect the supernatant. 3 ml of the supernatant liquid were combined with 2 ml of 0.5 M NaOH and the absorbance of the yellow colour was measured at 400 nm using a spectrophotometer. The p- nitrophenol released by the soil arylsulphatase enzyme was calculated by referring to a standard calibration curve developed using 10- 50 µg p- nitrophenol.

#### 3.4.3.6. Dehydrogenase Activity

Dehydrogenase activity was estimated as per the procedure described by Casida *et al.*, 1964.

Sixty g of the air dried soil was weighed to a 250 ml Erlen Meyer flask. One ml of 3 per cent Triphenyl Tetrazolium Chloride was added and incubated for 24 hrs at 27°C. After incubation, the soil was quantitatively transferred to a glass funnel and was given methanol washings consecutively till the volume reached 100 ml. The colour intensity was then read in a Spectrophotometer at 485 nm. A series of standards were used for preparing the calibration curve. The results were expressed in terms of Triphenyl Formazon hydrolysed g<sup>-1</sup> of soil 24 hrs<sup>-1</sup> in micrograms.

#### 3.4.3.7. β-D Glucosidase

β-D glucosidase activity was determined as described by Eivazi and Tabatabai, 1988.

One gram soil was placed in a test tube and treated with 4 ml of modified universal buffer (MUB, pH 6) and 1 ml of 0.5 M p- nitrophenyl β- D- glucopyranoside substrate solution. The solution was mixed thoroughly and allowed to incubate in the dark for 1 hour at 37°C. After incubation, the reaction product was stopped and the yellow colour from the p- nitrophenol was developed by the addition of 1 ml 0.5 M calcium chloride and 4 ml of Tris buffer (pH 10). The solution was mixed and filtered. The p- nitrophenol determined by



measuring the absorption on a spectrophotometer at a wavelength of 405 nm and quantified by comparison with a standard curve with 0 to 5 µg p- nitrophenyl.

#### **3.4.3.8. Soil Respiratory Activity**

The respiratory activity of the soil samples were estimated using the method outlined by Jenkinson and Powlson (1976).

#### **3.4.3.9. Microbial Load**

Microbial count in the soil was enumerated using serial dilution technique proposed by Timonin (1940). Composition of the media was presented in Appendix III.

| Sl no. | Microflora    | Medium                    |
|--------|---------------|---------------------------|
| 1      | Actinomycetes | Ken knight's agar         |
| 2      | Fungi         | Martins' Rose Bengal agar |
| 3      | Bacteria      | Nutrient agar             |

#### **3.5. Statistical analysis**

The data generated from these experiments were subjected to the analysis of variance as per the design and their significance was tested by the F test (Snedecor and Cochran, 1975). In the cases where the effects were found to be significant, CD was calculated using standard techniques.



Plate 6. A view of the soil respiratory activity study

## *Results*

## 4. RESULTS

A study entitled “Characterization, conversion and evaluation of selected lignocellulosic biomass” has been carried out at the Department of Soil Science and Agricultural Chemistry, College of Agriculture, Vellayani, Thiruvananthapuram during August 2013- February 2014. Four substrates and four inoculants were included in the study for evaluating their efficiency for composting, kind of inoculants on different types of substrates evaluation of each compost for its efficiency and effect of each compost on the yield and yield attributes of the test crop amaranthus. Results based on statistically analysed data pertaining to the experiment conducted during the course of investigation are presented in this chapter.

### 4.1. PROXIMATE ANALYSIS OF DIFFERENT SUBSTRATES

The data on the cellulose, hemicellulose, lignin, protein, C:N ratio, NPK content and heavy metal contents of different substrates are presented in Tables 6 to 8 respectively.

#### 4.1.1. Cellulose Content

Different substrates analyzed vary significantly in their cellulose content. The mean values ranged from 25.96 to 33.41 per cent. The substrate S<sub>2</sub> (coir pith) registered the highest mean value of 33.41 per cent which was on par with S<sub>3</sub> (water hyacinth) with mean value of 32.35 while lowest mean value was registered by S<sub>1</sub> (water cabbage) as 25.96 per cent cellulose content.

#### 4.1.2. Hemicellulose Content

All the four substrates varied significantly in hemicelluloses content (Table 6). The mean values ranged from 0.45 to 28.40 per cent. The substrate S<sub>3</sub> (water hyacinth) has recorded the highest mean value (28.40 per cent) which was significantly different from all other substrates followed by S<sub>4</sub> (farm waste) and S<sub>1</sub> (water cabbage) with, mean values of 12.02 and 4.4 respectively. The lowest hemicelluloses content of 0.45 per cent was recorded for S<sub>2</sub> (coir pith).

Table 6. Biochemical composition of different substrates, %

| Substrates     | Cellulose | Hemicelluloses | Lignin | Protein |
|----------------|-----------|----------------|--------|---------|
| S <sub>1</sub> | 25.96     | 4.40           | 33.20  | 17.11   |
| S <sub>2</sub> | 33.41     | 0.45           | 64.76  | 0.45    |
| S <sub>3</sub> | 32.35     | 28.40          | 17.00  | 9.90    |
| S <sub>4</sub> | 29.83     | 12.02          | 21.58  | 3.71    |
| CD             | 1.504     | 0.888          | 1.809  | 2.161   |

Table 7. Major nutrients of different substrates

| Substrates     | Total Nitrogen (%) | Total Phosphorous (%) | Total Potassium(%) | C:N Ratio |
|----------------|--------------------|-----------------------|--------------------|-----------|
| S <sub>1</sub> | 2.74               | 0.30                  | 0.33               | 16.98     |
| S <sub>2</sub> | 0.07               | 0.02                  | 0.01               | 243.59    |
| S <sub>3</sub> | 1.59               | 0.30                  | 0.25               | 27.49     |
| S <sub>4</sub> | 0.59               | 0.15                  | 0.24               | 86.95     |
| CD             | 0.035              | 0.025                 | 0.032              | 14.576    |

Table 8. Heavy metals of different substrates

| Substrates     | Pb (mg kg <sup>-1</sup> ) | Cd (µg kg <sup>-1</sup> ) | Ni (mg kg <sup>-1</sup> ) | Sn (µg kg <sup>-1</sup> ) |
|----------------|---------------------------|---------------------------|---------------------------|---------------------------|
| S <sub>1</sub> | Non detectable            | Non detectable            | 0.55                      | Non detectable            |
| S <sub>2</sub> | 0.01                      | 0.02                      | 0.97                      | Non detectable            |
| S <sub>3</sub> | 1.15                      | 0.06                      | 0.57                      | 0.56                      |
| S <sub>4</sub> | 0.03                      | 0.16                      | 0.51                      | Non detectable            |
| CD             | -                         | -                         | 0.288                     | -                         |

- S<sub>1</sub> Water cabbage  
 S<sub>2</sub> Coir pith  
 S<sub>3</sub> Water hyacinth  
 S<sub>4</sub> Farm wastes



#### 4.1.3. Lignin Content

Lignin content of substrates ranged from 17.00 to 64.76 per cent (Table 6). Substrate S<sub>2</sub> (coir pith) with highest mean value of 64.76 per cent was found to be significantly superior to all other substrates and was followed by S<sub>1</sub> (water cabbage) and S<sub>4</sub> (farm waste) with mean values of 33.20 and 21.58 per cent respectively. Substrate S<sub>3</sub> (water hyacinth) was found to have the lowest lignin content with mean value of 17 per cent.

#### 4.1.4. Proteins

The results of the chemical analysis of the substrate revealed that all the four substrates varied significantly in their protein content (Table 6) and mean values ranged from 0.45 to 17.11 per cent. The highest protein content was recorded by the substrate S<sub>1</sub> (water cabbage) with 17.11 per cent followed by S<sub>3</sub> (water hyacinth) with 9.90 per cent and S<sub>4</sub> (farm waste) (3.71 per cent) while the lowest protein content was recorded by S<sub>2</sub> (coir pith) with mean value of 0.45 per cent.

#### 4.1.5. NPK Content

Table 7 clearly depicts that all the four substrates were statistically different in total N content with mean value ranged from 0.07 to 2.74 per cent. Substrate S<sub>1</sub> (water cabbage) recorded the highest N content of 2.74 per cent followed by S<sub>3</sub> (water hyacinth) (1.59 per cent) and S<sub>4</sub> (farm waste) (0.59 per cent). S<sub>2</sub> (coir pith) recorded lowest mean value of 0.07 per cent.

Total P content, varied from 0.02 to 0.30 per cent and the highest value was recorded by S<sub>1</sub> (water cabbage) and S<sub>3</sub> (water hyacinth) with 0.30 per cent P content. S<sub>4</sub> (farm waste) has recorded a mean value of 0.15 per cent which was significantly superior to the lowest recorded value of S<sub>2</sub> (coir pith) (0.02 per cent) while inferior to S<sub>1</sub> (water cabbage) and S<sub>3</sub> (water hyacinth).

The total K content of the substrates ranged from 0.01 to 0.33 per cent. The highest mean value was recorded for the substrate S<sub>1</sub> (water cabbage) with 0.33 per cent K followed by S<sub>3</sub> (water hyacinth) (0.25 per cent) which was on par

with S<sub>4</sub> (farm waste) (0.24 per cent) while S<sub>2</sub> (coir pith) was recorded the lowest value of total K (0.01 per cent).

#### 4.1.6. C:N Ratio

The C:N ratio of different substrates ranged from 16.98 to 243.59 (Table 7). The wider C:N ratio of 243.59 was recorded by substrate S<sub>2</sub> (coir pith) which was significantly different from others followed by S<sub>4</sub> (farm waste) (86.95). The narrow C:N ratio was recorded by S<sub>1</sub> (water cabbage) with mean value of 16.98 and statistically on par with S<sub>3</sub> (water hyacinth) with mean value of 27.49.

#### 4.1.7. Heavy Metal Content

All the substrates were analysed for the heavy metals *viz.* Pb, Cd, Ni and Sn content. The data on heavy metal contents of the substrates were presented in the Table 8. All the heavy metal content did not differ significantly between the degradable wastes used however they significantly differ on Ni content. All the wastes used were on par with each other except coir pith which registered high Ni content of 0.97 mg kg<sup>-1</sup>.

##### 4.1.7.1. Lead Content

Pb content in the substrates were analyzed and found that substrate S<sub>3</sub> (water hyacinth) recorded the highest Pb content of 1.15 mg kg<sup>-1</sup>. Substrate S<sub>4</sub> (farm wastes) recorded the mean value of 0.03 mg kg<sup>-1</sup> followed by S<sub>2</sub> (0.01 mg kg<sup>-1</sup>). But water cabbage has no detectable levels of lead.

##### 4.1.7.2. Cadmium Content

With respect to Cd content of substrates, S<sub>4</sub> (farm waste) recorded the highest value of 0.16 µg kg<sup>-1</sup> followed by S<sub>3</sub> (water hyacinth) (0.06 µg kg<sup>-1</sup>), S<sub>2</sub> (farm waste) (0.02 µg kg<sup>-1</sup>) respectively. No detectable levels of Cd was found in water cabbage.

##### 4.1.7.3. Nickel Content

From Table 8 the Ni content of the substrates ranged from 0.51 mg kg<sup>-1</sup> to 0.97 mg kg<sup>-1</sup>. Coir pith (S<sub>2</sub>) has recorded the highest Ni content of 0.97 mg kg<sup>-1</sup>.

and significantly different from all other substrates which were on par with each other.

#### **4.1.7.4. Tin Content**

Substrate S<sub>3</sub> (water hyacinth) has recorded detectable levels of Sn (0.56 µg kg<sup>-1</sup>) and other substrates contain below the detectable level of Sn.

## **4.2. CHARACTERIZATION OF COMPOSTS**

### **4.2.1. Physico-Chemical Properties of Composts**

The physico chemical properties of composts are presented in the Table 9.

#### **4.2.1.1. Moisture Content**

Statistical analysis of the data (Table 9) on moisture content revealed that the moisture content was significantly influenced by the substrates, inoculants and their interactions. The highest moisture content was recorded by the S<sub>1</sub>I<sub>3</sub> (water cabbage+ Composting Inoculum) with mean value of 61.67 per cent and was on par with S<sub>2</sub>L<sub>4</sub> (59.76 per cent) and S<sub>3</sub>I<sub>2</sub> (58.93 per cent). The lowest moisture content was recorded by S<sub>4</sub>I<sub>1</sub> (11.74 per cent) which was significantly lower than all other treatments. Among the interactions, I<sub>3</sub> (Composting Inoculum) was found to be more effective on substrate S<sub>1</sub> (water cabbage) and S<sub>4</sub> (farm waste), for substrate S<sub>2</sub> (coir pith), L<sub>4</sub> (commercial enzyme cocktail) was found to be most effective and I<sub>2</sub> (*P. sajor caju*) was effective on S<sub>3</sub>(water hyacinth). Substrates were found to have individual effect on the moisture content. The highest value was recorded by S<sub>2</sub> with moisture content of 54.49 per cent followed by S<sub>3</sub> (54.29 per cent) while the lowest value was recorded for S<sub>4</sub> (18.72 per cent). Individual effects of four different inoculants on substrates were also found to be significant. I<sub>3</sub> recorded the highest value of 47.60 followed by L<sub>4</sub> (46.81 per cent) and I<sub>1</sub> recorded lowest mean value of 39.91 per cent.

#### **4.2.1.2. pH**

From Table 9 it was observed that all the four substrates and their interaction with inoculants were found to have significant effect on pH of the composts. All the composts have more or less neutral pH range however the

Table 9. Physico chemical properties of the composts as influenced by substrates, inoculants and their combinations.

| Treatments                    | Moisture content (%) | pH    | EC (dSm <sup>-1</sup> ) | Ash content (%) |
|-------------------------------|----------------------|-------|-------------------------|-----------------|
| S <sub>1</sub> I <sub>1</sub> | 40.70                | 7.39  | 1.89                    | 24.13           |
| S <sub>1</sub> I <sub>2</sub> | 37.59                | 7.45  | 2.41                    | 38.13           |
| S <sub>1</sub> I <sub>3</sub> | 61.67                | 6.89  | 0.88                    | 59.69           |
| S <sub>1</sub> I <sub>4</sub> | 54.92                | 6.66  | 1.72                    | 61.76           |
| S <sub>2</sub> I <sub>1</sub> | 53.88                | 6.74  | 0.35                    | 36.02           |
| S <sub>2</sub> I <sub>2</sub> | 54.50                | 6.72  | 0.42                    | 35.48           |
| S <sub>2</sub> I <sub>3</sub> | 49.82                | 6.31  | 0.76                    | 74.69           |
| S <sub>2</sub> I <sub>4</sub> | 59.76                | 6.70  | 0.39                    | 73.23           |
| S <sub>3</sub> I <sub>1</sub> | 53.33                | 7.03  | 2.80                    | 48.89           |
| S <sub>3</sub> I <sub>2</sub> | 58.93                | 7.17  | 1.80                    | 63.51           |
| S <sub>3</sub> I <sub>3</sub> | 55.08                | 7.57  | 3.79                    | 56.58           |
| S <sub>3</sub> I <sub>4</sub> | 49.82                | 7.47  | 1.80                    | 53.00           |
| S <sub>4</sub> I <sub>1</sub> | 11.74                | 7.31  | 1.03                    | 44.41           |
| S <sub>4</sub> I <sub>2</sub> | 16.53                | 7.64  | 1.22                    | 52.97           |
| S <sub>4</sub> I <sub>3</sub> | 23.85                | 7.57  | 0.69                    | 48.55           |
| S <sub>4</sub> I <sub>4</sub> | 22.75                | 7.47  | 1.12                    | 56.94           |
| S <sub>1</sub>                | 48.72                | 7.10  | 1.72                    | 45.77           |
| S <sub>2</sub>                | 54.49                | 6.62  | 0.48                    | 45.93           |
| S <sub>3</sub>                | 54.29                | 7.31  | 2.55                    | 54.86           |
| S <sub>4</sub>                | 18.72                | 7.50  | 1.01                    | 55.49           |
| I <sub>1</sub>                | 39.91                | 7.12  | 1.52                    | 38.37           |
| I <sub>2</sub>                | 41.89                | 7.25  | 1.46                    | 47.52           |
| I <sub>3</sub>                | 47.60                | 7.08  | 1.53                    | 59.88           |
| I <sub>4</sub>                | 46.81                | 7.07  | 1.26                    | 61.23           |
| CD-S I(0.05)                  | 3.105                | 0.428 | 0.115                   | 3.694           |
| CD -S(0.05)                   | 1.552                | 0.214 | 0.058                   | 1.847           |
| CD-I(0.05)                    | 1.552                | 0.214 | 0.058                   | 1.847           |

S<sub>1</sub> Water cabbage  
 S<sub>2</sub> Coir pith  
 S<sub>3</sub> Water hyacinth  
 S<sub>4</sub> Farm wastes

I<sub>1</sub> *Trichoderma reesei*  
 I<sub>2</sub> *Pleurotus sajorcaju*  
 I<sub>3</sub> Composting inoculum  
 I<sub>4</sub> Commercial enzyme cocktail

treatment S<sub>4</sub>I<sub>2</sub> (farm waste+ *P. sajor caju*) recorded the highest pH value of 7.64 which was on par with S<sub>3</sub>I<sub>3</sub> (7.57), S<sub>4</sub>I<sub>3</sub> (7.57), S<sub>4</sub>I<sub>4</sub> (7.47), S<sub>3</sub>I<sub>4</sub> (7.47), S<sub>1</sub>I<sub>2</sub> (7.45) and S<sub>1</sub>I<sub>1</sub> (7.39) and the lowest pH was recorded by the interaction S<sub>2</sub>I<sub>3</sub> (6.31). On substrates S<sub>1</sub> (water cabbage) and S<sub>4</sub> (farm waste) inoculant I<sub>2</sub> (*P. sajor caju*) was most effective and for S<sub>2</sub> (coir pith), I<sub>1</sub> (*T.reesei*) was effective and for S<sub>3</sub>(water hyacinth), I<sub>3</sub> (composting Inoculum) was found to be effective. The substrates have significant individual effect on pH as S<sub>4</sub> was recorded with the highest mean value of 7.50 and S<sub>2</sub> with lowest value of 6.62. The individual effect of inoculants on substrates were found to be non significant, even though the highest mean value was recorded for I<sub>2</sub> with mean value of 7.25.

#### 4.2.1.3. EC

In the case of EC (Table 9) all the four substrates, inoculants and their interactions were found to have significant effect on EC of the respective composts. The highest mean value for EC was recorded for the treatment S<sub>3</sub>I<sub>3</sub> (water hyacinth + Composting Inoculum) with mean value of 3.79 dS m<sup>-1</sup> while second highest mean value was recorded by S<sub>3</sub>I<sub>1</sub> (2.80 dS m<sup>-1</sup>) which was significantly different from the former. The lowest EC was recorded for the treatment S<sub>2</sub>I<sub>1</sub> with mean value of 0.35 dS m<sup>-1</sup>. On substrates S<sub>1</sub> (water cabbage) and S<sub>4</sub> (farm waste), inoculant I<sub>2</sub> (*P. sajor-caju*) was found to be effective, and for S<sub>2</sub> (coir pith) and S<sub>3</sub>(water hyacinth), inoculant I<sub>3</sub> (Composting Inoculum) was effective. The substrates also had individual effect on EC and the substrate S<sub>3</sub> was recorded the highest EC of 2.55 dS m<sup>-1</sup> and lowest value for EC was recorded with the substrate S<sub>2</sub> (0.48 dS m<sup>-1</sup>). The inoculants also have individual effect on EC of the compost. I<sub>3</sub> was reported to have the highest influence on EC as the mean value was recorded as 1.53 dS m<sup>-1</sup> and I<sub>4</sub> recorded the lowest mean value of 1.26 dS m<sup>-1</sup>.

#### 4.2.1.4. Ash Content

Ash content of the composts were statistically analysed and it was noticed from Table 9 that the ash content of composts were significantly influenced by the substrates, inoculants and their interactions. The highest mean value was recorded

by the interaction S<sub>2</sub>I<sub>3</sub> (coir pith+ Composting Inoculum) with mean value of 74.69 per cent followed by S<sub>2</sub>I<sub>4</sub> (73.23 per cent) which was on par with the former while the lowest ash content was recorded for S<sub>1</sub>I<sub>1</sub> (24.13 per cent). Regarding the effect of inoculants, substrates S<sub>1</sub> (water cabbage) and S<sub>4</sub> (farm waste) are influenced mostly by I<sub>4</sub> (commercial enzyme cocktail), and for S<sub>2</sub> (coir pith), I<sub>3</sub> (Composting Inoculum) was found to be most effective and I<sub>2</sub> (*P. sajor-caju*) was effective on S<sub>3</sub> (water hyacinth). Considering the individual effect of substrates, it was found to have significant effect on ash content. S<sub>4</sub> with 55.49 per cent ash content was recorded as the highest value and the S<sub>1</sub> with 45.77 per cent was recorded with the lowest ash content. It was obvious from the data that the inoculants were found to have significant role in the ash content of the composts as I<sub>4</sub> recorded the highest ash content of 61.23 per cent and I<sub>1</sub> recorded the lowest ash content of 38.37 per cent.

#### 4.2.1.5. Organic Matter

Statistical analysis of the data indicated that organic matter content (Table 10) of the composts was significantly influenced by the substrates, inoculants and their interactions. The highest organic matter content was recorded by the interaction of S<sub>1</sub>I<sub>1</sub> (water cabbage+ *T. reesei*) with mean value of 76.75 per cent and second highest value was recorded by S<sub>2</sub>I<sub>2</sub> (64.71 per cent) which was on par with S<sub>2</sub>I<sub>1</sub> (63.78 per cent) and S<sub>1</sub>I<sub>2</sub> (63.00 per cent). But both treatments varied significantly from S<sub>1</sub>I<sub>1</sub>. The lowest mean value was recorded by S<sub>2</sub>I<sub>3</sub> (26.24 per cent) which was on par with S<sub>2</sub>I<sub>4</sub> (26.97 per cent). Inoculant I<sub>1</sub> (*T. reesei*) was found to be effective on substrates S<sub>1</sub> (water cabbage), S<sub>3</sub> (water hyacinth) and S<sub>4</sub> (farm waste) and for S<sub>2</sub> (coir pith), I<sub>2</sub> (*P. sajor-caju*) was most effective. Considering the individual effects of substrates and inoculants, both were found to have significant influence on organic matter content. The substrate S<sub>1</sub> (water cabbage) recorded the highest value of 54.57 per cent and S<sub>3</sub> (water hyacinth) recorded the lowest organic matter content of 44.57 per cent. Regarding the individual effect of inoculants on substrates for organic matter was found to be significant. The highest mean value was recorded by the inoculants I<sub>1</sub>

Table 10. Organic matter and NPK content of the composts as influenced by substrates, inoculants and their interactions.

| Treatments                    | Organic matter (%) | Total N (%) | Total P (%) | Total K (%) |
|-------------------------------|--------------------|-------------|-------------|-------------|
| S <sub>1</sub> I <sub>1</sub> | 76.75              | 2.12        | 0.50        | 0.47        |
| S <sub>1</sub> I <sub>2</sub> | 63.00              | 2.58        | 0.48        | 0.67        |
| S <sub>1</sub> I <sub>3</sub> | 40.30              | 3.83        | 0.41        | 0.37        |
| S <sub>1</sub> I <sub>4</sub> | 38.24              | 2.40        | 0.38        | 0.79        |
| S <sub>2</sub> I <sub>1</sub> | 63.78              | 1.21        | 0.17        | 0.16        |
| S <sub>2</sub> I <sub>2</sub> | 64.71              | 1.22        | 0.15        | 0.12        |
| S <sub>2</sub> I <sub>3</sub> | 26.24              | 1.16        | 0.08        | 0.19        |
| S <sub>2</sub> I <sub>4</sub> | 26.97              | 1.13        | 0.13        | 0.13        |
| S <sub>3</sub> I <sub>1</sub> | 51.11              | 1.77        | 0.48        | 0.72        |
| S <sub>3</sub> I <sub>2</sub> | 37.83              | 2.03        | 0.51        | 0.69        |
| S <sub>3</sub> I <sub>3</sub> | 42.38              | 2.98        | 0.75        | 1.87        |
| S <sub>3</sub> I <sub>4</sub> | 46.96              | 2.81        | 0.50        | 0.75        |
| S <sub>4</sub> I <sub>1</sub> | 55.24              | 1.58        | 0.27        | 0.56        |
| S <sub>4</sub> I <sub>2</sub> | 46.51              | 1.69        | 0.40        | 0.79        |
| S <sub>4</sub> I <sub>3</sub> | 51.96              | 2.21        | 0.43        | 0.43        |
| S <sub>4</sub> I <sub>4</sub> | 44.76              | 1.74        | 0.39        | 0.75        |
| S <sub>1</sub>                | 54.57              | 2.73        | 0.44        | 0.57        |
| S <sub>2</sub>                | 45.43              | 1.18        | 0.13        | 0.15        |
| S <sub>3</sub>                | 44.57              | 2.40        | 0.56        | 1.01        |
| S <sub>4</sub>                | 49.62              | 1.81        | 0.37        | 0.63        |
| I <sub>1</sub>                | 61.72              | 1.67        | 0.35        | 0.48        |
| I <sub>2</sub>                | 53.01              | 1.88        | 0.38        | 0.57        |
| I <sub>3</sub>                | 40.22              | 2.55        | 0.42        | 0.71        |
| I <sub>4</sub>                | 39.23              | 2.02        | 0.35        | 0.63        |
| CD-S I(0.05)                  | 3.840              | 0.252       | 0.076       | 0.174       |
| CD -S(0.05)                   | 1.920              | 0.126       | 0.038       | 0.087       |
| CD-I(0.05)                    | 1.920              | 0.126       | 0.038       | 0.087       |

S<sub>1</sub> Water cabbage  
 S<sub>2</sub> Coir pith  
 S<sub>3</sub> Water hyacinth  
 S<sub>4</sub> Farm wastes

I<sub>1</sub> *Trichoderma reesei*  
 I<sub>2</sub> *Pleurotus sajorcaju*  
 I<sub>3</sub> Composting inoculum  
 I<sub>4</sub> Commercial enzyme cocktail

(*T. reesei*) with mean value 61.72 per cent and lowest value was recorded by I<sub>4</sub> (commercial enzyme cocktail) with mean value 39.23 per cent.

#### 4.2.1.6. Total Nitrogen

The total N content was significantly influenced by the individual effects of substrate, inoculants and their interactions (Table 10). The highest N content was recorded by the interaction S<sub>1</sub>I<sub>3</sub> (water cabbage+ Composting Inoculum) with mean value of 3.83 per cent. Second highest N content was recorded by S<sub>3</sub>I<sub>3</sub> (2.98 per cent) which was on par with S<sub>3</sub>I<sub>4</sub> (2.81 per cent). The lowest mean value was recorded by S<sub>2</sub>I<sub>4</sub> (1.13 per cent). Regarding the effect of inoculants on substrates, I<sub>3</sub> (Composting Inoculum) was found to be most effective on S<sub>1</sub> (water cabbage), S<sub>3</sub> (water hyacinth) and S<sub>4</sub> (farm waste) and on S<sub>2</sub> (coir pith), I<sub>2</sub> (*P. sajor-caju*) was effective. The individual effects of substrates were also found to be significant and S<sub>1</sub> (water cabbage) recorded the highest N content of 2.73 per cent and S<sub>2</sub> (coir pith) was recorded the lowest value of 1.18 per cent. Inoculants were also have effect on N content as I<sub>3</sub> (Composting Inoculum) recorded the highest value of 2.55 per cent and I<sub>1</sub> (*T. reesei*) recorded the lowest value of 1.67 per cent.

#### 4.2.1.7. Total Phosphorous

The total P (Table 10) content of composts was significantly influenced by the individual effects of substrates, inoculants and by their interaction also. The highest P content was recorded by S<sub>3</sub>I<sub>3</sub> (water hyacinth+ Composting Inoculum) with mean value of 0.75 per cent and second highest value was recorded by S<sub>3</sub>I<sub>2</sub> (0.51 per cent) which was on par with S<sub>1</sub>I<sub>1</sub> (0.50 per cent) and the lowest mean value was recorded by S<sub>2</sub>I<sub>3</sub> (0.08 per cent). Among the interactions, on S<sub>1</sub> (water cabbage) and S<sub>2</sub> (coir pith) inoculant I<sub>1</sub> (*T. reesei*) was found to be effective and on S<sub>3</sub> (water hyacinth) and S<sub>4</sub> (farm waste) inoculants I<sub>3</sub> (Composting Inoculum) was most effective. Regarding the individual effect of substrates and inoculants, S<sub>3</sub> (water hyacinth) recorded the highest mean value of 0.56 per cent and S<sub>2</sub> (coir pith) recorded the minimum value of 0.13 and for inoculants, I<sub>3</sub> (Composting Inoculum) recorded the highest mean value 0.42 and I<sub>4</sub> (commercial enzyme cocktail) and I<sub>1</sub> (*T. reesei*) recorded the minimum value of 0.35 per cent.



#### 4.2.1.8. Total Potassium

Perusal of the data (Table 10) indicated that substrates, inoculants and their interactions have significant effect on the total K content of composts. The highest mean value was recorded by S<sub>3</sub>I<sub>3</sub> (water hyacinth+ Composting Inoculum) (1.87 per cent) and it was followed by S<sub>4</sub>I<sub>2</sub> (0.79 per cent). But both were statistically different. The lowest K content was recorded by S<sub>2</sub>I<sub>2</sub> with mean value of 0.12 per cent. Regarding the interactions, on substrate S<sub>1</sub> (water cabbage), inoculants I<sub>4</sub> (commercial enzyme cocktail) was found to be most effective, and on S<sub>2</sub> (coir pith) and S<sub>3</sub> (water hyacinth), inoculant I<sub>3</sub> (Composting Inoculum) was most effective and for S<sub>4</sub> (farm waste), inoculants I<sub>2</sub> (*P. sajor-caju*) was effective on total K content of the compost. When the individual effects were considered, the substrate S<sub>3</sub> was noticed with highest mean value 1.01 per cent and lowest mean value was recorded with S<sub>2</sub> (0.15 per cent) and for inoculant, the highest mean value was recorded for I<sub>3</sub> (0.71 per cent) and lowest was recorded for I<sub>1</sub> (0.48per cent).

#### 4.2.2. Maturity Indices of Composts

Maturity indices of composts are presented in the Table 11.

##### 4.2.2.1. C:N ratio

The data analysis showed that the C:N ratio of the composts were significantly affected by the substrates, inoculants as well as their interactions (Table 11). The widest C:N ratio was recorded by the treatment S<sub>2</sub>I<sub>1</sub> (coir pith+ *T. reesei*) with the mean value of 30.64 which was on par with S<sub>2</sub>I<sub>2</sub>(coir pith+ *P. sajor-caju*) (30.35) and the narrow C:N ratio was reported by S<sub>1</sub>I<sub>3</sub> with 5.86 followed by S<sub>3</sub>I<sub>3</sub> with mean value of 8.24 which was significantly different from the former treatment. By analyzing the interaction effect of the substrates on different inoculants, inoculant I<sub>1</sub> (*T. reesei*) has recorded with the widest C:N ratio on all the substrates and inoculants I<sub>3</sub> (Composting Inoculum) has recorded the narrowest C:N ratios for all the four substrates. Considering the individual effect of substrates, S<sub>2</sub> (coir pith) (22.36) recorded the widest C:N and S<sub>3</sub>(water hyacinth) (11.33) with the narrowest C:N ratio and in case of individual effect of

Table I. Maturity indices of composts

| Treatments                    | C:N ratio | Dehydrogenase activity | Cellulase activity | Maturity period(days) |
|-------------------------------|-----------|------------------------|--------------------|-----------------------|
| S <sub>1</sub> I <sub>1</sub> | 20.93     | 717.51                 | 308.14             | 49.00                 |
| S <sub>1</sub> I <sub>2</sub> | 12.94     | 775.05                 | 119.15             | 46.33                 |
| S <sub>1</sub> I <sub>3</sub> | 5.86      | 567.71                 | 407.68             | 44.33                 |
| S <sub>1</sub> I <sub>4</sub> | 9.78      | 491.94                 | 316.86             | 47.67                 |
| S <sub>2</sub> I <sub>1</sub> | 30.64     | 82.97                  | 345.80             | 80.33                 |
| S <sub>2</sub> I <sub>2</sub> | 30.35     | 68.25                  | 387.42             | 83.33                 |
| S <sub>2</sub> I <sub>3</sub> | 13.86     | 114.27                 | 401.39             | 79.67                 |
| S <sub>2</sub> I <sub>4</sub> | 14.58     | 72.04                  | 394.37             | 86.67                 |
| S <sub>3</sub> I <sub>1</sub> | 16.82     | 491.66                 | 381.45             | 65.67                 |
| S <sub>3</sub> I <sub>2</sub> | 10.56     | 427.24                 | 322.87             | 66.00                 |
| S <sub>3</sub> I <sub>3</sub> | 8.24      | 610.38                 | 232.76             | 63.00                 |
| S <sub>3</sub> I <sub>4</sub> | 9.69      | 479.94                 | 304.50             | 67.67                 |
| S <sub>4</sub> I <sub>1</sub> | 20.31     | 414.02                 | 293.02             | 77.33                 |
| S <sub>4</sub> I <sub>2</sub> | 16.91     | 510.12                 | 200.60             | 78.33                 |
| S <sub>4</sub> I <sub>3</sub> | 13.64     | 562.74                 | 390.38             | 74.67                 |
| S <sub>4</sub> I <sub>4</sub> | 14.94     | 443.44                 | 352.57             | 78.00                 |
| S <sub>1</sub>                | 12.38     | 638.05                 | 262.96             | 46.83                 |
| S <sub>2</sub>                | 22.36     | 84.38                  | 382.25             | 82.50                 |
| S <sub>3</sub>                | 11.33     | 502.30                 | 310.39             | 65.58                 |
| S <sub>4</sub>                | 16.45     | 482.58                 | 309.14             | 77.08                 |
| I <sub>1</sub>                | 22.17     | 426.54                 | 332.10             | 68.08                 |
| I <sub>2</sub>                | 17.69     | 445.16                 | 232.51             | 68.50                 |
| I <sub>3</sub>                | 10.40     | 463.77                 | 358.05             | 65.42                 |
| I <sub>4</sub>                | 12.25     | 371.84                 | 342.07             | 70.00                 |
| CD-S I(0.05)                  | 2.319     | 54.157                 | 61.622             | 1.248                 |
| CD -S(0.05)                   | 1.159     | 27.078                 | 30.811             | 0.624                 |
| CD-I(0.05)                    | 1.159     | 27.078                 | 30.811             | 0.624                 |

Dehydrogenase activity - $\mu\text{g TPFhydrolysed g}^{-1}\text{soil 24 hrs}^{-1}$

Cellulase activity-  $\mu\text{g glucose hydrolyzed g}^{-1}\text{hr}^{-1}$

S<sub>1</sub> Water cabbage

S<sub>2</sub> Coir pith

S<sub>3</sub> Water hyacinth

S<sub>4</sub> Farm wastes

I<sub>1</sub> *Trichoderma reesei*

I<sub>2</sub> *Pleurotus sajor-caju*

I<sub>3</sub> Composting inoculum

I<sub>4</sub> Commercial enzyme cocktail

inoculants, I<sub>1</sub> (*T. reesei*) (22.17) recorded the widest C:N ratio and I<sub>3</sub> (Composting Inoculum) (10.40) has recorded the narrowest value.

#### 4.2.2.2. Dehydrogenase Enzyme Activity

Statistical analysis of the data indicated that the dehydrogenase enzyme activity (Table 11) was significantly influenced by the substrates, inoculants and their interactions. The highest mean value was recorded by S<sub>1</sub>I<sub>2</sub> (water cabbage + *T. reesei*) (775.05 µg TPF hydrolyzed g<sup>-1</sup>24 hr<sup>-1</sup>) and the second highest value was recorded by S<sub>1</sub>I<sub>1</sub> (717.51 µg TPF hydrolyzed g<sup>-1</sup>24 hr<sup>-1</sup>). S<sub>2</sub>I<sub>2</sub> was found to have lowest mean value of 68.25 µg TPF hydrolyzed g<sup>-1</sup>24 hr<sup>-1</sup>. Regarding the interactions, all the substrates except S<sub>1</sub> (water cabbage), were influenced by inoculant I<sub>3</sub> (Composting Inoculum) and inoculant I<sub>2</sub> (*P. sajor-caju*) was found to be effective on substrate S<sub>1</sub>(water cabbage). Regarding the individual effect of substrates on dehydrogenase activity, S<sub>1</sub> (water cabbage), was found to have highest mean value of 638.05 µg TPF hydrolyzed g<sup>-1</sup>24 hr<sup>-1</sup> and lowest value was recorded by S<sub>2</sub> (coir pith) as 84.38 µg TPF hydrolyzed g<sup>-1</sup>24 hr<sup>-1</sup>. For the individual effect of inoculants, I<sub>3</sub> ( Composting Inoculum) was recorded with highest dehydrogenase activity of 463.77 µg TPF hydrolyzed g<sup>-1</sup>24 hr<sup>-1</sup> and lowest mean value was recorded by I<sub>4</sub> (commercial enzyme cocktail) (371.84 µg TPF hydrolyzed g<sup>-1</sup>24 hr<sup>-1</sup>).

#### 4.2.2.3. Cellulase Activity

Statistical analysis of the data revealed that the cellulase enzyme activity (Table 11) was significantly influenced by the individual effects of substrates, inoculants and also by their interactions. The more cellulase activity was observed under S<sub>1</sub>I<sub>3</sub> (water cabbage + Composting Inoculum) with mean value of 407.68 µg glucose hydrolyzed g<sup>-1</sup>hr<sup>-1</sup> followed by S<sub>2</sub>I<sub>3</sub> (401.39 µg glucose hydrolyzed g<sup>-1</sup>hr<sup>-1</sup>). The lowest cellulase activity was recorded by S<sub>1</sub>I<sub>2</sub> with mean value of 119.15 µg glucose hydrolyzed g<sup>-1</sup>hr<sup>-1</sup>. Regarding the interactions, substrates S<sub>1</sub> (water cabbage), S<sub>2</sub> (coir pith) and S<sub>4</sub> (farm waste), were influenced mostly by the inoculant I<sub>3</sub> (Composting Inoculum) and for S<sub>3</sub> (water hyacinth) inoculant I<sub>1</sub> (*T. reesei*) was found to be effective. The individual effects of substrates and

inoculants were found to be significant and substrate S<sub>2</sub> (coir pith) recorded the highest activity (382.25 µg glucose hydrolyzed g<sup>-1</sup>hr<sup>-1</sup>) and S<sub>1</sub> (water cabbage) recorded the lowest activity of 262.96 (µg glucose hydrolyzed g<sup>-1</sup>hr<sup>-1</sup>) and regarding the individual effect of inoculants, I<sub>3</sub> (Composting Inoculum) (358.05 µg glucose hydrolyzed g<sup>-1</sup>hr<sup>-1</sup>) was recorded highest mean value and I<sub>2</sub> (*P. sajor-caju*) (232.51 µg of glucose hydrolyzed g<sup>-1</sup>hr<sup>-1</sup>) was found to have lowest value.

#### 4.2.2.4. Maturity Period

In the case of maturity period, the perusal of data from Table 11 indicates that the individual effect of substrate, inoculants and their interactions were found to be significant. The S<sub>2</sub>I<sub>4</sub> (coir pith+ commercial enzyme cocktail) took 86.67 days to composts the wastes, and was significantly superior to all other treatments followed by S<sub>2</sub>I<sub>2</sub> (83.33). The S<sub>1</sub>I<sub>3</sub> took minimum period of 44.33 days to compost the wastes. By analyzing the interaction of substrates by inoculants, the highest maturity period was recorded for substrate S<sub>1</sub> (water cabbage) by inoculants I<sub>1</sub> (*T.reesei*). For S<sub>2</sub> (coir pith) and S<sub>3</sub> (water hyacinth), inoculant I<sub>4</sub> (commercial enzyme cocktail) has reported more maturity period and for S<sub>4</sub> (farm waste), inoculant I<sub>2</sub> (*P. sajor-caju*) reported the highest maturity period and the lowest maturity period was reported by I<sub>3</sub> (composting Inoculum) for all the substrates. The individual effect of substrates was found to be significant and S<sub>2</sub> (coir pith) recorded the highest maturity period of 82.50 days and S<sub>1</sub> (water cabbage) was recorded with lowest maturity period (46.83 days). Regarding the individual effect of inoculants, commercial enzyme cocktail as an inefficient inoculant with mean value of 70 days and I<sub>3</sub> was recorded the lowest maturity period of 65.42 days.

#### 4.2.3. Heavy Metal Content.

The result of the analysis of the heavy metal content of the composts are presented in the Table 12.

#### 4.2.3.1. Lead Content

From the data it is inferred that S<sub>4</sub>I<sub>2</sub> (farm waste+ *P. sajor-caju*) has recorded the highest Pb content (0.208 mg kg<sup>-1</sup>) followed by S<sub>4</sub> I<sub>1</sub> (farm waste+ *T. reesei*) (0.205 mg kg<sup>-1</sup>) and S<sub>2</sub>I<sub>3</sub> (coir pith+ Composting Inoculum) has recorded the lowest mean value 0.024 mg kg<sup>-1</sup>. However the four composts prepared from the water cabbage had no detectable levels of Pb.

#### 4.2.3.2. Cadmium Content

Cd content of the different composts was also analyzed and S<sub>4</sub>I<sub>2</sub> (farm waste+ *P.sajor-caju*) recorded the highest Cd content of (6.423 µg kg<sup>-1</sup>) and all the composts prepared from the substrate S<sub>4</sub> were recorded the higher Cd content. S<sub>4</sub>I<sub>2</sub> was followed by S<sub>4</sub>I<sub>1</sub> (5.649 µg kg<sup>-1</sup> of Cd), S<sub>4</sub>I<sub>4</sub> (5.458 µg kg<sup>-1</sup>) and S<sub>4</sub>I<sub>3</sub> (5.115 µg kg<sup>-1</sup>). Under composts prepared from S<sub>3</sub> (water hyacinth), S<sub>3</sub>I<sub>4</sub> (water hyacinth + commercial enzyme cocktail) has recorded the highest mean value 3.583 µg kg<sup>-1</sup> and S<sub>3</sub>I<sub>3</sub> (water hyacinth + Composting Inoculum) has recorded the lowest value (2.526 µg kg<sup>-1</sup>). All the composts prepared from S<sub>1</sub> (water cabbage) and S<sub>2</sub> (coir pith) had no detectable amounts of Cd.

#### 4.2.3.3. Nickel Content

Among the heavy metals Ni alone significantly influenced by substrates, inoculants and their interactions. S<sub>2</sub>I<sub>3</sub> (coir pith+ Composting Inoculum) recorded the highest Ni content of 0.520 mg kg<sup>-1</sup> and which was on par with S<sub>1</sub>I<sub>1</sub> (0.510 mg kg<sup>-1</sup>). The lowest mean value was recorded by the S<sub>3</sub>I<sub>2</sub> (0.005 mg kg<sup>-1</sup>). Regarding the individual effect of substrates, S<sub>1</sub> (water cabbage) has recorded the highest mean value of 0.285 mg kg<sup>-1</sup> and the lowest mean value by the substrate S<sub>3</sub> (water hyacinth) (0.131 mg kg<sup>-1</sup>). Considering the effect of inoculants, I<sub>3</sub> (Composting Inoculum) has recorded the highest Ni content of 0.393 mg kg<sup>-1</sup>, followed by I<sub>1</sub> (*T.reesei*) (0.182 mg kg<sup>-1</sup>) and I<sub>2</sub> (*P. sajor-caju*) recorded the lowest mean value of 0.050 mg kg<sup>-1</sup>.

#### 4.2.3.4. Tin Content

No detectable amounts of Sn was observed in the substrates S<sub>1</sub> (water

Table 12. Heavy metal content of the composts as influenced by substrates, inoculants and their interactions

| Treatments                    | Pb (mg kg <sup>-1</sup> ) | Cd (µg kg <sup>-1</sup> ) | Ni (mg kg <sup>-1</sup> ) | Sn (µg kg <sup>-1</sup> ) |
|-------------------------------|---------------------------|---------------------------|---------------------------|---------------------------|
| S <sub>1</sub> I <sub>1</sub> | N.D                       | N.D                       | 0.510                     | N.D                       |
| S <sub>1</sub> I <sub>2</sub> | N.D                       | N.D                       | 0.039                     | N.D                       |
| S <sub>1</sub> I <sub>3</sub> | N.D                       | N.D                       | 0.414                     | N.D                       |
| S <sub>1</sub> I <sub>4</sub> | N.D                       | N.D                       | 0.178                     | N.D                       |
| S <sub>2</sub> I <sub>1</sub> | 0.027                     | N.D                       | 0.053                     | N.D                       |
| S <sub>2</sub> I <sub>2</sub> | 0.041                     | N.D                       | 0.082                     | N.D                       |
| S <sub>2</sub> I <sub>3</sub> | 0.024                     | N.D                       | 0.520                     | N.D                       |
| S <sub>2</sub> I <sub>4</sub> | 0.028                     | N.D                       | 0.132                     | N.D                       |
| S <sub>3</sub> I <sub>1</sub> | 0.193                     | 2.607                     | 0.127                     | 0.678                     |
| S <sub>3</sub> I <sub>2</sub> | 0.095                     | 3.430                     | 0.005                     | 0.578                     |
| S <sub>3</sub> I <sub>3</sub> | 0.071                     | 2.526                     | 0.214                     | 0.712                     |
| S <sub>3</sub> I <sub>4</sub> | 0.124                     | 3.583                     | 0.177                     | 0.877                     |
| S <sub>4</sub> I <sub>1</sub> | 0.205                     | 5.649                     | 0.040                     | N.D                       |
| S <sub>4</sub> I <sub>2</sub> | 0.208                     | 6.423                     | 0.074                     | N.D                       |
| S <sub>4</sub> I <sub>3</sub> | 0.108                     | 5.115                     | 0.422                     | N.D                       |
| S <sub>4</sub> I <sub>4</sub> | 0.158                     | 5.458                     | 0.082                     | N.D                       |
| S <sub>1</sub>                | -                         | -                         | 0.285                     | -                         |
| S <sub>2</sub>                | -                         | -                         | 0.197                     | -                         |
| S <sub>3</sub>                | -                         | -                         | 0.131                     | -                         |
| S <sub>4</sub>                | -                         | -                         | 0.155                     | -                         |
| I <sub>1</sub>                | -                         | -                         | 0.182                     | -                         |
| I <sub>2</sub>                | -                         | -                         | 0.050                     | -                         |
| I <sub>3</sub>                | -                         | -                         | 0.393                     | -                         |
| I <sub>4</sub>                | -                         | -                         | 0.143                     | -                         |
| CD- SI(0.05)                  | -                         | -                         | 0.0248                    | -                         |
| CD-S(0.05)                    | -                         | -                         | 0.0124                    | -                         |
| CD-I(0.05)                    | -                         | -                         | 0.0124                    | -                         |

N.D : Not detectable

S<sub>1</sub> Water cabbage  
 S<sub>2</sub> Coir pith  
 S<sub>3</sub> Water hyacinth  
 S<sub>4</sub> Farm wastes

I<sub>1</sub> *Trichoderma reesei*  
 I<sub>2</sub> *Pleurotus sajor-caju*  
 I<sub>3</sub> Composting inoculum  
 I<sub>4</sub> Commercial enzyme cocktail

cabbage), S<sub>2</sub> (coir pith) and S<sub>4</sub> farm waste as inferred from the Table 12 and only S<sub>3</sub> (water hyacinth) contain detectable levels of Sn. S<sub>3</sub>I<sub>4</sub> has recorded with 0.877  $\mu\text{g kg}^{-1}$ , S<sub>3</sub>I<sub>3</sub> (0.712  $\mu\text{g kg}^{-1}$ ), S<sub>3</sub>I<sub>1</sub> (0.678  $\mu\text{g kg}^{-1}$ ) and the lowest content was recorded by the interaction S<sub>3</sub>I<sub>2</sub> (0.578  $\mu\text{g kg}^{-1}$ ).

### 4.3. POT CULTURE EXPERIMENT

#### 4.3.1. Effect of Treatments on Yield Attributes and Yield

The data on plant height, no of leaves per branches, number of branches, girth of stem and yield are presented in the Table 13.

##### 4.3.1.1. Plant Height

Statistical analysis of the data revealed that the plant height did not vary significantly with respect to the different treatments, substrates, inoculants and their interactions. However the plant varied from 70.00 cm to 45.00 cm. The tallest plant was recorded by T<sub>13</sub> (100 per cent N as compost from water hyacinth and Composting Inoculum) (70.00 cm) and the shortest plant was recorded by T<sub>19</sub> (absolute control) (42.33 cm). While considering the individual effect of substrates, S<sub>3</sub> (water hyacinth) produced the tallest plant with mean height of 58.50 cm and S<sub>1</sub> (water cabbage) produced the shortest plant with 53.83 cm height. However the individual effect of substrates were not significant. Considering the effect Inoculants, it was found to be significant on plant height as I<sub>3</sub> (Composting Inoculum) recorded the highest mean value of 59.08 cm and I<sub>2</sub> (*P. sajor-caju*) recorded the lowest mean value of 51.33 cm.

##### 4.3.1.2. Number of Leaves per Branch

The treatments varied significantly with respect number of leaves per branch as inferred from Table 13. Substrates, inoculants and their interactions were found to have significant effect on the number of leaves per branches. The treatment T<sub>5</sub> (100 per cent N as compost from water cabbage and Composting Inoculum) has recorded the highest number of leaves per branch of 7.10 which was on par with treatment T<sub>7</sub> (6.33) while the lowest value was recorded by the treatment T<sub>8</sub> (2.75), but it was higher than absolute control T<sub>19</sub> (2.65). Regarding

Table 13. Effect of treatments on yield attributes and yield

| Treatments                                       | Plant height (cm) | Number of leaves branch <sup>-1</sup> | Number of branches | Girth of the stem (cm) | Yield (g /plant) |
|--|-------------------|---------------------------------------|--------------------|------------------------|------------------|
| T <sub>1</sub>                                   | 63.33             | 4.80                                  | 4.80               | 4.43                   | 128.30           |
| T <sub>2</sub>                                   | 49.67             | 3.63                                  | 3.80               | 4.07                   | 84.70            |
| T <sub>3</sub> (S <sub>1</sub> I <sub>1</sub> )  | 55.67             | 5.45                                  | 2.50               | 4.10                   | 103.30           |
| T <sub>4</sub> (S <sub>1</sub> I <sub>2</sub> )  | 45.00             | 4.06                                  | 4.80               | 2.95                   | 82.20            |
| T <sub>5</sub> (S <sub>1</sub> I <sub>3</sub> )  | 61.33             | 7.10                                  | 5.70               | 4.33                   | 154.50           |
| T <sub>6</sub> (S <sub>1</sub> I <sub>4</sub> )  | 53.33             | 3.57                                  | 4.80               | 3.35                   | 96.50            |
| T <sub>7</sub> (S <sub>2</sub> I <sub>1</sub> )  | 57.67             | 6.33                                  | 5.50               | 3.05                   | 86.00            |
| T <sub>8</sub> (S <sub>2</sub> I <sub>2</sub> )  | 56.00             | 2.75                                  | 3.50               | 3.17                   | 76.40            |
| T <sub>9</sub> (S <sub>2</sub> I <sub>3</sub> )  | 51.33             | 4.27                                  | 3.50               | 2.78                   | 101.20           |
| T <sub>10</sub> (S <sub>2</sub> I <sub>4</sub> ) | 55.00             | 4.77                                  | 4.00               | 3.47                   | 97.20            |
| T <sub>11</sub> (S <sub>3</sub> I <sub>1</sub> ) | 52.67             | 4.62                                  | 4.30               | 3.28                   | 100.80           |
| T <sub>12</sub> (S <sub>3</sub> I <sub>2</sub> ) | 53.33             | 4.23                                  | 5.10               | 3.70                   | 97.40            |
| T <sub>13</sub> (S <sub>3</sub> I <sub>3</sub> ) | 70.00             | 4.67                                  | 5.70               | 4.55                   | 159.54           |
| T <sub>14</sub> (S <sub>3</sub> I <sub>4</sub> ) | 58.00             | 4.99                                  | 5.30               | 3.38                   | 99.70            |
| T <sub>15</sub> (S <sub>4</sub> I <sub>1</sub> ) | 64.67             | 4.43                                  | 3.80               | 4.32                   | 106.20           |
| T <sub>16</sub> (S <sub>4</sub> I <sub>2</sub> ) | 51.00             | 4.39                                  | 3.20               | 3.15                   | 90.20            |
| T <sub>17</sub> (S <sub>4</sub> I <sub>3</sub> ) | 53.67             | 2.93                                  | 2.70               | 3.67                   | 101.40           |
| T <sub>18</sub> (S <sub>4</sub> I <sub>4</sub> ) | 52.00             | 4.15                                  | 4.00               | 3.08                   | 99.30            |
| T <sub>19</sub>                                  | 42.33             | 2.65                                  | 2.20               | 2.48                   | 49.50            |
| S <sub>1</sub>                                   | 53.83             | 5.05                                  | 4.46               | 3.68                   | 109.12           |
| S <sub>2</sub>                                   | 55.00             | 4.53                                  | 4.13               | 3.12                   | 90.17            |
| S <sub>3</sub>                                   | 58.50             | 4.63                                  | 5.10               | 3.73                   | 114.36           |
| S <sub>4</sub>                                   | 55.33             | 3.97                                  | 3.42               | 3.55                   | 99.28            |
| I <sub>1</sub>                                   | 57.67             | 5.21                                  | 4.04               | 3.67                   | 99.07            |
| I <sub>2</sub>                                   | 51.33             | 3.86                                  | 4.14               | 3.24                   | 86.55            |
| I <sub>3</sub>                                   | 59.08             | 4.74                                  | 4.38               | 3.83                   | 127.17           |
| I <sub>4</sub>                                   | 54.58             | 4.37                                  | 4.54               | 3.32                   | 98.14            |
| CD - T/<br>SI(0.05)                              | 13.212            | 1.757                                 | 2.192              | 1.507                  | 29.98            |
| CD -S(0.05)                                      | 6.606             | 0.878                                 | 1.096              | 0.754                  | 14.99            |
| CD-I(0.05)                                       | 6.606             | 0.878                                 | 1.096              | 0.754                  | 14.99            |



the individual effect of substrate, S<sub>1</sub>(water cabbage) has produced number of leaves with the highest mean value of 5.05 and S<sub>4</sub> (farm waste) recorded the lowest value of 3.97. Considering the effect of inoculants I<sub>1</sub> (*T. reesei*) has recorded the highest mean value 5.21 and I<sub>2</sub> (*P. sajor- caju*) recorded the lowest mean value 3.86.

#### **4.3.1.3. Number of Branches**

Critical appraisal of the data presented in Table 13 revealed that the treatments, inoculants and their interactions did not vary significantly from the controls while the individual effects of substrates were significant. Treatment T<sub>5</sub> (100 per cent N as compost from water cabbage and Composting Inoculum) and T<sub>13</sub> (100 per cent N as compost from water hyacinth and Composting Inoculum) recorded the highest mean value of 5.70 and the lowest mean value was recorded by the treatment T<sub>3</sub> (2.50) while all the 16 treatments were superior to the absolute control. Considering the individual effect of substrates, S<sub>3</sub> (water hyacinth )has (5.10) recorded the highest number of branches and S<sub>4</sub> (farm waste) (3.42) recorded the lowest mean value. The individual effect of inoculants, did not vary significantly from each other. However L<sub>4</sub> (commercial enzyme cocktail) has recorded the highest mean value 4.54 while I<sub>1</sub> (*T. reesei*) recorded the lowest mean value 4.04.

#### **4.3.1.4. Girth of the Stem**

Treatments, substrates, inoculants and their interactions had no significant effect on girth of the stem when compared with the controls. However the treatment T<sub>13</sub> (100 per cent N as compost from water hyacinth and Composting Inoculum) has recorded the highest stem girth of 4.55 cm and T<sub>9</sub> recorded the lowest girth of 2.78 cm while all the treatments were superior to the absolute control. When the individual effect of substrates were considered, S<sub>3</sub> (water hyacinth) recorded the highest mean value of 3.73 cm and S<sub>2</sub> (coir pith) recorded the lowest mean value of 3.12 cm. Regarding the individual effect of inoculants, I<sub>3</sub> (Composting Inoculum) recorded the highest value (3.83 cm) while I<sub>2</sub> (*P. sajor caju*) (3.24 cm) recorded the lowest mean value.

#### 4.3.1.5. Yield

Treatments, inoculants, substrates and their interactions significantly influenced the yield of amaranthus. Treatment T<sub>13</sub> (100 per cent N as compost from water hyacinth and Composting Inoculum) (159.54 g) has recorded the highest yield which was on par with T<sub>5</sub> (100 per cent N as compost from water cabbage and Composting Inoculum) (154.51 g) while T<sub>8</sub> (76.40 g) has recorded the lowest yield. However all the treatments were found to be significantly superior to the absolute control T<sub>19</sub> (49.48 g). Considering the individual effect of substrates, S<sub>3</sub> (water hyacinth) (114.36 g) recorded the highest yield and S<sub>2</sub> (coir pith) (90.17 g) recorded the lowest yield. Among the individual effect of inoculants on yield I<sub>3</sub> (Composting Inoculum) (127.17 g) recorded the highest mean value and I<sub>2</sub> (*P. sajor-caju*) (86.55 g) has recorded the lowest value.

#### 4.3.2. Effect of Treatments on Physico- Chemical Properties of the Soil

The data on physico chemical properties like bulk density, particle density, and water holding capacity, soil pH and EC are presented in the Table 14.

##### 4.3.2.1. Bulk Density

Various treatments, substrates, and their interactions significantly influenced the bulk density of the soil while the influence of the inoculants were found to be non significant. T<sub>16</sub> (100 per cent N as compost from farm waste and *P. sajor-caju*) recorded the highest mean value of 1.48 Mg m<sup>-3</sup> among all the 16 treatments while the absolute control T<sub>19</sub> has recorded the highest bulk density (1.50 Mg m<sup>-3</sup>). While T<sub>8</sub> recorded the lowest bulk density of 1.18 Mg m<sup>-3</sup>. Considering the individual effect of substrates, S<sub>4</sub> (farm waste) (1.44 Mg m<sup>-3</sup>) was noticed with the highest value and S<sub>2</sub> (coir pith) (1.28 Mg m<sup>-3</sup>) recorded the lowest mean value while among the inoculants, I<sub>1</sub> (*T. reesei*) recorded the highest bulk density of 1.36 Mg m<sup>-3</sup> and the lowest value recorded by I<sub>3</sub> (Composting Inoculum) (1.30 Mg m<sup>-3</sup>).

Table 14. Effect of treatments on physico-chemical properties of the post harvest soil

| Treatments                                       | Bulk Density (Mg m <sup>-3</sup> ) | Particle Density (Mg m <sup>-3</sup> ) | Water Holding Capacity (%) | pH    | EC (μSm <sup>-1</sup> ) |
|--|------------------------------------|--|----------------------------|-------|-------------------------|
| T <sub>1</sub>                                   | 1.35                               | 2.29                                   | 30.05                      | 6.62  | 178.40                  |
| T <sub>2</sub>                                   | 1.30                               | 1.98                                   | 31.23                      | 6.55  | 166.69                  |
| T <sub>3</sub> (S <sub>1</sub> I <sub>1</sub> )  | 1.24                               | 2.06                                   | 31.47                      | 6.75  | 176.70                  |
| T <sub>4</sub> (S <sub>1</sub> I <sub>2</sub> )  | 1.37                               | 2.07                                   | 33.12                      | 6.89  | 194.84                  |
| T <sub>5</sub> (S <sub>1</sub> I <sub>3</sub> )  | 1.21                               | 2.07                                   | 31.04                      | 6.72  | 171.87                  |
| T <sub>6</sub> (S <sub>1</sub> I <sub>4</sub> )  | 1.30                               | 2.09                                   | 30.53                      | 6.76  | 182.63                  |
| T <sub>7</sub> (S <sub>2</sub> I <sub>1</sub> )  | 1.41                               | 2.04                                   | 30.91                      | 6.74  | 162.68                  |
| T <sub>8</sub> (S <sub>2</sub> I <sub>2</sub> )  | 1.18                               | 2.08                                   | 39.29                      | 6.74  | 171.45                  |
| T <sub>9</sub> (S <sub>2</sub> I <sub>3</sub> )  | 1.30                               | 1.97                                   | 34.28                      | 6.57  | 135.11                  |
| T <sub>10</sub> (S <sub>2</sub> I <sub>4</sub> ) | 1.22                               | 1.88                                   | 34.13                      | 6.71  | 177.39                  |
| T <sub>11</sub> (S <sub>3</sub> I <sub>1</sub> ) | 1.33                               | 2.18                                   | 32.14                      | 6.73  | 178.99                  |
| T <sub>12</sub> (S <sub>3</sub> I <sub>2</sub> ) | 1.28                               | 2.11                                   | 31.16                      | 6.90  | 181.41                  |
| T <sub>13</sub> (S <sub>3</sub> I <sub>3</sub> ) | 1.28                               | 2.01                                   | 35.07                      | 6.72  | 156.90                  |
| T <sub>14</sub> (S <sub>3</sub> I <sub>4</sub> ) | 1.42                               | 1.97                                   | 32.51                      | 6.75  | 166.30                  |
| T <sub>15</sub> (S <sub>4</sub> I <sub>1</sub> ) | 1.45                               | 2.01                                   | 30.98                      | 6.98  | 181.44                  |
| T <sub>16</sub> (S <sub>4</sub> I <sub>2</sub> ) | 1.48                               | 2.08                                   | 30.92                      | 6.80  | 180.69                  |
| T <sub>17</sub> (S <sub>4</sub> I <sub>3</sub> ) | 1.40                               | 1.97                                   | 31.76                      | 6.76  | 159.63                  |
| T <sub>18</sub> (S <sub>4</sub> I <sub>4</sub> ) | 1.45                               | 1.92                                   | 34.05                      | 6.91  | 196.36                  |
| T <sub>19</sub>                                  | 1.50                               | 2.10                                   | 30.02                      | 6.40  | 119.35                  |
| S <sub>1</sub>                                   | 1.28                               | 2.07                                   | 31.54                      | 6.78  | 181.51                  |
| S <sub>2</sub>                                   | 1.28                               | 1.99                                   | 34.65                      | 6.69  | 161.67                  |
| S <sub>3</sub>                                   | 1.33                               | 2.07                                   | 32.72                      | 6.78  | 170.90                  |
| S <sub>4</sub>                                   | 1.44                               | 1.99                                   | 31.93                      | 6.86  | 179.53                  |
| I <sub>1</sub>                                   | 1.36                               | 2.07                                   | 31.38                      | 6.80  | 174.95                  |
| I <sub>2</sub>                                   | 1.33                               | 2.09                                   | 33.62                      | 6.83  | 182.10                  |
| I <sub>3</sub>                                   | 1.30                               | 2.00                                   | 33.04                      | 6.70  | 155.58                  |
| I <sub>4</sub>                                   | 1.35                               | 1.96                                   | 32.81                      | 6.78  | 180.67                  |
| CD- T/<br>S I (0.05)                             | 0.122                              | 0.216                                  | 1.594                      | 0.187 | NS                      |
| CD -S(0.05)                                      | 0.061                              | 0.108                                  | 0.797                      | 0.094 | NS                      |
| CD-I(0.05)                                       | 0.061                              | 0.108                                  | 0.797                      | NS    | 16.217                  |

NS- not significant

#### 4.3.2.2. Particle Density

Effect of various treatments, substrates, inoculants and their interactions were found not significant on particle density. However, treatment T<sub>11</sub> (100 per cent N as compost from water hyacinth and *T.reesei*) recorded the highest particle density of 2.18 Mg m<sup>-3</sup> while T<sub>10</sub> recorded the lowest mean value of 1.88 Mg m<sup>-3</sup>. Considering 19 treatments as a whole, treatment T<sub>1</sub> has recorded the highest value of 2.29 Mg m<sup>-3</sup> which was on par with T<sub>11</sub>. Even though the individual effects were not significant, substrate S<sub>1</sub> recorded the highest mean value of 2.07 Mg m<sup>-3</sup> and S<sub>2</sub> recorded the lowest mean value of 1.99 Mg m<sup>-3</sup> while among the inoculants I<sub>2</sub> has recorded the highest value of 2.09 Mg m<sup>-3</sup> and I<sub>4</sub> of 1.96 Mg m<sup>-3</sup> has recorded the lowest mean value.

#### 4.3.2.3. Water Holding Capacity

Water holding capacity of soil ranged from 30.02 per cent to 39.29 per cent. The various treatments, substrates, inoculants and their interactions were found to influence significantly the water holding capacity of the treated soils. Treatment T<sub>8</sub> (100 per cent N as compost from coir pith and *P. sajor-caju*) recorded the highest mean value of 39.29 per cent. While the lowest mean value was recorded by T<sub>6</sub> (30.53 per cent). However all the treatments were superior to the absolute control T<sub>19</sub> (30.02 per cent) and T<sub>1</sub> (30.05 per cent).

#### 4.3.2.4. pH

Statistical analysis of the data revealed that the pH of the soil was significantly influenced by the treatments, substrates and inoculants however their interactions were found to be non significant. pH of the soils ranged from 6.40 to 6.98. The highest pH was recorded by the treatment T<sub>15</sub> (100 per cent N as compost from farm waste and *T.reesei*) (6.98) which was on par with T<sub>18</sub> (6.91), T<sub>12</sub> (6.90) and T<sub>4</sub> (6.89) while T<sub>9</sub> (6.57) recorded the lowest pH value and all the treatments were found to be superior than the absolute control T<sub>19</sub> (6.40) which was on par with other controls T<sub>1</sub>(6.62) and T<sub>2</sub>(6.55). Regarding the individual effects, S<sub>4</sub> (farm waste) (6.86) has recorded the highest pH value while S<sub>2</sub> (coir pith) (6.69) recorded the lowest mean value. Considering the individual effect of

inoculants, there was significant effect on pH. Inoculant I<sub>2</sub> (*P. sajor-caju*) (6.83) recorded the highest mean value while I<sub>4</sub> (commercial enzyme cocktail) (6.70) recorded the lowest value.

#### 4.3.2.5. EC

Various treatments imposed variations on the EC of the soil however treatments, substrates and interaction of substrates on inoculants were not at all significant. It was obvious from the data that the individual effect of inoculants had significant effect on soil EC. The mean values of EC ranged from 119.35  $\mu\text{Sm}^{-1}$  to 196.36  $\mu\text{Sm}^{-1}$ . Treatment T<sub>18</sub> (100 per cent N as compost from farm waste and commercial enzyme cocktail) recorded the highest EC of 196.36  $\mu\text{Sm}^{-1}$  while treatment T<sub>9</sub> recorded the lowest value of 135.11  $\mu\text{Sm}^{-1}$ . But all the treatments were superior to the absolute control T<sub>19</sub> (119.35  $\mu\text{Sm}^{-1}$ ). While taking in consideration of the individual effects of substrates, they were statistically non significant and S<sub>1</sub> (water cabbage) recorded the highest mean value (181.51  $\mu\text{Sm}^{-1}$ ) and S<sub>2</sub> (coir pith) recorded the lowest mean value (161.67  $\mu\text{Sm}^{-1}$ ). The inoculants, have significant effect on EC as I<sub>2</sub> (*P. sajor-caju*) recorded the highest value of 182.10  $\mu\text{Sm}^{-1}$  and I<sub>3</sub> (Composting Inoculum) recorded the lowest mean of 155.58  $\mu\text{Sm}^{-1}$ .

#### 4.3.2.6. Major Nutrients

Effect of treatments on major nutrients viz. available N, available P, available K, and organic carbon of the soil is presented on Table 15.

##### 4.3.2.6.1. Available Nitrogen

It was observed from the data that the treatments, substrates, and inoculants imposed significant effect on available N content of the soil however their interaction effects were found to be non significant. The mean values ranged from 167.25 kg ha<sup>-1</sup> to 275.97 kg ha<sup>-1</sup>. The highest value recorded as 275.97 kg ha<sup>-1</sup> by T<sub>5</sub> (100 per cent N as compost from water cabbage and Composting Inoculum) which was on par with the treatments T<sub>6</sub> (250.88 kg ha<sup>-1</sup>), T<sub>13</sub> (246.70 kg ha<sup>-1</sup>), T<sub>17</sub> (246.70 kg ha<sup>-1</sup>) while the lowest mean value among the treatments

Table 15. Effect of treatments on available content of major nutrients in the soil

| Treatments                                       | Available N<br>(kg ha <sup>-1</sup> ) | Available P<br>(kg ha <sup>-1</sup> ) | Available K<br>(kg ha <sup>-1</sup> ) | Organic<br>Carbon (%) |
|--|---------------------------------------|---------------------------------------|---------------------------------------|-----------------------|
| T <sub>1</sub>                                   | 167.25                                | 71.89                                 | 290.08                                | 1.16                  |
| T <sub>2</sub>                                   | 209.07                                | 63.79                                 | 255.26                                | 1.65                  |
| T <sub>3</sub> (S <sub>1</sub> I <sub>1</sub> )  | 209.07                                | 73.17                                 | 290.48                                | 1.37                  |
| T <sub>4</sub> (S <sub>1</sub> I <sub>2</sub> )  | 209.07                                | 79.97                                 | 290.85                                | 1.97                  |
| T <sub>5</sub> (S <sub>1</sub> I <sub>3</sub> )  | 275.97                                | 73.10                                 | 282.38                                | 1.51                  |
| T <sub>6</sub> (S <sub>1</sub> I <sub>4</sub> )  | 250.88                                | 60.24                                 | 256.79                                | 1.76                  |
| T <sub>7</sub> (S <sub>2</sub> I <sub>1</sub> )  | 192.34                                | 65.39                                 | 299.93                                | 2.31                  |
| T <sub>8</sub> (S <sub>2</sub> I <sub>2</sub> )  | 188.16                                | 55.84                                 | 221.56                                | 2.30                  |
| T <sub>9</sub> (S <sub>2</sub> I <sub>3</sub> )  | 188.16                                | 48.01                                 | 236.80                                | 2.41                  |
| T <sub>10</sub> (S <sub>2</sub> I <sub>4</sub> ) | 183.98                                | 54.83                                 | 189.65                                | 2.33                  |
| T <sub>11</sub> (S <sub>3</sub> I <sub>1</sub> ) | 192.34                                | 63.79                                 | 223.87                                | 2.60                  |
| T <sub>12</sub> (S <sub>3</sub> I <sub>2</sub> ) | 225.79                                | 90.10                                 | 211.47                                | 2.13                  |
| T <sub>13</sub> (S <sub>3</sub> I <sub>3</sub> ) | 246.70                                | 97.70                                 | 396.28                                | 2.23                  |
| T <sub>14</sub> (S <sub>3</sub> I <sub>4</sub> ) | 234.15                                | 77.21                                 | 201.90                                | 2.46                  |
| T <sub>15</sub> (S <sub>4</sub> I <sub>1</sub> ) | 221.61                                | 82.94                                 | 259.63                                | 2.34                  |
| T <sub>16</sub> (S <sub>4</sub> I <sub>2</sub> ) | 209.07                                | 76.10                                 | 256.07                                | 2.00                  |
| T <sub>17</sub> (S <sub>4</sub> I <sub>3</sub> ) | 246.70                                | 81.24                                 | 211.88                                | 2.12                  |
| T <sub>18</sub> (S <sub>4</sub> I <sub>4</sub> ) | 225.79                                | 79.04                                 | 269.36                                | 2.22                  |
| T <sub>19</sub>                                  | 136.25                                | 44.63                                 | 171.06                                | 1.23                  |
| S <sub>1</sub>                                   | 236.24                                | 71.62                                 | 280.13                                | 1.65                  |
| S <sub>2</sub>                                   | 188.16                                | 56.03                                 | 236.99                                | 2.34                  |
| S <sub>3</sub>                                   | 224.75                                | 83.70                                 | 258.38                                | 2.36                  |
| S <sub>4</sub>                                   | 225.79                                | 79.81                                 | 249.24                                | 2.17                  |
| I <sub>1</sub>                                   | 203.84                                | 71.32                                 | 268.48                                | 2.15                  |
| I <sub>2</sub>                                   | 208.02                                | 76.98                                 | 244.99                                | 2.10                  |
| I <sub>3</sub>                                   | 239.38                                | 75.03                                 | 281.83                                | 2.07                  |
| I <sub>4</sub>                                   | 223.70                                | 67.83                                 | 229.42                                | 2.19                  |
| CD- T /CD-<br>SI(0.05)                           | 36.956                                | 16.07                                 | 40.845                                | 0.664                 |
| CD -S(0.05)                                      | 18.478                                | 7.304                                 | 20.422                                | 0.332                 |
| CD-I(0.05)                                       | 18.478                                | 7.304                                 | 20.422                                | NS                    |

NS- not significant

was recorded by T<sub>10</sub> (183.98 kg ha<sup>-1</sup>). The absolute control T<sub>19</sub> recorded as the lowest value of 167.25 kg ha<sup>-1</sup>. Individual effects of substrates were found to have significant effect on available N content. Substrate S<sub>1</sub> (water cabbage) has recorded the highest value of 236.24 kg ha<sup>-1</sup> and was on par with S<sub>3</sub> (water hyacinth) and S<sub>4</sub> (farm waste) while S<sub>2</sub> (coir pith) recorded the lowest mean value 188.16 kg ha<sup>-1</sup>. Regarding the individual effect of inoculants, I<sub>3</sub> (Composting Inoculum) recorded the highest mean value of 239.38 kg ha<sup>-1</sup> and I<sub>1</sub> (*T. reesei*) registered the lowest mean value 208.02 kg ha<sup>-1</sup>.

#### 4.3.2.6.2. Available Phosphorous

The data revealed that the available P content of the soil was significantly varied with the treatments. The mean value of available P content of soils under different treatments ranged from 44.63 kg ha<sup>-1</sup> to 97.70 kg ha<sup>-1</sup>. Among the treatments, T<sub>13</sub> (100 per cent N as compost from water hyacinth and Composting Inoculum) recorded the highest value 97.70 kg ha<sup>-1</sup> which was on par with T<sub>12</sub> (90.10 kg ha<sup>-1</sup>), T<sub>15</sub> (82.94 kg ha<sup>-1</sup>) and T<sub>17</sub> (81.24 kg ha<sup>-1</sup>) while the lowest mean value recorded by the treatment T<sub>9</sub> (48.01 kg ha<sup>-1</sup>). All the treatments were found to be superior to the absolute control T<sub>19</sub> which recorded the mean value of 44.63 kg ha<sup>-1</sup>. Considering the individual effect of substrates, a significant effect on soil available P content was noticed as S<sub>3</sub> registered the highest value of 83.70 kg ha<sup>-1</sup> and S<sub>2</sub> has recorded the lowest value of 71.32 kg ha<sup>-1</sup>. Though the effect of inoculants was not significant on available soil P, the highest mean value was recorded by the inoculant I<sub>2</sub> (*P. sajor-caju*) (76.98 kg ha<sup>-1</sup>) and the lowest mean value was recorded by the inoculant I<sub>4</sub> commercial enzyme cocktail (67.83 kg ha<sup>-1</sup>).

#### 4.3.2.6.3. Available Potassium

Critical appraisal of the data revealed that the available K was significantly influenced by the treatments. The mean values ranged from 171.06 kg ha<sup>-1</sup> to 396.28 kg ha<sup>-1</sup> while the highest value was recorded by T<sub>13</sub> (100 per cent N as compost from water hyacinth and Composting Inoculum) (396.28 kg ha<sup>-1</sup>) which was followed by the treatment T<sub>7</sub> (299.93 kg ha<sup>-1</sup>). The lowest mean

value was reported by the treatment T<sub>10</sub> (189.65 kg ha<sup>-1</sup>). However all the treatments were superior to the absolute control T<sub>19</sub> 171.06 kg ha<sup>-1</sup>. Considering the individual effect of substrates, it was found that available K content was varied significantly. Substrate S<sub>1</sub> (water cabbage) recorded the highest K content of 280.13 kg ha<sup>-1</sup> while the substrate S<sub>2</sub> (coir pith) (236.99 kg ha<sup>-1</sup>) recorded the lowest value. The individual effect of inoculants was not significant on the available K content, however I<sub>3</sub> (Composting Inoculum) recorded the highest mean value (281.83 kg ha<sup>-1</sup>) and I<sub>4</sub> (commercial enzyme cocktail) (229.42 kg ha<sup>-1</sup>) recorded the lowest mean value.

#### **4.3.2.6.4. Organic Carbon Content**

It was obvious from the data that the organic carbon content of the soils was significantly influenced by the treatments. The mean values ranged from 1.16 per cent to 2.60 per cent. The treatment T<sub>11</sub> (100 per cent N as compost from water hyacinth and *T. reesei*) recorded the highest organic C content 2.60 per cent and T<sub>3</sub> recorded the lowest mean value of 1.37 per cent. But all the treatments were found to be superior to the absolute control T<sub>19</sub> (1.22 per cent) and T<sub>1</sub> (1.16 per cent). Considering the individual effect of substrates, it was found to have significant effect on soil organic carbon. S<sub>2</sub> (coir pith) recorded the highest value of 2.34 per cent and S<sub>1</sub> (water cabbage) (1.65 per cent) recorded the lowest mean value. Regarding the individual effect of inoculants on soil organic carbon content, it was not significant. However inoculant I<sub>4</sub> (commercial enzyme cocktail) recorded the highest value 2.19 and I<sub>3</sub> (Composting Inoculum) recorded the lowest mean value of 2.07 per cent.

#### **4.3.2.7. Micro Nutrients**

Available micro nutrient content of the soils are presented in the Table 16.

##### **4.3.2.7.1. Iron Content**

Critical appraisal of the data shows that the treatments influenced the available Fe content of the soil significantly. The mean value ranged from 10.91 mg kg<sup>-1</sup> to 41.69 mg kg<sup>-1</sup>. The treatment T<sub>9</sub> (100 per cent N as compost from coir



Table 16. Effect of treatments on available content of micro nutrients in the soil, mg kg<sup>-1</sup>.

| Treatments                                       | Fe    | Mn    | Zn    | Cu    | B     |
|--|-------|-------|-------|-------|-------|
| T <sub>1</sub>                                   | 18.30 | 14.40 | 2.86  | 0.96  | 0.85  |
| T <sub>2</sub>                                   | 28.24 | 24.57 | 3.50  | 0.51  | 0.90  |
| T <sub>3</sub> (S <sub>1</sub> I <sub>1</sub> )  | 23.56 | 17.71 | 4.84  | 0.81  | 0.63  |
| T <sub>4</sub> (S <sub>1</sub> I <sub>2</sub> )  | 18.02 | 29.00 | 3.80  | 1.05  | 0.25  |
| T <sub>5</sub> (S <sub>1</sub> I <sub>3</sub> )  | 23.72 | 26.95 | 4.09  | 1.12  | 0.22  |
| T <sub>6</sub> (S <sub>1</sub> I <sub>4</sub> )  | 10.91 | 30.82 | 3.89  | 0.84  | 0.18  |
| T <sub>7</sub> (S <sub>2</sub> I <sub>1</sub> )  | 30.13 | 25.54 | 4.55  | 0.92  | 0.15  |
| T <sub>8</sub> (S <sub>2</sub> I <sub>2</sub> )  | 31.64 | 36.17 | 5.12  | 1.11  | 0.23  |
| T <sub>9</sub> (S <sub>2</sub> I <sub>3</sub> )  | 41.69 | 34.83 | 2.60  | 0.91  | 0.15  |
| T <sub>10</sub> (S <sub>2</sub> I <sub>4</sub> ) | 21.67 | 25.81 | 4.90  | 0.83  | 0.15  |
| T <sub>11</sub> (S <sub>3</sub> I <sub>1</sub> ) | 11.19 | 28.62 | 4.57  | 0.61  | 0.78  |
| T <sub>12</sub> (S <sub>3</sub> I <sub>2</sub> ) | 36.49 | 34.87 | 3.34  | 0.62  | 0.62  |
| T <sub>13</sub> (S <sub>3</sub> I <sub>3</sub> ) | 37.81 | 42.57 | 3.60  | 1.12  | 0.14  |
| T <sub>14</sub> (S <sub>3</sub> I <sub>4</sub> ) | 18.46 | 30.52 | 5.46  | 0.88  | 0.21  |
| T <sub>15</sub> (S <sub>4</sub> I <sub>1</sub> ) | 28.84 | 31.65 | 4.93  | 0.57  | 0.15  |
| T <sub>16</sub> (S <sub>4</sub> I <sub>2</sub> ) | 22.50 | 28.24 | 3.92  | 0.62  | 0.62  |
| T <sub>17</sub> (S <sub>4</sub> I <sub>3</sub> ) | 37.56 | 25.75 | 3.44  | 0.79  | 0.64  |
| T <sub>18</sub> (S <sub>4</sub> I <sub>4</sub> ) | 22.15 | 20.17 | 4.07  | 0.65  | 0.19  |
| T <sub>19</sub>                                  | 9.53  | 11.25 | 1.59  | 0.27  | 0.09  |
| S <sub>1</sub>                                   | 19.05 | 26.12 | 4.15  | 0.96  | 0.32  |
| S <sub>2</sub>                                   | 31.22 | 30.59 | 4.29  | 0.94  | 0.17  |
| S <sub>3</sub>                                   | 25.99 | 34.15 | 4.24  | 0.81  | 0.44  |
| S <sub>4</sub>                                   | 27.76 | 26.45 | 4.09  | 0.66  | 0.40  |
| I <sub>1</sub>                                   | 23.43 | 25.88 | 4.72  | 0.73  | 0.43  |
| I <sub>2</sub>                                   | 27.16 | 32.07 | 4.04  | 0.85  | 0.43  |
| I <sub>3</sub>                                   | 35.19 | 32.53 | 3.43  | 0.99  | 0.29  |
| I <sub>4</sub>                                   | 18.30 | 26.83 | 4.58  | 0.80  | 0.18  |
| CD- T/<br>S I(0.05)                              | 9.238 | NS    | NS    | NS    | 0.062 |
| CD -S(0.05)                                      | 4.619 | NS    | NS    | 0.228 | 0.031 |
| CD-I(0.05)                                       | 4.619 | NS    | 0.898 | NS    | 0.031 |

NS-not significant

pith and Composting Inoculum) recorded the highest mean value of 41.69 mg kg<sup>-1</sup> which was on par with the treatments T<sub>13</sub> (37.81 mg kg<sup>-1</sup>), T<sub>17</sub> (37.56 mg kg<sup>-1</sup>) and T<sub>12</sub> (36.49 mg kg<sup>-1</sup>) while treatment T<sub>6</sub> recorded the lowest mean value of 10.91 mg kg<sup>-1</sup>. All the treatments were noted as superior than the absolute control T<sub>19</sub> (9.53 mg kg<sup>-1</sup>). Regarding the individual effect of substrates on available Fe content, it was found to be significant. Substrate S<sub>2</sub> (coir pith) recorded the highest mean value of 31.22 mg kg<sup>-1</sup> and the substrate S<sub>1</sub> (water cabbage) recorded the lowest mean value of 19.05 mg kg<sup>-1</sup>. Considering the individual effect of inoculants on available Fe content, it was also significant. Inoculant I<sub>3</sub> (Composting Inoculum) was found with highest mean value of 35.19 mg kg<sup>-1</sup> and the inoculant I<sub>4</sub> (commercial enzyme cocktail) recorded the lowest mean value of 18.30 mg kg<sup>-1</sup>.

#### 4.3.2.7.2. Manganese Content

Application of various treatments did not impose any significant effect on the available Mn content of the treated soils. The mean values ranged from 17.71 mg kg<sup>-1</sup> to 42.57 mg kg<sup>-1</sup>. T<sub>13</sub> (100 per cent N as compost from water cabbage and Composting Inoculum) recorded the highest Mn content of 42.57 mg kg<sup>-1</sup> and the treatment I<sub>3</sub> recorded the lowest mean value of 17.71 mg kg<sup>-1</sup>. But all the treatments were found to be superior to the absolute control T<sub>19</sub> (11.25 mg kg<sup>-1</sup>). Though the individual effect of substrates and inoculants had no significant effect the substrate S<sub>3</sub> (water hyacinth) recorded the highest mean value of 34.15 mg kg<sup>-1</sup> and substrate S<sub>1</sub> (water cabbage) has recorded the lowest mean value of 26.12 mg kg<sup>-1</sup>. I<sub>3</sub> (Composting Inoculum) has recorded the highest Mn content (32.35 mg kg<sup>-1</sup>) among the inoculants, while the lowest mean value of 25.88 mg kg<sup>-1</sup> was recorded by I<sub>1</sub> (*T. reesei*).

#### 4.3.2.7.3. Zinc Content

It was obvious from the data that available Zn content of the soil was not influenced by the treatments and substrates but the inoculant had statistically significant effect. The mean values were ranged from 2.60 mg kg<sup>-1</sup> to 5.46 mg kg<sup>-1</sup> and the highest value was registered by the treatment T<sub>14</sub> (100 per cent N as

compost from water hyacinth and commercial enzyme cocktail) with mean value of  $5.46 \text{ mg kg}^{-1}$  and the lowest value was recorded by the treatment  $T_9$  ( $2.60 \text{ mg kg}^{-1}$ ). But all the treatments were superior to the absolute control  $T_{19}$  ( $1.59 \text{ mg kg}^{-1}$ ). Regarding the individual effect of substrates, it was also found to be not significant. Substrate  $S_2$  (coir pith) recorded the highest mean value of  $4.29 \text{ mg kg}^{-1}$  and the lowest value was recorded by  $S_4$  (farm waste) ( $4.09 \text{ mg kg}^{-1}$ ). Considering the individual effect of inoculants, it could be observed from the data that the available Zn content of the soil was found to be significant.  $I_1$  (*T.reesei*) registered the highest value of  $4.72 \text{ mg kg}^{-1}$  which was on par with  $I_2$  (*P. sajor-caju*) and  $I_4$  (commercial enzyme cocktail) and the inoculant  $I_3$  (Composting Inoculum) recorded the lowest mean value of  $3.43 \text{ mg kg}^{-1}$ .

#### 4.3.2.7.4. Copper Content

Perusal of the data revealed that the available Cu content in the soil was not significantly varied with treatments; variation was significant only for the substrates. The average values ranged from  $0.57 \text{ mg kg}^{-1}$  to  $1.12 \text{ mg kg}^{-1}$ . Treatments  $T_{13}$  (100 per cent N as compost from water hyacinth and Composting Inoculum) and  $T_5$  (100 per cent N as compost from water cabbage and Composting Inoculum) recorded  $1.12 \text{ mg kg}^{-1}$  of available Cu and the treatment  $T_{15}$  recorded the lowest value of  $0.57 \text{ mg kg}^{-1}$ . However all the sixteen treatments were found to be superior to the absolute control  $T_{19}$  with mean value of  $0.27 \text{ mg kg}^{-1}$ . Considering the individual effect of substrates, it was found to be significant on the available Cu content of the treated soils. Substrate  $S_1$  (water cabbage) has recorded the highest mean value of  $0.96 \text{ mg kg}^{-1}$  which was on par with  $S_2$  and  $S_3$  and the substrate  $S_4$  recorded the lowest mean value of  $0.66 \text{ mg kg}^{-1}$ . The individual effect of inoculants on substrate for Cu content was not statistically significant, though the values varied with inoculants.  $I_3$  (Composting Inoculum) recorded the highest Cu content of  $0.99 \text{ mg kg}^{-1}$  while the lowest content was recorded by the inoculants  $I_1$  (*T. reesei*) ( $0.73 \text{ mg kg}^{-1}$ ).

#### 4.3.2.7.5. Boron Content

From the study it is inferred that the B content in the soil was significantly influenced by the treatments, substrates and the inoculants. The treatment T<sub>2</sub> (100 per cent N as compost from vermin compost) recorded the highest B content with mean value 0.90 mg kg<sup>-1</sup> which was on par with T<sub>1</sub> (0.85 mg kg<sup>-1</sup>). However among the other sixteen treatments, T<sub>11</sub> recorded the highest B content (0.78 mg kg<sup>-1</sup>) which was significantly superior to all other treatments. Absolute control recorded by the lowest B content of 0.09 mg kg<sup>-1</sup>. With regard to the individual effect, substrates imposed significant influence on the B content. S<sub>3</sub>(water hyacinth (0.44 mg kg<sup>-1</sup>) recorded the highest B content which was on par with S<sub>4</sub> (farm waste) (0.40 mg kg<sup>-1</sup>). Among the inoculants I<sub>1</sub> (*T. reesei*) and I<sub>2</sub> (*P. sajor-caju*) were recorded as the best with mean value of 0.43 mg kg<sup>-1</sup> and were on par with each other.

#### 4.3.2.8. Heavy Metal Content of the Soil

Heavy metal content in the soil which was treated by the composts are presented in the Table 17.

##### 4.3.2.8.1. Lead

Regarding the Pb content in the treated soils, T<sub>16</sub> (100 per cent N as compost from farm waste and *P.sajor-caju*) recorded the highest Pb content of 0.279 mg kg<sup>-1</sup> and T<sub>7</sub> recorded the value of 0.053 mg kg<sup>-1</sup>. However Pb content in the absolute control T<sub>19</sub> recorded the lowest value (0.018 mg kg<sup>-1</sup>) than the treatments. Soil treated with compost prepared from S<sub>1</sub> (water cabbage) had Pb below the detectable limit and all other substrates resulted in varying levels of lead accumulation in treated soils.

##### 4.3.2.8.2. Cadmium

In the case of Cd, the highest content was recorded in the treatment T<sub>9</sub> (100 per cent N as compost from coir pith and Composting Inoculum) (1.369 µg kg<sup>-1</sup>) followed by T<sub>15</sub> (1.366 µg kg<sup>-1</sup>). Treatments T<sub>4</sub> and T<sub>5</sub> were reported to have Cd below the detectable level.

Table 17. Effect of treatments on heavy metal content of the soil.

| Treatments                                       | Pb<br>(mg kg <sup>-1</sup> ) | Cd<br>(µg kg <sup>-1</sup> ) | Ni<br>(mg kg <sup>-1</sup> ) | Sn<br>(µg kg <sup>-1</sup> ) |
|--|------------------------------|------------------------------|------------------------------|------------------------------|
| T <sub>1</sub>                                   | 0.941                        | 1.166                        | 0.406                        | N.D                          |
| T <sub>2</sub>                                   | 0.564                        | 0.466                        | 0.432                        | N.D                          |
| T <sub>3</sub> (S <sub>1</sub> I <sub>1</sub> )  | N.D                          | 0.088                        | 0.441                        | N.D                          |
| T <sub>4</sub> (S <sub>1</sub> I <sub>2</sub> )  | N.D                          | N.D                          | 0.461                        | N.D                          |
| T <sub>5</sub> (S <sub>1</sub> I <sub>3</sub> )  | N.D                          | N.D                          | 0.512                        | N.D                          |
| T <sub>6</sub> (S <sub>1</sub> I <sub>4</sub> )  | N.D                          | 0.064                        | 0.539                        | N.D                          |
| T <sub>7</sub> (S <sub>2</sub> I <sub>1</sub> )  | 0.053                        | 0.823                        | 0.616                        | N.D                          |
| T <sub>8</sub> (S <sub>2</sub> I <sub>2</sub> )  | 0.185                        | 0.575                        | 0.027                        | N.D                          |
| T <sub>9</sub> (S <sub>2</sub> I <sub>3</sub> )  | 0.194                        | 1.369                        | 0.032                        | N.D                          |
| T <sub>10</sub> (S <sub>2</sub> I <sub>4</sub> ) | 0.187                        | 1.164                        | 0.036                        | N.D                          |
| T <sub>11</sub> (S <sub>3</sub> I <sub>1</sub> ) | 0.066                        | 0.653                        | 0.012                        | N.D                          |
| T <sub>12</sub> (S <sub>3</sub> I <sub>2</sub> ) | 0.192                        | 0.293                        | 0.067                        | N.D                          |
| T <sub>13</sub> (S <sub>3</sub> I <sub>3</sub> ) | 0.260                        | 0.796                        | 0.073                        | N.D                          |
| T <sub>14</sub> (S <sub>3</sub> I <sub>4</sub> ) | 0.219                        | 0.773                        | 0.052                        | N.D                          |
| T <sub>15</sub> (S <sub>4</sub> I <sub>1</sub> ) | 0.219                        | 1.366                        | 0.017                        | N.D                          |
| T <sub>16</sub> (S <sub>4</sub> I <sub>2</sub> ) | 0.279                        | 0.808                        | 0.077                        | N.D                          |
| T <sub>17</sub> (S <sub>4</sub> I <sub>3</sub> ) | 0.168                        | 0.531                        | 0.014                        | N.D                          |
| T <sub>18</sub> (S <sub>4</sub> I <sub>4</sub> ) | 0.212                        | 0.956                        | 0.070                        | N.D                          |
| T <sub>19</sub>                                  | 0.018                        | 0.062                        | 0.037                        | N.D                          |
| S <sub>1</sub>                                   | -                            | -                            | 0.49                         | -                            |
| S <sub>2</sub>                                   | -                            | -                            | 0.18                         | -                            |
| S <sub>3</sub>                                   | -                            | -                            | 0.05                         | -                            |
| S <sub>4</sub>                                   | -                            | -                            | 0.04                         | -                            |
| I <sub>1</sub>                                   | -                            | -                            | 0.27                         | -                            |
| I <sub>2</sub>                                   | -                            | -                            | 0.16                         | -                            |
| I <sub>3</sub>                                   | -                            | -                            | 0.16                         | -                            |
| I <sub>4</sub>                                   | -                            | -                            | 0.17                         | -                            |
| CD- T/<br>S I(0.05)                              | -                            | -                            | 0.024                        | -                            |
| CD -S(0.05)                                      | -                            | -                            | 0.012                        | -                            |
| CD-I(0.05)                                       | -                            | -                            | 0.012                        | -                            |

ND- not detectable

#### 4.3.2.8.3. Nickel

Ni content in the soil ranged from 0.616 mg kg<sup>-1</sup> to 0.012 mg kg<sup>-1</sup> and the highest mean value was recorded by the treatment T<sub>7</sub> (100 per cent N as compost from coir pith and *T. reesei*) while the lowest mean value was recorded by the Treatment T<sub>11</sub> (0.012) respectively.

#### 4.3.2.8.4. Tin

Regarding the Sn content, the treated soils were found to have below the detectable limit.

### 4.3.3. Effect of Treatments on Biological Properties of the Soil

The different treatments applied had significant effect on the biological properties of the soil. Important data on soil enzymes *viz.* urease, phosphatase, arylsulphatase, dehydrogenase,  $\beta$ - glucosidase and other biological properties like soil respiratory activity and viable count of microflora are presented on the Table 18.

#### 4.3.3.1. Urease Activity

Regarding the urease activity, the data showed that the urease enzyme activity varied significantly with the treatments, substrates and the inoculants (Table 18). The mean values ranged from 137.91 to 248.68  $\mu\text{g}$  urea hydrolysed g<sup>-1</sup> soil hr<sup>-1</sup>. The highest urease activity was recorded by the treatment T<sub>13</sub> (100 per cent N as compost from water hyacinth and Composting Inoculum) (248.68  $\mu\text{g}$  kg<sup>-1</sup> urea hydrolysed g<sup>-1</sup> soil hr<sup>-1</sup>) and the lowest mean value was recorded by the treatment T<sub>7</sub> (137.91  $\mu\text{g}$  kg<sup>-1</sup> urea hydrolysed g<sup>-1</sup> soil hr<sup>-1</sup>). Considering the individual effect of substrates and inoculants, a significant effect was noticed on urease enzyme activity. The substrate S<sub>1</sub> recorded the highest mean value of 205.49  $\mu\text{g}$  kg<sup>-1</sup> urea hydrolysed g<sup>-1</sup> soil hr<sup>-1</sup> while the substrate S<sub>2</sub> recorded the lowest mean value (162.65  $\mu\text{g}$  kg<sup>-1</sup> urea hydrolysed g<sup>-1</sup> soil hr<sup>-1</sup>). Considering the individual effect of inoculant, I<sub>3</sub> recorded the highest activity of 210.84  $\mu\text{g}$  kg<sup>-1</sup> urea hydrolysed g<sup>-1</sup> soil hr<sup>-1</sup> and I<sub>1</sub> recorded the lowest mean value (163.42  $\mu\text{g}$  kg<sup>-1</sup> urea hydrolysed g<sup>-1</sup> soil hr<sup>-1</sup>).

Table 18. Effect of treatments on biological properties of the soil.

| Treatments                                       | Urease activity | Phosphatase activity | Dehydrogenase activity | Aryl sulphatase | $\beta$ -D glucosidase |
|--|-----------------|----------------------|------------------------|-----------------|------------------------|
| T <sub>1</sub>                                   | 152.96          | 18.81                | 198.46                 | 0.022           | 2.10                   |
| T <sub>2</sub>                                   | 233.02          | 14.45                | 216.38                 | 0.020           | 3.07                   |
| T <sub>3</sub> (S <sub>1</sub> I <sub>1</sub> )  | 217.95          | 18.46                | 158.59                 | 0.018           | 3.40                   |
| T <sub>4</sub> (S <sub>1</sub> I <sub>2</sub> )  | 203.82          | 17.95                | 129.34                 | 0.032           | 2.70                   |
| T <sub>5</sub> (S <sub>1</sub> I <sub>3</sub> )  | 209.59          | 15.39                | 248.45                 | 0.015           | 3.03                   |
| T <sub>6</sub> (S <sub>1</sub> I <sub>4</sub> )  | 190.61          | 18.33                | 139.74                 | 0.028           | 3.63                   |
| T <sub>7</sub> (S <sub>2</sub> I <sub>1</sub> )  | 137.91          | 18.62                | 165.35                 | 0.029           | 4.10                   |
| T <sub>8</sub> (S <sub>2</sub> I <sub>2</sub> )  | 156.63          | 20.14                | 159.16                 | 0.031           | 4.17                   |
| T <sub>9</sub> (S <sub>2</sub> I <sub>3</sub> )  | 184.78          | 23.13                | 168.71                 | 0.028           | 4.03                   |
| T <sub>10</sub> (S <sub>2</sub> I <sub>4</sub> ) | 171.29          | 15.75                | 92.22                  | 0.017           | 4.37                   |
| T <sub>11</sub> (S <sub>3</sub> I <sub>1</sub> ) | 149.19          | 16.24                | 133.31                 | 0.016           | 3.47                   |
| T <sub>12</sub> (S <sub>3</sub> I <sub>2</sub> ) | 152.36          | 15.67                | 192.39                 | 0.023           | 4.33                   |
| T <sub>13</sub> (S <sub>3</sub> I <sub>3</sub> ) | 248.68          | 14.68                | 252.34                 | 0.037           | 3.47                   |
| T <sub>14</sub> (S <sub>3</sub> I <sub>4</sub> ) | 187.45          | 16.45                | 138.48                 | 0.017           | 2.77                   |
| T <sub>15</sub> (S <sub>4</sub> I <sub>1</sub> ) | 148.62          | 18.08                | 160.74                 | 0.023           | 4.07                   |
| T <sub>16</sub> (S <sub>4</sub> I <sub>2</sub> ) | 148.13          | 18.19                | 132.06                 | 0.016           | 3.40                   |
| T <sub>17</sub> (S <sub>4</sub> I <sub>3</sub> ) | 200.30          | 13.22                | 151.26                 | 0.017           | 4.77                   |
| T <sub>18</sub> (S <sub>4</sub> I <sub>4</sub> ) | 195.68          | 20.23                | 195.94                 | 0.018           | 4.07                   |
| T <sub>19</sub>                                  | 140.02          | 22.84                | 67.54                  | 0.016           | 5.20                   |
| S <sub>1</sub>                                   | 205.49          | 17.53                | 169.03                 | 0.021           | 3.19                   |
| S <sub>2</sub>                                   | 162.65          | 19.41                | 146.36                 | 0.03            | 4.17                   |
| S <sub>3</sub>                                   | 184.42          | 15.76                | 179.13                 | 0.023           | 3.51                   |
| S <sub>4</sub>                                   | 172.18          | 17.43                | 160.00                 | 0.019           | 4.08                   |
| I <sub>1</sub>                                   | 163.42          | 17.85                | 154.50                 | 0.022           | 3.76                   |
| I <sub>2</sub>                                   | 165.23          | 17.99                | 153.24                 | 0.026           | 3.65                   |
| I <sub>3</sub>                                   | 210.84          | 16.61                | 205.19                 | 0.025           | 3.83                   |
| I <sub>4</sub>                                   | 186.26          | 17.69                | 141.60                 | 0.020           | 3.708                  |
| CD- T/ S I                                       | 22.766          | NS                   | 61.937                 | 0.004           | 1.066                  |
| CD -S  | 11.383          | NS                   | NS                     | 0.002           | 0.533                  |
| CD-I   | 11.383          | NS                   | 30.968                 | 0.002           | NS                     |

NS-not significant

Urease activity-  $\mu\text{g}$  urea hydrolysed  $\text{g}^{-1}$  soil  $\text{hr}^{-1}$ Phosphatase activity-  $\mu\text{g}$  pnp released  $\text{g}^{-1}$  soil  $\text{hr}^{-1}$ Dehydrogenase activity -  $\mu\text{g}$  TPF hydrolysed  $\text{g}^{-1}$  soil 24 hrs<sup>-1</sup>Arylsulphates activity-  $\mu\text{M}$  pnp released  $\text{g}^{-1}$  soil  $\text{hr}^{-1}$  $\beta$ -D glucosidase activity-  $\mu\text{g}$  pnp  $\beta$ - D Glucosidase  $\text{g}^{-1}$  soil  $\text{hr}^{-1} \times 10^{-4}$

#### 4.3.6.2. Phosphatase Activity

It is inferred from the Table 18 that the phosphatase enzyme activity was not significantly influenced by the treatments. However the mean values ranged from 13.22  $\mu\text{g pnp released g}^{-1} \text{ soil hr}^{-1}$  to 23.13  $\mu\text{g pnp released g}^{-1} \text{ soil hr}^{-1}$  and T<sub>9</sub> (100 per cent N as compost from coir pith and Composting Inoculum) recorded the highest value which was on par with T<sub>19</sub> (22.84  $\mu\text{g pnp released g}^{-1} \text{ soil hr}^{-1}$ ). The lowest mean value was recorded by the treatment T<sub>17</sub> with mean value of 13.22 ( $\mu\text{g pnp released g}^{-1} \text{ soil hr}^{-1}$ ) and it was inferred as the best treatment due to the inverse relationship of phosphatase enzyme activity with the inorganic phosphorous content. Considering the individual effects of substrate on phosphatase activity the substrate S<sub>2</sub> recorded the highest value (19.41  $\mu\text{g pnp released g}^{-1} \text{ soil hr}^{-1}$ ) while S<sub>3</sub> recorded the lowest mean value of 15.76  $\mu\text{g pnp released g}^{-1} \text{ soil hr}^{-1}$ . Regarding the effect of inoculants, I<sub>2</sub> recorded the highest mean value of 17.99  $\mu\text{g pnp released g}^{-1} \text{ soil hr}^{-1}$  and I<sub>3</sub> recorded the lowest mean value of 16.61  $\mu\text{g pnp released g}^{-1} \text{ soil hr}^{-1}$ .

#### 4.3.6.3. Dehydrogenase Activity

It is obvious from the data that the treatments and inoculants imposed significant influence on the dehydrogenase enzyme activity of the treated soils. The mean values ranged from 67.54 ( $\mu\text{g TPF hydrolysed g}^{-1} \text{ soil 24 hrs}^{-1}$ ) to 252.34 ( $\mu\text{g TPF hydrolysed g}^{-1} \text{ soil 24 hrs}^{-1}$ ) and the highest mean value was recorded by the treatment T<sub>13</sub> (100 per cent N as compost from water hyacinth and Composting Inoculum) (252.34  $\mu\text{g TPF hydrolysed g}^{-1} \text{ soil 24 hrs}^{-1}$ ) which was on par the treatments T<sub>5</sub> (248.45  $\mu\text{g TPF hydrolysed g}^{-1} \text{ soil 24 hrs}^{-1}$ ), T<sub>2</sub> (216.38  $\mu\text{g TPF hydrolysed g}^{-1} \text{ soil 24 hrs}^{-1}$ ), T<sub>1</sub> (198.46  $\mu\text{g TPF hydrolysed g}^{-1} \text{ soil 24 hrs}^{-1}$ ), T<sub>18</sub> (195.94  $\mu\text{g TPF hydrolysed g}^{-1} \text{ soil 24 hrs}^{-1}$ ), and T<sub>12</sub> (192.39  $\mu\text{g TPF hydrolysed g}^{-1} \text{ soil 24 hrs}^{-1}$ ) and the lowest mean value was recorded by the treatment T<sub>10</sub> with mean value 92.22 ( $\mu\text{g TPF hydrolysed g}^{-1} \text{ soil 24 hrs}^{-1}$ ). However all the treatments were noticed as superior to the absolute control T<sub>19</sub> (67.54  $\mu\text{g TPF hydrolysed g}^{-1} \text{ soil 24 hrs}^{-1}$ ). Considering the individual effect of the substrates on the dehydrogenase enzyme activity, it was found that the effect



of substrates was not significant. Even though the highest mean value was recorded by the substrate S<sub>3</sub> (179.13 µg TPF hydrolysed g<sup>-1</sup> soil 24 hrs<sup>-1</sup>) and the lowest mean value was recorded for S<sub>2</sub> (146.36 µg TPF hydrolysed g<sup>-1</sup> soil per 24 hrs). Effect of inoculants on the dehydrogenase enzyme activity was found to be significant and the inoculant I<sub>3</sub> has recorded the highest mean value of 205.19 µg TPF hydrolysed g<sup>-1</sup> soil 24 hrs<sup>-1</sup>. Inoculant I<sub>4</sub> has recorded the lowest mean value 141.60 µg TPF hydrolysed g<sup>-1</sup> soil 24 hrs<sup>-1</sup> and were on par with I<sub>1</sub> and I<sub>2</sub>.

#### 4.3.6.4. Arylsulphatase Activity

Activity of arylsulphatase enzyme significantly varied with different treatments. The mean values ranged from 0.16 µM pnp released g<sup>-1</sup> soil hr<sup>-1</sup> to 0.037 µM pnp released g<sup>-1</sup> soil hr<sup>-1</sup>. Among the treatments T<sub>13</sub> (100 per cent N as compost from water hyacinth and Composting Inoculum) has recorded the highest mean value (0.037 µM pnp released g<sup>-1</sup> soil hr<sup>-1</sup>) while T<sub>5</sub> has recorded the lowest mean value (0.015 µM pnp released g<sup>-1</sup> soil hr<sup>-1</sup>). All the treatments were found to be superior to the absolute control (0.016 µM pnp released g<sup>-1</sup> soil hr<sup>-1</sup>) except for T<sub>5</sub> (0.015 µM pnp released g<sup>-1</sup> soil hr<sup>-1</sup>) which was lower than T<sub>19</sub>. Different substrates and inoculants also influenced the arylsulphatase enzyme activity significantly. Considering the individual effect of substrates, S<sub>2</sub> recorded the highest mean value of 0.03 µM pnp released g<sup>-1</sup> soil hr<sup>-1</sup> S<sub>4</sub> recorded the lowest mean value 0.019 µM pnp released g<sup>-1</sup> soil hr<sup>-1</sup>. Among the inoculants, the I<sub>2</sub> recorded the highest mean value of 0.026 µM pnp released g<sup>-1</sup> soil hr<sup>-1</sup> and I<sub>4</sub> recorded the lowest mean value 0.020 µM pnp released g<sup>-1</sup> soil hr<sup>-1</sup>.

#### 4.3.6.5. β- Glucosidase Activity

Critical appraisal of the data revealed that the β- Glucosidase enzyme activity was found to be significantly influenced by the treatments. The mean values were ranged from 2.1x10<sup>-4</sup> to 5.2 x10<sup>-4</sup> pnp β- D Glucosidase g<sup>-1</sup> soil hr<sup>-1</sup>. The highest mean value was recorded by the treatment T<sub>17</sub> (100 per cent N as compost from farm waste and Composting Inoculum) (4.767 x10<sup>-4</sup> pnp β-D Glucosidase g<sup>-1</sup> soil hr<sup>-1</sup>). But absolute control T<sub>19</sub> recorded a mean value of 5.2x10<sup>-4</sup> pnp β-D Glucosidase g<sup>-1</sup> soil hr<sup>-1</sup> which was significantly superior to all

Table 18. Effect of treatments on biological properties of the soil continued.

| Treatments                                       | Soil respiratory activity ( $\mu\text{g CO}_2$ evolved $\text{g}^{-1}$ soil $\text{hr}^{-1}$ ) | Microbial load (Viable count- cfu $\text{g}^{-1}$ soil) |                     |                             |
|--|--|---|---------------------|-----------------------------|
|  |  | Bacteria $\times 10^6$                                  | Fungi $\times 10^4$ | Actinomycetes $\times 10^3$ |
| T <sub>1</sub>                                   | 8.07   | 35.67   | 5.00                | 14.00                       |
| T <sub>2</sub>                                   | 8.73   | 124.67  | 7.33                | 18.00                       |
| T <sub>3</sub> (S <sub>1</sub> I <sub>1</sub> )  | 6.67   | 137.67  | 11.33               | 13.00                       |
| T <sub>4</sub> (S <sub>1</sub> I <sub>2</sub> )  | 7.77   | 126.00  | 10.00               | 13.67                       |
| T <sub>5</sub> (S <sub>1</sub> I <sub>3</sub> )  | 8.36   | 137.33  | 9.33                | 18.67                       |
| T <sub>6</sub> (S <sub>1</sub> I <sub>4</sub> )  | 8.14   | 113.33  | 8.67                | 8.33                        |
| T <sub>7</sub> (S <sub>2</sub> I <sub>1</sub> )  | 8.95   | 90.00   | 7.33                | 10.67                       |
| T <sub>8</sub> (S <sub>2</sub> I <sub>2</sub> )  | 8.07   | 82.00   | 7.33                | 18.00                       |
| T <sub>9</sub> (S <sub>2</sub> I <sub>3</sub> )  | 6.60   | 118.00  | 6.67                | 14.00                       |
| T <sub>10</sub> (S <sub>2</sub> I <sub>4</sub> ) | 7.55   | 97.00   | 7.67                | 20.67                       |
| T <sub>11</sub> (S <sub>3</sub> I <sub>1</sub> ) | 7.92   | 111.00  | 10.33               | 21.33                       |
| T <sub>12</sub> (S <sub>3</sub> I <sub>2</sub> ) | 7.33   | 98.67   | 9.00                | 10.33                       |
| T <sub>13</sub> (S <sub>3</sub> I <sub>3</sub> ) | 7.85   | 146.33  | 10.67               | 13.67                       |
| T <sub>14</sub> (S <sub>3</sub> I <sub>4</sub> ) | 7.70   | 128.67  | 7.67                | 16.67                       |
| T <sub>15</sub> (S <sub>4</sub> I <sub>1</sub> ) | 6.38   | 94.00   | 6.00                | 12.67                       |
| T <sub>16</sub> (S <sub>4</sub> I <sub>2</sub> ) | 7.41   | 89.00   | 7.00                | 16.00                       |
| T <sub>17</sub> (S <sub>4</sub> I <sub>3</sub> ) | 8.73   | 100.67  | 7.33                | 16.00                       |
| T <sub>18</sub> (S <sub>4</sub> I <sub>4</sub> ) | 7.92   | 93.00   | 6.00                | 13.33                       |
| T <sub>19</sub>                                  | 7.04   | 25.00   | 3.33                | 7.00                        |
| S <sub>1</sub>                                   | 7.74   | 128.58  | 9.83                | 13.42                       |
| S <sub>2</sub>                                   | 7.79   | 96.75   | 7.25                | 15.83                       |
| S <sub>3</sub>                                   | 7.70   | 121.17  | 9.42                | 15.50                       |
| S <sub>4</sub>                                   | 7.61   | 94.17   | 6.58                | 14.50                       |
| I <sub>1</sub>                                   | 7.48   | 108.17  | 8.75                | 14.42                       |
| I <sub>2</sub>                                   | 7.65   | 98.92   | 8.33                | 14.50                       |
| I <sub>3</sub>                                   | 7.89   | 125.58  | 8.50                | 15.58                       |
| I <sub>4</sub>                                   | 7.83   | 108.00  | 7.50                | 14.75                       |
| CD- T/<br>S I(0.05)                              | 2.358  | 39.322  | 2.554               | 7.065                       |
| CD -S(0.05)                                      | 1.178  | 19.66   | 1.277               | NS                          |
| CD-I(0.05)                                       | NS   | NS  | NS                  | NS                          |

NS- not significant

the sixteen treatments and the lowest value was recorded by T<sub>1</sub> ( $2.1 \times 10^{-4}$  pnp  $\beta$ -D Glucosidase g<sup>-1</sup> soil hr<sup>-1</sup>). The individual effect of the substrates, were found to be significant on  $\beta$ - Glucosidase activity. Among the substrates, S<sub>2</sub> recorded the highest mean value of  $4.17 \times 10^{-4}$  and the lowest mean value was recorded by the substrate S<sub>1</sub> ( $3.19 \times 10^{-4}$  pnp  $\beta$ -D Glucosidase g<sup>-1</sup> soil hr<sup>-1</sup>). Considering the individual effect of inoculants, variation was not statistically significant. Inoculant I<sub>3</sub> recorded the highest mean value ( $3.83 \times 10^{-4}$  pnp  $\beta$ -D Glucosidase g<sup>-1</sup> soil hr<sup>-1</sup>) and I<sub>2</sub> ( $3.65 \times 10^{-4}$  pnp  $\beta$ -D Glucosidase g<sup>-1</sup> soil hr<sup>-1</sup>) recorded the lowest mean value.

#### 4.3.6.6. Soil Respiratory Activity

Table 18(continued) presented the effect of treatments on soil respiratory activity and the mean values ranged from 6.38 to 8.95  $\mu$ g CO<sub>2</sub> evolved g<sup>-1</sup>soil hr<sup>-1</sup>. Various treatments had no significant effect on soil respiratory activity. The treatment T<sub>7</sub> (100 per cent N as compost from coir pith and *T.reesei*) recorded the highest mean value of 8.95  $\mu$ g CO<sub>2</sub> evolved g<sup>-1</sup>soil hr<sup>-1</sup> while treatment T<sub>15</sub> recorded the lowest mean value 6.38  $\mu$ g of CO<sub>2</sub> evolved g<sup>-1</sup> soil hr<sup>-1</sup>. Among the individual effect of substrates and inoculants, were found to be non significant on soil respiratory activity. However the substrate S<sub>2</sub> ( $7.79 \mu$ g CO<sub>2</sub> evolved g<sup>-1</sup>soil hr<sup>-1</sup>) recorded the highest respiratory activity and lowest mean value was recorded by S<sub>4</sub> with mean value of  $7.61 \mu$ g of CO<sub>2</sub> evolved g<sup>-1</sup> of soil hr<sup>-1</sup>. Regarding the effect of inoculants on respiratory activity of the soil, it was also found to be not significant. The highest mean value was recorded by I<sub>3</sub> ( $7.89 \mu$ g CO<sub>2</sub> evolved g<sup>-1</sup>soil hr<sup>-1</sup>) and lowest mean value was recorded by I<sub>1</sub> ( $7.48 \mu$ g CO<sub>2</sub> evolved g<sup>-1</sup>soil hr<sup>-1</sup>).

#### 4.3.6.7. Microbial Load

The population of bacteria, fungi and actinomycetes were recorded as the total viable count and the data was presented in the Table 18(continued).

The effect of treatments on count of bacteria was found to be significant. The mean value ranged from  $82 \times 10^6$ cfu g<sup>-1</sup>soil to  $146.33 \times 10^6$  cfu g<sup>-1</sup>soil. Maximum significant bacterial population of  $146.33 \times 10^6$  cfu g<sup>-1</sup> soil was

recorded in the treatment T<sub>13</sub> (100 per cent N as compost from water hyacinth and Composting Inoculum) and the lowest population was observed in the treatment T<sub>8</sub> (82 x 10<sup>6</sup> cfu g<sup>-1</sup> soil) however all the treatments were superior to the absolute control. The effect of individual substrates on count of bacteria was also significant. The substrate S<sub>1</sub> has recorded the highest bacteria count of 128.58 x 10<sup>6</sup> cfu g<sup>-1</sup> soil and the substrate S<sub>4</sub> recorded the lowest mean value of 94.17 x 10<sup>6</sup> cfu g<sup>-1</sup> soil. The data on effect of inoculants was not statistically significant. However the highest count of bacteria was recorded by the inoculant I<sub>3</sub> (125.58 x 10<sup>6</sup> cfu g<sup>-1</sup> soil) and the lowest value was recorded by the inoculant I<sub>2</sub> (98.92 x 10<sup>6</sup> cfu g<sup>-1</sup> soil).

In case of fungi, the mean values ranged from 6 x 10<sup>4</sup> cfu g<sup>-1</sup> soil to 11.33 x 10<sup>4</sup> cfu g<sup>-1</sup> soil and the highest mean value was recorded by the treatment T<sub>3</sub> (100 per cent N as compost from water cabbage and *T. reesei*) and the lowest mean value was recorded by the treatment T<sub>15</sub>. However all the treatments were found to be significantly superior to the absolute control T<sub>19</sub> (3.33 x 10<sup>4</sup> cfu g<sup>-1</sup> soil). Regarding the individual effect of substrates, it was found to have significant effect on fungal count. S<sub>1</sub> has recorded the highest fungal count of 9.83 x 10<sup>4</sup> cfu g<sup>-1</sup> soil and substrate S<sub>4</sub> the lowest mean value of 6.58 x 10<sup>4</sup> cfu g<sup>-1</sup>. The effect of different of inoculants on fungal count was not statistically significant eventhough the highest mean value was recorded by the inoculant I<sub>1</sub> (8.75 x 10<sup>4</sup> cfu g<sup>-1</sup> soil) and the lowest mean value was recorded by the inoculant I<sub>4</sub> (7.50 x 10<sup>4</sup> cfu g<sup>-1</sup> soil).

Considering the count of actinomycetes, the mean values ranged from 7x10<sup>3</sup>cfu g<sup>-1</sup> soil to 21.33 x10<sup>3</sup>cfu g<sup>-1</sup> soil and the highest mean value was recorded by the treatment T<sub>11</sub> (100 per cent N as compost from water hyacinth and *T. reesei*) (21.33 x10<sup>3</sup>cfu g<sup>-1</sup> soil) and the lowest mean value of (8.333 x10<sup>3</sup>cfu g<sup>-1</sup> soil) was recorded by T<sub>6</sub> however, all the treatments were found to be superior than the absolute control T<sub>19</sub> (7 x10<sup>3</sup>cfu g<sup>-1</sup> soil). The individual effect of substrates was not statistically significant however the highest actinomycete count was recorded by the substrate S<sub>2</sub>(15.83 x10<sup>3</sup> cfu g<sup>-1</sup> soil) and lowest actinomycete count was recorded by the substrate S<sub>1</sub> (13.42 x10<sup>3</sup> cfu g<sup>-1</sup> soil). Individual effect

of inoculants were also non significant eventhough I<sub>3</sub> recorded the highest count of actinomycetes ( $15.58 \times 10^3$  cfu g<sup>-1</sup> soil) and the lowest count was recorded by the inoculant I<sub>1</sub>( $14.42 \times 10^3$  cfu g<sup>-1</sup> soil<sup>3</sup>).

#### **4.3.7. Effect of Treatments on Total Content of Major Nutrients in Amaranthus**

Table 19 presents the data on influence of the treatments on plant content of major nutrients viz. nitrogen, phosphorous and potassium.

##### **4.3.7.1. Total Nitrogen**

Regarding the total N content of the amaranthus, it was obvious from the data that treatments imposed significant influence and the mean values ranged from 1.36 to 2.93 per cent. The treatment T<sub>15</sub> (100 per cent N as compost from farm waste and *T.reesei*) recorded the highest mean value of 2.93 per cent and treatment T<sub>7</sub> recorded the lowest mean value of 1.36 per cent. All the treatments recorded were found to be superior to the absolute control T<sub>19</sub> (0.95 per cent). The individual substrates imposed significant effect on N content. Substrate S<sub>4</sub> has recorded the highest mean value of 2.60 per cent and the lowest value was recorded by the substrate S<sub>2</sub> (1.78 per cent). Regarding the individual effect of inoculants, even though their effect were found to be non significant inoculant I<sub>3</sub> recorded the highest N content of 2.20 per cent and inoculant I<sub>1</sub> recorded the lowest content of 1.98 per cent.

##### **4.3.7.2. Total Phosphorous**

Total phosphorous content of the amaranthus crop was not significantly influenced by the treatments, substrates or by their inoculants. The mean values ranged from 0.020 to 0.124 per cent and the highest mean value was recorded by the treatment T<sub>15</sub> (100 per cent N as compost from farm waste and *T.reesei*) (0.124 per cent) and the lowest mean value was recorded by the treatment T<sub>16</sub> (0.020 per cent). Regarding the individual effect of the substrate S<sub>4</sub> recorded the highest mean value (0.050 per cent) and the substrate S<sub>2</sub> recorded the lowest mean value of 0.023 per cent. Considering the effect of inoculants on total phosphorous

Table 19. Effect of treatments on the total content of major nutrients in amaranthus, %.

| Treatments                                       | Total Nitrogen | Total Posphorous | Total Potassium |
|--|----------------|------------------|-----------------|
| T <sub>1</sub>                                   | 1.40           | 0.035            | 5.63            |
| T <sub>2</sub>                                   | 2.69           | 0.024            | 4.30            |
| T <sub>3</sub> (S <sub>1</sub> I <sub>1</sub> )  | 1.75           | 0.031            | 3.87            |
| T <sub>4</sub> (S <sub>1</sub> I <sub>2</sub> )  | 2.02           | 0.028            | 4.40            |
| T <sub>5</sub> (S <sub>1</sub> I <sub>3</sub> )  | 2.46           | 0.046            | 5.30            |
| T <sub>6</sub> (S <sub>1</sub> I <sub>4</sub> )  | 2.13           | 0.030            | 4.80            |
| T <sub>7</sub> (S <sub>2</sub> I <sub>1</sub> )  | 1.36           | 0.021            | 3.67            |
| T <sub>8</sub> (S <sub>2</sub> I <sub>2</sub> )  | 1.90           | 0.023            | 4.07            |
| T <sub>9</sub> (S <sub>2</sub> I <sub>3</sub> )  | 2.02           | 0.023            | 3.50            |
| T <sub>10</sub> (S <sub>2</sub> I <sub>4</sub> ) | 1.83           | 0.024            | 3.80            |
| T <sub>11</sub> (S <sub>3</sub> I <sub>1</sub> ) | 1.89           | 0.042            | 3.83            |
| T <sub>12</sub> (S <sub>3</sub> I <sub>2</sub> ) | 1.85           | 0.033            | 4.97            |
| T <sub>13</sub> (S <sub>3</sub> I <sub>3</sub> ) | 1.90           | 0.050            | 4.17            |
| T <sub>14</sub> (S <sub>3</sub> I <sub>4</sub> ) | 2.09           | 0.030            | 4.13            |
| T <sub>15</sub> (S <sub>4</sub> I <sub>1</sub> ) | 2.93           | 0.124            | 4.97            |
| T <sub>16</sub> (S <sub>4</sub> I <sub>2</sub> ) | 2.46           | 0.020            | 3.97            |
| T <sub>17</sub> (S <sub>4</sub> I <sub>3</sub> ) | 2.41           | 0.025            | 4.60            |
| T <sub>18</sub> (S <sub>4</sub> I <sub>4</sub> ) | 2.61           | 0.031            | 4.53            |
| T <sub>19</sub>                                  | 0.95           | 0.016            | 4.10            |
| S <sub>1</sub>                                   | 2.09           | 0.034            | 4.59            |
| S <sub>2</sub>                                   | 1.78           | 0.023            | 3.76            |
| S <sub>3</sub>                                   | 1.93           | 0.039            | 4.28            |
| S <sub>4</sub>                                   | 2.60           | 0.050            | 4.52            |
| I <sub>1</sub>                                   | 1.98           | 0.054            | 4.08            |
| I <sub>2</sub>                                   | 2.06           | 0.026            | 4.35            |
| I <sub>3</sub>                                   | 2.20           | 0.036            | 4.39            |
| I <sub>4</sub>                                   | 2.17           | 0.029            | 4.32            |
| CD- T/<br>S I(0.05)                              | 0.617          | NS               | 1.233           |
| CD -S(0.05)                                      | 0.309          | NS               | 0.617           |
| CD-I(0.05)                                       | NS             | NS               | NS              |

NS-not significant

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content  $I_1$  recorded the highest mean value 0.054 per cent and the inoculant  $I_2$  recorded the lowest mean value 0.026 per cent.

#### **4.3.7.3. Total Potassium**

Total K content of the amaranthus is presented in the Table 19 and the critical appraisal of the data showed that the imposed treatments had significant effect on total K content of plants and the mean values ranged from 3.50 per cent to 5.63 per cent. Between the sixteen treatments the highest mean value was recorded by the treatment  $T_5$  (100 per cent N as compost from water cabbage and Composting Inoculum) (5.3 per cent) while the treatment  $T_1$  (POP) recorded the highest K value of 5.63 per cent which was on par with  $T_5$ . The lowest K content was recorded by the treatment  $T_9$  (3.50 per cent). Regarding the individual effect of substrates effect was found to be significant on K content and  $S_1$  recorded the highest mean value of 4.59 per cent and  $S_2$  has recorded the lowest mean value 3.76 per cent. Considering the individual effect of inoculant on the total K content  $I_3$  has recorded the highest mean value (4.39 per cent) and  $I_1$  recorded the lowest mean value (4.08 per cent).

#### **4.3.8. Effect of Treatments on Total Content of Micro Nutrients in amaranthus.**

The micronutrients content of the plants are presented in the Table 20.

##### **4.3.8.1. Iron Content**

Perusal of the data revealed that the different treatments had significant effect on the total Fe content of the plants. The average values ranged between 274.90 mg kg<sup>-1</sup> to 482.57 mg kg<sup>-1</sup> and the highest Fe content was recorded by the treatment  $T_9$  (100 per cent N as compost from coir pith and Composting Inoculum) (482.57 mg kg<sup>-1</sup>) and the lowest content was recorded by the treatment  $T_{18}$  (274.90 mg kg<sup>-1</sup>). However all the treatments were found to be superior to the absolute control (185.93 mg kg<sup>-1</sup>). Regarding the individual effect of substrates,  $S_1$  recorded the highest mean value (463.61 mg kg<sup>-1</sup>) and the lowest mean value was recorded by the substrate  $S_4$  (309.59 mg kg<sup>-1</sup>). Among the inoculants,  $I_3$  has

recorded the highest Fe content of 415.61 mg kg<sup>-1</sup> and the inoculant I<sub>1</sub> recorded the lowest mean value of 368.42 mg kg<sup>-1</sup>.

#### 4.3.8.2. Manganese Content

The treatments imposed significant effect on the total Mn content of the plant. Average values ranged between 82.55 mg kg<sup>-1</sup> and 157.17 mg kg<sup>-1</sup>. The highest mean value of Mn content was recorded by the treatment T<sub>13</sub> (100 per cent N as compost from water hyacinth and Composting Inoculum) (157.17 mg kg<sup>-1</sup>) and the lowest mean value was recorded by the treatment T<sub>8</sub> (82.55 mg kg<sup>-1</sup>). However all the sixteen treatments were found to be significantly superior than the absolute control T<sub>19</sub> (68.23 mg kg<sup>-1</sup>). Regarding the individual effect of substrates on the Mn content, S<sub>1</sub> has recorded the highest mean value of 132.00 mg kg<sup>-1</sup> and the substrate S<sub>2</sub> has recorded the lowest mean value of 94.07 mg kg<sup>-1</sup>. Considering the effect of inoculants, I<sub>3</sub> has recorded the highest mean value of 135.88 mg kg<sup>-1</sup> and I<sub>4</sub> has recorded the lowest mean value of 99.12 mg kg<sup>-1</sup>.

#### 4.3.8.3. Zinc Content

Zn content of the plants was found to be significantly influenced by the treatments, substrates and inoculants. Average content of the plants ranged from 17.86 mg kg<sup>-1</sup> to 54.03 mg kg<sup>-1</sup> while the highest mean value 54.03 mg kg<sup>-1</sup> recorded by the treatment T<sub>7</sub> (100 per cent N as compost from water cabbage and *T. reesei*) which was on par with the treatment T<sub>9</sub> (100 per cent N as compost from water cabbage and Composting Inoculum) (50.00 mg kg<sup>-1</sup>). The lowest content of Zn (17.86 mg kg<sup>-1</sup>) was recorded by the treatment T<sub>18</sub>. But all the treatments were noticed as superior to the absolute control T<sub>19</sub> (14.80 mg kg<sup>-1</sup>). Considering the individual effects of substrates, it was obvious from the data that the highest mean value was recorded by the substrate S<sub>2</sub> (41.85 mg kg<sup>-1</sup>) and the lowest mean value was recorded by the substrate S<sub>4</sub> (28.90 mg kg<sup>-1</sup>). Regarding the effect of inoculants I<sub>3</sub> has recorded the highest mean value 39.68 mg kg<sup>-1</sup> and the I<sub>4</sub> recorded the lowest mean value (30.83 mg kg<sup>-1</sup>).



Table 20. Effect of treatments on micronutrient content of amaranthus, mg kg<sup>-1</sup>.

| Treatments                                       | Fe      | Mn     | Zn     | Cu     | B     |
|--|---------|--------|--------|--------|-------|
| T <sub>1</sub>                                   | 473.43  | 105.49 | 29.87  | 25.60  | 42.55 |
| T <sub>2</sub>                                   | 322.19  | 135.38 | 29.69  | 30.26  | 59.87 |
| T <sub>3</sub> (S <sub>1</sub> I <sub>1</sub> )  | 463.42  | 139.11 | 31.43  | 43.43  | 49.45 |
| T <sub>4</sub> (S <sub>1</sub> I <sub>2</sub> )  | 463.03  | 132.49 | 29.68  | 30.10  | 45.98 |
| T <sub>5</sub> (S <sub>1</sub> I <sub>3</sub> )  | 468.63  | 149.12 | 40.50  | 49.21  | 57.46 |
| T <sub>6</sub> (S <sub>1</sub> I <sub>4</sub> )  | 459.37  | 107.27 | 37.90  | 38.10  | 44.79 |
| T <sub>7</sub> (S <sub>2</sub> I <sub>1</sub> )  | 320.13  | 95.37  | 54.03  | 43.88  | 39.08 |
| T <sub>8</sub> (S <sub>2</sub> I <sub>2</sub> )  | 298.00  | 82.55  | 35.53  | 33.84  | 34.61 |
| T <sub>9</sub> (S <sub>2</sub> I <sub>3</sub> )  | 482.57  | 110.23 | 50.00  | 26.89  | 52.28 |
| T <sub>10</sub> (S <sub>2</sub> I <sub>4</sub> ) | 348.00  | 88.11  | 27.84  | 35.11  | 30.02 |
| T <sub>11</sub> (S <sub>3</sub> I <sub>1</sub> ) | 358.83  | 120.76 | 37.38  | 25.12  | 39.79 |
| T <sub>12</sub> (S <sub>3</sub> I <sub>2</sub> ) | 437.27  | 106.94 | 34.43  | 46.49  | 54.43 |
| T <sub>13</sub> (S <sub>3</sub> I <sub>3</sub> ) | 408.67  | 157.17 | 32.54  | 49.22  | 63.12 |
| T <sub>14</sub> (S <sub>3</sub> I <sub>4</sub> ) | 408.50  | 106.33 | 39.72  | 36.85  | 42.96 |
| T <sub>15</sub> (S <sub>4</sub> I <sub>1</sub> ) | 331.30  | 107.03 | 33.06  | 17.73  | 44.42 |
| T <sub>16</sub> (S <sub>4</sub> I <sub>2</sub> ) | 329.60  | 117.66 | 29.01  | 28.44  | 52.45 |
| T <sub>17</sub> (S <sub>4</sub> I <sub>3</sub> ) | 302.57  | 127.00 | 35.66  | 25.90  | 56.79 |
| T <sub>18</sub> (S <sub>4</sub> I <sub>4</sub> ) | 274.90  | 94.78  | 17.86  | 14.64  | 38.76 |
| T <sub>19</sub>                                  | 185.93  | 68.23  | 14.80  | 11.61  | 30.44 |
| S <sub>1</sub>                                   | 463.61  | 132.00 | 34.88  | 40.21  | 49.42 |
| S <sub>2</sub>                                   | 362.18  | 94.07  | 41.85  | 34.93  | 39.00 |
| S <sub>3</sub>                                   | 403.32  | 122.80 | 36.02  | 39.42  | 50.07 |
| S <sub>4</sub>                                   | 309.59  | 111.62 | 28.90  | 21.68  | 48.10 |
| I <sub>1</sub>                                   | 368.42  | 115.57 | 38.97  | 32.54  | 43.18 |
| I <sub>2</sub>                                   | 381.98  | 109.91 | 32.16  | 34.72  | 46.87 |
| I <sub>3</sub>                                   | 415.61  | 135.88 | 39.68  | 37.80  | 57.41 |
| I <sub>4</sub>                                   | 372.69  | 99.12  | 30.83  | 31.17  | 39.13 |
| CD- T/<br>SI(0.05)                               | 164.807 | 47.236 | 13.776 | 10.444 | 6.883 |
| CD -S(0.05)                                      | 82.403  | 23.618 | 6.888  | 5.222  | 3.442 |
| CD-I(0.05)                                       | NS      | 23.618 | 6.888  | 5.222  | 3.442 |

NS-not significant

#### 4.3.8.4. Copper Content

The total Cu content of the plants were ranged from 14.64 mg kg<sup>-1</sup> to 49.22 mg kg<sup>-1</sup> and the highest value was recorded by the treatment T<sub>13</sub> (100 per cent N as compost from water hyacinth and Composting Inoculum) and the lowest mean value was recorded by the treatment T<sub>18</sub> respectively. T<sub>13</sub> was found to be on par with the treatments T<sub>5</sub> (49.21 mg kg<sup>-1</sup>), T<sub>12</sub> (46.49 mg kg<sup>-1</sup>), T<sub>7</sub> (43.88 mg kg<sup>-1</sup>), T<sub>3</sub> (43.43 mg kg<sup>-1</sup>) while the treatment T<sub>18</sub> has recorded the lowest mean value 14.64 mg kg<sup>-1</sup>. However all the treatments were found to be significantly superior than the absolute control T<sub>19</sub> (11.613 mg kg<sup>-1</sup>). Regarding the individual effect of substrates, S<sub>1</sub> (40.21 mg kg<sup>-1</sup>) recorded the highest mean value and the substrate S<sub>4</sub> (21.68 mg kg<sup>-1</sup>) recorded the lowest mean value. While inoculants were also found to have impose their individual effect on Cu content of plants. I<sub>3</sub> recorded the highest Cu content of 37.80 mg kg<sup>-1</sup> and the lowest mean value was recorded by the inoculant I<sub>4</sub> (31.17 mg kg<sup>-1</sup>).

#### 4.3.8.5. Boron Content

Regarding the B content of the plants, the treatments, substrates and inoculants had significant effect. The mean values of the treatments ranged from 30.02 mg kg<sup>-1</sup> to 63.12 mg kg<sup>-1</sup> were recorded by the treatments T<sub>10</sub> and T<sub>13</sub>(100 per cent N as compost from water hyacinth and Composting Inoculum) respectively. T<sub>13</sub> was on par with the treatments T<sub>2</sub> ( 59.87 mg kg<sup>-1</sup>), T<sub>5</sub> (57.46 mg kg<sup>-1</sup>) and T<sub>17</sub> (56.79 mg kg<sup>-1</sup>). The lowest mean value was recorded by the treatment T<sub>10</sub> (30.02 mg kg<sup>-1</sup>). Regarding the individual effect of substrates, it was found to be significant on B content. Substrate S<sub>3</sub> has recorded the highest mean value of 50.07 mg kg<sup>-1</sup> and the lowest mean value was recorded by the substrate S<sub>2</sub> (39.00 mg kg<sup>-1</sup>) Regarding the effect of inoculants, I<sub>3</sub> has recorded the highest mean value 57.41 mg kg<sup>-1</sup> and the lowest mean value was recorded by the inoculants I<sub>4</sub> (39.13 mg kg<sup>-1</sup>).

Table 21. Nutrient balance sheet- Nitrogen

| Treatments      | Dry matter production(g) | Uptake (g/pot) | Expected balance(g/pot) |
|-----------------|--------------------------|----------------|-------------------------|
| T <sub>1</sub>  | 64                       | 0.896          | 1.749                   |
| T <sub>2</sub>  | 42.1                     | 1.13           | 1.515                   |
| T <sub>3</sub>  | 51.6                     | 0.903          | 1.742                   |
| T <sub>4</sub>  | 42.1                     | 0.842          | 1.803                   |
| T <sub>5</sub>  | 77.25                    | 0.18           | 2.465                   |
| T <sub>6</sub>  | 48.25                    | 1.02           | 1.625                   |
| T <sub>7</sub>  | 43                       | 0.58           | 2.065                   |
| T <sub>8</sub>  | 38.2                     | 0.72           | 1.925                   |
| T <sub>9</sub>  | 50.6                     | 1.01           | 1.635                   |
| T <sub>10</sub> | 48.6                     | 0.88           | 1.765                   |
| T <sub>11</sub> | 50.4                     | 0.90           | 1.745                   |
| T <sub>12</sub> | 48.7                     | 0.92           | 1.725                   |
| T <sub>13</sub> | 79.5                     | 1.59           | 1.055                   |
| T <sub>14</sub> | 49.85                    | 0.99           | 1.655                   |
| T <sub>15</sub> | 53.1                     | 1.55           | 1.095                   |
| T <sub>16</sub> | 45.1                     | 0.90           | 1.745                   |
| T <sub>17</sub> | 50.7                     | 1.22           | 1.425                   |
| T <sub>18</sub> | 49.65                    | 1.29           | 1.355                   |
| T <sub>19</sub> | 24.24                    | 0.23           | 2.415                   |

Table 22. Nutrient balance sheet- Phosphorous

| Treatments`     | Dry matter production(g) | Uptake (g/pot) | Expected balance(g/pot) |
|-----------------|--------------------------|----------------|-------------------------|
| T <sub>1</sub>  | 64                       | 0.224          | 1.076                   |
| T <sub>2</sub>  | 42.1                     | 0.10           | 1.2                     |
| T <sub>3</sub>  | 51.6                     | 0.15           | 1.15                    |
| T <sub>4</sub>  | 42.1                     | 0.11           | 1.19                    |
| T <sub>5</sub>  | 77.25                    | 0.37           | 0.93                    |
| T <sub>6</sub>  | 48.25                    | 0.14           | 1.16                    |
| T <sub>7</sub>  | 43                       | 0.09           | 1.21                    |
| T <sub>8</sub>  | 38.2                     | 0.08           | 1.22                    |
| T <sub>9</sub>  | 50.6                     | 0.11           | 1.19                    |
| T <sub>10</sub> | 48.6                     | 0.11           | 1.19                    |
| T <sub>11</sub> | 50.4                     | 0.21           | 1.09                    |
| T <sub>12</sub> | 48.7                     | 0.61           | 1.09                    |
| T <sub>13</sub> | 79.5                     | 0.39           | 0.91                    |
| T <sub>14</sub> | 49.85                    | 0.14           | 1.16                    |
| T <sub>15</sub> | 53.1                     | 0.65           | 0.65                    |
| T <sub>16</sub> | 45.1                     | 0.09           | 1.21                    |
| T <sub>17</sub> | 50.7                     | 0.12           | 1.18                    |
| T <sub>18</sub> | 49.65                    | 0.15           | 1.15                    |
| T <sub>19</sub> | 24.75                    | 0.03           | 1.26                    |

Table 23. Balance sheet of major nutrient –Potassium

| Treatment       | Dry matter production(g) | Uptake (g/pot) | Expected balance(g/pot) |
|-----------------|--------------------------|----------------|-------------------------|
| T <sub>1</sub>  | 64                       | 3.6            | 3.38                    |
| T <sub>2</sub>  | 42.1                     | 2.7            | 2.48                    |
| T <sub>3</sub>  | 51.6                     | 1.99           | 1.77                    |
| T <sub>4</sub>  | 42.1                     | 1.85           | 1.63                    |
| T <sub>5</sub>  | 77.25                    | 4.09           | 3.87                    |
| T <sub>6</sub>  | 48.25                    | 2.31           | 2.09                    |
| T <sub>7</sub>  | 43                       | 1.57           | 1.35                    |
| T <sub>8</sub>  | 38.2                     | 1.55           | 1.33                    |
| T <sub>9</sub>  | 50.6                     | 1.77           | 1.55                    |
| T <sub>10</sub> | 48.6                     | 1.84           | 1.62                    |
| T <sub>11</sub> | 50.4                     | 1.93           | 1.71                    |
| T <sub>12</sub> | 48.7                     | 2.42           | 2.2                     |
| T <sub>13</sub> | 79.5                     | 3.31           | 3.09                    |
| T <sub>14</sub> | 49.85                    | 2.05           | 1.83                    |
| T <sub>15</sub> | 53.1                     | 2.63           | 2.41                    |
| T <sub>16</sub> | 45.1                     | 1.79           | 1.57                    |
| T <sub>17</sub> | 50.7                     | 2.33           | 2.11                    |
| T <sub>18</sub> | 49.65                    | 2.28           | 2.06                    |
| T <sub>19</sub> | 24.75                    | 1.01           | 0.79                    |

## *Discussion*

## 5. DISCUSSION

A study entitled “Characterization, conversion and evaluation of selected lignocellulosic biomass” was undertaken in the Department of Soil Science and Agricultural Chemistry, College of Agriculture Vellayani during August 2013-February 2014. The study involved the characterization of biomass viz. water cabbage, coir pith, water hyacinth and farm waste containing banana pseudostem and dry leaves and evaluating the efficiency of the inoculants viz. *T. reesei*, *P. sajor-caju*, Composting Inoculum and commercial enzyme cocktail on these biomass. The compost formed as a result of these inoculants on these substrate was then evaluated through a pot culture experiment using amaranthus as test the crop.

The results pertaining to the study are discussed in this chapter.

### 5.1. PROXIMATE ANALYSIS OF THE DIFFERENT SUBSTRATES

The different substrates viz. water cabbage, coir pith, water hyacinth and farm waste containing banana pseudostem and dry leaves involved in this study were analyzed for its proximate constituents viz. cellulose, hemicelluloses, lignin, proteins, C:N ratio, NPK content and heavy metals (Pb,Cd,Ni, and Sn) .

In the case of cellulose content, significant variations were noticed between the substrates (Figure 3). The highest value was noticed in the coir pith and was on par with water hyacinth. Similar results were reported by Thomas *et al.* (2013) who observed higher cellulose content in coir pith. Raji *et al.* (2008) have also reported the highest content of cellulose in water hyacinth up to 18.12 per cent which was then utilized for ethanol production.

While with respect to the hemicellulose content, a wide variation was noticed as read from (Table 6). Water hyacinth recorded the highest value for hemicellulose (28.4 per cent). Nigam (2002) has also observed the highest hemicellulose content in water hyacinth to an extent of 55 percent of dry weight and also reported that the hemicellulose content could be converted to fuel bio

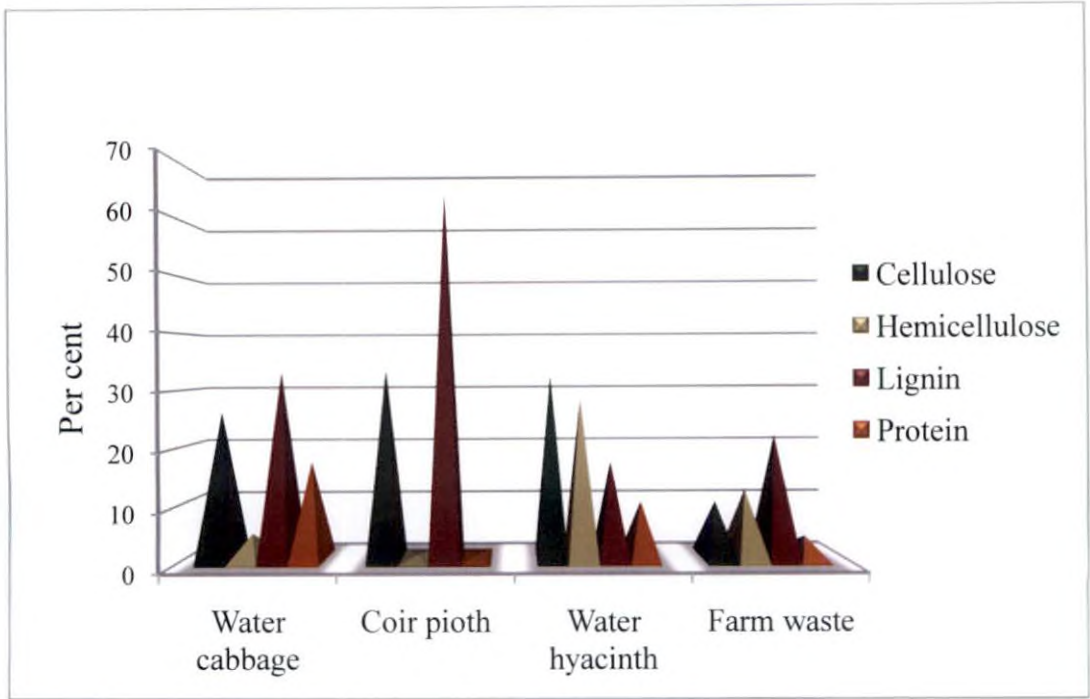


Figure 3. Proximate analysis for cellulose, hemicelluloses, lignin and protein content in the substrates.

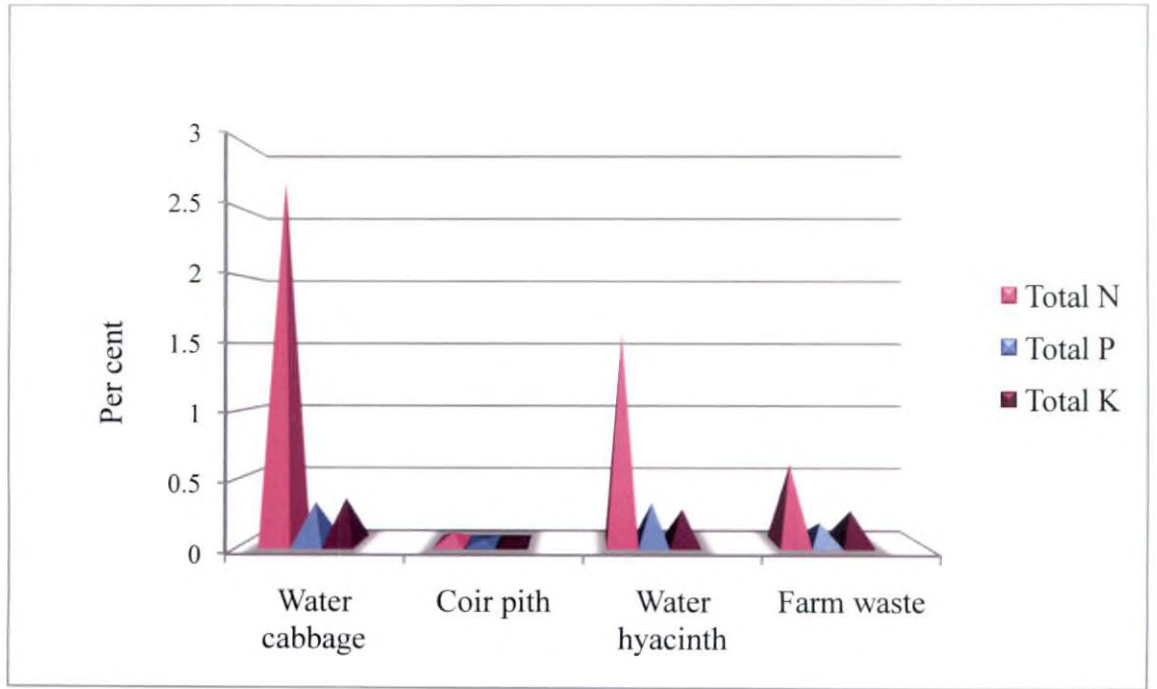


Figure 4. Proximate analysis for N, P and K content in the substrates.



ethanol production via hydrolysis. The lowest value for hemicelluloses was noticed in coir pith in contrary to cellulose content.

Coir pith is thus an agro industrial by product which is considered to be a waste in the coir factory in India. Natural coir pith is rich in K but low in N and P and has high lignin, cellulose and low hemicellulose (Shashirekha and Rajaratnam, 2007).

With respect to lignin content also, the highest value was noticed in coir pith (64.76 per cent) while water hyacinth recorded the lowest value. High lignin content upto 88 per cent was also reported by Shashirekha and Rajaratnam (2007) and Nattudurai *et al.* (2014).

A significant variation was noticed among the substrates with respect to protein content. Water cabbage reported the highest value for protein content (17.10 per cent) followed by water hyacinth and pseudostem and dried leaves. Similar results were also reported by Saupi *et al.* (2009) in their study on the protein content from aquatic weeds. This might be also due to the extractability of proteins in aquatic weeds.

With respect to the nutrient content of the various substrates *viz.* water cabbage, coir pith, water hyacinth and farm waste, a significant difference was noticed. Water cabbage recorded the highest value of N, P and K while coir pith was found to be low as inferred from the Table 7 and Figure 4. The highest nutrient content of aquatic weed *L. flava* might be due to the accumulation of the nutrients from wet land ecosystem. This might be also due to the availability of mineral elements in water and hydro soil. Species of plant and time of year influence the nutrient content of aquatic weeds like water cabbage (Sutton and Portier, 1998). A high mineral content of 3.03 per cent N, 0.42 per cent P and 2.81 per cent K were also reported by Linn (1975).

Regarding the C:N ratio, the widest value was reported by the substrate coir pith. The wide C:N ratio coupled with low N, P and K content, presence of

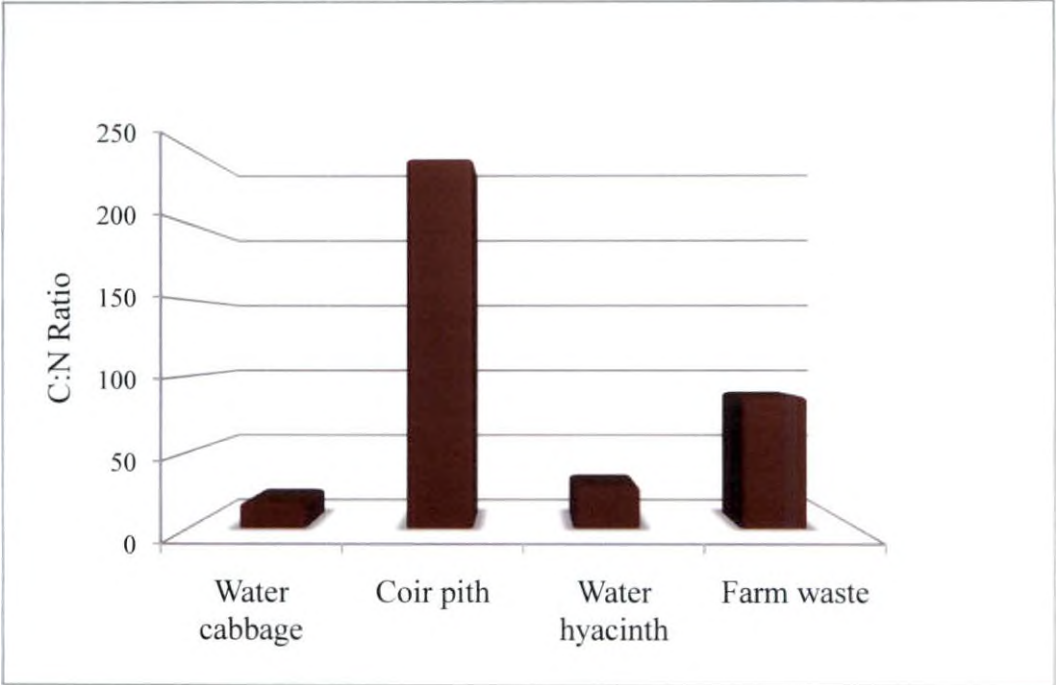


Figure 5. C:N ratio of four substrates.

soluble tannin related phenolic compounds (8-12 percent), low biodegradability can be attributed to the low N, P and K content of coir pith (Parthasarathy, 2005). C:N ratio is one of the most important factors affecting composting process and compost quality (Golueke, 1991). The narrowest C:N ratio was noticed under water cabbage favoured early composting. A wide C: N ratio of coir pith was also reported by Srinivasan *et al.* (2005). The widest C: N ratio was noticed with coir pith had significantly differed from other substrates (Figure.5). The narrowest mean value was noticed for water cabbage favoured early composting. A wide C: N ratio of 162:1 by coir pith was reported by Muthurayar and Dhanarajan (2013). Prabhu and Thomas (2002) have also reported that coir pith has high C: N ratio, lignin and polyphenol content and it was highly resistant to easy decomposition and could be stabilized by suitable Composting techniques.

A heavy metal is a metal or metalloid of environmental concern. Soils may become contaminated by the accumulation of heavy metals and metalloids through emissions from the application of fertilizers, sewage sludges, pesticides, waste water irrigation. Most commonly found heavy metals are Pb, Cr, As, Zn, Cd, Cu, Hg and Ni (Wuana and Oikiemen, 2011). A maximum bio concentration factor (BCF) for Pb was 1531 suggesting that the water hyacinth is a good accumulator of Zn and could be used to treat waste water contaminated with metals like Zn, Pb and Cd (Chunkao *et al.*, 2012). From the Table 8. Figure.6, it was inferred that water hyacinth has recorded the highest lead content while water cabbage has recorded no traces of lead.

The highest content of Pb was noticed in water hyacinth might be due to the phytoremediation potential of some heavy metals by water hyacinth as reported by Okunoiva and Ogunkanni (2010). They have also suggested water hyacinth could effectively phytoremediate contaminated H<sub>2</sub>O containing metals such as K, Na, Zn, Pb, Fe and Ca.

With regard to Cadmium content as inferred from Figure 7 variations were noticed. Though the content of Cd was found to be in traces, substrate S<sub>4</sub> (farm



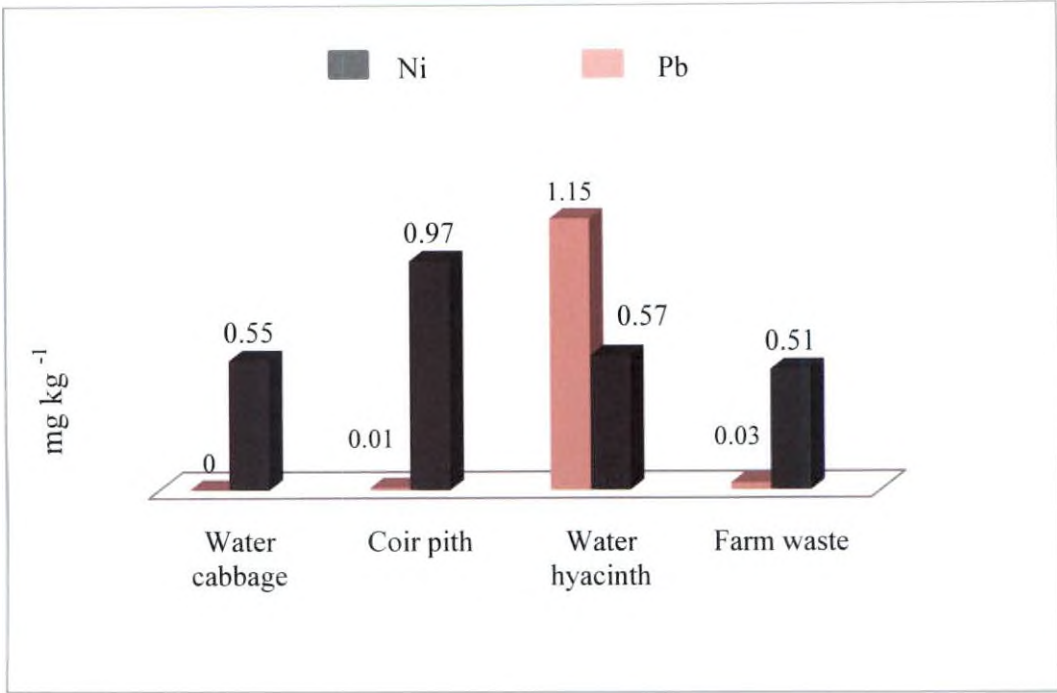


Figure 6. Proximate analysis for Pb and Ni content in the substrates.

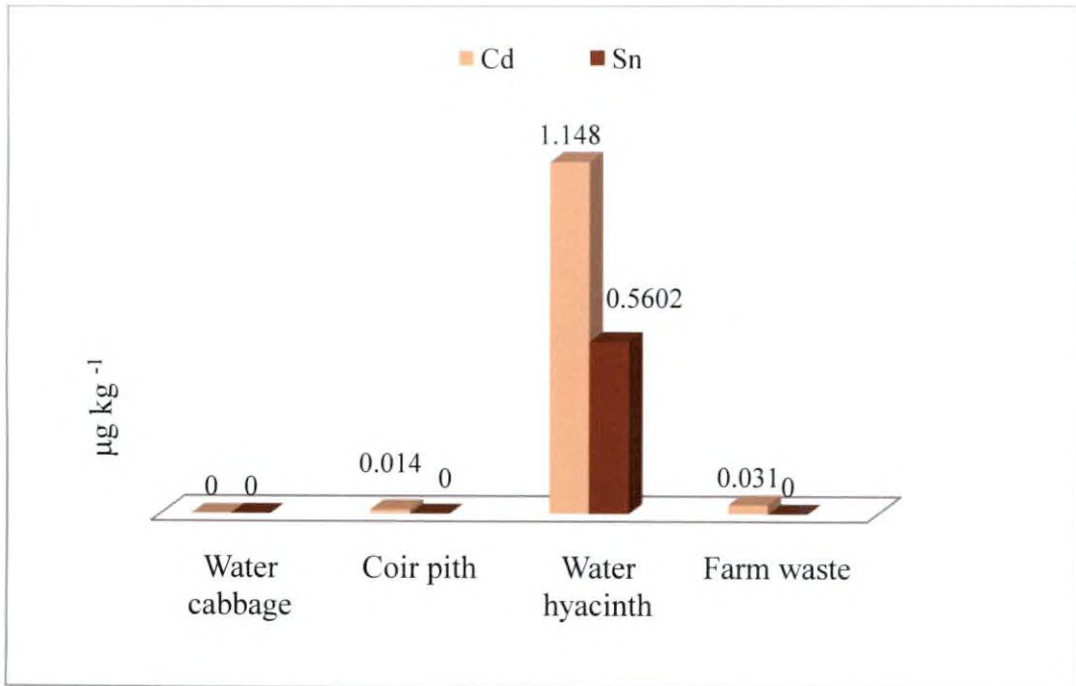


Figure 7. Proximate analysis for Cd and Sn content in the substrates.

waste) recorded the highest value. This might be due to the absorption of Cd from the soils in which the crop was grown. All crop residues contain Cd and the harvested portion will serve to remove Cd from soil and the amounts removed from the soil are quite small compared to the amount present in the soil originally. These findings corroborated with the findings of Page *et al.* (1987). The lowest content of Cd as observed in water cabbage might be due to the poor absorption of Cd from the soil, water system and also due to the non preferred selection of Cd ions.

In the case of Nickel a wide variation in the content was observed among the substrates. From Table 8 it was inferred that the highest content of Ni was recorded by coir pith which was significantly different from all other substrates. This might be attributed to the higher absorption of Nickel in the soil- water ecosystem where the coconut husks were put for retting for extraction. These results corroborated with the findings of Sathyakeerthy (1999).

## 5.2. CHARACTERIZATION OF COMPOSTS

### 5.2.1. Physico-chemical Properties of the Compost

The second phase of the study was the preparation of compost of the substrates *viz.* water cabbage, coir pith, water hyacinth, and farm wastes by the action of inoculants *viz.* *T. reesei*, *P. sajor-caju*, Composting Inoculum and commercial enzyme cocktail. Composting has become a preferred method for variety of substrates or organic matter for application as soil conditioner and amendment (Butler *et al.*, 2001). One of the most important advantages of using composted manures for agricultural purpose is its stability and maturity. Composted manures have gained a wide acceptance as organic amendment for sustainable agriculture as they have shown to increase soil organic matter levels, improve soil physical properties and modify soil microbial communities thereby enhancing microbial biomass, activity and diversity (Eneji *et al.*, 2001).

It is of great importance to focus on the information regarding the nutritional and toxicity status of manure amendments, as a wide range of physico-



chemical and bio chemical changes do occur during composting. Since the total concentration of elements in manure may not provide the best indication of their bio availability, characterizing the compost is essential. The resultant composts were analyzed for physico-chemical and biological properties such as moisture content, pH, EC, ash content, organic matter, N, P and K, dehydrogenase activity, cellulase activity, maturity period, C/N ratio, heavy metal content etc.

From the data presented in Table 9 and Figure 8 significant variation was observed with respect to moisture content of the various composts. Composts produced with the combination of water cabbage and Composting Inoculum recorded the highest moisture content and was similar to compost prepared with the combination of coir pith + commercial enzyme cocktail and water hyacinth + *P. sajor-caju*. A compost of 60 per cent of moisture content was reported to be of a good quality as reported by Singh *et al.* (2004). Similar results were reported by Edwards and Bater (1992) and reported that the optimum moisture content is around 65 per cent. According to Liang *et al.* (2003), the moisture content in the range of 60 to 70 per cent was proved having maximal microbial activity while 50 per cent moisture content was minimal requirement for rapid rise in microbial activity. Compost produced out of the combination of water cabbage and Composting Inoculum showed higher moisture content than the other composts which might be due to their higher absorption capacity (Bhattacharyya, 2007.). The interaction effects were also found to be significant as Composting Inoculum was effective on water cabbage and farm waste, for coir pith commercial enzyme cocktail and *P. sajor-caju* on water hyacinth. A comparatively higher value recorded in coir pith might be due to the higher absorption capacity and moisture retaining capacity. Similar findings were reported by Jeyaseeli and Raj (2010). In the case of individual effect of inoculants, the Composting Inoculum has a prominent effect than the others due to the moisture retaining capacity of the Composting Inoculum which act as a cocktail of various cellulolytic fungi.

The electrical conductivity of the resultant compost was significantly influenced by all the four substrates, inoculants and their interactions. The

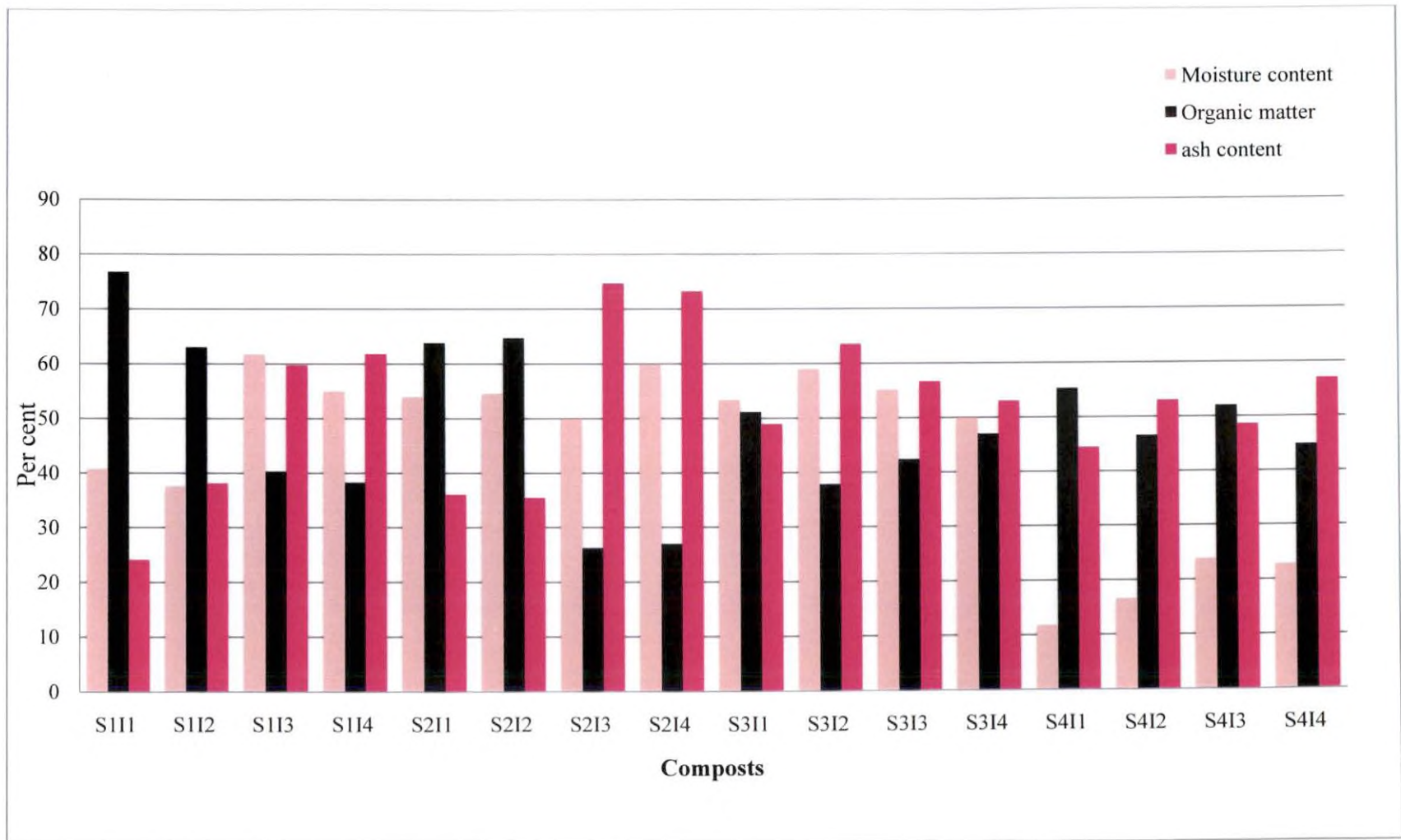


Figure 8. Physico-chemical properties (moisture content, organic matter and ash content) of the composts.



compost prepared out of water hyacinth and Composting Inoculum recorded the highest EC value followed by compost from water hyacinth and *T. reesei*. It was also observed from the study that water hyacinth had a significant role in the recording highest value of EC. The EC value reflected that the compost does not exhibit any phyto inhibitory effect on the growth of the plant as it was less than 4 dSm<sup>-1</sup>. Similar results also reported by the Dhal *et al.* (2012) while individual effect of substrates was also significant and water hyacinth recorded the highest value for EC. The Composting Inoculum has reported the highest value for EC while considering the individual effect of inoculants. Hence the combination water hyacinth and Composting Inoculum has reported a higher significant value for EC.

Figure 8 displays the result of ash content of the compost. The highest ash content was recorded in the compost prepared out of coir pith and Composting Inoculum which was similar to the compost produced with coir pith and commercial enzyme cocktail. The highest ash content of 80 per cent was also reported by Muthurayar and Dhanarajan (2013). *Psuedostem* with dried leaves was reported have a high ash content as might be due to the highest biomass per dry matter where as in the case of individual inoculants, commercial enzyme cocktail was found to produce highest ash content.

Organic matter content of the composts was influenced by substrates, inoculants and their interactions as evident from Table 10; Figure 8. The compost produced out of water cabbage and *T. reesei* recorded the highest organic matter content and was observed to be the best in quality as reported by Brinton (2000). Similarly a significant effect of water cabbage was noticed in the study with the highest organic matter content. Thus the substrate water cabbage in combination with *T. reesei* has contributed to high quality compost with high organic matter content. Different inoculants (I<sub>1</sub>, I<sub>2</sub>, I<sub>3</sub>, and I<sub>4</sub>) were found to have a significant effect on organic matter content. Thus in combination with substrate water cabbage was reported to produce good quality compost. The lowest value was



noticed in the commercial enzyme cocktail which indicate lesser organic carbon content.

### 5.2.2. Nutrient Content of Compost

The nutrient content of the compost was significantly influenced by the substrates, inoculants and their interactions as evident from Table 10; Figure 9. Compost prepared from water cabbage and Composting Inoculum reported a high total N content due to the positive interaction between the substrate and inoculum. This might be attributed to the highest mineralization in to N accelerated by the Composting Inoculum (Amirkhan and Ishaq, 2011). Generally the total N concentration increases during compost due to the concentration effect (Paredes *et al.*, 1996) whereas the highest P content was reported in water hyacinth and Composting Inoculum. Conversion of inorganic P of the compost might be due to the process of microbial decomposition which produced organic acids which dissolved the inorganic P in the compost produced (Bernal *et al.*, 2009). In the case of K also, a significant variation was noticed with respect to substrate, inoculants and their interactions. The role of Composting Inoculum in mineralizing the organic form of nutrients N, P and K was evident as the composting by Composting Inoculum on different substrates had increased the total N, P and K content. Since the nutrient content of the compost is an important factor influencing the plant growth, the Composting Inoculum can be used for this purpose. This might be due to the action of various cellulolytic organisms resulting in mineralization of nutrients thereby increase N, P and K content which strongly agreed with Verma *et al.* (2009).

### 5.2.3. Maturity Indices of Compost

The maturity of the composts can be determined by assessing the various physical, chemical and bio chemical parameters (Chen, 2003). Maturity is one of the most important aspects of compost quality, particularly for composts used in high value agricultural crops. Trends in dehydrogenase activity, cellulase activity, maturity period and C: N ratios were presented in Table 11; Figure 10. All organic matter is made up of substantial amounts of carbon (C) combined with lesser

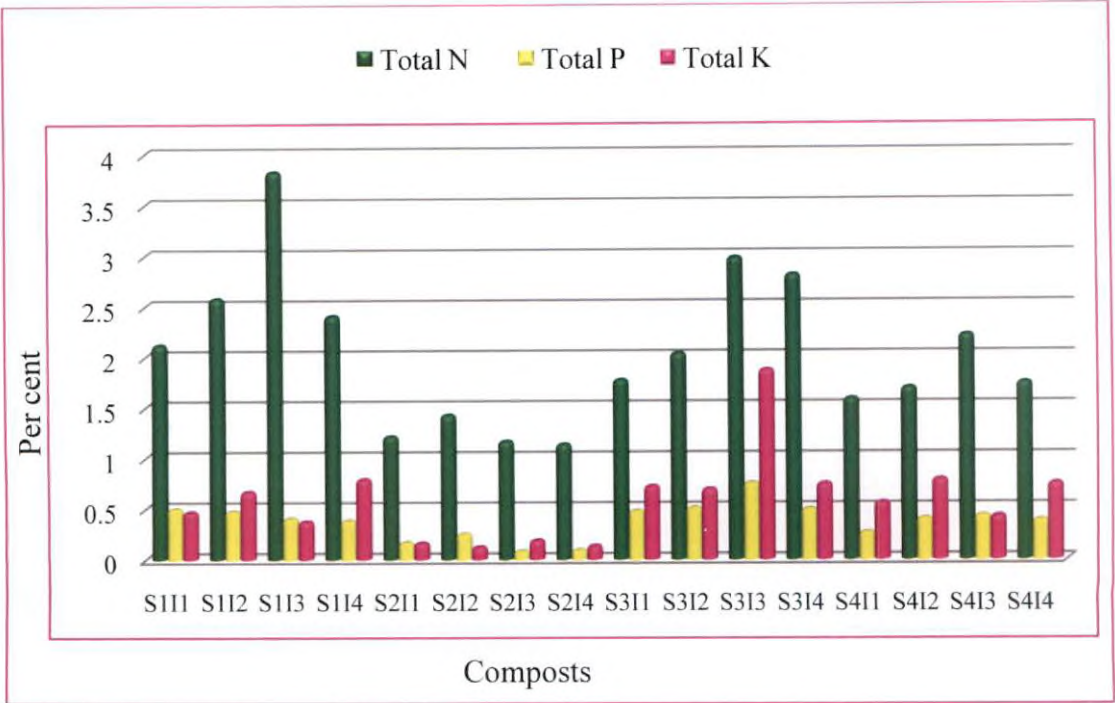


Figure 9. Major nutrient (N, P and K) content of the composts.

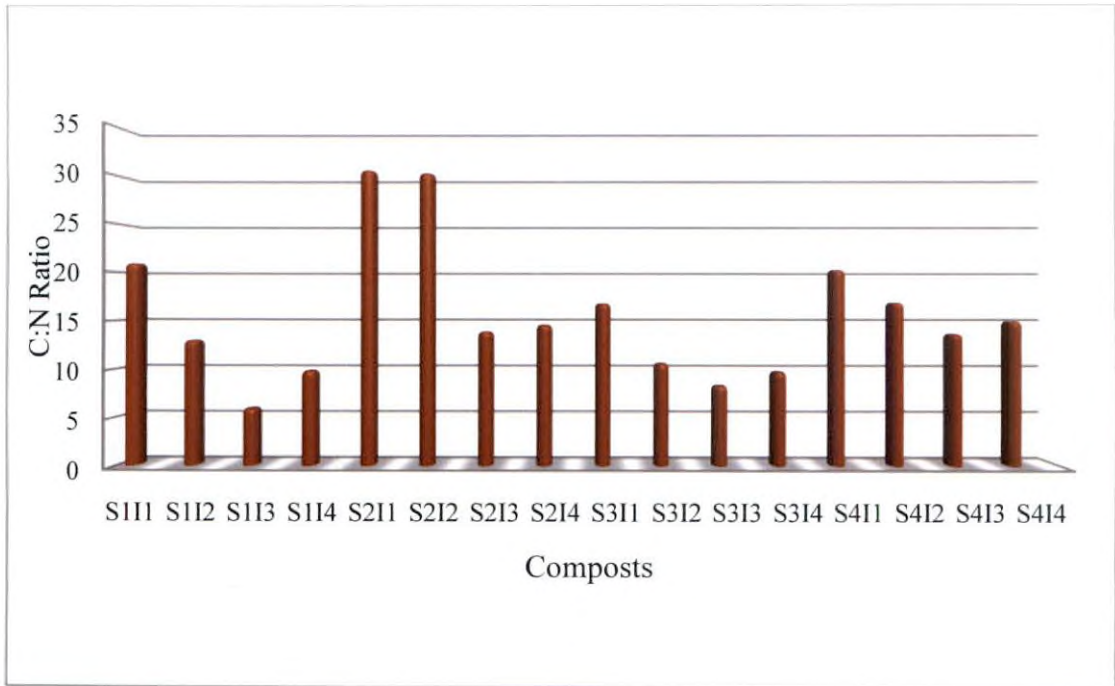


Figure 10. C:N ratio of the composts.



amount of nitrogen and the balance of these two elements is called C: N ratio. It is an important factor deciding the availability of nutrients. From the figure 10. depicting the C: N ratio , the narrowest ratio was recorded in the compost produced out of the combination of water cabbage + Composting Inoculum and water hyacinth + Composting Inoculum. The decrease in C:N ratio as compared to other composts using different substrates and inoculants might be due to relative increase in total nitrogen or loss of dry matter (organic carbon) as CO<sub>2</sub> as well as water loss by evaporation during mineralization process. The decrease in C: N ratio over time might have been also attributed to the rapid decrease in C-substrates by the utilization of fungus in composting Inoculum. Similar results were reported by Nedagwa *et al.* (2000). In the case of individual effect, the performance of Composting Inoculum was found to be the best.

The decrease in C: N ratio could be due to the respiratory activity of micro organisms and an increase in total N by mineralization of organic matter. This might also due to faster decomposition of organic matter which leads to the reduction of C as CO<sub>2</sub>. Similar results were reported by Jadia and Fulekar (2008) in vegetable market waste and Shwetha *et al.* (2010) in sugarcane byproducts.

The application of Composting Inoculum which is a consortium of various cellulolytic organisms can effectively degrade lignocellulosic components to simpler compound within a short periods thus decreasing C: N ratio. These results also indicated the cellulolytic capacity of Composting Inoculum and its potential in degradation of organic waste as well as recycling the waste for the use of compost other than the fungi itself, degradable organic compounds in raw materials might also stimulate the degradation process (Sherief *et al.*, 2010). Thus for these reasons, the Composting Inoculum was found to be the best source for composting or degrading organic materials.

Microbial parameters such as dehydrogenase (Figure 11) and cellulase (Figure 12.) activity can be considered as indicators of compost maturity. Water cabbage when composted using *T. reesei* was found to report higher values for

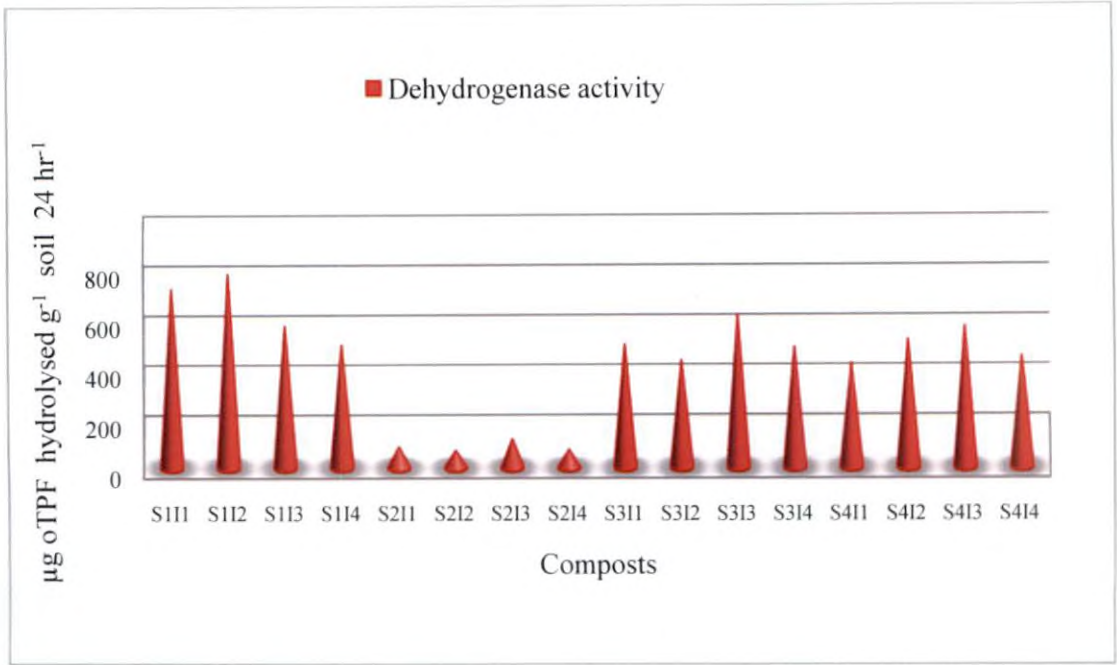


Figure 11. Dehydrogenase enzyme activity of composts.

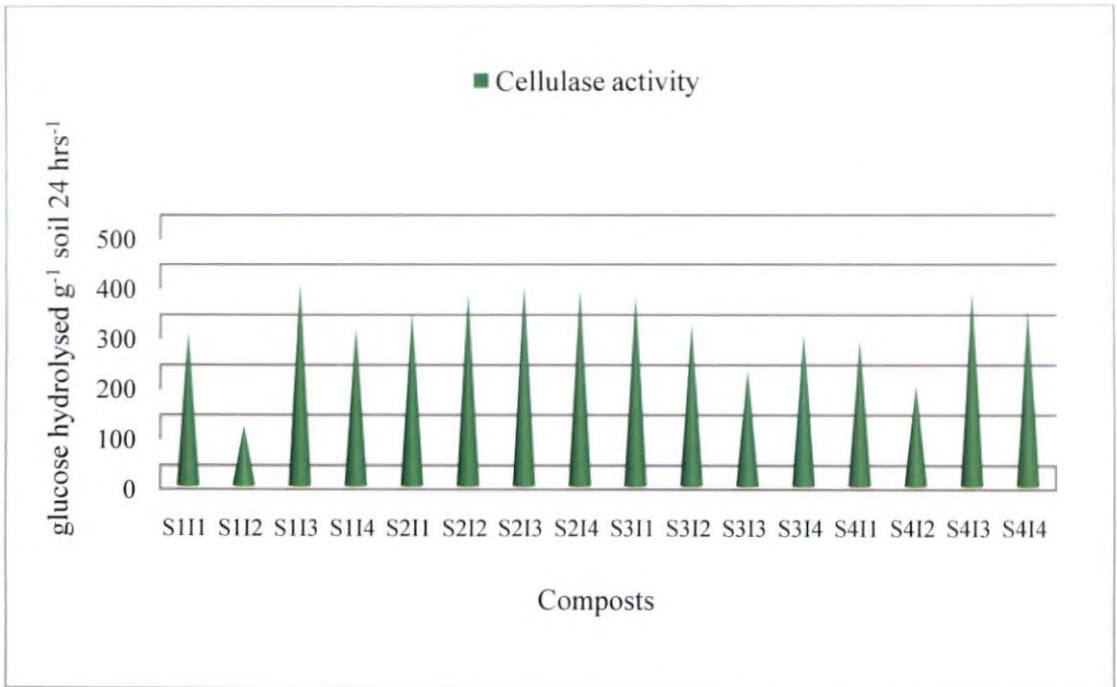


Figure 12. Cellulase enzyme activity of the composts.



dehydrogenase while coir pith as substrate and *P. sajor- caju* as inoculum is reported to have the lowest activity. These results corroborated with the finding of Tiquia (2005). Water cabbage as substrate had profound effect on the activity of dehydrogenase enzyme. The effect of Composting Inoculum was also evident from the study conducted.

Cellulase is an another important parameter in deciding the maturity of compost. From the result it was noticed that the water cabbage with Composting Inoculum resulted in good quality compost with high cellulose activity. The individual effect of Composting Inoculum was also evident from study (Figure 12). Thus the increased cellulose activity depicts an increased rate of cellulose degradation and thus the maturity of compost. This indicates the lignocellulolytic activity of the fungus present in the Composting Inoculum and it could have degraded cellulose and speeded up composting process. Similar findings were reported by Singh and Sharma (2003).

The lowest maturity period in water cabbage and Composting Inoculum compost (44 days) indicated that the time taken for composting to reach maturity was less due to the existence of different fungi which helps in the degradation of each component. A two fold increase in the degradation potential was noticed in this compost when compared to the compost produced out of coir pith with commercial enzyme cocktail which recorded the more maturity period. Even though the combination of coir pith and commercial enzyme cocktail reported to have high maturity periods, the individual effect of Composting Inoculum on other substrate was found to be significant (Table 11).

#### **5.2.4. Heavy Metal Content**

The production of compost from agricultural and industrial waste and municipal by-products is an important means of recovering organic matter and an essential method of disposal. However the presence of heavy metals in compost is causing adverse effects on animal and human health, transmitted through the food chain from the soil, ground water and plants

(Senesei *et al.*, 1999). The production and application of compost potentially contaminate the environment with heavy metals. The heavy metal content of compost must be exactly determined for the quality manure production.

From this study it was observed that the compost produced with the combination of banana pseudostem +dry leaves and *P. sajor-caju* reported to have detectable levels of Pb and Cd. Whereas in the case Ni content, coir pith and Composting Inoculum has reported highest Ni content. In the case of tin the highest value was noticed with water hyacinth and commercial enzyme cocktail. Although all the compost showed different levels of Pb, Cd, Ni, and Sn all the composts were reported to have heavy metals below the critical limits as per the specifications of FAO (Table 12).

Irrespective of substrate and inoculum used, these composts can be used as a quality organic manure for crop production. It was also observed that the process of composting promotes the complexation of heavy metals whose mobility and availability tends to decrease with reduced toxicity. Thus availability factors also varied from one metal to another and varies with the substrates used for composting (Wong and Selvam, 2006).

### 5.3. POT CULTURE EXPERIMENT

A pot culture study was initiated during December 2013 to assess the performance of various composts produced using different substrates and inoculants *viz.* *T. reesei*, *P. sajor-caju*, Composting Inoculum and commercial enzyme cocktail. The test crop amaranthus variety Arun was grown on pots. The post harvest analysis of soil was done for various physical, chemical and biological parameters. A detailed discussion on yield, yield attributes, post harvest soil and plant analysis are given below.

#### 5.3.1. Yield and Yield Attributes

The composts prepared from different substrates in combination with four inoculants were applied to the test crop amaranthus variety Arun. The yield and yield attributes were significantly varied with treatments as shown in the Table 13



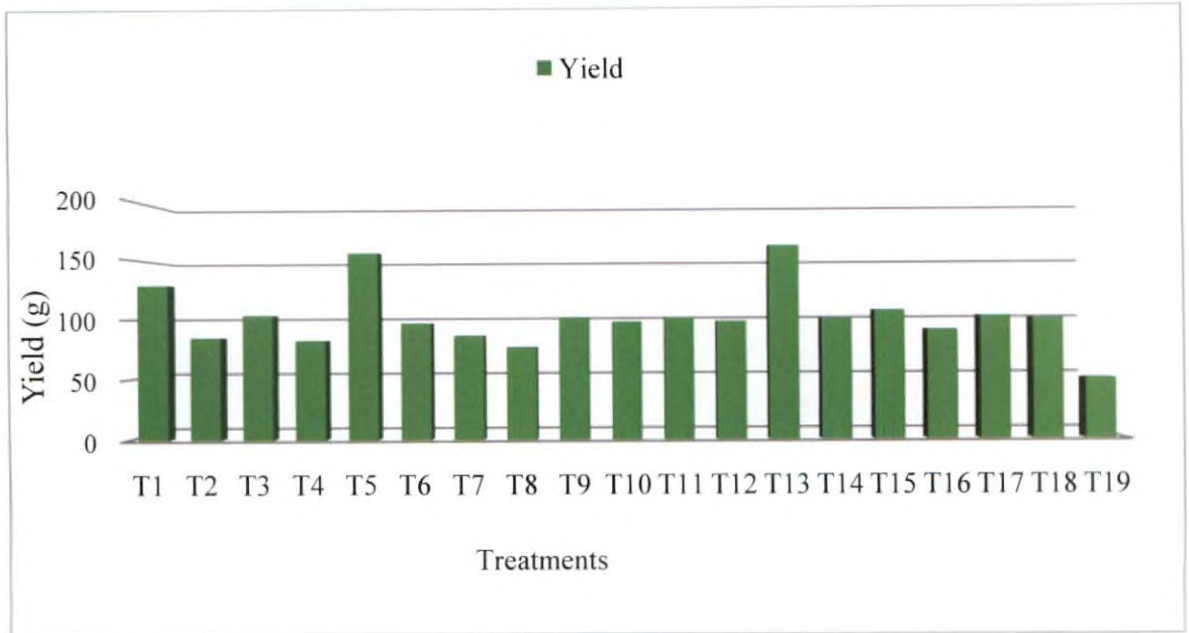


Figure13. Effect of various compost preparations on yield of amaranthus.

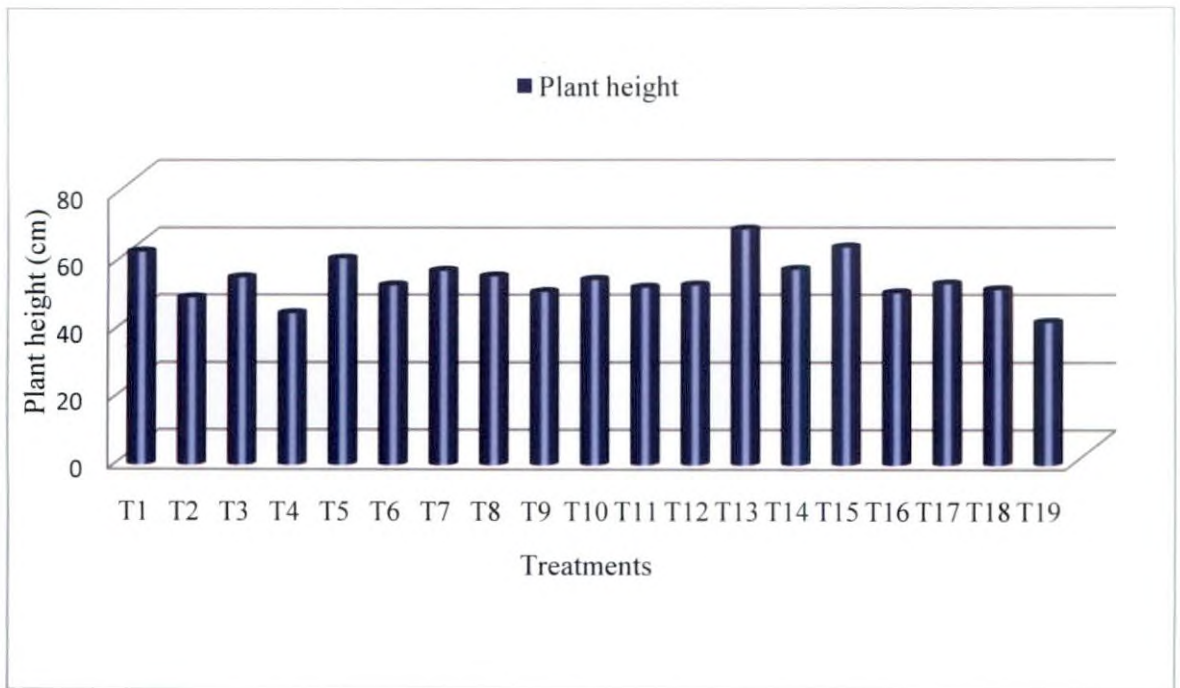


Figure 14. Effect various compost preparations on height of amaranthus.

and Figures 13, 14 and,15. In case of plant height, the tallest plant was produced by the application of compost prepared out of the combination of water hyacinth and Composting Inoculum. Tindall (1975) reported that amaranthus required soil with high organic content, and adequate mineral nutrients. This may be the reason for the observed increase in plant height. Considering the effect of substrate alone on the plant height, water hyacinth was noticed as the best. This might be due to the high nutrient content in the substrate as well as in compost. This strongly agreed with the findings of Gajalakshmi and Abbasi (2002) who observed that the pots containing soil amended with water hyacinth compost with *Crossandra* plants achieved significantly better height, larger number of leaves, more favourable shoot: root ratio, greater biomass per unit time and larger length of inflorescence. Among inoculants, Composting Inoculum was found to be the best. Composting Inoculum contains a consortium of microbes which produce ligno- cellulolytic enzymes as well as various growth promoting factors. This might be the reason for the enhanced plant height observed with the Composting Inoculum than the other inoculants.

While considering the number of leaves per branch, significant variation was observed between the treatments. The treatment received 100 per cent N as compost prepared from the combination of water cabbage and Composting Inoculum has showed the highest number of leaves per branch. Application of compost prepared from water cabbage and Composting Inoculum also had the similar effect. Even though the substrates alone had no significant influence on the number of leaves per branch water cabbage recorded the highest value. In the case of effect of inoculants, *T. reesii* was noticed as the best inoculum having positive influence on the number of leaves per branch. Oworu *et al.* (2010) also reported a similar result with the application of 12 t ha<sup>-1</sup>compost resulted the highest leaf yield in amaranthus and the lowest yield was observed in plots where no compost was applied. It was evident that the applied composts materials with sufficient N boosted leaf production by enhancing the production of chlorophyll, protoplasm, protein, nucleic acid etc.



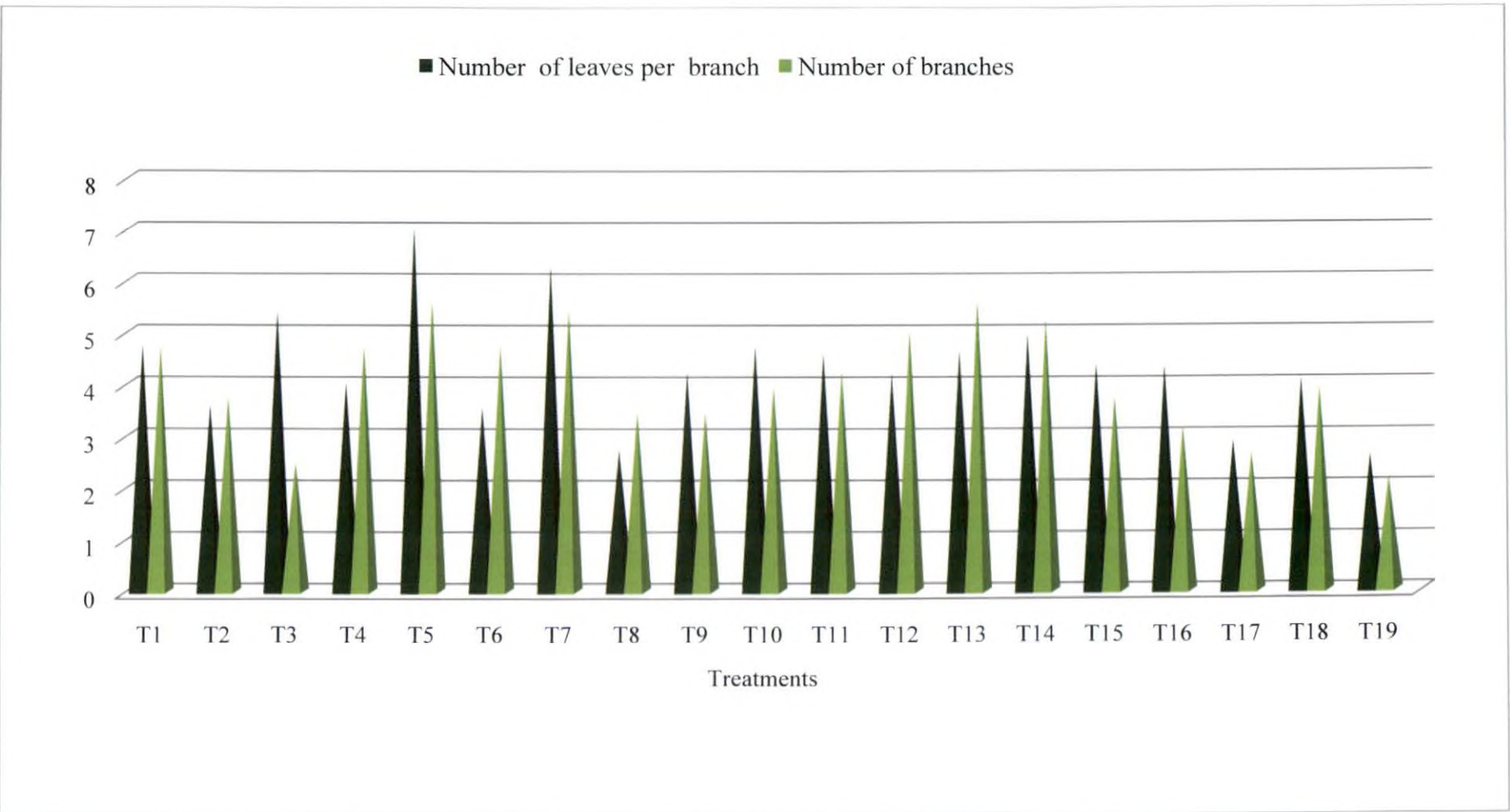


Figure 15. Effect of various compost preparation on the yield attributes (Number of leaves per branch and Number of leaves) of amaranthus

With regard to the number of branches, no significant influence was observed either by treatments or by inoculants. Application of composts prepared from the combination Composting Inoculum with water cabbage and water hyacinth recorded the highest number of branches. Among inoculants, commercial enzyme cocktail reported the highest value. Substrate was also found to had significant effect on number of branches and the water hyacinth was noticed as the best substrate with highest number of branches. Eventhough treatments, substrates and inoculants had no significant effect on the girth of stem, more girth was observed under 100 per cent N as compost prepared out of the combination of water hyacinth and Composting Inoculum. Substrates as well as inoculants were not significant on the girth of stem, however substrate water hyacinth and Composting Inoculum recorded as the highest girth individually.

With respect to the yield, treatments imparted significant effect on the yield of the crop. The treatment receiving 100 per cent N as compost prepared from water hyacinth and Composting Inoculum combination recorded the highest yield. The compost prepared from water cabbage and Composting Inoculum also recorded very similar yield. This might be due to the similar nutrient status and other biochemical properties of the two composts. Considering the individual effects, both substrates and inoculants had significant effect on the yield of the crop. Water hyacinth has recorded the highest yield among the substrates and Composting Inoculum was reported as the best among inoculants. These observations are strongly agreed with Ainika *et al.* (2012) reported that the growth and yield parameters of amaranthus accessed were significantly increased in response to the application of organic manure at the highest rate of 10 t ha<sup>-1</sup> and might be attributed to the fact that farmyard manure and other organic manures contain all essential plant nutrients though in lower concentrations compared with inorganic fertilizer.

### **5.3.2. Physico-Chemical Characters of the Soil**

Though the physical parameters like bulk density, particle density and water holding capacity are not much influenced by the application of composts in

pot culture experiment significance was noticed on certain physical properties especially water holding capacity (Table 14). In the case of bulk density the lowest value was recorded with the application of compost (coir pith + *P. sajor-caju*) and identified as the best treatment. This might be attributed to the effectiveness of coir pith compost in decreasing bulk density and increasing macro porosity. This was in conformity with the findings of Rivenshield and Bassak, 2007.

According to Richard (2006) the bulk density and particle density is one of the factors determining the successful functioning of growth indices. In the case of individual effects of substrate and inoculum, coir pith and Composting Inoculum have influenced the bulk density significantly.

In the case of particle density, the application of compost (coir pith+ commercial enzyme cocktail) recorded the lowest value. The substrate coir pith had some significant effect on reducing the bulk density and particle density on composting. A similar result was reported by Sreenarayanan and Chatopadhyay (1986). Generally during Composting of coir pith, bulk density and particle density decreased which might be due to the faster decomposition and breaking activity of heavy particles of coir pith into smaller particles.

The water holding capacity of soil was also influenced to a greater extent by the application of various treatments. Application of compost (coir pith and *P. sajor-caju*) was found to impart favourable effect on soil moisture content. It has a high water holding capacity of eight times its weight. It is a fluffy, light, spongy material with increased water-holding capacity and extremely compressive as reported by Ghosh *et al.* (2007). In all the three physical parameters *viz.* bulk density, particle density and water holding capacity compost as coir pith series, the best substrate as with the improvement noticed in the pot experiment. This can be thus utilized as organic amendment as reported by Kumar and Ganesh (2013).

pH is another important factor that decides the availability of nutrients. Compost (Farm waste and *T. reesei*) recorded the pH of 6.98 and similar in its

effect with compost from Farm waste and commercial enzyme cocktail, water hyacinth and *P. sajor-caju*, coir pith and *P. sajor-caju*. The addition of compost to soil may modify the pH of the soil and has the capacity to buffer or stabilize soil pH. The increase in pH could be due to the higher pH value of the amendment or the compost and this increase is not considered dangerous to soil quality because the values remained close to neutrality. Similar findings were reported by Romaniuk *et al.* (2011). Regarding the individual effect of substrates, farm waste recorded the highest value while in the case of inoculants, *P. sajor-caju* was the highest with respect to pH.

Soil electrical conductivity was significantly affected by different composts. The highest value of for EC was reported with compost (farm waste and commercial enzyme cocktail). This result can be interpreted that the application of composts did not drastically increase the EC as the values noted were below 4 dSm<sup>-1</sup>. The individual effect of substrates were found to be non significant on EC. Considering the individual effect of inoculants, *P. sajor-caju* was significantly superior to others.

### 5.3.3. Major Nutrients

The major nutrients presented in Figure 16 revealed that different composts effected on soil available N, P and K. Compost S<sub>1</sub>I<sub>3</sub> (water cabbage and Composting Inoculum) was found to be efficient in mineralizing the N from organic sources whose effects are similar to composts from water cabbage + commercial enzyme cocktail, water hyacinth + Composting Inoculum and farm waste + Composting Inoculum. From this study it was evident that Composting Inoculum has a profound effect in influencing soil available N and recorded the highest value. Similarly the substrate water cabbage also significantly influenced the available N status. This might be due to the conversion of organic nitrogen by the process of mineralization by nitrifying bacteria present in compost and soil as reported by Monedero *et al.* (2001).

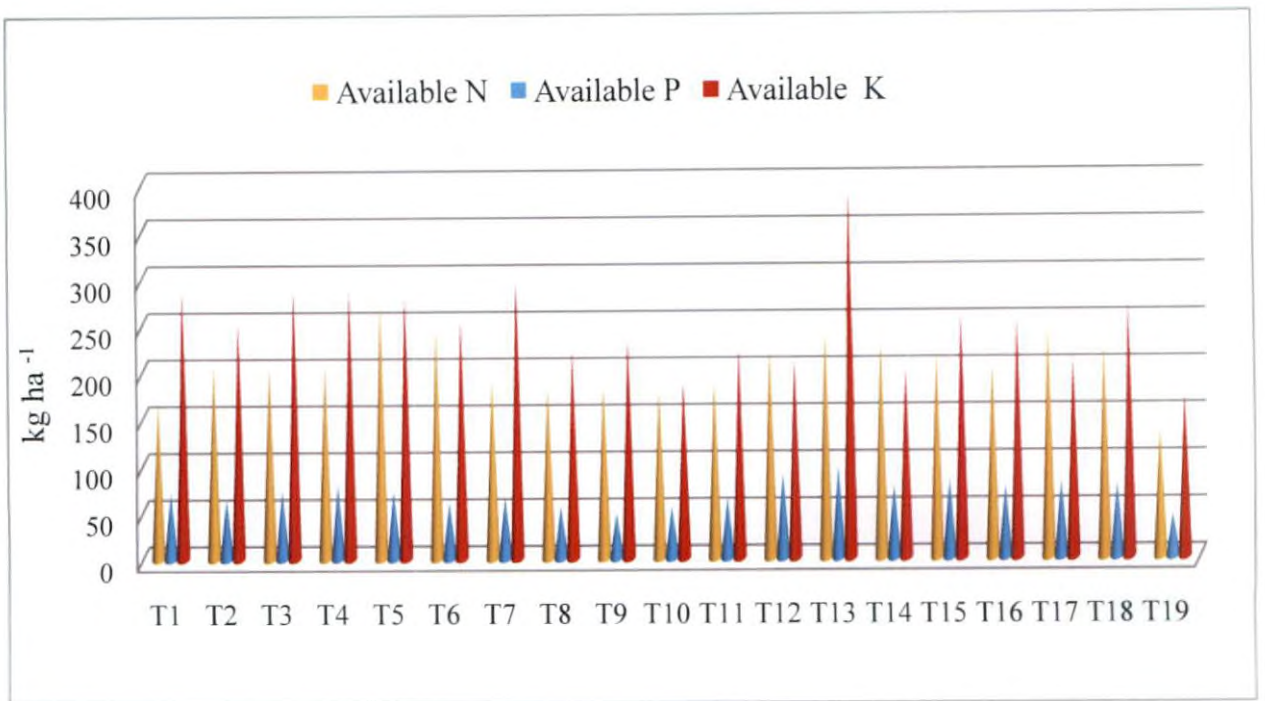


Figure 16. Effect of treatments on available content of major nutrients (N, P and K) in the soil.

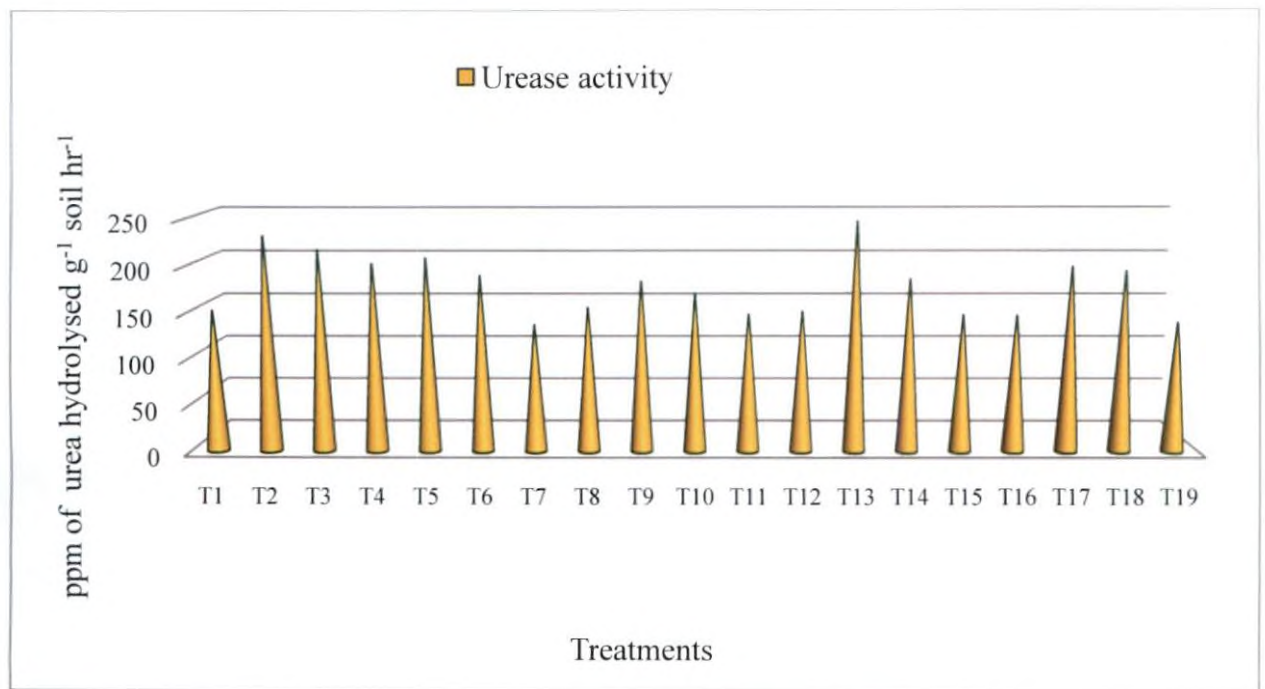


Figure 17. Effect of treatments on urease enzyme activity in the post harvest soil.



Available P content decreased generally with the application of various composts. Application of composts viz. water cabbage + Composting Inoculum, water cabbage + *P. sajor-caju*, farmwaste + *T. reesei*, farm waste + Composting Inoculum reported to have significant influence on the available P status of the soil. The dramatic increase in available P status of the soil might be due to the solubilisation of P from the unavailable fixed forms by the action of various organisms in the inoculum used. These findings were in agreement with the finding of Kaushik and Garg (2004). This might be also due to increased microbial activity causing greater mineralization of added compost and production of organic acids which solubilise fixed P in soil. In the case of individual effects, water cabbage in influencing the available P status was significantly maintained higher while the individual effects of inoculants was not significant.

The highest value for available K in the soil was recorded by the application of compost prepared out of water hyacinth and Composting Inoculum. The build up of soil available K was due to the additional K supplied through it and also solubilisation action of certain organic acids produced during decomposition and a greater capacity to hold K in available form. Kumar *et al.* (2007) also reported a soil build up of K due to the addition of composts. Substrate water cabbage significantly affected the available K content individually while the significant effect of inoculants was not reported.

Like the major nutrients, organic carbon status of the soil was also affected by the application of various composts. The compost prepared out of water hyacinth and *Trichoderma* found to increase the organic carbon content of soil. These results are in agreement with of Lynch *et al.* (2005) who reported the application of water hyacinth alone or coupled with other materials increased soil C:N ratio and the organic carbon status. The effect of inoculants used in the study was not found to be significant.

#### 5.3.4. Micro Nutrients

Availability of micronutrients to crop plants is affected by many soil factors *viz.* soil reaction, pH, soil organic matter content (Lindsay and Norvell, 1978). From the present study it was observed that all the micronutrients such as Fe, Mn, Zn, and Cu were influenced by the application of composts (Table 15). Application of composts of coir pith and Composting Inoculum, water hyacinth and commercial enzyme cocktail, water cabbage and Composting Inoculum were reported to have increased the soil Fe, Zn, and Cu content respectively. This might be due to the addition of organic matter by these composts which complexes with micronutrients and thereby increasing micronutrient mobility. This is in agreement with the findings of Kadalli *et al.* (2000). The increase in the micronutrient status such as Fe, Zn and Cu with the application of composts might be due to the improved soil characters and microbial activities as reported by Mortvedt (2010).

Non significant effect on Mn concentration was observed which was supported by the findings of Hue *et al.* (1988) who reported that the organic matter can regulate the availability of micronutrients forming stable complexes which become a part of solid phase and thus reduces the availability leading to a non significant effect.

In the case of Fe content of soil, coir pith and Composting Inoculum were found to influence mostly, there by their interaction effect were also significant. Water hyacinth and Composting Inoculum recorded highest Mn content, Coir pith and *T. reesei* recorded highest Zn content individually and Cu content was highest for water cabbage and Composting Inoculum. B content was significantly influenced by the treatments. Among the sixteen treatments, compost from water hyacinth and *T. reesei* has recorded the highest B content in soil. Individual effects were also significant and water hyacinth reported the highest B content and on par with farm waste. Among inoculants, *T. reesei* and *P.sajor- caju* were reported as the best. Similar findings were also reported by Wei and Liu (2005) where the increase in soil available Fe, Zn and Cu was due to the application of



sewage sludge compost. From this study it was evident that the application of compost increased the micronutrient status of the soil and however the increase was below the phytotoxic levels of these micronutrients as reported by Alloway and Ayers (1997).

### 5.3.5. Heavy Metals

The presence of heavy metals in soil after compost application was presented in Table 17. It was inferred that Pb, Cd, Ni were influenced by the application of composts. No detectable levels of Pb in soil was reported in this study by the application of composts prepared from water cabbage. This might be due to the absence of this toxic metal in the substrate itself. Coir pith composts resulted in lower value for Pb because it might have been successfully removed by adsorption on coir pith .

With respect to Cd, compost prepared from coir pith and Composting Inoculum was reported to have contributed to Cd content where as in Ni, coir pith and *T. reesei*. Even though some detectable levels of heavy metals are reported, their concentration in the post harvest soil samples are very below the permissible tolerance levels as suggested by Mellsted (1973) and Naidu *et al.* (1996).

### 5.3.6. Biological Parameters

Composting basically being a microbiological process, enzyme activities can be a part of reliable measure of compost stability and maturity. Soil enzymes increase the reaction rate at which plant residues decompose and release plant available nutrients. Enzymes respond to soil management changes long before other soil quality indicator changes are detectable (Sitaramalakshmi *et al.*, 2013). Soil is a dynamic system where all biochemical activities processed through enzymatic processes (Tabatabai, 1982). The effects of treatments on biological properties are presented in Table 18.

Enzymes such as urease, phosphatase, dehydrogenase, aryl sulphatase and cellulase were measured indirectly by determining their activity in the laboratory, which reflect potential activity and do not represent true in-situ activity levels and



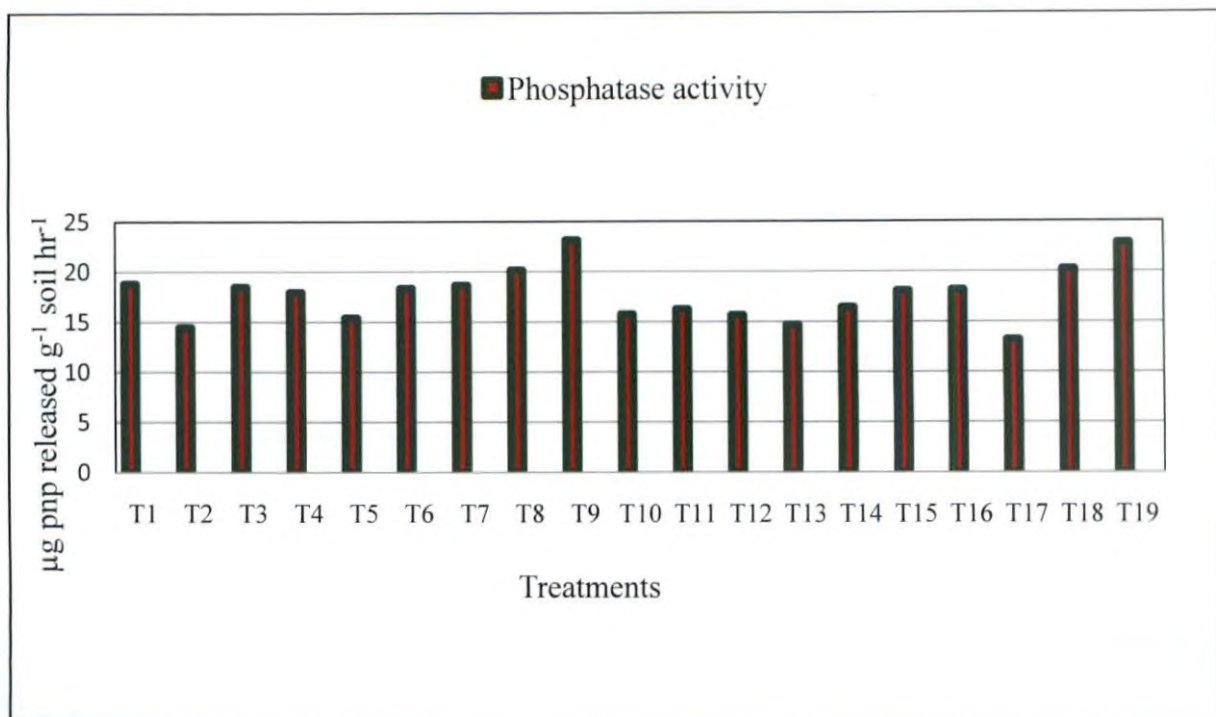


Figure 18. Effect of treatments on phosphatase enzyme activity in the post harvest soil.

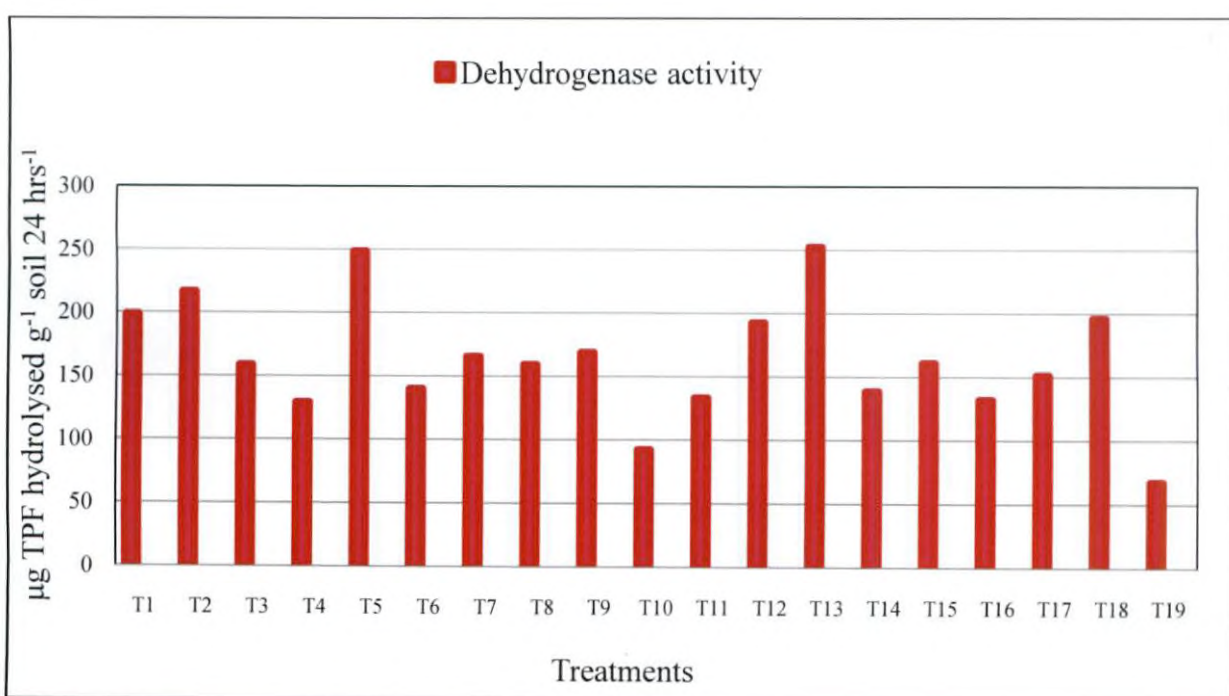


Figure 19. Effect of treatments on dehydrogenase enzyme activity in the post harvest soil.

can be viewed as index (Bendick and Dick, 1994). With regard to urease activity as observed from Figure 17. Application of compost using water hyacinth with Composting Inoculum recorded the highest value for urease and Composting Inoculum has proved its supremacy in influencing the urease activity. The lowest value for absolute control might be due to lack of sufficient organic carbon compounds for micro organisms which lead to decrease in synthesis of urease enzymes. Similar results were reported by Raut *et al.* (2008) and Devi *et al.* (2009) in their studies on composting of different organic residues. The highest activity with the application of water hyacinth compost might be attributed to high availability of easily degradable substrate and varying nutrient contents which ultimately resulted in the spurt of ureolytic bacteria and ultimately increasing the urease activity.

Phosphatase are enzymes of agronomic importance because it hydrolyses compounds of organic P and transforms them into inorganic P. Phosphatase or phosphomonoesterase catalyses the hydrolysis of esters of phosphoric acid to release  $PO_4$  and it is of paramount importance as a soil quality indicator (Traser-Cepeda *et al.*, 2008). In the present study (Figure 18) the application of compost (coir pith + Composting Inoculum) was observed to have increased phosphatase activity which was similar to activity reported in control pots. The increased activity of phosphatase enzyme might be probably due to the release of organically bound P as the synthesis of this enzyme stimulated by the presence of organic substrates (Biswas and Narayanasamy, 2006). The phosphatase activity thus extensively and positively influenced the yield thus signifying the role of compost (coir pith and Composting Inoculum). Kumar *et al.* (2007) have attributed to this increase in phosphatase activity to the large fungal population in the soil and increased rate of hydrolysis of the organic P in the soil.

Dehydrogenase exists as an integral part of intact cell involved in oxidative phosphorylation and reflects the total oxidative potential of soil microbial community by transferring hydrogen and electrons from substrates to acceptors. From the study it was evident that the application of water hyacinth



and Composting Inoculum had reported to increase the dehydrogenase activity in soil which was similar in effect to water cabbage and Composting Inoculum (Figure 19). This might be due to the significant individual effect of water hyacinth and Composting Inoculum on the soil dehydrogenase activity. Similar effects were reported by Bundela *et al.* (2010) described a generalized short term to medium term increase in dehydrogenase enzyme with organic inputs. As a consequence organic compost generally enhances the development of microflora and increases the total activity of soil. The increased production and release of growth promoting substance by the application of water hyacinth and Composting Inoculum compost might have contributed to the intense proliferation of microbial growth resulting in higher dehydrogenase activity (Castaldi *et al.*, 2008)

With regard to arylsulphatase (an enzyme catalyzing the hydrolysis of organic sulphate esters), a significant effect of compost was noticed. In the present study from (Figure 20) it was observed that compost prepared from water hyacinth and Composting Inoculum was reported to have increased aryl sulphates activity in post harvest soil sample. The substrate coir pith and inoculum *Pleurotus* have influenced the aryl sulphatase activity individually. Similar effects with the application of compost were reported by Eivazi *et al.* (2003).

The  $\beta$ -glucosidase activity has found to be sensitive to soil management and has been proposed as a soil quality indicator because it provides an early indicator of change in organic matter status. From the study it was evident that the treatments influenced  $\beta$ -glucosidase activity (Figure 21). In contrast with the other observations absolute control has recorded the highest value for this enzyme. This might be due to the presence of other enzymes in other treatments involving the application of various composts that have interfered with the activity of  $\beta$ -glucosidase. Since the absolute control recorded the lowest value for enzyme activities, the interference was found to be less. These results corroborated with the findings of (Tejeda *et al.*, 2009).

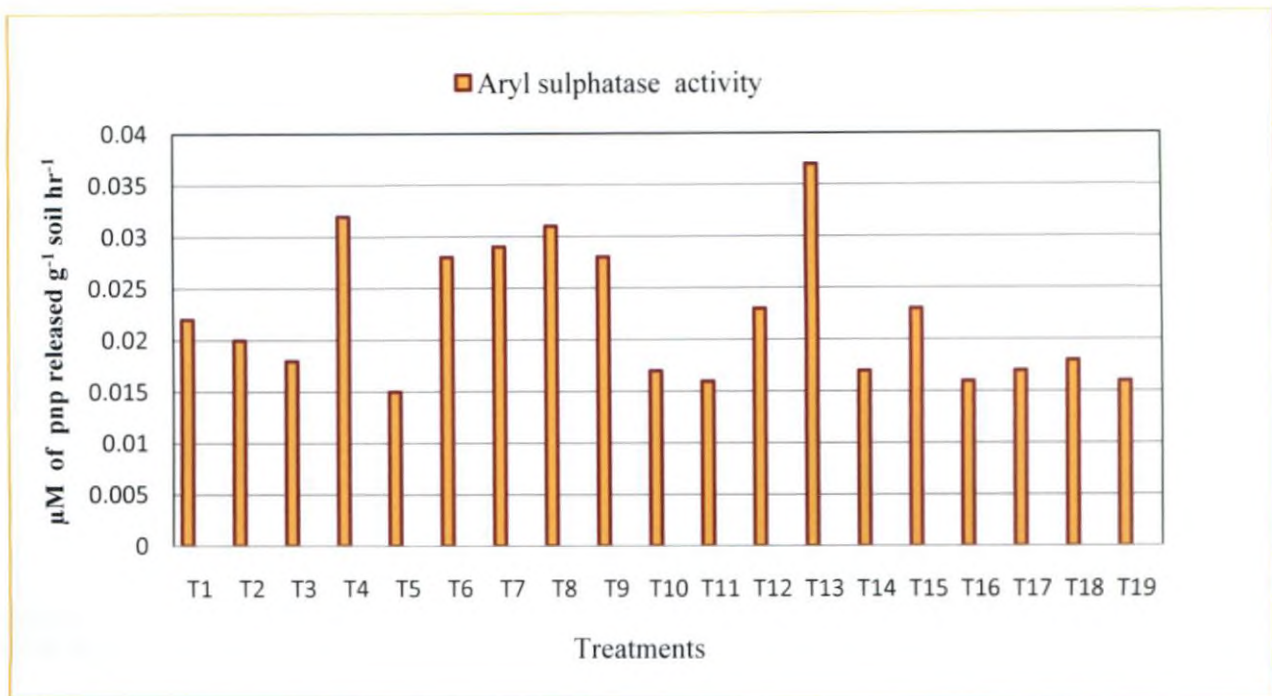


Figure 20. Effect of treatments on aryl sulphatase enzyme activity in the post harvest soil.

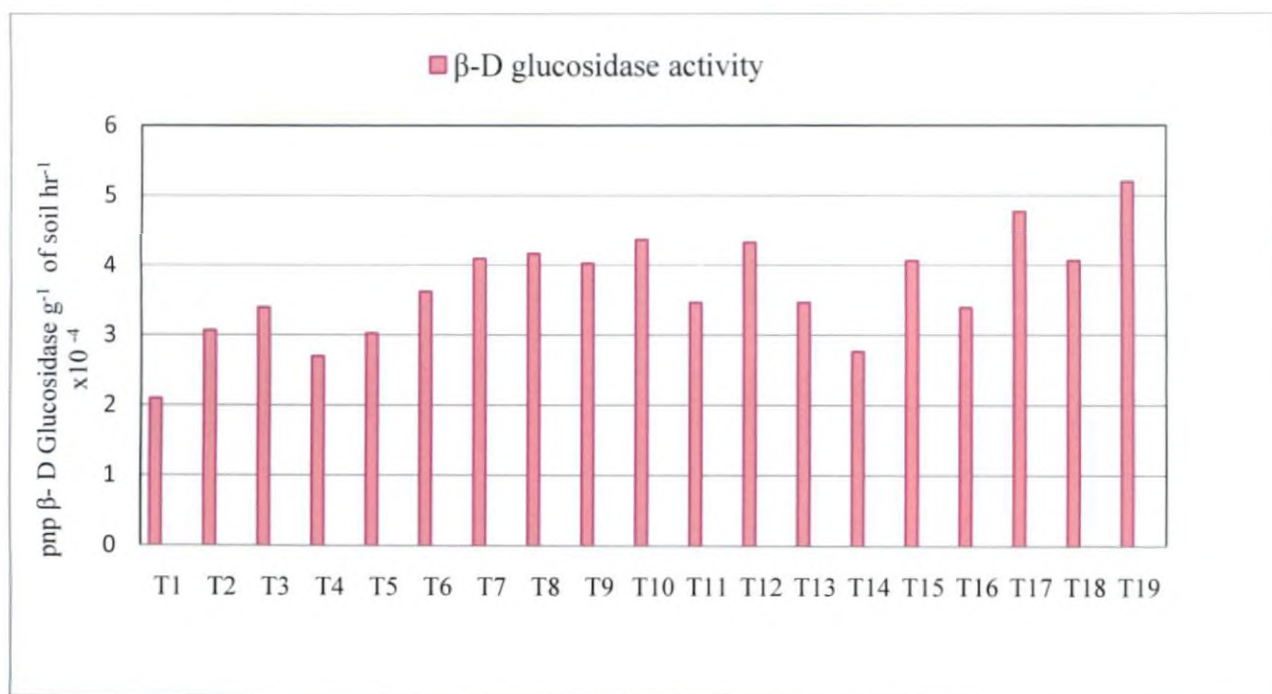


Figure 21. Effect of treatments on β D- Glucosidase enzyme activity in the post harvest soil.

Respiratory activity of soil is an important index of microbial growth and activity in soil (Frankenberger and Dick, 1982). From this study, no significant variation was observed with the application of various substrates. However the compost from coir pith and *T. reesei* recorded the highest value for respiratory activity. This might have been due to the spurt in the microbial population with the addition of composts. A higher soil respiration supports the notion that the rapid decomposition of plant residues (coir pith) that might have increased the nutrients available for stimulation of heterotrophic microorganisms. Absolute control recorded the lowest value due to the lack of microorganisms and lower soil microbial metabolite of CO<sub>2</sub> resulting in lower CO<sub>2</sub> production. However it was inferred that application of compost from coir pith and *T. reesei* has a special mechanism or secretions that increases the microbial respiration in soil. These findings were in agreement with the finding of Zhang *et al.* (2003).

Soil microbial communities are the driving force in regulating soil processes such as organic matter decomposition and nutrient cycling. The role that microbial activity plays in an ecosystem process is significant because approximately 80 per cent to 90 per cent of soil processes are mediated by microorganisms (Nannipieri *et al.*, 1978). Since microbial activities are important in regulating soil properties (Dick, 1992) an integral role that play in biochemical cycles, a better understanding of the structure and function of microbial communities in soil amended with manure is needed and hence the present study was initiated in this aspect.

In the case of bacteria the compost water cabbage and Composting Inoculum has proved their efficiency individually in increasing bacterial count. The effect of substrate was found to be significant on the bacterial population while inoculant did not affect. Addition of composted water cabbage increased the bacterial activity representing a shift in microbial response resulting in changes in substrate availability.

In the case of fungi, the highest value recorded with the addition of compost prepared out of water cabbage and *T. reesei* and the individual effect of substrates were found to be significant and reflected in the interaction. Bohme *et al.* (2005) reported that the fungal activity in the soil was highest with the application of compost. However the effect of inoculum was insignificant under this study.

Highest population of actinomycetes was noticed with compost produced from water hyacinth and *T. reesei* while the lowest value recorded with the application of compost from water cabbage and commercial enzyme cocktail. The substrates did not reveal any individual effect on actinomycetes. Similar observations were noticed with the individual effect of inoculants. This might be due to the more stable and readily available substrate which supported higher level of actinomycetes by the compost acted upon by *Trichoderma*. Similar results were reported by Dick (1992).

#### 5.4. NUTRIENT CONTENT OF AMARANTHUS

From the study, total nutrient content of amaranthus are presented in the Tables 19 and 20.

Nitrogen is the most important element needed for the growth and development of the plants. Composts are considered as one the best sources of nitrogen. Present study reveals that the N content of the plants was significantly influenced by the treatments. Application of compost prepared from farm waste and *T. reesei* has resulted the highest N content in the plants. This might be due the high N content of the compost as well as the readily available form of ions due to the particular interaction of inoculant and the substrate. Saxena *et al.* (2001) found that the application of N helped in increasing the vegetative growth of plants and also plant dry matter in soybean. Among the substrates, farm waste imposed significant effect on the N content. This might be due to the presence of relatively more available form of N in composts.

P is an essential nutrient required for the plant growth, nutrient uptake especially for the root development. Compost prepared from farm waste and

*T. reesei* resulted in the highest P content in the crop. El-Din *et al.* (2000) reported that the compost produced by highly effective cellulose decomposing micro organisms like *T. viridae* or *Streptomyces auerofaciens* induced a significant increase in plant dry matter, N and P content and fruit yield in Tomato. Considering the total K content, compost from water cabbage and Composting Inoculum has resulted the highest K content in the test plant.

With respect to the micro nutrient status, the highest Fe content was recorded by compost prepared from coir pith and Composting Inoculum. Substrates imposed significant influence on Fe content. Regarding Mn content, water hyacinth and Composting Inoculum has resulted in the highest value. Substrate as well as inoculants imparted significant variations. Zn is one of the most important micronutrient needed for the metabolic activities of the plants. The highest Zn content was recorded by coir pith and *T. reesei* which was on par with coir pith and Composting Inoculum. Substrates and inoculants also significantly influenced the Zn content. Individual effect of coir pith recorded the highest Zn content to the plants and among inoculants Composting Inoculum recorded the highest value. Considering the Cu content treatments and substrates significantly influenced and treatment with water hyacinth and Composting Inoculum had recorded the highest Cu content and the substrate water cabbage was noticed as the best. Another important micronutrient B was also significantly varied with the treatments, substrates and inoculants. The best treatment recorded the highest B content by the compost prepared from water hyacinth and Composting Inoculum which was on par with water cabbage and Composting Inoculum. Kler *et al.* (2002) also reported similar results as on micro nutrients *viz.* Fe, Cu, Zn and Mn content in plants under organic farming over the chemical fertilizer and control. More extractable micronutrients might be attributed to chelating action of organic compounds released during decomposition of organic sources, which increased the availability of micronutrients by preventing fixation, oxidation, precipitation and leaching. Similar results have been reported by Hao and Chang (2003).

# *Summary*



## 6. SUMMARY

The study entitled "Characterization, conversion and evaluation of selected lignocellulosic biomass" was carried out in the Department of Soil Science and Agricultural Chemistry at College of Agriculture, Vellayani, Thiruvananthapuram during August 2013 to February 2014. The study was envisaged for the characterization of the lignocellulosic biomass from selected plant sources, assessment of various microbial and enzymatic sources for degrading the lignocellulosic biomass into compost and evaluation of the resultant compost for assessing manurial value.

The salient findings of the first phase involving the characterization are furnished below.

- The substrate S<sub>2</sub> (coir pith) registered the highest cellulose content with S<sub>3</sub> (water hyacinth) while the lowest content was recorded by S<sub>1</sub> (water cabbage).
- The substrate S<sub>3</sub> (water hyacinth) recorded the highest hemicellulose content and S<sub>2</sub> (coir pith) recorded the lowest content. The highest lignin content was recorded by S<sub>2</sub> (coir pith) while S<sub>3</sub> (water hyacinth) recorded the lowest content. S<sub>2</sub> (coir pith) recorded the widest C:N ratio and the lowest was recorded by S<sub>1</sub>(water cabbage).
- Regarding the NPK content, S<sub>1</sub> (water cabbage) recorded the highest N and K content, while S<sub>3</sub> (water hyacinth) recorded the highest P content. S<sub>2</sub> (coir pith) recorded the lowest contents of all the nutrients.
- Heavy metal contents in all substrates were low inspite of this S<sub>3</sub> (water hyacinth) recorded comparatively higher content of Pb and Sn, while Cd content was highest in S<sub>4</sub> (pseudostem and dried leaves of banana). Coir pith recorded the highest Ni content while Pb, Cd and Sn content in water cabbage were below the detectable levels.

The salient findings of second phase involving the production of composts with four substrates and four inoculants are summerized as follows:

- The highest moisture content was recorded by S<sub>1</sub>I<sub>3</sub> (water cabbage and Composting Inoculum). Regarding pH, S<sub>4</sub>I<sub>2</sub> (farm waste+ *P. sajor-caju*) recorded the highest value. All the biodegradable wastes maintained neutral pH. However the substrates, S<sub>4</sub> (farm waste) recorded the highest pH. Compost produced from water hyacinth and Composting Inoculum recorded the highest value of EC. Considering the ash content, S<sub>2</sub>I<sub>3</sub> (coir pith +Composting Inoculum) recorded the highest value, while S<sub>4</sub> (farm waste) and I<sub>4</sub> (commercial enzyme cocktail) registered the highest values individually.
- Compost S<sub>1</sub>I<sub>1</sub> (water cabbage + *T. reesei*) recorded the highest organic matter content. S<sub>1</sub> (water cabbage) and I<sub>1</sub> (*T. reesei*) recorded the highest value of organic matter.
- Regarding the nutrient content S<sub>1</sub>I<sub>3</sub> (water cabbage + Composting Inoculum) recorded the highest N content followed by S<sub>3</sub>I<sub>3</sub> (water hyacinth + Composting Inoculum). Water cabbage was the best among the substrates followed by water hyacinth. Composting Inoculum was the best among the inoculants. S<sub>2</sub> (coir pith) and I<sub>1</sub> (*T. reesei*) registered the lowest nitrogen contents individually.
- Total content of P and K were highest in compost S<sub>3</sub>I<sub>3</sub> (water hyacinth + Composting Inoculum) and S<sub>3</sub> (water hyacinth) was the best among substrates and I<sub>3</sub> (Composting Inoculum) was the best among inoculants.
- The dehydrogenase enzyme activity was highest in the compost S<sub>1</sub>I<sub>2</sub> (water cabbage+ *P. sajor-caju*). Regarding the individual effects, substrate S<sub>1</sub> (water cabbage) and I<sub>3</sub> (Composting Inoculum) recorded the highest value. S<sub>1</sub>I<sub>3</sub> (water cabbage +Composting Inoculum) recorded the highest cellulase activity. S<sub>2</sub> (coir pith) recorded the highest activity among substrates and I<sub>3</sub> (Composting Inoculum) among the inoculants.
- In the case of maturity period, S<sub>2</sub>I<sub>4</sub> (coir pith + *P. sajor-caju*) noticed with the longest duration and S<sub>1</sub>I<sub>3</sub> (water cabbage + Composting Inoculum) with the shortest period. Substrate S<sub>2</sub> (coir pith) and inoculant I<sub>4</sub> (commercial enzyme cocktail) recorded the longest maturity period

individually while S<sub>1</sub> (water cabbage) and I<sub>3</sub> (Composting Inoculum) recorded the shortest maturity periods.

- The widest C: N ratio was recorded by the treatment S<sub>2</sub>I<sub>1</sub> (coir pith + *P. sajor-caju*) and the lowest was reported by S<sub>1</sub>I<sub>3</sub> (water cabbage + Composting Inoculum). Considering the individual effect of substrates, S<sub>2</sub> (coir pith) recorded the widest value and the lowest value by S<sub>3</sub> (water hyacinth) . In case of individual effect of inoculants, I<sub>1</sub> (*T. reesei*) had recorded the widest C:N ratio and I<sub>3</sub> (Composting Inoculum) has recorded the lowest value.
- All the composts prepared were reported to have heavy metals lower than the permissible limit. However S<sub>4</sub>I<sub>2</sub> (farm waste + *P. sajor-caju*) recorded the highest Pb content and composts prepared from S<sub>4</sub> (water cabbage) recorded no detectable levels of Pb. Composts prepared from farm waste by the action of *P. sajor-caju* recorded the highest Cd content. S<sub>2</sub>I<sub>3</sub> (coir pith + Composting Inoculum) has recorded the highest Ni content. The compost S<sub>3</sub>I<sub>4</sub> (water hyacinth + commercial enzyme cocktail) recorded the highest Sn content.

With regard to biometric observations in pot culture experiment,

- Plant height variation was not significant. However the highest plant height was recorded by T<sub>13</sub> (100 per cent N as compost from water hyacinth and Composting Inoculum). S<sub>3</sub> (water hyacinth) and I<sub>3</sub> (Composting Inoculum) recorded the highest mean values individually.
- The treatment T<sub>5</sub> (100 per cent N as compost from water cabbage and Composting Inoculum) recorded the highest number of leaves per branches. Among the substrates, S<sub>1</sub> (water cabbage) recorded the highest value and S<sub>4</sub> (farm waste) recorded the lowest value. Inoculant I<sub>1</sub> (*T. reesei*) was recorded as the best in the case of inoculants.
- Treatments, inoculants and their interactions did not significantly influence the number of branches, only substrates had significant effect.

Among substrates, S<sub>3</sub> (water hyacinth) recorded the highest number of branches.

- Treatments, substrates, inoculants and their interactions have no significant effect on girth of the stem however the treatment T<sub>13</sub> (100 per cent N as compost from water hyacinth and Composting Inoculum) recorded the highest girth of stem.
- With regard to yield of amaranthus T<sub>13</sub> (100 per cent N as compost from water hyacinth and Composting Inoculum) recorded the highest yield which was on par with T<sub>5</sub> (100 per cent N as compost from water cabbage and Composting Inoculum). Considering the individual effect of substrates, S<sub>3</sub> (water hyacinth) and among inoculants I<sub>3</sub> (Composting Inoculum) recorded the highest yields respectively.
- In the case of physical properties *viz.* particle density, bulk density and water holding capacity, compost application imparted positive variations in the soil.
- Treatments imposed significant effect on available N content of the soil however their interaction was found to be non significant. The highest value recorded by T<sub>5</sub> (100 per cent N as compost from water cabbage and Composting Inoculum) which was on par with the treatments T<sub>6</sub> (100 per cent N as compost from water cabbage + commercial enzyme cocktail), T<sub>13</sub> (100 per cent N as compost from water hyacinth and Composting Inoculum) , and T<sub>17</sub> (farm waste + Composting Inoculum). S<sub>1</sub> (water cabbage) recorded the highest value among substrates. Regarding inoculants, I<sub>3</sub> (Composting Inoculum) recorded the highest value
- T<sub>13</sub> (100 per cent N as compost from water hyacinth and Composting Inoculum) recorded the highest value of available P content. Considering the individual effect S<sub>3</sub> (water hyacinth) recorded the highest value while the effects of inoculants were found to be non significant.
- In the case of available K, the highest value was recorded by T<sub>13</sub> (100 per cent N as compost from water hyacinth and Composting Inoculum). S<sub>1</sub>

(water cabbage) recorded the highest K among substrates while the effect of inoculants was not significant.

- Treatment T<sub>11</sub> (100 per cent N as compost from water hyacinth and *T. reesei*) recorded the highest organic carbon content. Considering the individual effect of substrates, S<sub>2</sub> (coir pith) recorded the highest value and inoculants did not influence significantly.
- Available Fe content of the soil significantly influenced and T<sub>9</sub> (coir pith + Composting Inoculum) recorded the highest value which was on par with T<sub>13</sub> (100 per cent N as compost from water hyacinth and Composting Inoculum), T<sub>17</sub> (100 per cent N as compost from farm waste and Composting Inoculum) and T<sub>12</sub> (100 per cent N as compost from water hyacinth and *P. sajor-caju*). S<sub>2</sub> (coir pith) recorded the highest value among substrates and I<sub>3</sub> (Composting Inoculum) was found with the highest Fe content among inoculants.
- Application of various treatments did not impose significant effect on the available Mn content of the treated soils even though T<sub>13</sub> (100 per cent N as compost from water hyacinth and Composting Inoculum) recorded the highest Mn. S<sub>3</sub> (water hyacinth) and I<sub>3</sub> (Composting Inoculum) recorded the highest mean values among substrates and inoculants respectively.
- Considering the Zn content, treatments and substrates had no significant effect but the inoculants imposed significant effect and I<sub>1</sub> (*T. reesei*) recorded the highest value.
- Available Cu content was not significantly influenced by the treatment however T<sub>5</sub> (100 per cent N as compost from water cabbage and Composting Inoculum) and T<sub>13</sub> (100 per cent N as compost from water hyacinth and Composting Inoculum) recorded the highest Cu content.
- Treatments, interactions, substrates and the inoculants significantly influenced the available B content in the soil. Among the sixteen treatments, T<sub>11</sub> (100 per cent N as compost from water hyacinth and *T. reesei*) recorded the highest content of B. With regard to substrates S<sub>3</sub> (water hyacinth) and S<sub>4</sub> (farmwaste) were on par and among the inoculants

I<sub>1</sub> (*T. reesei*) and I<sub>2</sub> (*P. sajor-caju*) recorded the highest and equal B content.

- Regarding the content of heavy metals viz. Pb, Cd, Ni and Sn in the treated soils, it was found that all were below critical levels and the application of various composts did not result heavy metal accumulation in the soil.
- Different treatments had imposed significant effect on biological properties of the soil. The highest activity of urease was recorded by the treatment T<sub>13</sub> (100 per cent N as compost from water hyacinth and Composting Inoculum). Substrate S<sub>1</sub> (water cabbage) and Inoculum I<sub>3</sub> (Composting Inoculum) recorded the highest activity individually.
- The phosphatase enzyme activity was not significantly influenced by the treatments however T<sub>9</sub> (100 per cent N as compost from coir pith and Composting Inoculum) recorded the highest value.
- In the case of dehydrogenase enzyme activity the highest mean value was recorded by the treatment T<sub>13</sub> (100 per cent N as compost from water hyacinth and Composting Inoculum) which was on par the treatment T<sub>5</sub> (100 per cent N as compost from water cabbage and Composting Inoculum). Individual effect of the substrates was not significant while effect of inoculants was significant and I<sub>3</sub>(Composting Inoculum) registered highest dehydrogenase activity.
- Arylsulphatase enzyme activity was significantly varied between treatments however T<sub>13</sub> (100 per cent N as compost from water hyacinth and Composting Inoculum) recorded the highest value.
- $\beta$ - Glucosidase enzyme activity was highest in absolute control T<sub>19</sub>. Effect of the substrates, were significant and S<sub>2</sub> (coir pith) recorded the highest activity while the effect of inoculants was not significant.
- Regarding the other biological properties, treatment T<sub>7</sub> (100 per cent N as compost from coir pith and *T. reesei*) recorded the highest respiratory activity. Maximum bacterial population of was recorded in the treatment T<sub>13</sub>. (100 per cent N as compost from water hyacinth and Composting Inoculum). In the case of fungi, the highest mean value was recorded by

the treatment T<sub>3</sub> (water cabbage+ Composting Inoculum). Considering the population of actinomycetes the highest number of viable colonies were recorded by the treatment T<sub>11</sub> (100 per cent N as compost from water hyacinth and *T. reesei*). Individual effect of substrates and inoculants were not statistically significant with respect to the population of bacteria, fungi and actinomycetes.

- Treatments imposed significant influence on total N content of the plants and T<sub>15</sub> (100 per cent N as compost from farm waste and *T. reesei*) recorded the highest content. Substrates imposed significant influence and S<sub>4</sub> (farm waste) recorded the highest value while the effect of inoculants was not significant.
- Total P and K contents were not significantly influenced by the treatments, substrates or by their inoculants. Substrates imposed significant effect on K content and S<sub>1</sub> (water cabbage) recorded the K content.
- Different treatments had no significant effect on the total Fe content of the plants while the individual effect of substrates was significant and, S<sub>1</sub> (water cabbage) recorded the highest mean value. Effect of inoculants was not at all significant on Fe content.
- Various treatments had no significant effect on the total Mn content of the plants however the highest content was recorded by the treatment T<sub>13</sub> (100 per cent N as compost from water hyacinth and Composting Inoculum). S<sub>1</sub> (water cabbage) and I<sub>3</sub> (Composting Inoculum) recorded the highest Mn content individually.
- Zn, Cu and B content of the plants was found to be significantly influenced by the treatments, substrates and inoculants. The highest Zn content was recorded by the treatment T<sub>7</sub> (100 per cent N as compost from coir pith and *T. reesei*). In the case of individual effects of substrates, the highest value was recorded by the substrate S<sub>2</sub> (coir pith) and among inoculants I<sub>3</sub> (Composting Inoculum) recorded the highest value. The highest value of total Cu was recorded by the treatment T<sub>13</sub> (100 per cent N as compost from water hyacinth and Composting Inoculum) which was

found to be on par with the treatments T<sub>5</sub> (100 per cent N as compost from water cabbage and Composting Inoculum), T<sub>12</sub> (100 per cent N as compost from water hyacinth + *P. sajor-caju*) and T<sub>7</sub> (coir pith+ *T. reesei*) and T<sub>3</sub> (100 per cent N as compost from water cabbage and *T. reesei*). Regarding the individual effect of substrates and inoculants, S<sub>1</sub> (water cabbage) and I<sub>3</sub> (Composting Inoculum) recorded the highest mean values. T<sub>13</sub> (100 per cent N as compost from water hyacinth and Composting Inoculum) recorded the highest B content and was on par with the treatments T<sub>5</sub> (100 per cent N as compost from water cabbage and Composting Inoculum) and T<sub>17</sub> (100 per cent N as compost from farmwaste and Composting Inoculum). Individual effects of substrates and inoculants were significant and S<sub>3</sub> (water hyacinth) and I<sub>3</sub> (Composting Inoculum) recorded the highest B content respectively.

## CONCLUSION

Thus it was inferred from the study that water cabbage was the best substrate in terms of its chemical composition followed by water hyacinth. S<sub>1</sub>I<sub>3</sub> (water cabbage + Composting Inoculum) and S<sub>3</sub>I<sub>3</sub> (water hyacinth+composting Inoculum) were yielded best composts. T<sub>13</sub>{100 % N as compost (water hyacinth + composting Inoculum)} and T<sub>5</sub>{100 % N as compost (water cabbage + composting Inoculum)} were performed better in pot culture. With regards to inoculants used on different substrates, composting Inoculum was found to be the most effective for composting the agrowastes.



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\* Originals not seen

# *Appendices*

## APPENDIX I

Weather Parameters during field experiment (December 2013- February 2014)

| Standard weeks | Maximum Temperature (°C) | Minimum Temperature (°C) | Maximum RH (%) | Minimum RH(%) | Rainfall (mm) |
|----------------|--------------------------|--------------------------|----------------|---------------|---------------|
| 49             | 30.9                     | 22.8                     | 98.6           | 69.9          | 1.4           |
| 50             | 30.3                     | 22.6                     | 96.7           | 69.6          | 26            |
| 51             | 31.2                     | 21.7                     | 97.7           | 72            | 47            |
| 52             | 31                       | 20.2                     | 96.6           | 59.1          | 0             |
| 1              | 30.9                     | 21.5                     | 94.9           | 77.6          | 0             |
| 2              | 29                       | 21.8                     | 94.4           | 77.4          | 14            |
| 3              | 31                       | 22.3                     | 94.1           | 76.1          | 0             |
| 4              | 31.3                     | 20.7                     | 90.4           | 69.9          | 0.5           |
| 5              | 31.4                     | 21.9                     | 92.3           | 68.6          | 0             |

## APPENDIX II

| Composts        | Details                                     |
|-----------------|---|
| C <sub>1</sub>  | Water cabbage + <i>T. reesei</i>            |
| C <sub>2</sub>  | Water cabbage + <i>P. sajor-caju</i>        |
| C <sub>3</sub>  | Water cabbage + Composting Inoculum         |
| C <sub>4</sub>  | Water cabbage + Commercial enzyme cocktail  |
| C <sub>5</sub>  | Coir pith + <i>T. reesei</i>                |
| C <sub>6</sub>  | Coir pith + <i>P. sajor-caju</i>            |
| C <sub>7</sub>  | Coir pith + Composting Inoculum             |
| C <sub>8</sub>  | Coir pith + Commercial enzyme cocktail      |
| C <sub>9</sub>  | Water hyacinth + <i>T. reesei</i>           |
| C <sub>10</sub> | Water hyacinth + <i>P. sajor-caju</i>       |
| C <sub>11</sub> | Water hyacinth + Composting Inoculum        |
| C <sub>12</sub> | Water hyacinth + Commercial enzyme cocktail |
| C <sub>13</sub> | Farm waste + <i>T. reesei</i>               |
| C <sub>14</sub> | Farm waste + <i>P. sajor-caju</i>           |
| C <sub>15</sub> | Farm waste + Composting Inoculum            |
| C <sub>16</sub> | Farm waste + Commercial enzyme cocktail     |

## APPENDIX III

### COMPOSITION OF MEDIA FOR MICROBIAL ENUMERATION

#### 1. Enumeration of Bacteria

Media: Nutrient Agar

Composition

|                     |   |         |
|---------------------|---|---------|
| 1. Peptone          | - | 5 gm    |
| 2. NaCl             | - | 5 gm    |
| 3. Beef extract     | - | 3 gm    |
| 4. Agar             | - | 20 gm   |
| 5. pH               | - | 7.0     |
| 6. Distilled water- |   | 1000 ml |

#### 2. Enumeration of Fungi

Media: Rose Bengal agar

Composition

|                                    |   |         |
|------------------------------------|---|---------|
| 1. Glucose                         | - | 3g      |
| 2. MgSO <sub>4</sub>               | - | 0.2 g   |
| 3. K <sub>2</sub> HPO <sub>4</sub> | - | 0.9 g   |
| 4. Rose Bengal                     | - | 0.5 g   |
| 5. Streptomycin                    | - | 0.25 g  |
| 6. Agar                            | - | 20 g    |
| 7. Distilled water-                |   | 1000 ml |

#### 3. Enumeration of Actinomycetes

Media: Kenknight's Agar

Composition

|                                    |   |         |
|------------------------------------|---|---------|
| 1. Dextrose                        | - | 1 g     |
| 2. KH <sub>2</sub> PO <sub>4</sub> | - | 0.1 g   |
| 3. NaNO <sub>3</sub>               | - | 0.1 g   |
| 4. KCl                             | - | 0.1 g   |
| 5. MgSO <sub>4</sub>               | - | 0.1 g   |
| 6. Agar                            | - | 15 g    |
| 7. Distilled water-                |   | 1000 ml |

# **Characterization, Conversion and Evaluation of Selected Lignocellulosic Biomass**

*by*

**ANUSHMA S.**

**(2012-11-111)**

**Abstract of the thesis**

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**DEPARTMENT OF SOIL SCIENCE AND AGRICULTURAL CHEMISTRY**

**COLLEGE OF AGRICULTURE**

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**2014**

## ABSTRACT

The study entitled "Characterization, conversion and evaluation of selected lignocellulosic biomass" was conducted during the period 2013-14 at the Department of Soil Science & Agricultural Chemistry, College of Agriculture, Vellayani. The main objectives of the study were the characterization of the lignocellulosic biomass from selected plant sources, assessment of various microbial and enzymatic sources for degrading the lignocellulosic biomass into compost and evaluation of the resultant compost for assessing its manurial value.

The experiment was completed in three stages. Representative samples of water cabbage (*L. flava*), coir pith, water hyacinth (*E. crassipes*), and farm wastes (dried leaves and pseudostem of banana) were collected and analysed for bio-chemical composition. Water cabbage recorded 2.74 per cent N, 0.30 per cent P, 0.33 per cent K, 16.98 C:N ratio, and no detectable levels of heavy metals except Ni (0.55 ppm) and inferred as the best substrate in terms of nutrient content followed by water hyacinth, farm wastes and coir pith respectively.

The substrates were converted to composts using various inoculants viz. *T. reesei*, *P. sajor-caju*, Composting Inoculum and commercial enzyme cocktail (cellulase/pectinase and lactase) and physico-chemical and biological characteristics were analyzed. A mixture of water cabbage and Composting Inoculum) was concluded as the best in terms of nitrogen content, cellulase activity, maturity period, C:N ratio and no detectable levels of heavy metals except Ni (0.414 ppm) followed by the mixture of Water hyacinth and Composting Inoculum had comparable N, P, K, EC, C:N ratio, and heavy metal content below the detectable limit. Considering the effect of inoculants on different substrates, Composting Inoculum was concluded as the best in terms of moisture content, EC, N, P, K, dehydrogenase activity, cellulase activity, maturity period and C:N ratio.

The resultant composts from the previous stage were evaluated for their performance as manure in a pot culture experiment with test crop amaranthus (variety - Arun). Application of 100 % N as compost water hyacinth and Composting Inoculum was noticed as the best treatment in terms of yield, plant height, number of branches, girth of stem, soil properties viz. water holding capacity, available N, available P, available K,

available Mn, available Cu, bacteria population and plant content of micronutrients viz. Mn, Cu, and B. Major enzymes viz. dehydrogenase, urease and aryl sulphatase imposed significance on yield and yield attributes. Application of 100 % N as compost from water cabbage and Composting Inoculum was found to be good and on par with T<sub>13</sub> in many of the characters. Application of various composts did not result any heavy metal accumulation in the soil

Thus it was inferred from the study that water cabbage was the best substrate in terms of their chemical composition followed by water hyacinth. water cabbage + Composting Inoculum was recorded as the best compost followed by water hyacinth + Composting Inoculum. 100 % N as compost (water hyacinth + Composting Inoculum) was noticed as the best treatment in pot culture. With regards to inoculants used on different substrates, Composting Inoculum was found to be the most effective for composting the agrowastes.

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