

**Optimization of biodegradation and nutrient release in food waste and goat manure
vermicompost amended with seaweed-biochar**

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DECLARATION

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ABSTRACT

Vermicomposting has been gaining momentum as a technology capable of generating a nutrient rich organic fertilizer. However, for effective nutrient release, there is need for the optimization of this biological process to allow repeatable production of organic nutrient sources. This study aimed to generate an optimized vermicompost produced from food waste, goat manure and seaweed biochar. This study was guided by the following specific objectives, (i) to characterise biochar produced from two seaweed species at different pyrolysis temperatures (ii) to optimize different combination of Food Waste (FW) and Goat Manure (GM) vermicompost (iii) to optimise the seaweed biochar incorporation into food waste-goat manure mixtures for enhanced biodegradation and nutrient mineralization. Biochar from *G. funicularis* had the highest macro elements with a total of C 38.3%; N 4.3% and P 6.3 g/kg contents, whilst *L. pallida* had the highest cations. A pyrolysis temperature of 400°C resulted in the best quality biochar in terms of macro elements, pH, and total C. *G. funicularis* biochar had significantly higher nutrient concentrations with a great potential for soil quality improvement. The 75 FW: 25 GM and 50 FW: 50 GM vermicompost combinations had significantly ($P \leq 0.05$) the highest final concentration of Olsen phosphorus of 0.98 and 0.96 g per kg of compost, respectively. Biochar incorporation of 4% may be the most optimal, as it enhanced biodegradation and nutrient release than all other treatments with biochar incorporation. Our study clearly indicated that local Namibian waste biomass can be used to develop nutrient rich organic fertilizer. However, the study was done under laboratory conditions further studies will need to evaluate the actual amount of beach cast biomass harvested as well as food waste produced in various industries in order to assess the actual feasibility of producing such fertilizers in practical terms.

Keywords: Seaweed biochar, nutrient release, biodegradation, humification parameters, macro-nutrients.

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PREFACE

This thesis is made up of six separate chapters. Chapter 1 gives a general introduction on food waste, biochar, goat manure as waste material with a good potential as a nutrient source. Chapter 2 contains literature review on food waste, biochar and goat manure, the use of these materials for vermicomposting and how to improve their fertilizer value. Chapter 3 presents the fertilizer value of biochar prepared from beach-cast seaweed (*Laminaria pallida* and *Gracilariopsis funicularis*) as a nutrient source in organic agriculture. Chapter 4 deals with the optimization of goat manure-food waste mixture vermicompost using *Eisenia fetida* for enhanced nutrient release. Chapter 5 evaluates the seaweed (*Gracilariopsis funicularis*) biochar incorporation into a goat manure-food waste vermicompost for optimized vermi-degradation and nutrient release. Chapter 6 gives the general discussion of all the research chapters, conclusions and recommendations for future research on biochar vermicomposting. The chapters in the thesis have been treated as separate though linked studies and thus some repetition between chapters could not completely be avoided.

DEDICATION

I dedicate this work to Pelagia Mwalonga Katakula, Immanuel Uupindi Katakula, Phillipus Nhinda, and Otto Mwetulundila, who encouraged and loved me.

CHAPTER ONE

GENERAL INTRODUCTION

1.0 INTRODUCTION

The increase in world population and rapid growth of urbanization has resulted in the intensification of agriculture and industrialization resulting in generation of huge waste materials with potential to pollute the environment (Bhat et al. 2018). As a result, several researchers are looking into sustainable methods of managing these wastes to avoid environmental pollution (Pandey et al. 2018). Thermophilic composting, also known as traditional composting, and vermicomposting are two of the best-known processes for the biological degradation of solid organic wastes, with vermicomposting having been shown to be superior in terms of nutrients over traditional composting (Hussain et al. 2018). Though traditional composting has been well established at industrial level, its major drawbacks have been the long duration required to degrade materials as well as the loss of nitrogen through volatilization during the thermophilic stage (Mupambwa et al. 2016). However, vermicomposting has been shown to be superior in terms of nutrients over traditional composting mainly due to the presence of earthworms and their gut associated microbes which drive organic matter decomposition at a faster rate yielding higher nutrient content compost (Hussain et al. 2018). Though vermicomposts have been promoted as organic fertilizer sources, their concentration of macro nutrients such as N, P and K, has remained very low relative to those of inorganic fertilizers, thus limiting their commercial adoption in agriculture (Mupambwa and Mnkeni 2018).

Due to very low nutrient concentrations in vermicomposts, several researchers have looked into the possibilities of using amendments as a way of enhancing the nutrient concentrations and biodegradation in vermicomposts (Mupambwa and Mkeni 2018). Adhami et al. (2014) incorporated 6% raw rock phosphate and 2% modified rock phosphate (RP) into leaf compost

and sheep dung during composting with or without earthworms. Relative to the control with no rock phosphate amendment, the results of this study indicated that without earthworms, rock phosphate incorporation at both levels in leaf and sheep compost increased NaHCO_3 extractable phosphorus by 45%. However, in the presence of earthworms, this only resulted in a 10% increase in NaHCO_3 extractable P. In another study, using fly ash as a nutrient amendment with earthworm inclusion at stocking densities between 12.5 to 37.5 g- worms per kg of biomass, Mupambwa and Mnkeni (2016) reported an average 51% net increase in Olsen extractable phosphorus. Wang et al, (2017) also evaluated the influence of incorporating medical stone between 0% and 10% into pig manure during normal composting on biodegradation and humification. In this study, Wang et al. (2017) observed that incorporation of medical stone; which is composed of calcium oxalate and calcium phosphate; by up to 10% resulted in a 27% increase in hemicelluloses biodegradation whilst humification also increased by 10%. However, though several researchers have looked into the influence of amendments to improve compost quality, much of the work has used inorganic amendments during traditional composting and there is limited research on use of organic amendments.

Biochar is one such organic material that has been recently promoted as a nutrient amendment for vermicomposts (Doan et al. 2015). Biochar is a solid, carbon rich material obtained from plant or animal biomass through thermal conversion in the absence of oxygen, a process called pyrolysis (Jain et al. 2018). Gong et al. (2018) indicated that during vermicomposting of pre-composted green waste amended with bamboo biochar at 0%, 3%, and 6%, with the 6% resulting in the fastest maturity and highest vermicompost quality. Moreover, bamboo biochar addition increased the lignin degradation by up to 14 %, and also increased earthworm biomass, juvenile and cocoon numbers of *Eisenia fetida*, and enzyme activities (Gong et al. 2018).

Marinska et al. (2016) amended a mixture of sewage sludge and wheat straw with biochar made from willow woodchips reporting a 66% increase in earthworm reproduction at 8% biochar incorporation. Their study concluded that biochar amendment could allow for faster conversion of sewage sludge into vermicomposts. In another study Jain et al. (2018) evaluated the effects of woody biomass biochar on a mixture of aquatic weeds, cow dung and saw dust on composting efficiency, reporting advanced thermophilic temperatures and a 45% increase in total nitrogen. Contrary to other reports, Pakalchik et al. (2018) observed high earthworm mortality when biochar was applied at 5% to soil, indicating the need for further studies on biochar incorporation rates. Moreover, much of the work on biochar has used materials derived from terrestrial ligno-cellulosic feedstock with high total carbon and low nutrient content (Robert et al. 2015). In contrast, recent research has demonstrated that biochar produced from marine plants yields material with lower carbon and high nutrient content, though few studies have evaluated seaweed biochar in vermicomposting (Robert et al. 2015).

In arid countries like Namibia, terrestrial production is very limited though the marine environment has high productivity which results in generation of high seaweed biomass (Burke et al. 1995). In Namibia, the high productivity of the Atlantic Ocean powered by the Benguela upwelling system results in production of huge quantities of beach cast seaweeds, which reduce the aesthetic value of the coastal areas whilst costing money and time to clean up (Sakko 1998). Furthermore, though being a semi-arid arid country, Namibia generates huge quantities of animal and municipal waste in the form of goat manure and food waste, which have potential as organic matter and nutrient sources in agriculture. Within municipal wastes, food waste alone is not rich in nutrients thus combining it with goat manure and biochar can improve the nutrient concentration of the compost whilst reducing the concentration of heavy metals (Hussain et al. 2018). Though vermicomposting of goat manure and food waste has been done,

there is no work that looked at optimizing the combined vermicomposting of the food waste and goat manure amended with seaweed biochar. Against this background, the aim of the study was to optimize the biodegradation and nutrient release of food waste and goat manure vermicompost amended with seaweed biochar for development of effective organic nutrient sources.

MAIN OBJECTIVE

To optimize the biodegradation and nutrient release of food waste and goat manure vermicompost through the addition of seaweed biochar for development of effective organic nutrient sources.

SPECIFIC OBJECTIVES

1. To evaluate the influence of seaweed species and pyrolysis temperature on biochar chemical characteristics (pH, EC, Total C and N, Extractable NO₂, NO₃, NH₄, Olsen-extractable P, Heavy metals (Cd, Hg, Pb, Cr, Ni, Zn and As), Cations (Ca, Mg, Na, K).
2. To determine the optimum combination of goat manure and food waste to achieve a vermicompost with optimum biodegradation and high nutrient content.
3. To determine the optimum inclusion ratio of seaweed biochar into goat manure and food waste vermicompost for achieving effective biodegradation and nutrient mineralization.

Specific Hypothesis

- There are no significant differences among bio-chemical characteristics (pH, EC, Total C and N, Extractable NO₂, NO₃, NH₄, Olsen-extractable P, Heavy metals (Cd, Hg, Pb, Cr, Ni, Zn and As), Cations (Ca, Mg, Na, and K) obtained from biochar produced using different seaweed species collected from different sites under different temperatures.

- There are no significant differences among nutrient concentration in vermicompost produced using different combinations of goat manure and food waste.
- There are no significant differences between biodegradation among nutrient mineralisation in vermicompost produced from seaweed biochar, goat manure and food waste mixture.

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Vermicomposting technology

The increase in population, urbanization, industrialization and agricultural production results in the increase of organic wastes in the environment. These solid wastes contain valuable plant nutrients and organic matter which are essential for crop production and soil fertility (Yatoo et al. 2020). The application of these organic wastes to the soil may be a recycling alternative for these wastes. However, their direct land application may be harmful due to heavy metals, toxic organic compounds, and pathogenic microorganisms into the environment (Subbulakshmi and Bharathi 2020). In order to mitigate these problems, the solid waste should be converted effectively by composting and vermicomposting (Piya et.al 2018). Composting is a biological process that is facilitated by microorganisms to break down organic waste materials into a useable product. Thermophilic composting has been widely used to kill weed seeds through promotion of microbial activity and the high temperatures. Though composting has been widely used, the process takes a long-time and could result in nitrogen losses due to ammonification (Mawonga, 2016). In order to reduce these nitrogen losses, vermicomposting has been promoted as one of the processes that accelerate the breakdown of the wastes (Mupambwa and Mnkeni 2018). Vermicomposting is therefore defined as a bio-oxidative process in which earthworms interact with microorganisms within the decomposer community, thus accelerating the stabilization of organic matter and greatly modifying its physical and biochemical properties (Mupambwa and Mnkeni 2018). Although microorganisms are responsible for the biochemical degradation of organic matter, earthworms are crucial drivers of the process, by fragmenting and conditioning the substrate and dramatically altering its biological activity (Yangchan et al, 2019). According to the earthworms physiological,

morphological and behavioural characteristics, earthworms are classified into three main ecological categories: epigeic, anecic and endogeic species (Hoeffner et al, 2019; Mupambwa and Mnkeni 2018). More detailed description of the ecological categories is presented in Table 2.1. However, many studies have cited *Eisenia fetida* earthworms from the epigeic specie as the most preferred species for vermicomposting, because they tolerate harsh environmental conditions, high reproduction rates and their feed consumption rates are high (Saranraj and Stella 2012; Piya et al, 2018).

Table 2.1: Earthworm species used for vermicomposting.

Epigeic specie: <i>Lumbricus rubellus</i> , <i>Eisenia fetida</i> , <i>Eisenia andrei</i> , <i>Dendrobdena rubida</i> , <i>Eudrilus eugeniae</i> , <i>Perionyx excavatus</i> and <i>Eiseniella tetraedra</i> .	Litter dwellers; they live in organic horizons, in or near the surface litter and feed primarily on coarse particulate organic matter, ingesting large amounts of undecomposed litter. High feed consumption rates, high reproduction rates, and high tolerance to a wide range of environmental conditions.
Anecic specie: <i>Lumbricus terrestris</i> , <i>Aporrectodea trapezoides</i> and <i>Allolobophora longa</i>	They cast at the soil surface and emerge at night to feed primarily on surface litter, manure and other partially decomposed organic matter which they pull down into their burrows. Some anecic species may also create heaps of cast material termed at the burrow entrance, consisting of a mixture of cast, soil and partially-incorporated surface litter.
Endogeic specie: <i>Allolobophora caliginosa</i> , <i>A. rosea</i> and <i>Octoasion cyaneum</i>	Deep branching burrow systems which are filled with cast material as they move through the organic-mineral layer of the soil. No major importance in litter incorporation and decomposition.

They feed on subsurface material and are important in other soil formation processes, including root decomposition, soil mixing, and aeration.

Based on (Gupta et al. 2019), (Mupambwa and Mnkeni 2018)

Earthworms feed on organic wastes and are unique as they consume only a small portion from these wastes for their growth and excrete a major proportion of wastes in a partially-digested form (Joseph 2019). This is because the intestine of earthworms contains an array of micro-organisms, hydrolytic enzymes such as cellulase, amylase, lipase, protease, urease and chitinase, which help in rapid decomposition of partially digested materials (Nagavallema et al. 2004). The enzymes transform the complex organic matter into vermi-compost in a relatively smaller duration of 1–2 months as compared to traditional composting process which takes the longer duration of nearly 5 months (Sanchez-Monedero et al. 2001). The mechanism of action of digestion and formation of vermicompost by earthworms occurs in many steps. The organic matter passes through the gizzard of the earthworm where it is grounded into fine powder. Thereafter the hydrolytic enzymes, micro-organisms and other fermenting substances further help in their breakdown within the gut, that finally passes out in the form of casts. These are referred to as the vermicomposts (Munnoli et al. 2010).

2.2 Conditions of vermicomposting

2.2.1 C/N ratio

There are conditions that are required during vermicomposting process. The carbon to nitrogen (C/N) ratio is one of the important factors affecting the composting process as well as the properties of the end product. A C/N ratio between 25 and 30 is usually considered as the optimum ratio for vermicomposting (Mupambwa and Mnkeni 2018; Atalia et al, 2018). Different initial mix ratios will ensure different C/N ratios to obtain optimum mix ratio for composting. This is because the intestine of earthworms contains an array of micro-organisms, hydrolytic enzymes and hormones which help in rapid decomposition of partially digested materials thereby transforming the complex organic matter into vermi-compost in a relatively smaller duration of 1–2 months (Nagavallema et al. 2004) as compared to traditional

composting process which takes the longer duration of nearly 5 months (Sanchez-Monedero et al. 2001).

Microorganisms utilize the C as a source of energy and the N for building cell structure. Microorganisms utilize C and N at a ratio of about 30:1 (Atalia et al, 2018). High or low C/N ratios can be adjusted by adding high nitrogen or carbon rich wastes, respectively. Sawdust, wheat straw, grass clippings, dry leaves, etc. are examples of carbon-rich materials, whereas poultry manure, slaughterhouse waste, sewage sludge, etc. are nitrogen-rich. The C/N ratio decreases during the composting process as carbon is lost in the form of carbon dioxide. The temperature of the earthworm feed should be in the range of 20–35 °C along with relative humidity between 60–80% (Gupta et al, 2019).

2.2.2 Stocking density

It is reported that stocking density critically affects the rate of vermicomposting, though different stocking densities have been used for vermicomposting a stocking density of 25 g worms/kg was recorded to be appropriate for faster degradation and nutrient rich during fly ash-cow dung-waste paper mixture (Mupambwa and Mnkeni 2016). According to Ndegwa et al. (2000) a stoking density of 1.60 kg of worms and a feeding rate of 1.25 kg feed during paper waste mulch resulted in the highest bioconversion of the substrate.

2.2.3 Temperature

It is important to monitor the temperatures of the vermi reactors or bins used for vermicomposting as the materials used may heat up the vermi reactors as they decay and may kill the earthworms. The most commonly used *Eisenia fetida*, feed most rapidly at temperatures of 15-25 °C but they may also survive at 10 °C, however it is reported that temperatures above 30 °C may harm the earthworms (Yuvaraj and Mahendran 2020).

2.2.4 Moisture

Moisture is a critical factor for the survival of earthworms because it facilitates the absorption of oxygen. Moisture is important because it evaporates easily inside the body cavities of earthworms, especially in dry environments (Bosede et al. 2020)

For an effective vermicomposting, moisture content within a range of 50-70% must be maintained and moisture content less than 40% will result in slow composting. However, if the moisture content exceeds 70%, it may disrupt oxygen movement and create anaerobic conditions (Bagastyo et al. 2020). The moisture content ranging from 30.3% to 34.1% of food waste was recorded during the vermicomposting of food waste and corn cobs to produce a nutrient –rich fertilizer for organic farming (Boonna et al. 2020). The initial moisture content for the pre-composted of fresh and dried potato waste was 55 and 50% respectively, which maintained humidity between 50 and 57% during vermicomposting (Bosede et al. 2020).

2.3 Qualities of a mature vermicompost

Humification parameters such as humic and fulvic acid fractions are important for compost quality, because they are responsible for organic fertility functions in the soil and most resistant to microbial degradation (Bernal et al. 2009). Various indices were used by researchers for evaluation of humification level for the materials during composting and the established humification limit was: humification ratio ≥ 7.0 ; humification index ≥ 3.5 ; percent of humic acids ≥ 50 ; and polymerisation index ≥ 1.0 (Bernal et al. 2009). According to Sharma and Garg (2018) a humification index value less than five indicates a high level of organic material. Humification and humic acid content in vermicompost was found to be 28% higher than normal compost, the higher content in humic acid was because earthworms fragment the soil more rapidly. Hence, finer particles react and retain humic acid in a high amount. Campitelli et al. (2012) suggested that a ratio of polymerization index greater than 1.6 indicated a good

maturity degree achieved by the organic matter during rabbit manure vermicomposting. The germination index of > 50% has been reported as ideal index (Bernal et al 2009). Apart from germination index, there are parameters that are also important to measure such as phytotoxicity test which involves seed germination, relative seed germination and relative root elongation (Bernal et al 2009).

2.3.1 Organic matter content

Organic matter degradation in composting is totally a biological process, because different types of microorganisms degrade the organic matter (Bernal et al. 2017). The types of microorganisms responsible for organic matter degradation depend on the compost's temperature. Bacteria are active during the initial activation phase, fungi are active throughout the entire process, and *Actinomyces* sp. dominate the maturation phase (Bernal et al., 2009).

2.3.2 C/N Ratio

The carbon to nitrogen (C/N) ratio is important in composting because the microorganisms need a good balance of carbon and nitrogen ranging from 25:1 C/N ratio to 35:1 C/N ratio in order to remain active. High C/N ratios prolong composting duration with low C/N ratios that reduces nitrogen loss as indicated in Table 2.2 (Mupambwa and Mnkeni 2016). The C/N ratio can be regulated by selecting the most suitable combination of compost materials and added bulking agents to ensure a final ratio within the optimum range (Bernal et al. 2017). During the vermicomposting process of most materials, a decrease in C/N ratio is observed due to the release of organic matter content, however, compost is reported to be characterized as mature only when the C/N ratio is below 20 and nitrogen content is above 3% (Bernal et al. 2017).

2.3.4 Compost phytotoxicity

Phytotoxicity is a critical factor that has to be evaluated before applying compost into the soil. This is especially significant for composts derived from organic matter wastes as their high phenolic compound contents could reduce their quality (Bernal et al, 2017). There are various tests and indicators to be evaluated to avoid possible phytotoxicity effects of final composts. Among the others, the most commonly used test for phytotoxicity evaluation is the determination of the germination index, which can also be used as an indicator of compost maturity. Germination index values above 80% indicate the absence of phytotoxicity Gong et al. (2018).

Table 2.2: Nutrient release during vermicomposting of different organic materials.

Raw materials used	Experiment set up	Observation on nutrient release
Cow dung and paper waste mixed to achieve a C/N of 30:1	<p>Fly ash incorporated into the cow dung-waste paper at a ratio of 1:2 (w/w).</p> <p>Materials were vermicomposted for 7 days.</p> <p>Four treatment combinations based on with worms or not.</p> <p>Materials were composted for 10 weeks.</p>	<p>Earthworm and effective microorganisms showed the highest increase in Olsen P (43.8% more than the control) (Mupambwa et al. 2016)</p>
Chicken manure mixed with waste paper.	<p>Materials were pre-composted for 20 days.</p> <p>Six treatments of C/N ratio 20, 30,40,50,60 and 70 achieved by mixing chicken manure with shredded paper.</p>	<p>The total N, total P, and total K concentrations increased with time while total carbon, C/N ratio decreased. (Ravindran and Mkeni 2016),</p>

Materials were composted for 7 weeks.

Excess activated sludge on
vermicomposting of fruit and
vegetable wastes.

100 g (wet basis) bed material was added
10 adult earthworms with an individual weight of
350–500 mg were inoculated.

Nitrogen and phosphorus content
was improved in the final
vermicompost. (Ajibade et al. 2020)

The substrate compartments of six reactors, 100 g
(wet basis) of food waste were added.

, ,

2.4 Organic materials suitable for vermicompost

The application of chemical fertilizers in high quantities degrades soil quality in long run. Several researchers have reported positive changes in soil quality and soil productivity by application of vermicompost from various organic materials compared to chemical fertilizers (Piya et al, 2018). There is an increased interest in the potential of vermicomposts, which are products of a non-thermophilic biodegradation of organic materials through interactions between earthworms and microorganisms. Vermicomposts are used for plant growth media and soil amendments. Vermicomposts are finely divided peat-like materials with high porosity, aeration, drainage, water-holding capacity and microbial activity, which make them excellent soil amendments or conditioners as indicated in Table 2.3 (Arancon et al. 2004). Organic materials that are suitable for vermicomposting include vegetable waste, cow dung, poultry manure and sewage sludge among others as discussed in the next section.

2.4.1 Food waste

The huge quantities of food wastes generated are mostly found in urban areas and these need proper management. A study in which lawn waste and kitchen waste amended with buffalo dung through composting and vermicomposting over 3 months observed that at the final days of vermicomposting process, there was a decline in pH from 8.17 to 6.74, total organic carbon from 36.12 to 28.04) and C: N from 39 to 16), whereas the increase was observed in parameters like electrical conductivity from 1.19 to 3.22), *N* from 1.35% to 1.89%, *P* from 0.18% to 0.44%, *K* from 0.78% to 1.06% (Karwal et al. 2020). In the same study, it was reported that the concentration of toxic heavy metals (Co, Cd, Cu, As, Cr, and Pb) declined in the final vermicompost. The study also showed that buffalo dung, kitchen waste, and lawn waste (6:1:3 ratio) gave the best quality vermicompost Karwal et al. 2020).

2.4.2 Goat manure

Goat manure and cattle manure were used for culturing of the *Eisenia fetida* earthworm and it was reported that the concentrations of total C, P and K in goat manure vermicompost was higher than those in cattle manure vermicompost. It was also observed that cattle vermicompost had a higher N content than goat vermicompost but the C: N ratio of fresh manure was higher than that of vermicomposts for both materials (Loh et al. 2005). Other researchers used goat manure-based vermicompost in organic garlic production. In this study, they observed that goat manure vermicompost had very high total N (1.79%), very high available P (52 ppm), very high exchangeable K (1.75 mg/kg) and it was moderately alkaline (pH 7.73) (Gichaba et al. 2020).

2.4.3 Cattle dung

Cattle dung is one of the abandoned type of manure and have been evaluated in several studies as compost during vermicomposting. Cattle dung was mixed with garden waste for vermicomposting and it was observed that total nitrogen, total phosphorus, and total potassium contents and the electrical conductivity increased, while total organic carbon and the C/N ratio decreased in all the mixtures after vermicomposting Li et al. (2020). Other researchers used cattle dung vermicompost and they have observed an increased in plant nutrients with a minimum C: N ratio (from 22.13 to 14.38) and a maximum increase in nitrogen concentrations (1.77 to 29.15mg/kg) (Raza et al., 2020).

Poultry manure

Chicken manure contains high levels of nitrogen because of the protein feeds the chickens are eating and chicken vermicomposting need to be done because there is ammonia accumulating in their manure. Chicken manure vermicompost was compared with pine bark compost at 0, 25, 50, 75,100% media and used on tomato and cabbage seedlings. The results showed that

chicken manure vermicompost did not prove to be the most ideal medium and its incorporation should be limited to 50% for effective seedling emergence and early development of crops (Mupambwa et al., 2017).

Table 2.3: Quality of vermicompost prepared from different organic materials.

Substrate used	Earthworm species, stocking density and duration	Observation and research gaps
<ul style="list-style-type: none"> Rice straw and kitchen waste mixture 	<ul style="list-style-type: none"> <i>Eisenia fetida</i> 5 earthworms of average size 300 mg per incubator, for 45 days 	<ul style="list-style-type: none"> The vermicomposting resulted in a significant increase in phosphorus (31.38–55.89%) and potassium (33.40–63.15%). An increase was observed in total organic carbon (38.24–43.49%) and total nitrogen (9.01–32.52%). <p>Future studies may need to investigate the effect of earthworm growth on the indigenous microbial community in the soil.</p>
<ul style="list-style-type: none"> Vegetable waste and goat dung 	<ul style="list-style-type: none"> <i>Perionyx excavatus</i> 45 days 	<ul style="list-style-type: none"> Increase in NPK

-
- Poultry manure
 - *Perionyx excavatus*
 - 45 days
 - High ammonical nitrogen concentration, auto heating and high bulk density should be addressed while vermicomposting with poultry litter.
 - Poultry wastes contain significant amount of salts and ammonia that kill worms.
 - It is necessary to neutralize the fresh wastes with calcium carbonate.
-

Based on (Zhi-wei et al. 2019), (Joseph 2019), (Joshi et al. 2020).

2.5 Amendments for vermicompost

Due to very low nutrient concentrations in vermicomposts, several researchers such as Mupambwa and Mnkeni (2018) have looked into the possibilities of using amendments as a way of enhancing the nutrient concentrations and biodegradation in vermicompost. However, much of the work used inorganic amendments such as fly ash and rock phosphate during traditional composting and there is limited research on use of organic amendments.

2.5.1 Rock phosphate

Enrichment of vermicompost with rock phosphate has been found to increase the nutrient contents, humification parameters, efficiency of decomposition and phosphorus fractions in the vermicompost. Phosphate rock or rock phosphate is a term that describes any naturally occurring geological material that contains high concentrations of phosphate-bearing minerals with a phosphate content of 15 to 20% (Mupambwa and Mnkeni 2018). A study by Ajibade et al. (2020) reported that the addition of *Eisenia fetida* and *Pseudomonas fluorescense* during cow and pig manure-waste paper with rock phosphate enriched, showed that final vermicompost of pig manure treatments achieved greater maturity with a lower C/N ratio of 10.3, high humification index (HI) of 7.6%, and humification ratio (HR) of 12.89% in the treatment with *E. fetida* and *P. fluorescense*. In the same study, the addition of *E. fetida* alone in cow manure treatment resulted in the highest Olsen P of 2600 mg/kg, followed by the pig manure treatment with *E. fetida* only (2246.15 mg/kg) (Ajibade et al. 2020).

2.5.2 Fly ash

Fly ash is a product of coal combustion, which is captured through electrostatic precipitators at thermal power stations, and there are different types depending on the origin of the coal materials (Mupambwa and Mnkeni 2016). According to Lukashe et al. (2019) inoculation of fly ash–cow dung–waste paper vermicompost with phosphate solubilizing bacteria increased

the biodegradation process with a decrease in C/N ratio which resulted in a final C/N ratio of 11 compared to the control which had C/N ratio of 18. Inoculation with phosphate solubilizing bacteria resulted in improved availability of Olsen P which amounted to 48.3% more Olsen P relative to the control (Lukashe et al. 2019).

2.5.3 Biochar

Apart from inorganic materials that are being promoted as amendments, biochar is one such organic material that has recently been promoted as a nutrient amendment for vermicomposting (Doan et al. 2015). A study by Gong et al. (2018) indicated that during vermicomposting of pre-composted green waste amended with bamboo biochar at 0%, 3%, and 6%, with the 6% resulted in the fastest maturity and highest vermicompost quality. In another study biochar prepared from woody biomass of *Prosopis juliflora* was added into a mixture of *Hydrilla verticillata*, cow dung and sawdust having ratio of 8:1:1 (control), respectively. Addition of biochar resulted in advanced thermophilic temperatures (59 °C) and improved the physical properties of composting process. Addition of 5% biochar as a bulking agent in composting mixture, increased total nitrogen by 45% compared to the other trials, and air-filled porosity decreased by 39% and was found to be within recommended range from literature studies. Considering temperature, degradation rate and nitrogen transformation with an amendment of 5% biochar was recommended for *Hydrilla verticillata* composting (Jain et al. 2018). Liu et.al (2014) reported that the sewage sludge biochar can improve soil fertility and enhance plant growth while not increasing plant uptake of heavy metals, and remedied contaminated soil by reducing the plant availability of heavy metals.

In arid countries like Namibia, terrestrial production is very limited though the marine environment has high productivity which results in generation of high seaweed biomass (Burke et al. 1995). In Namibia, the high productivity of the Atlantic Ocean powered by the Benguela

upwelling system results in production of huge quantities of beach cast seaweeds, which reduce the aesthetic value of the coastal areas whilst costing money and time to clean up (Sakko 1998). Therefore, there is a need to evaluate the potential nutrient values of this beach cast seaweed for crop production.

2.6 Heavy metal changes during vermicomposting

Soil pollution from heavy metal contamination has increased to the point that it endangers human life in some areas; therefore, there is a need for the reduction of this pollution (Piya et al, 2018). Vermicomposting is one of the methods that has been promoted to clean up the soil from various pollutants, such as heavy metals, by using earthworms. During the vermicomposting process, the earthworms metabolize and excrete a mixture of soil and organic matter. Within the digestive system of these earthworms, the microorganisms transform organics such as: proteins, nucleic acids, fats and carbohydrates into more stable products known as vermicompost. Therefore, earthworms are able to clean up the soil from various pollutants, and are able to accumulate heavy metals in their tissues from the soil (Aleagha et al. 2011). According to Abu et al. (2015) heavy metal concentration through vermicomposting varies according to the feed used. Vermicomposting of four treatments Cow dung, Spent Mushroom Compost, Goat manure and Goat manure + spent mushroom compost spiked with 2 litres of landfill leachate each for 75 days resulted in major reduction of heavy metals. Chromium was removed at highest level ranging from 95-99.81%. Cadmium and Lead (Pb) were reduced by 90% and 80% in all treatments respectively (Piya et al, 2018). Meanwhile, Copper concentration increased in Cow dung and Zinc also showed an increase but only in Goat manure + Spent mushroom compost. Furthermore, the heavy metal concentration was found within the permissible compost limits given by different organizations. It has also been reported that vermicomposting reduces heavy metal concentration in higher amount than

normal composting and thus can be approached as an environmentally friendly method to reduce the toxicity from the soil. However, further studies should be done to evaluate the importance of the process (Piya et al, 2018).

2.7 Conclusion

The review has highlighted that, there is an opportunity to use alternative processes such as vermicomposting to transform and recycle organic waste materials into nutrient sources and there is need for optimizing this process. Improving the fertilizer value of food waste and goat manure with the incorporation of biochar from seaweed is important as the biochar has high essential macro and micro elements that can be used for plant growth. Studies that look at using waste materials such as goat manure and food waste can be important in the development of organic fertilizers.

CHAPTER THREE

3.0 THE POTENTIAL FERTILIZER VALUE OF NAMIBIAN BEACH-CAST SEAWEED (*LAMINARIA PALLIDA* AND *GRACILARIOPSIS FUNICULARIS*) BIOCHAR AS A NUTRIENT SOURCE IN ORGANIC AGRICULTURE

3.1 ABSTRACT

Biochar has been reported as an efficient tool for carbon sequestration and soil quality improvement. Most research in the past, evaluated the quality of biochar from terrestrial based materials. On the other hand, this study evaluated the macro; micro nutrients and heavy metal concentrations of biochar from seaweeds of the Namibian coast. Seaweed species *Laminaria pallida* and *Gracilariopsis funicularis* were pyrolysed at different temperatures i.e. control (uncharred); 200; 400; 600 and 800 °C in order to determine the chemical and physical properties of the biochar produced. There were significant interactions ($P \leq 0.05$) between temperature and seaweed species on most parameters measured. A pyrolysis temperature of 400 °C resulted in an almost 50% reduction in biochar yield, with higher temperatures resulting in even more yield loss whilst producing biochar with higher alkaline pH above 7 and high EC above 20 mS/cm. *G. funicularis* biochar resulted in the highest macro element with total C 38.3%; N 4.3% and P 6.3 g/kg contents, whilst *L. pallida* biochar had the highest contents of cations of 16.2 g/kg Ca; 6.4 g/kg Mg; 151 g/kg K and 45 g/kg Na. Higher pyrolysis temperatures did not result in consistent changes in total heavy metal content, with only Cd content being above the maximum permissible limits of 3.9 mg/kg for biochar. Generally, a pyrolysis temperature of 400 °C gave the best quality biochar in terms of macro elements, pH and total C. *G. funicularis* biochar showed significantly higher nutrient concentrations, therefore has great potential in soil quality improvement.

Keywords: heavy metals; marine biomass; Namibian coast; plant nutrients; Pyrolysis temperature; seaweeds.

3.2 INTRODUCTION

The increase in sea surface temperature, as a result of climate change is most likely to result in changes in marine populations. Warming sea surface temperatures, pollution, and other anthropogenic disturbances result in production of large quantities of seaweeds which end up being washed out onto the shores (Wade et al., 2020). These seaweeds have potential value as animal feed or as soil amendments in hyper-arid countries like Namibia. The turnover of terrestrial biomass in arid regions is generally low resulting in poor quality soils with low organic matter. However, Namibia, which is the most arid country in Southern Africa, is endowed with a highly productive marine environment which is powered by the Benguela upwelling system. This results in the production of huge quantities of seaweed biomass. These seaweeds tend to accumulate as beach cast on the Namibian coast line. However, there is very limited information being available on the actual quantities of this biomass produced (Critchley et al 1991). However, there are recent cases where several entrepreneurs are trying to exploit this marine resource for preparing animal feed. This beach cast seaweed biomass along the Namibian coast is mainly composed of seaweeds like *L. pallida*, *Ulva lactuca* and *G. funicularis* which reduce the aesthetic value of the beach whilst costing money to clean out (Iyer et al., 2005). Apart from using such algal biomass directly as soil amendment, one of the technologies that is gaining momentum due to its ability to sequester plant biomass carbon in the soil is the conversion of such biomass into biochar.

Biochar is currently being evaluated and promoted as a better soil amendment compared to other organic amendments like animal manure, to improve soil quality, reduce greenhouse gas emissions, whilst sequestering more plant-based carbon in the soil (Karbout et al., 2019). However, the physical and chemical properties of biochar depend on several factors which include pyrolysis temperature, pyrolysis time, pyrolysis method and the biomass feedstock material used (Li et al., 2017). If biochar is to be effectively used as a soil amendment, it is

critical to understand how the production processes influence the physical and chemical properties of the biochar such as volatile matter, fixed carbon, ash content and elemental composition (Hernandez et al., 2017). There are various biomass materials that have been evaluated for biochar preparation which include sugarcane bagasse, sewage sludge and cattle manure; among others, all having different physico-chemical properties (Figueredo et al., 2017; Munoz et al., 2019).

Munoz et al (2019) reported that biochar from cattle and pig manures increased soil carbon content, favouring the persistence of carbon in the soil under an incubation study. Figueredo et al (2017) used different organic materials like sewage sludge and sugar cane bagasse and concluded that, biochar produced at higher temperatures had lower yield as compared to the one produced at lower temperatures. In one of the first studies to prepare biochar from marine resources, Roberts et al (2015) evaluated the various physico-chemical properties of red and brown seaweed species which had been commercially cultivated and reported that the red seaweed species contained higher potassium, sulphur and had lower carbon and pH. In a review, Zacharia et al (2015) also indicated that when seaweeds are converted into biochar, the biochar yield is high and the content of macro-elements such as N, P and K is also high.

The background presented informs our current study and points to the importance of marine biomass in biochar production, though the studies highlighted focused on artificially cultured seaweed biomass relative to that which exists in the natural environment. Based on available literature, there is no research that has evaluated the potential nutrient value of biochar prepared from the natural and nutrient rich Southern African Benguela powered marine environment. Apart from focusing on the beneficial characteristics of biochar in soil, it is also important to evaluate the concentration of potentially toxic elements such as Ni; Zn; Cr; Cd and Pb in the biochar before applying it to the soil. It is thus of paramount importance to evaluate the

environmental safety associated with the soil application of biochar derived from marine environments, where limited research is available on this. Therefore, the objectives of the study were to evaluate the (i) biochar yield, pH, EC; (ii) potentially beneficial extractable nutrients (C, N, P, K, Mg, Ca, Na); (iii) potentially toxic heavy metals (Cd, Pb, Cr, As, Zn, Ni) in beach-cast sea weed (*Gracilariopsis funicularis*, *Laminaria pallida*) biochar prepared under different pyrolysis temperatures.

3.3 MATERIALS AND METHODS

3.3.1 Source of materials

The experiment was conducted at the Sam Nujoma Campus of the University of Namibia, located in Henties Bay in the Erongo Region of Namibia. Two species of seaweed; i.e. *L. pallida* and *G. funicularis* were collected from the Henties Bay and Walvis Bay coast as indicated in the map in Figure 3.1 during low tides. The full description of these seaweeds can be obtained from Iyer et al (2005) and Branch et al (2010).

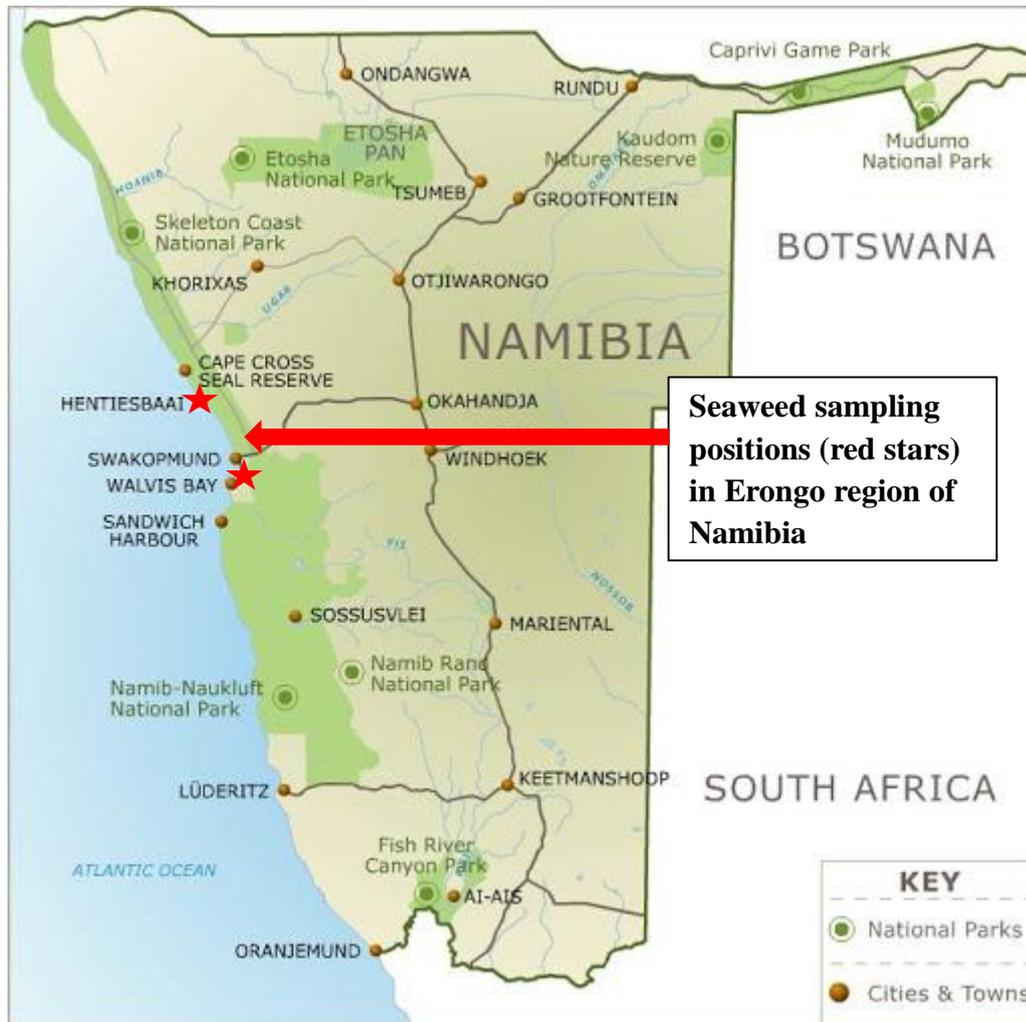


Figure 3.1 Seaweed sampling positions along the Henties Bay and Walvis Bay coastline

3.3.2 Pre-treatment of the materials

The collected seaweeds were rinsed with fresh water thoroughly to get rid of foreign objects and sand. The washed seaweed species were then air-dried and then mechanically ground using the industrial grinder (TRAPP-TRF- 400 manufactured in Brazil) with a 25 mm mesh size. The full pictorial description of the seaweed harvesting and processing is shown in y Figure 3.2 The ground samples were then oven dried at 60 degrees Celsius for 48 hours, to allow them to have constant moisture content, before being used for biochar preparation.

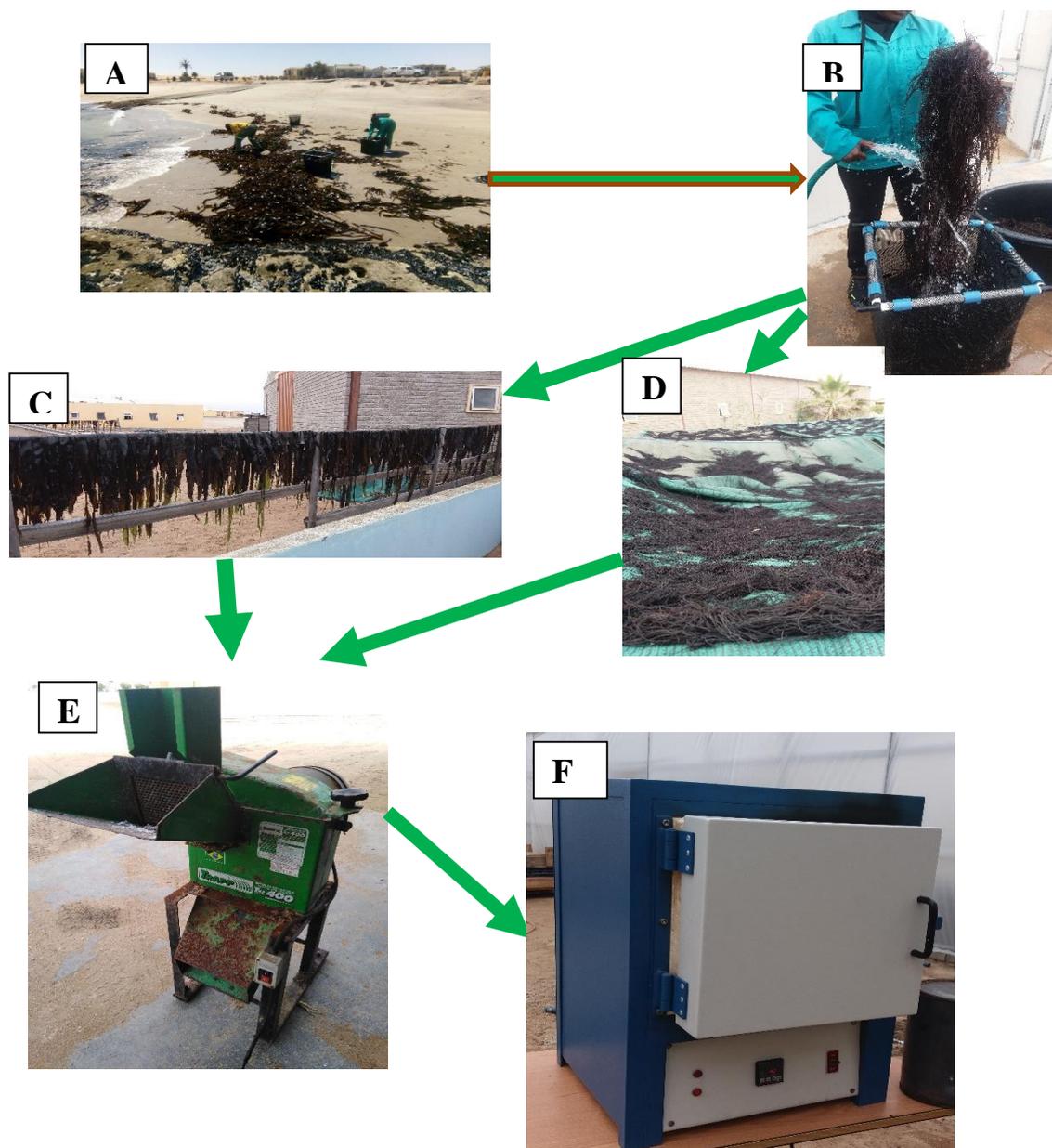


Figure 3.2: A schematic diagram showing the sequential processes involved in seaweed biochar preparation. **A:** seaweed harvesting from Langstrand in Namibia’s coast; **B:** washing of the seaweeds using fresh tap water; **C** and **D:** air drying of the seaweeds at Sam Nujoma Campus of UNAM; **E:** mechanical grinder used for grinding seaweeds; **F:** muffle furnace used to prepare the biochar at different temperatures.

3.3.3 Treatments and experimental design

A factorial experiment with two factors; i.e. the two seaweed species i.e. *Laminaria pallida* and *Gracilariopsis funicularis* with five temperature levels, namely, control (room temperature), 200, 400, 600 and 800 degrees Celsius was established. This gave a 2×5 factorial design which was laid in a completely randomized design with three replications.

3.3.4 Biochar preparation

The biochar that was used in this study was produced from the seaweeds through slow pyrolysis using a muffle furnace with a digital temperature controller. To achieve a uniform volume, for *Gracilariopsis funicularis* an amount of 300g was used and for *Laminaria pallida* a 100g quantity of seaweed was weighed into a sterile metal cylindrical container with a lid and this was then placed into a muffle furnace set at the prescribed temperatures as described by the treatments and pyrolysed for 1 hour. The two types of biochar produced were further ground into fine powder using mortar and pestle. The biochar samples were then subjected to different soil analyses as seen below.

3.3.5 pH and Electrical conductivity (EC)

Electrical conductivity and pH of the biochar were measured in water using a multi-meter fitted with a glass electrode (Lovibond Water Testing, Senso Direct 150). The pH was measured in a 1:10 (w/v) in deionized water with a multi – meter with the ratio being recommended for organic materials (Hendershot and Lalonde 2006). This ratio allows for pH and EC measurement as materials containing high amounts of organic matter like biochar tend to form a thick dry paste when the ratio is kept the same as for mineral samples. Therefore, a wider ratio of sample to water was used 1:10 (w: v). An amount of 5 g of biochar sample was measured and placed in a 100 mL plastic container and 50 mL of water was added and then

shaken with a horizontally reciprocating shaker for 30 minutes. After shaking, the pH and EC were determined from the suspension using a multi-meter (AgriLASA 2004).

3.3.6 Total Carbon and Nitrogen

Total C and N were determined with the dry combustion method using a LECO CHN628 auto analyzer (LECO, USA). An amount of 0.1 g of soil (sieved through a 0.5 mm mesh sieve) was measured into an aluminium foil and then was placed into the LECO for automatic total C and N determination.

3.3.7 Total Phosphorus

To determine the total phosphorus concentration in the biochar samples, the samples were initially digested using the wet digestion method which involves perchloric acid (65%) and nitric acid (60%) as described by AgriLASA (2004). An amount of 0.5 g biochar samples were weighed into 50 mL digestion tubes. 10 mL of nitric acid was then added to the digestion tubes and was allowed to boil gently at 150°C for 30 minutes. After 30 minutes, 4 mL of perchloric acid was added and allowed to boil for 30 minutes at 150°C. After the digestion, the digestion tubes were removed and left to cool and the mixture was diluted with deionized water to 100mL (AgriLASA 2004). The diluted samples were then filtered using Whatman number 2 filter paper into 100 mL volumetric flasks and filled up to the mark with de-ionized water.

After the digestion process, the samples were then stored for analysis to measure phosphorus by using the ascorbic acid method (Kuo 1996). An aliquot of biochar sample or P standard solution that contains 2 mL of P was transferred into a 50 mL volumetric flask and was diluted with deionized water to 25 mL. A mixed reagent of 8 mL was then added to mix well. The solution was then diluted to volume and after 10 minutes, the absorbance of P at 880 nm was measured and a blank that contains all reagents was then prepared except the P solution.

3.3.8 Extractable Phosphorus

To extract the phosphorus in the biochar samples, the Olsen method was used (Schoenau and Halloran 2006). This method involved the use of a solution of 0.5 mol Sodium bicarbonate, adjusted to pH 8.5 using. 2.5 g of biochar was placed in a 100 mL extraction bottle to which 50 mL of 0.5 M Sodium bicarbonate solution was added. The solution was then shaken for 30 minutes on a reciprocal shaker at 180 rpm. The extracts were then filtered using Whatman No. 2 filter paper. The extracts were then analyzed for P using the Ascorbic acid method as described by Kuo (1996). Both total P and extractable P were determined because total P is the total amount of P in the seaweed biochar and the extractable P represents the soil bioavailable P that can be absorbed by plants from the seaweed biochar.

3.3.9 Extractable Ammonium, Nitrate and Nitrite

Extractable inorganic nitrogen fractions ($\text{NH}_4\text{-N}$ and $\text{NO}_2/\text{NO}_3\text{-N}$) were extracted using 0.5 M potassium sulphate (Okalebo et al., 2002). 5 g of biochar samples were shaken for an hour on a mechanical reciprocal shaker at 180 rpm and filtered through Whatman No.2 filter paper for colorimetric analysis using a UV spectrophotometer (Thermo Scientific, Genesys 105 UV - Vis). The preparation of samples for $\text{NH}_4\text{-N}$ colorimetric determination involved transfer of 0.2 mL of sample extract into a test tube with 5 mL of N1 reagent (prepared by mixing specific quantities of sodium salicylate, sodium citrate, sodium tartrate and sodium nitroprusside) followed by 5 mL of N2 reagent (prepared by mixing specific quantities of sodium hydroxide and sodium hypochlorite). The samples with reagent mixtures were allowed to stand for an hour and the resulting blue colour intensity was measured at 655 nm wavelength using a UV spectrophotometer (Thermo Scientific, Genesys 105UV -Vis) that was calibrated using standard range of 0 – 25 $\mu\text{g NH}_4^+\text{-N / mL}$ treated in the same manner as the samples. Colorimetric sample preparation for $\text{NO}_2/\text{NO}_3\text{-N}$ was done by transferring 1.0 mL 5% salicylic acid to 0.5 mL sample extract in a test tube followed by 10 mL 4 M NaOH. After an hour of

standing the resulting yellow colour intensity was measured at 419 nm wavelength using a UV spectrophotometer (Thermo Scientific, Genesys 105UV -Vis) that was calibrated using standards range of 0 – 10 µg NO₃-N/mL treated in the same manner as the samples.

3.3.10 Total heavy metals

To measure the total concentrations of Cd, Pb, Cr, Zn, Ni and As, the biochar samples were digested using the wet digestion method which involves perchloric acid and nitric acid (AgriLASA 2004). Total concentration of heavy metals in the extracts was determined using inductively coupled plasma - optical emission spectrometer (ICP-OES) (iCAP 6000 SERIES).

3.3.11 Extractable cations (Ca, Mg, Na, K)

Cations were extracted using the ammonium acetate method (AgriLASA 2004). A solution of 1 Mol ammonium acetate adjusted to pH 7 using 1M NaOH, was used to extract the cations. 5 g of biochar was placed in 100 cm³ extraction bottles and 50 cm³ of ammonium acetate solution was added into the extraction bottle and the mixture was shaken horizontally on a reciprocating shaker at 180 rpm for 30 minutes. The extracts were filtered using Whatman No. 2 filter paper and the cation concentrations in the solution were determined using the ICP-OES (iCAP 6000 SERIES).

3.3.12 Statistical Analysis

Analysis of variance (ANOVA) for a factorial experiment was done using JMP version 14.0.0 Statistical software (SAS Institute, Inc., Cary, North Carolina, USA, 2010). Microsoft Excel was used for plotting of all graphs. All data collected met the ANOVA assumptions which are: the samples were normally distributed, independent and had common variance. The level of significance used was 5%.

3.4 RESULTS

3.4.1 Effect of pyrolysis temperature on biochar yield from two seaweed species.

There was a significant effect of the two-way interaction between pyrolysis temperature and seaweed species (Table 3.1) on biochar yield, implying that the effect of pyrolysis temperature on biochar yield was not consistent when averaged across the effect of species. Apart from the control, the *L. pallida* species had the highest biochar yield of 92.6 % \pm 1.08 at a pyrolysis temperature of 200 °C whilst *G. funicularis* also had the highest yield 82.3 % \pm 1.08 at 200 °C. Both species *G. funicularis* and *L. pallida* had the lowest biochar yield of 36 % \pm 1.08 and 41.2 % \pm 1.08 at 800 °C, respectively. It was observed that at a pyrolysis temperature of 400 °C, biochar yield decreased in both seaweed species by almost 50 % (Figure 3.2). Relative to the control with uncharred seaweeds, the increase in pyrolysis temperature from 200; 400; 600 and 800 °C resulted in a decrease in biochar yield by average of 18%, 49%, 54% and 62%, respectively. On average, across all pyrolysis temperatures, *L. pallida* resulted in 22.1% significantly higher ($P \leq 0.05$) biochar yield compared to *G. funicularis*.

Table 3. 1 Repeated measures ANOVA for changes in several parameters following 12 weeks of food waste- goat manure vermicomposting.

Parameter	Treatment		Time (weeks)		Treatments × Time	
	<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>
EC (mS/cm)	699.5	< .0001	63.32	< .0001	7.14	0.0007
pH	63.5	< .0001	46.46	< .0001	4.70	0.0051
C/N	4.64	0.0525	57.62	< .0001	2.96	ns
Extractable ammonium (g/kg)	101.85	< .0001	94.50	< .0001	3.5	0.0106
Extractable Nitrate & Nitrite (g/kg)	0.39	ns	15.91	< .0001	2.01	ns
Extractable Olsen P (g/kg)	533.2	< .0001	392.9	< .0001	15.22	< .0001
Extractable Ca (g/kg)	74.46	< .0001	17.81	0.0027	4.98	0.0308
Extractable K (g/kg)	4.623	< .0529	13.42	0.0080	5.48	0.0305
Extractable Mg (g/kg)	71.33	< .0001	59.73	< .0001	3.35	0.0524
Extractable Na (g/kg)	17.77	0.0022	28.36	< .0001	4.61	0.0110

ns = not significant at $P \leq 0.05$; *F* = *F*- test ; *P* = Probability

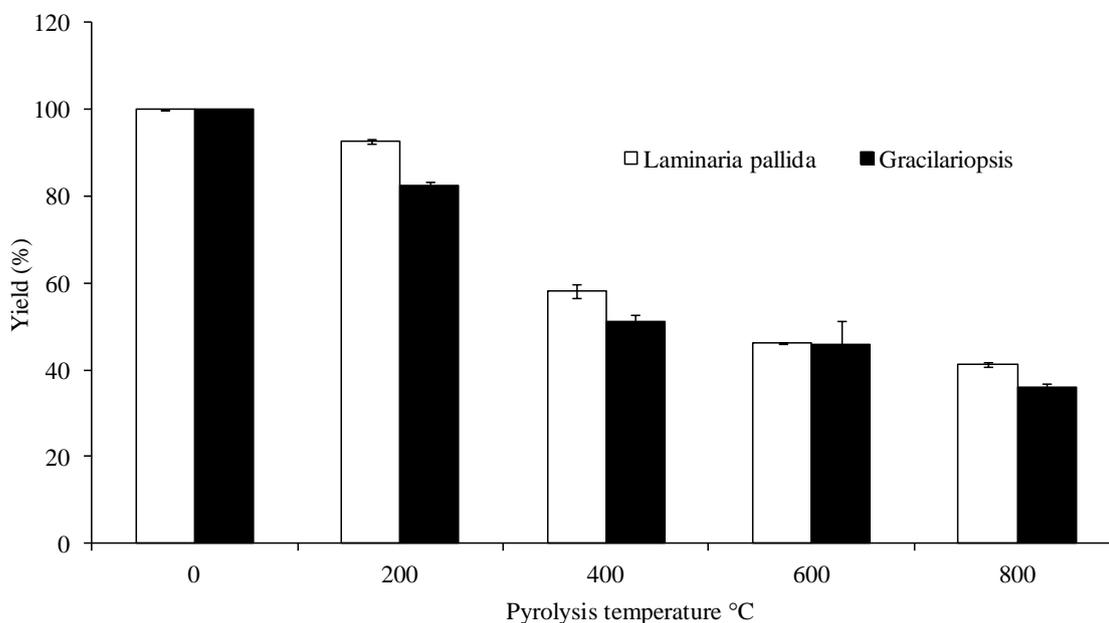


Figure 3.2 Biochar yield prepared using *L. pallida* and *G. funicularis* seaweeds at different temperatures. Error bars indicate standard deviation.

3.2 Effect of pyrolysis temperature on pH and EC of biochar from two seaweeds species

There was a significant effect of the two-way interaction between pyrolysis temperature and seaweed species on pH and EC (Table 3.1). *L. pallida* biochar had the highest pH of 10.7 ± 0.196 at a pyrolysis temperature of 800 °C as compared to *G. funicularis* biochar which had a pH of 9.7 ± 0.196 at the same temperature (Figure 3.3 A). The biochar from *G. funicularis* had the lowest pH across all pyrolysis temperatures with an average pH of 7.03 ± 0.09 whilst the biochar from *L. pallida* had an average pH of 8.4 ± 0.09 representing a 19.3% difference. At a pyrolysis temperature of 400 °C, *G. funicularis* biochar pH was observed to be close to neutral with the pH value of 6.8 ± 0.196 whilst below this temperature it was acidic and above this temperature it was alkaline.

Of the two seaweed species, biochar from *L. pallida* had significantly the highest EC ($P \leq 0.05$) of $110.2 \text{ mS/cm} \pm 3.52$ at a pyrolysis temperature of $800 \text{ }^\circ\text{C}$ whilst *G. funicularis* biochar had EC of $85.2 \text{ mS/cm} \pm 3.52$ (Figure 3.3 B). On average, across all pyrolysis temperatures, *G. funicularis* biochar had an average EC of $43.8 \text{ mS/cm} \pm 1.57$, whilst *L. pallida* biochar had an average EC of $64.9 \text{ mS/cm} \pm 1.57$. *G. funicularis* biochar had the lowest EC which was observed at $400 \text{ }^\circ\text{C}$ with an EC of $24.3 \text{ mS/cm} \pm 3.52$. However, this was not the case for biochar from *L. pallida* where the lowest EC ($37.36 \text{ mS/cm} \pm 3.52$) was at the control.

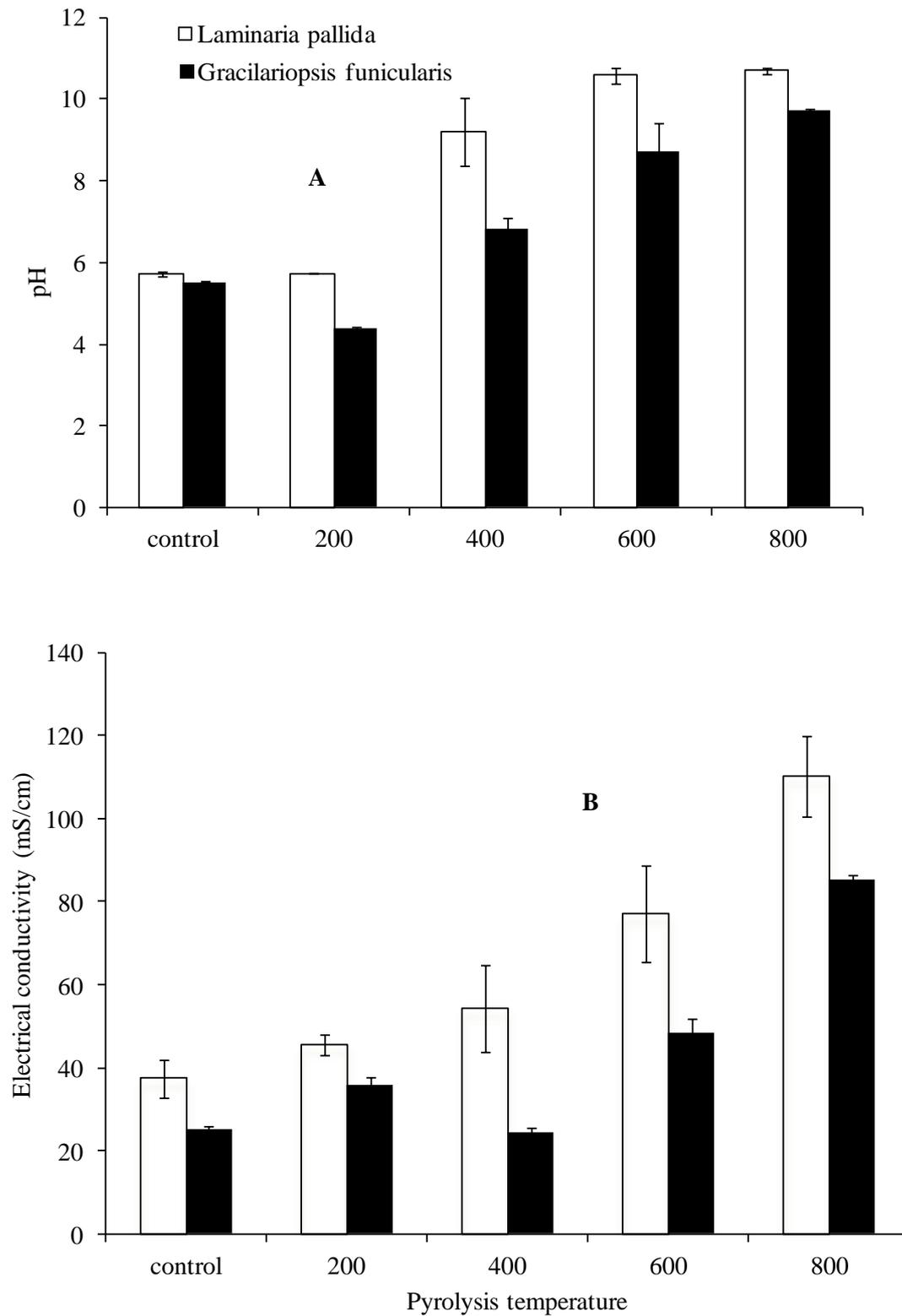


Figure 3.3 Changes in pH (A) and electrical conductivity (B) in biochar samples prepared using *L.pallida* and *G. funicularis* seaweeds at different temperatures. Error bars indicate standard deviation.

3.3 Effect of pyrolysis temperature on total and extractable P and N of biochar from two seaweeds species

There was a significant effect of the two-way interaction between pyrolysis temperature and seaweed species on total and extractable P and N (Table 3.1). The highest total P in the biochar samples was $6.33 \text{ g/kg} \pm 0.14$ from *L. pallida* at a pyrolysis temperature of $600 \text{ }^\circ\text{C}$, whilst at the same temperature, biochar from *G. funicularis* had a total P concentration of $6.29 \text{ g/kg} \pm 0.14$ (Figure 3.4 A). The lowest total P in biochar samples was obtained from *L. pallida* at the control with $2.73 \text{ g/kg} \pm 0.14$ total P which was the same for the biochar from *G. funicularis* with a total P of $3.38 \text{ g/kg} \pm 0.14$ at the same temperature. There was a significant effect of the two-way interaction between pyrolysis temperature and seaweed species on Olsen extractable P. Biochar from *G. funicularis* had the highest concentration of Olsen extractable P which was observed at the control with $1.2 \text{ g/kg} \pm 0.02$ whilst the lowest concentration of Olsen P was in *L. pallida* biochar at $600 \text{ }^\circ\text{C}$ with a concentration of $0.14 \text{ g/kg} \pm 0.02$ (Figure 3.4 B). On average across all temperatures, for *G. funicularis*, P concentration of extractable fraction represented 18.08 % of the total concentration, whilst for *L. pallida* the extractable P concentration was 13.62 % of total P concentration.

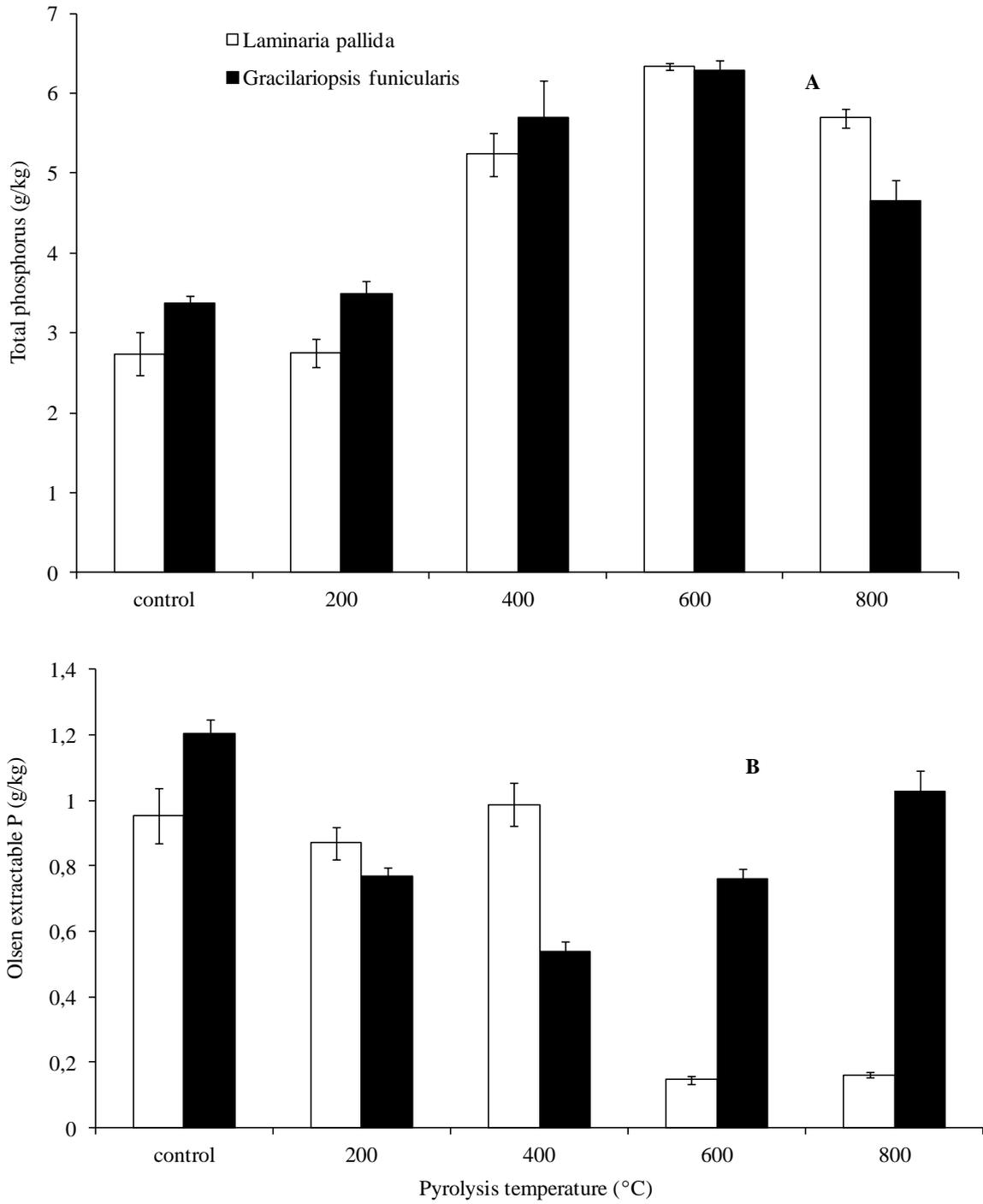


Figure 3.4 Total phosphorus (A) and Olsen extractable P (B) in biochar samples prepared using *L. pallida* and *G. funicularis* seaweeds at different temperatures. Error bars indicate standard deviation.

There was a significant effect of the two-way interaction between pyrolysis temperature and seaweed species on total N (table 3.1). For biochar from both the seaweed species, the highest total N content was obtained at a pyrolysis temperature of 400 °C which was 4.37 % \pm 0.04 from *G. funicularis* biochar and 2.15 % \pm 0.04 from *L. pallida* biochar. For all the seaweed species, the lowest concentration of total N in biochar samples was observed at 800 °C, with *L. pallida* biochar having a concentration of 1.25 % \pm 0.04 whilst *G. funicularis* biochar had 1.86% \pm 0.04.

There was a significant effect of the two-way interaction between pyrolysis temperature and seaweed species on extractable NO₂/NO₃ as indicated in Table 3.1. Biochar from *G. funicularis* had the highest concentration of extractable NO₂/NO₃ of 1.23 g/kg \pm 0.03 at a pyrolysis temperature of 200 °C, whilst extractable concentration of NO₂/NO₃ biochar from *L. pallida* was the highest at a pyrolysis temperature of 400 °C with 0.81 g/kg \pm 0.03. The lowest extractable concentration of NO₂/NO₃ was the same for the two seaweed species with 0.01 g/kg \pm 0.03 at 400 and 600 °C though this was at different temperatures.

There was a significant effect of the two-way interaction between pyrolysis temperature and seaweed species on extractable ammonium as indicated in the ANOVA table (Table 3.1). The highest concentration of extractable ammonium in biochar among both seaweed species was 0.99 g/kg \pm 0.02 from *L. pallida* at the control (Figure 3.4 B). However, for biochar from *G. funicularis* the highest extractable concentration of ammonium was observed at 400 °C pyrolysis temperature with 0.39 g/kg \pm 0.02. It was noteworthy that at 800 °C, there was no nitrate, nitrite and ammonium detected in biochar samples from both seaweeds (Figure 3.5 B and C).

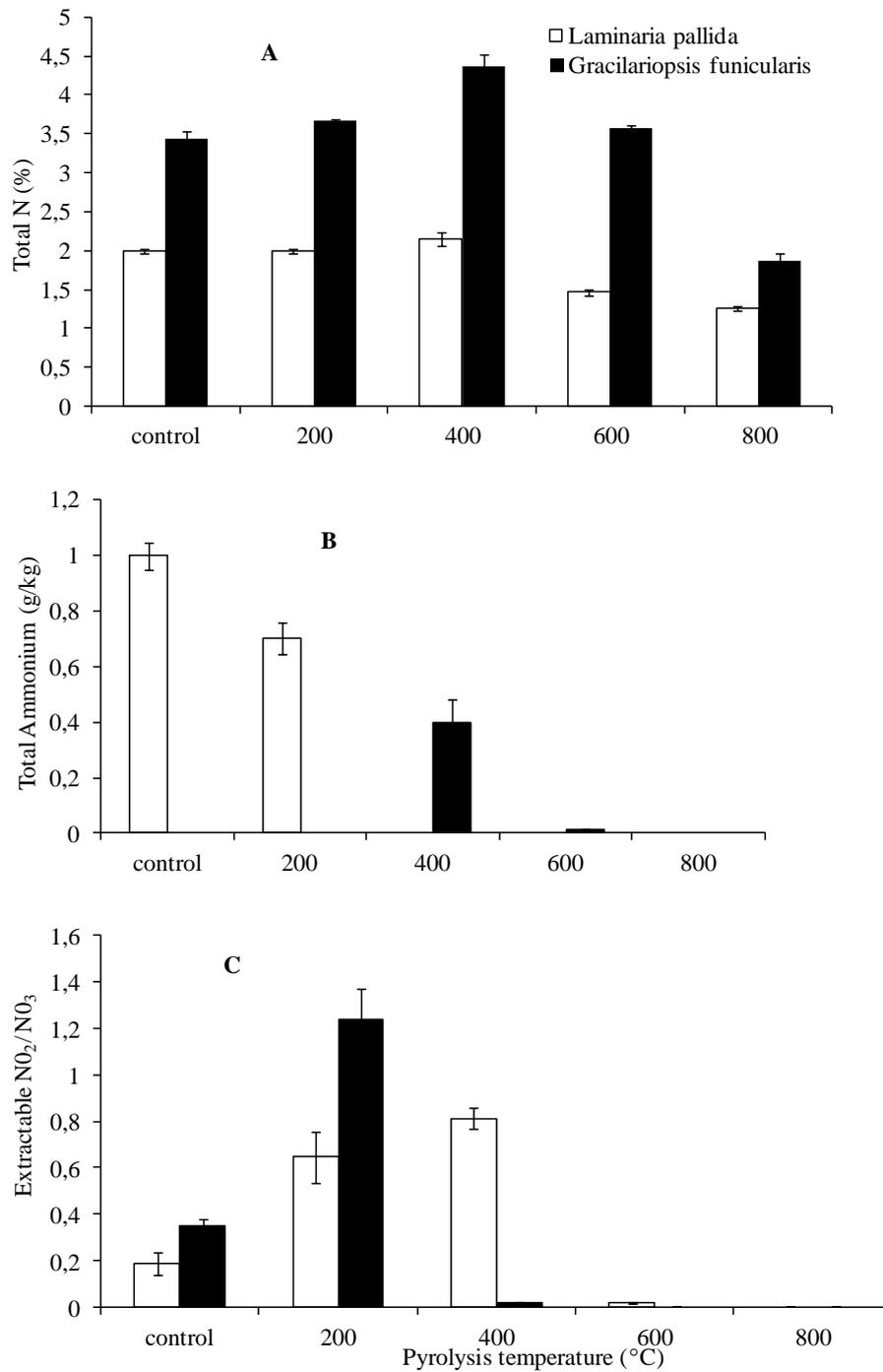


Figure 3.5 Total nitrogen (A); extractable ammonium (B); extractable nitrate/nitrite (C) in biochar samples prepared using *L.pallida* and *G. funicularis* seaweeds at different temperatures. Error bars indicate standard deviation.

3.4 Effect of pyrolysis temperature on total C of biochar from two seaweed species

There was a significant effect of the two-way interaction between pyrolysis temperature and seaweed species on total C (Table 3.1). It was interesting to observe that at a pyrolysis temperature of 400 °C biochar samples of *G. funicularis* and *L. pallida* resulted in the highest total C of 38.3% ± 0.31 and 26.1% ± 0.31, respectively. The lowest total C was observed at a pyrolysis temperature of 800 °C in biochar from both seaweed species (Figure 3.6). Across all pyrolysis temperatures, biochar from *G. funicularis* recorded the highest average total C of 33.3 % ± 0.14 whilst *L. pallida* biochar had an average of 22.9 % ± 0.14 representing 31.2 % difference of total C between *G. funicularis* and *L. pallida*.

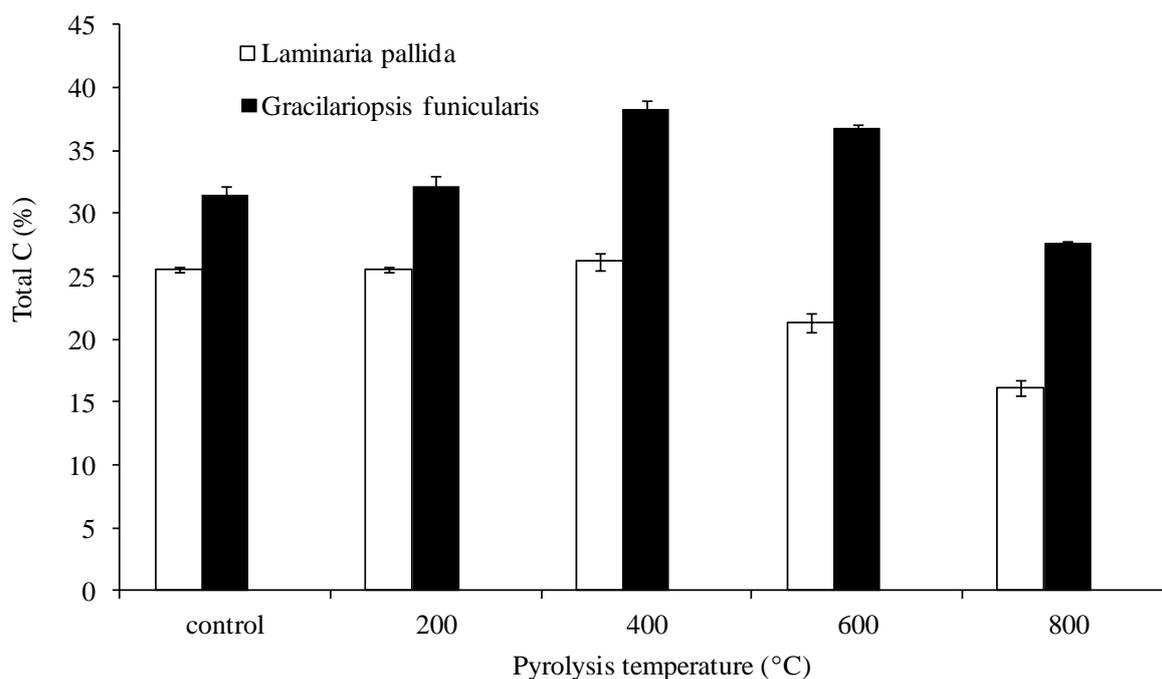


Figure 3.6 Total C in biochar samples prepared using *L.pallida* and *G. funicularis* seaweeds at different temperatures. Error bars indicate standard deviation.

3.5 Effect of pyrolysis temperature on extractable cations of biochar from two seaweed species.

There was a significant effect of the two-way interaction between pyrolysis temperature and seaweed species on extractable cations (Ca, Na, Mg, K) from the biochar produced from the two seaweed species (Table 3.1). A trend was observed where the concentration of extractable Ca increased from the control to 400 °C in biochar from both seaweed species and thereafter decreased (Figure 3.7 A). *L. pallida* biochar had the highest extractable Ca concentration of 7.8 g/kg \pm 0.16 at 400 °C with the lowest extractable Ca of 0.7 g/kg \pm 0.16 at 800 °C. On another note, the *G. funicularis* extractable Ca content ranged from 1.1 \pm 0.16 to 2.0 g/kg \pm 0.16, with the highest content at 200 °C.

Similar to extractable Ca, *G. funicularis* biochar had the lowest extractable Na concentration across all pyrolysis temperatures, with the highest extractable Na content of 16.7 g/kg \pm 0.42 being recorded at a pyrolysis temperature of 800 °C whilst the lowest biochar extractable Na was at a pyrolysis temperature of 400 °C with 7.85 g/kg \pm 0.42 (Figure 3.7 B). It was interesting to note that there was a consistent trend observed where, as the temperature increased, the Na content also increased. For *L. pallida*, the Na content ranged from 20.17 g/kg \pm 0.42 to 38.7 g/kg \pm 0.42 between the control and 800 °C. On average, extractable K increased with an increase in pyrolysis temperature (Figure 3.7 C). *L. pallida* biochar had the highest extractable concentration of K from control to 600 °C pyrolysis temperature. However, at a pyrolysis temperature of 800 °C *G. funicularis* biochar had the highest K content of 185 g/kg \pm 3.2 compared to *L. pallida* with 174.9 g/kg \pm 3.2. The highest extractable Mg (5.9 g/kg \pm 0.1) was observed in the biochar samples from *L. pallida* at 800 °C pyrolysis, whilst the samples from *G. funicularis* had the highest extractable Mg concentration in the control with a content of 1.8 g/kg \pm 0.1 (Figure 3.7 D). It was interesting to observe that the lowest extractable Mg was recorded at the same pyrolysis temperature of 600°C in biochar from both seaweed species,

with a concentration of $2.4 \text{ g/kg} \pm 0.1$ for *L. pallida* and $0.2 \text{ g/kg} \pm 0.1$ for *G. funicularis* biochar.

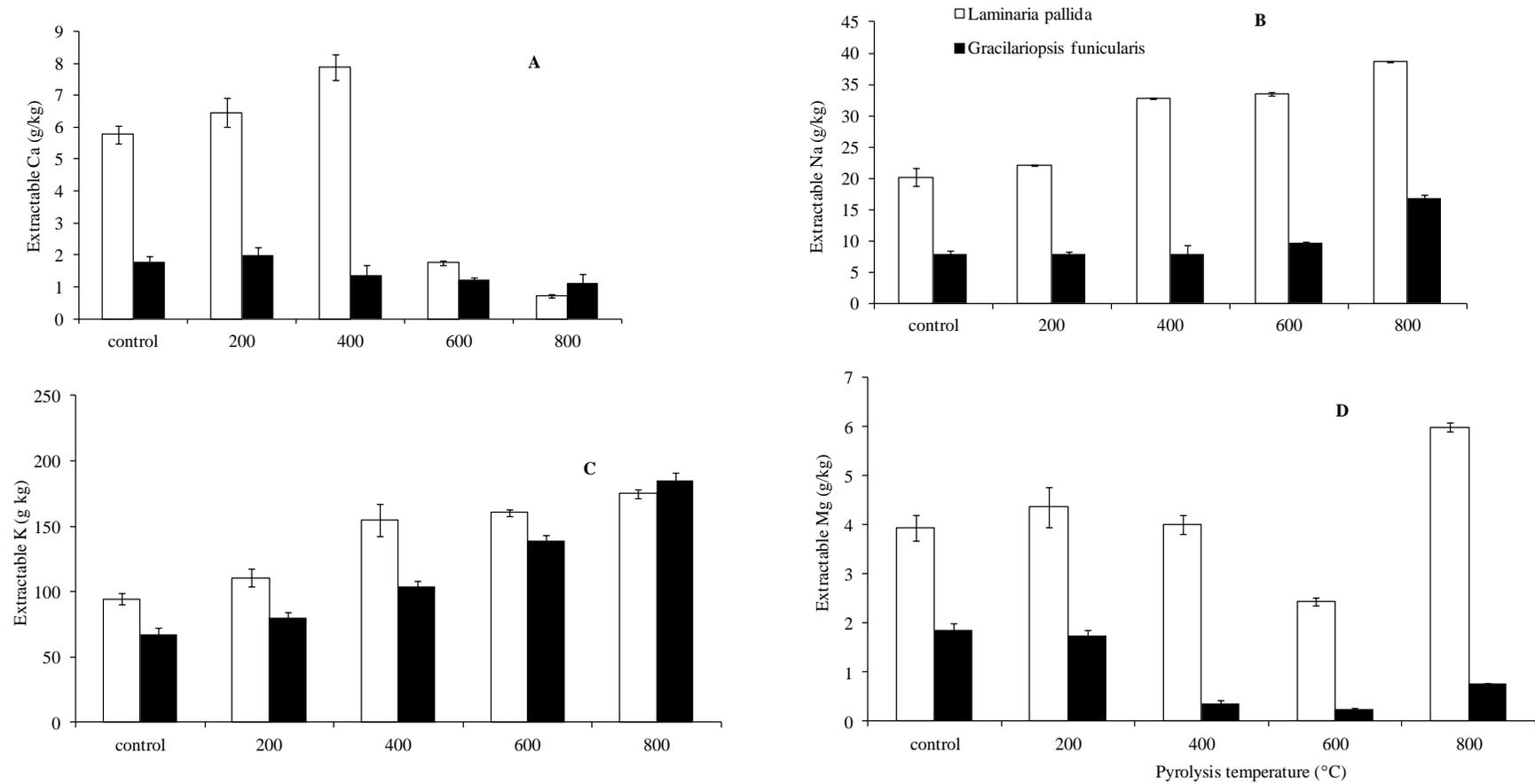


Figure 3.7 Extractable Ca (A), Extractable Na (B), Extractable K (C) and Extractable Mg (D) in biochar samples prepared using *L. pallida* and *G. funicularis* seaweeds at different temperatures. Error bars indicate standard deviation.

3.6 Effect of pyrolysis temperature on total heavy metals of biochar from two seaweed species

There was a significant two-way interaction ($P \leq 0.05$) between pyrolysis temperature and seaweed species differences on total Cd, As, Ni, Cr, Pb and Zn heavy metals as indicated in the ANOVA Table 3.1). Across all pyrolysis temperatures, the biochar samples from *G. funicularis* were observed with the highest total Cd content of $3.5 \text{ mg/kg} \pm 0.04$ while *L. pallida* had an average total of $0.4 \text{ mg/kg} \pm 0.04$ (Figure 3.7 A). For total Cd, *L. pallida* biochar resulted in a constant total Cd content of almost $0.5 \text{ mg/kg} \pm 0.09$ from control to 400°C pyrolysis temperature and thereafter it significantly decreased at 600°C and increased at 800°C pyrolysis temperatures. A similar trend was observed with biochar from *G. funicularis*, whereby the total Cd content was almost constant with a content of $4.5 \text{ mg/kg} \pm 0.09$ from the control to 200°C pyrolysis temperature and thereafter it increased to $7.5 \text{ mg/kg} \pm 0.09$ at a pyrolysis temperature of 400°C and decreased rapidly at 600°C pyrolysis temperature.

Total As content in the biochar decreased with an increase in pyrolysis temperature as influenced by seaweed species (Figure 3.8 B). Biochar yield from *L. pallida* had the highest total As content with $48 \text{ mg/kg} \pm 0.77$ at the control whilst *G. funicularis* biochar had $5 \text{ mg/kg} \pm 0.77$ total As at the same temperature. The lowest total As content was $1.35 \text{ mg/kg} \pm 0.77$ from *G. funicularis* biochar samples whilst the lowest from *L. pallida* biochar was $7.86 \text{ mg/kg} \pm 0.77$, all being recorded at a pyrolysis temperature of 400°C .

The highest total Ni content was recorded from *L. pallida* biochar with an average of $2.6 \text{ mg/kg} \pm 0.12$ while *G. funicularis* had an average of $1.08 \text{ mg/kg} \pm 0.12$ across all pyrolysis temperatures (Figure 3.8 C). The highest total Ni content of $5.17 \text{ mg/kg} \pm 0.26$ was recorded from *L. pallida* biochar at a pyrolysis temperature of 400°C whilst the highest total Ni of 2.02

mg/kg \pm 0.26 for *G. funicularis* was observed at control. The lowest total Ni content was recorded at 800 °C for both seaweed species with *L. pallida* biochar having 0.56 mg/kg \pm 0.26 and biochar from *G. funicularis* with the content of 0.47 mg/kg \pm 0.26.

Biochar from *G. funicularis* had the highest total Cr content with 4.60 mg/kg \pm 0.34 at 800 °C whilst *L. pallida* biochar had the highest content of 4.58 mg/kg \pm 0.34 at 200 °C. The lowest total Cr content (1.84 mg/kg \pm 0.34) from *L. pallida* biochar and *G. funicularis* biochar (2.42 mg/kg \pm 0.34) were recorded at a pyrolysis temperature of 600 °C and 200 °C, respectively (Figure 3.8 D).

The highest total Pb content was recorded from *G. funicularis* with 7.38 mg/kg \pm 0.66 at 200 °C whilst the lowest total Pb content was 0.79 mg/kg \pm 0.66 from *G. funicularis* at the control. Interestingly, for *L. pallida* biochar, the lowest content of Pb was observed at a pyrolysis temperature of 200 °C, whilst the highest was observed at 800 °C.

The highest total Zn content was recorded from *L. pallida* biochar with a content of 167.9 mg/kg \pm 4.12 at 800 °C (Figure 3.9 A), while *G. funicularis* biochar had the highest total Zn content of 28.1 mg/kg \pm 4.12 at a pyrolysis temperature of 400°C. The lowest total Zn content was recorded at 600 °C from *G. funicularis* with 5.25 mg/kg \pm 4.12 whilst biochar content from *L. pallida* had a total Zn content of 5.8 mg/kg \pm 4.12 at a pyrolysis temperature of 800°C.

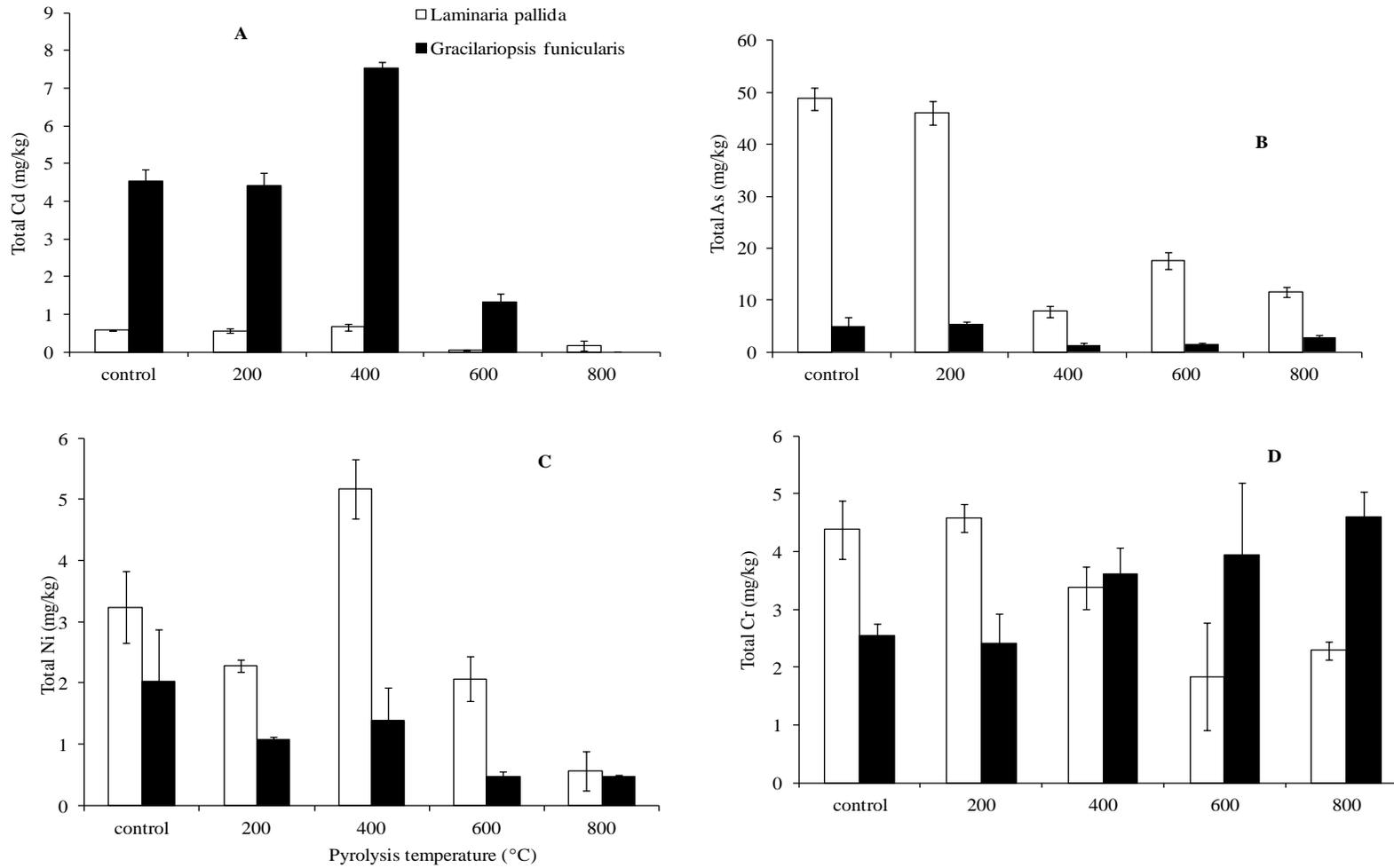


Figure 3.8 Total Cd (A), total As (B), total (Ni), total Cr (D) in biochar samples prepared using *L. pallida* and *G. funicularis* seaweeds at different pyrolysis temperatures. Error bars indicate standard deviation.

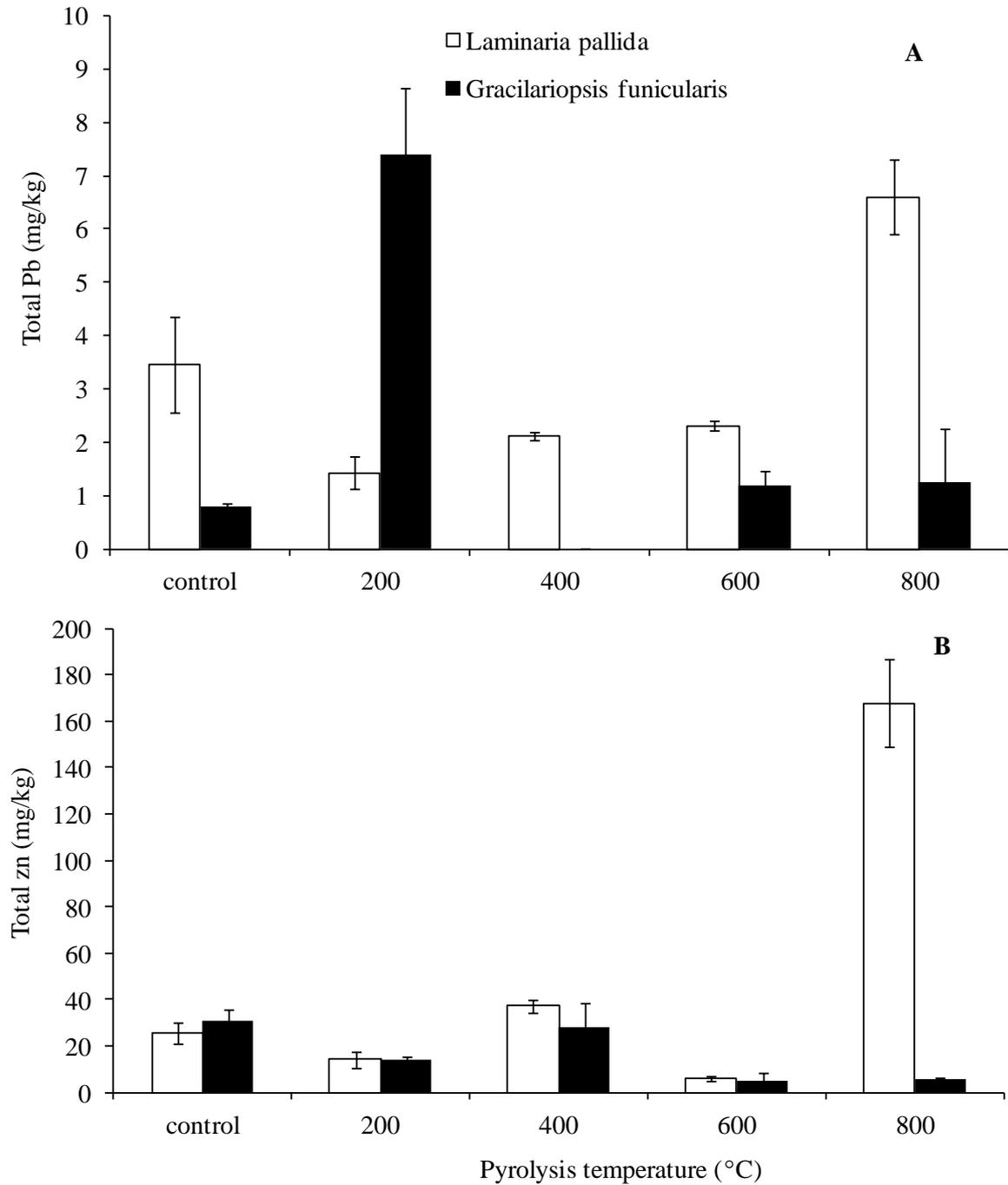


Figure 3.9 Total Pb (A) and total Zn (B) in biochar samples prepared using *L. pallida* and *G. funicularis* seaweeds at different pyrolysis temperatures. Error bars indicate standard deviation.

3.5 DISCUSSION

3.5.1 Biochar physico-chemical parameters

Seaweeds or macro-algae within the marine environment play a critical role in providing essential ecosystem services, food and shelter for marine and terrestrial organisms, whilst also providing a source of phycocolloids, soil additives and nutraceuticals (Bird et al., 2011). Furthermore, seaweeds are considered an important resource due to their rapid growth, high ability to fix atmospheric carbon as well as their high plant nutrient composition when compared to terrestrial plants (Yu et al., 2017). One potential use for seaweed biomass that has been gaining research interest is by converting it into biochar, which has been demonstrated as a tool for carbon sequestration and as a soil amendment with potential in improving soil quality (Bird et al., 2011). The study therefore presents one of the first critical steps in evaluating the potential fertilizer value of biochar from two abundant seaweeds in the Southern African Benguela powered marine environment. Among the critical parameters that can determine the sustainability of using a certain feedstock for biomass production is its biochar yield per unit biomass. In this study, though biochar yield decreased with increase in temperature, the two seaweed species (*L. pallida* and *G. funicularis*) showed slightly different responses to pyrolysis temperature on biochar yield.

The biochar yield differences could be due to the variations in the two seaweed cellular structure and composition (Yu et al., 2017). To achieve biochar yields above 50%, it was observed that pyrolysis temperatures of 400 °C, with the rapid decrease in biochar yield beyond this temperature being due to the high ash content in seaweeds compared to terrestrial biomass (Bird et al. 2011; Yu et al. 2017). It was noteworthy that the biochar yields from the *L. pallida* and *G. funicularis* observed in the study compared well with results from other studies which used both macro-algae and terrestrial biomass (Tomczyk et al., 2020; Rodriguez et al., 2020; Roberts et al., 2016).

This observation on the biochar yield therefore implies that biochar from seaweeds presented in our study can produce sustainable yields that will not require huge biomass to be harvested for the practical application of this biochar as a soil amendment. The decrease in biochar yield which was attributed to the higher ash content in seaweeds further explains the concomitant increase in electrical conductivity observed in our study. However, from an agronomic perspective, the electrical conductivity which increased with pyrolysis temperatures, indicating the increase in concentration of soluble salts in the different biochars, was way above the critical limit recommended for non-salt tolerant crops of 4 mS/cm (Angelova et al., 2013). Bird et al (2011) observed similar high electrical conductivity results in algal biochar prepared at 450 °C. The biochar from both seaweed species showed pH that was slightly acidic to highly alkaline under the increasing pyrolysis temperature. It was interesting to note that for *G. funicularis* biochar, at 400 °C, where yield loss was not below 50%, the pH was also neutral, which is important for the agronomic use of this biochar. Furthermore, the alkaline pH of the two seaweed biochars at pyrolysis temperatures of 600 °C and 800 °C could make it possible to use these biochars as liming materials in acidic soils, similar to the use of alkaline fly ash reported by Mupambwa and Mnkeni (2015).

3.5.2 Seaweed biochar as a source of organic carbon

One of the major benefits of biochar as a soil amendment is its ability to sequester carbon in soils due to the presence of rich stable carbon in biochar compared to other organic amendments like composts (Pariyar et al., 2020). As it was the case with pH, the highest carbon content in the seaweed biochars in our study was observed at a pyrolysis temperature of 400 °C. The values of carbon reported in our study compare very well with those of other organic soil amendment like cow manure used in preparation of vermicomposts (Lukashe et al., (2019). Though comparable to natural organic materials like animal manures, the total C in the seaweed biochars in this study were quite low compared to those reported for lignocellulosic terrestrial

materials such as saw dust (78.6%); sugarcane bagasse (81.6%) among others (Tomczyk et al., 2020).

3.5.3 Seaweed biochar as a source of macro-nutrients

In organic soil fertility management, one of the main limitations to the high uptake of the organic nutrient sources is their low macro nutrient composition compared to synthetic fertilizers (Mupambwa and Mnkeni 2018). In a recent review, Yu et al (2017) indicated that relative to terrestrial biomass, seaweed/algal biochar yields higher concentrations of important plant macro nutrients (P, N, K, Ca). In this study, it was noted that the total content of P and N increased with an increase in pyrolysis temperature, though the content of the extractable (plant available fraction) decreased. The content of extractable P in the seaweed biochars was significantly higher than those reported for other natural organic nutrient sources.

The pronounced variation between the total and extractable P and N may require further research to evaluate if processes like vermicomposting can be used to further increase the conversion of the total available N and P into more available forms as indicated by Godlewska et al (2017). Though higher essential plant nutrient concentrations are usually preferred in most soil amendments like N and P, there is need to also monitor the contents of macro nutrients like Na as such elements can result in soil dispersion or plant toxicity if present in higher than those recommended for plants or soils and this is critical with cations and trace or heavy metals. The high concentrations of the cations K and Na in the biochars could also explain the high electrical conductivities observed in our study. In soil fertility management, Na concentration is very critical to monitor as this element can result in loss in soil quality as it causes soil dispersion which destroys the structural stability of the soil whilst increasing salinity (Roberts et al., 2015). Bird et al (2011) has indicated that the Na component in seaweed biochar can be leached out with time, which can be done during co-composting of biochar with other organic

materials. In this study the levels of extractable cations were below those reported for other algae based seaweeds (Bird et al., 2011; Roberts et al., 2015).

3.5.4 Potentially toxic heavy metals and seaweed biochar

When organic nutrient sources are used as soil amendments, there is a need to monitor the level of potentially toxic elements, as these natural nutrient sources can be a source of pollution in soils if not carefully monitored (Guo et al., 2020). In this study, pyrolysis temperature and seaweed species did not clearly influence the content of all heavy metals measured. However, the decrease in Cd, As observed in our study as pyrolysis temperature increased has been attributed to their lower boiling points hence they elute from the reactor faster than other elements like Cr and Pb with higher boiling points (Pariyar et al., 2020).

According to the International Biochar Initiative (2015), there are maximum permissible limits for various heavy metals for biochar to be considered safe for use as a soil amendment and these are Cd (3.9 mg/kg); As (100 mg/kg); Ni (420 mg/kg); Cr (1200mg/kg); Pb (300mg/kg); Zn (7400 mg/kg). It was noteworthy that for all the biochars evaluated in our study, only Cd in *G. funicularis* at the pyrolysis temperature of 400 °C, 200 °C and the control was above the maximum permissible limits for biochar. Processes like vermicomposting have been reported to have the ability to reduce the concentration of heavy metals in composts, and studies that further evaluate such in algal biochar require attention.

3.5.6 Conclusions

The study has demonstrated that biochar derived from marine biomass presents an opportunity of converting algal biomass into potentially beneficial organic nutrient sources. For the two seaweeds evaluated in our study, it is clear that, though growing under the same marine conditions, the different seaweeds produce biochar with different chemical characteristics.

Based on the results from this study, we can further conclude that the best biochar may be produced from *G. funicularis* at a pyrolysis temperature of 400 °C, as this gave the highest concentration of most macro elements and total C. However, it will be critical to further evaluate the effects of inclusion of these seaweed biochar as soil or compost amendments on dissolved salts and conductivity of the soil or compost. Technologies like vermicomposting need to be evaluated on their ability to reduce the content of potentially toxic heavy metals and cations in algal biochar. Further characterization of the seaweed biochar is recommended to evaluate the actual presence of the recalcitrant carbon molecules before wide-spread recommendation of this biochar as superior soil carbon source.

CHAPTER FOUR

4.0 OPTIMIZING VERMICOMPOSTING OF GOAT MANURE -VEGETABLE FOOD WASTE MIXTURE FOR NUTRIENT RELEASE.

4.1 ABSTRACT

In arid countries, goats are the major household livestock and these generate underutilized manure. Furthermore, in arid countries, a lot of the fruits and vegetables are imported, which contributes to generation of huge post-harvest losses. Vermicomposting is a technology with potential to convert such organic wastes into nutrient rich organic fertilizers. This study evaluated the influence of various mixtures of vegetable food wastes (FW) with goat manure on vermi-degradation and plant nutrient release. No significant differences ($P < 0.05$) were observed on changes in C/N ratio, though significant differences were observed on humification parameters. The biodegradation measured as changes in C/N ratio and humification properties followed the general order of 75FW:25GM > 50FW:50GM > control (100GM) > 25FW:75GM. Similarly, the 75FW:25GM and 50FW:50GM treatments showed significantly ($P < 0.05$) the highest final concentration of Olsen extracted phosphorus of 0.98 and 0.96 g per kg of compost. Relative to the control, the treatments with 75%; 50% and 25% FW resulted in a 113%; 109% and 60% more phosphorus, respectively, after 12 weeks. There were no significant differences ($P > 0.05$) between all treatments on nitrate/nitrite concentrations. It was noteworthy that the changes in P and N during vermicomposting were highly predictable using second order polynomial equations. The control treatment with 100GM resulted in the highest EC of 6.7 mS/m whilst the 50FW:50GM treatment having the lowest EC of 4.3 mS/m. Based on our study and a composting mixture of between 75FW:25GM or 50FW:50GM will give superior compost with high macro-nutrient composition.

Keywords: Nutrient release; arid country; Biodegradation; Namibia; macro-nutrients; humification parameters.

4.2 INTRODUCTION

The increase in world human population has resulted in intensified agricultural production and industrialization to meet the demands of this growing population (Mupambwa et al. 2019). These intensified activities have however resulted in overproduction of waste materials such as animal manures, food wastes and other industrial wastes, leading to inappropriate and enormous disposal of these wastes (Lukashe et al. 2019; Piya et al. 2018; Mupambwa and Mnkeni 2018). Such inappropriate disposals pose huge environmental challenges such as introduction of harmful trace metals, inorganic salts and pathogens into the soil as well as releasing potentially toxic greenhouse gases (Esmaeili et al. 2020; Awasthi et al. 2020; Lazcano et al. 2008). There is a need for identifying effective and sustainable technologies that can improve the value of these waste materials as a way of mitigating against the present environmental challenges they pose.

Among the technologies that have been gaining momentum in waste management due to their limited environmental footprints and ability to improve the value of the waste materials are composting and vermicomposting (Hussain et al. 2018; Ravindran et al. 2019). Such technologies have also been promoted due to the growing realization of the potential of organic nutrient sources such as vermicomposts in improving soil quality compared to inorganic fertilizers (Mupambwa and Mnkeni, 2018). Therefore, vermicomposts are among the main organic nutrient sources that are being promoted due to their high potential to enhance soil biological, chemical and physical properties (Mupambwa and Mnkeni, 2018). Vermicomposting is an eco-friendly process that involves accelerated conversion of organic matter from waste materials into nutrient rich humus through interaction of earthworms and microorganisms (Piya et al. 2018; Mupambwa and Mnkeni, 2018). Unlike inorganic fertilizers that only feed the crop, improved organic fertilizers like vermicomposts are being promoted in

soil fertility management as they feed the soil to feed the crop, providing not only crop nutrients but also humic acids, plant growth hormones, N-fixing and P-solubilizing microbes, enzymes and vitamins (Gupta et al, 2019; Chatterjee et al. 2014).

These organic fertilizers can be critical in improving the resilience of physically degraded agricultural soils in hyper arid environments such as those prevalent in Namibia, Botswana and parts of South Africa, where terrestrial biomass production is very limited (Mupambwa et al. 2019). Arid countries are associated with poor quality soils due to their low organic matter contents and vermicomposts present an option to enhance the fertility of such degraded soils with not only organic matter but also nutrients whilst improving their ability to sequester more carbon (Verma et al. 2020; Waleed 2016). In arid countries like Namibia, though terrestrial plant biomass production is limited, there is production of measurable quantities of animal manures especially from goats, due to their high adaptability (Gichangi et al. 2009). However, this goat manure is not widely used as a soil fertilizer due to its low and poor bio-available nutrient quality and high concentrations of salts. It is therefore important to understand the effects of technologies like vermicomposting in enhancing the nutrient quality of waste materials like goat manure, where limited research has been undertaken.

Furthermore, due to the aridity of some countries in sub-Saharan Africa, countries like Namibia import more than 70% of their vegetable and fruit requirements from other countries (Mupambwa et al. 2019). Due to the large distances involved during the transportation of these fruits and vegetables, huge quantities of food wastes are generated due to transportation associated post-harvest losses. Unfortunately, much of the food wastes generated during transportation, are disposed of at dumpsites where they pose serious health challenges during the uncontrolled decomposition.

Though some researchers have looked at potentials of food waste vermicomposting, much research that has focused on vegetable food wastes has indicated that these composts are characterized by low nutrient bioavailability. This is mainly due to the fact that vegetable food wastes are composed of water with very small quantities of dry biomass which can be mineralized to create bio available nutrients (Srivastava et al 2020). During vegetable waste composting, it has been reported that much of the important nutrients are lost as leachates, with a 6000 kg of vegetable compost losing around 400L of nutrients containing leachates per day (Sall et al. 2019). The leachate losses can be reduced by mixing the vegetable food wastes with bulking materials that could absorb these liquid nutrients.

As highlighted in the preceding statements, Namibia as a hyper-arid country has materials that are usually considered wastes (i.e. goat manure and vegetable food wastes) whose fertilizer values could be improved by vermicomposting. The combined vermicomposting of these two waste materials thus presents an option of generating a nutrient rich organic fertilizer using the complementary chemical characteristics of the materials. It is therefore interesting to evaluate the co-composting of vegetable food wastes mixed with animal manures like goat manure to create nutrient rich organic composts for use in improving the quality of soils especially in arid environments. Furthermore, there is paucity of information on the optimized and combined vermicomposting of goat manure and food wastes fed by *Eisenia fetida* earthworms. Therefore, the objectives of this study were to evaluate the effects of different goat manure – vegetable food waste mixtures during vermicomposting by *Eisenia fetida* on (i) compost biodegradation (C/N ratio; pH; EC and humification parameters) (ii) nutrient release (Olsen extractable P; extractable cations and nitrogen).

4.3 MATERIALS AND METHODS

4.3.1 Source of materials used in the study

The experiment was conducted at the Sam Nujoma Campus of the University of Namibia, located in Henties Bay in the Erongo Region of Namibia. The vegetable food wastes were collected from local supermarkets in Henties Bay and these were frozen until further use. The dried goat manure was collected from a local farm in the Uis area (21°13'7"S 14°52'3"E) of Namibia. Plastic buckets of 20 L capacity were used as vermi-reactors for the process of vermicomposting, and holes were drilled at the bottom and on the lid for gaseous exchange and leachate drainage (Mupambwa 2015). The earthworms were obtained from a local wormery at the University of Namibia – Sam Nujoma Campus where the species *Eisenia fetida* was kept feeding on mainly vegetable food wastes and goat and cow manure wastes.

4.3.2 Treatments; experimental design and setup

Vegetable food waste (FW) was mixed with goat manure (GM) at 4 ratios on a w/w basis and this gave 4 treatment combinations which are: 25% FW + 75% GM; 50% FW + 50% GM; 75% FW + 25% GM and 100% GM (Control). The experiment was laid in a completely randomized design with three replications. The mixtures from the different treatments were allowed to pre compost for 2 weeks to moisten the materials and to get rid of volatile toxic gases (Mupambwa and Mnkeni 2016). After the 2-week pre-composting, earthworms (*E. fetida*) were introduced into each vermireactor at a stocking density of 25 g of worms per kg of compost following recommendations of Mupambwa and Mnkeni (2016). A total of 3 kg (dry mass basis) food waste and goat manure mixture was used for the experiment and this was kept at a moisture content of 70-80% under shade at room temperature. Sampling was done for each treatment at 0; 2; 4; 6; 8; 10; 12 weeks. The collected compost samples were air dried and ground using a mechanical grinder to analyse the selected parameters described below.

4.3.3 Electrical conductivity and pH

Electrical conductivity (EC) and pH were measured in water at a ratio 1:10 (w/v) as described by AgriLASA (2004). Briefly, a 5g vermicompost sample was mixed with 50 mL of deionized water and shaken with a horizontally reciprocating shaker at 120 rpm for 30 minutes and then pH and EC were measured using a calibrated multi meter (Lovibond Water Testing, Senso Direct 150).

4.3.4 Olsen extractable P

Olsen method was used to determine extractable phosphorus because it has been shown to be effective for both acidic and calcareous soils (Schoenau and Halloran 2006). A solution of 0.5M sodium hydrogen carbonate adjusted to a pH of 8.5 using 1M of Sodium hydroxide was used for extraction. Briefly, a 2.5g of the compost was shaken in 50 ml of the extracting solution for 30 minutes at 120 rpm and then filtered with Whatman Number 2 filter paper. The extracts were then analyzed for P using the Ascorbic acid method as described by Kuo (1996).

4.3.5 Total C and N

Total C and N were determined using the dry combustion method using a LECO CHN628 auto analyser (LECO Corporation, USA).

4.3.6 Extractable Cations (Ca, Mg, Na, and K)

Cations were extracted using the ammonium acetate method (AgriLASA 2004). A solution of 1 Mol ammonium acetate adjusted to pH 7 was used to extract the cations. To prepare this, an amount of 57 mL of glacial acetic acid was diluted with de-ionised water to a volume of 500 mL. An amount of 69 mL concentrated ammonia solution was then added to the diluted solution of acetic acid. The solution was mixed well and diluted to about 900 mL with deionised water and then pH was adjusted to 7 using either acetic acid or ammonia solution. A 5 g of compost was placed in 100 mL extraction bottle and 50 mL of ammonium acetate

solution was added into the extraction bottle and the mixture was shaken horizontally on a reciprocating shaker at 180 rpm for 30 minutes. The extracts were filtered using Whatman Number 2 filter paper and the cation concentrations in the solution were determined using a calibrated Inductively Coupled Plasma – Optical Emission Spectrometer (ICP-OES - iCAP 6000 SERIES).

4.3.7 Exchangeable Ammonium, Nitrate and Nitrite

Exchangeable inorganic nitrogen fractions ($\text{NH}_4\text{-N}$ and $\text{NO}_2/\text{NO}_3\text{-N}$) were extracted using 0.5 M potassium sulphate (Okalebo et al., 2002). A 5g sample of vermicompost was shaken for 1 hour on a reciprocal mechanical shaker at 180 rpm and filtered through Whatman Number 2 filter paper. The inorganic nitrogen fractions in the extracts were then determined using colorimetric analysis employing a UV spectrophotometer (Thermo Scientific, Genesys 105 UV -Vis). The preparation of samples for $\text{NH}_4\text{-N}$ colorimetric determination involved transfer of 0.2 mL of sample extract into a test tube with 5 mL of N1 reagent (prepared by mixing specific quantities of sodium salicylate, sodium citrate, sodium tartrate and sodium nitroprusside) followed by 5 mL of N2 reagent (prepared by mixing specific quantities of sodium hydroxide and sodium hypochlorite).

The samples with reagent mixtures were allowed to stand for an hour and the resulting blue colour intensity was measured at 655 nm wavelength using a UV spectrophotometer (Thermo Scientific, Genesys 105UV -Vis) that was calibrated using standards ranging between 0 – 25 $\mu\text{g NH}_4^+\text{-N/mL}$ treated in the same manner as the samples. Colorimetric sample preparation for $\text{NO}_2/\text{NO}_3\text{-N}$ was done by transferring 1.0 mL 5% salicylic acid to 0.5 mL sample extract in a test tube followed by 10 mL 4 M NaOH. After an hour of standing the resulting yellow colour intensity was measured at 419 nm wavelength using a UV spectrophotometer (Thermo Scientific, Genesys 105UV -Vis) that was calibrated using standards ranging from 0 – 10 $\mu\text{g NO}_3\text{-N/mL}$ treated in the same manner as the samples.

4.3.8 Humification parameters

The humic and fulvic acid fractions in the composts were extracted using a method described by Sanchez-Monedero et al, (1996). A 0.1 mol L⁻¹ NaOH solution was used at an extraction ratio of 2:40 (w/v), and shaken for 4 hours with a reciprocal shaker. The extracts were then centrifuged at 4000 rpm for 15 min. After centrifugation, the supernatant was divided into two fractions, with one half stored for analysis of total extractable C fraction (C_{tEX}).

The other half was acidified to pH 2 by adding drops of concentrated H₂SO₄, to form a precipitate representing the humic acid fractions (HA) whilst the liquid part represented the fulvic acid fraction. The acidified extracts were allowed to coagulate for 24 hours at 4 °C and further centrifuged at 4000 rpm to separate the humic and fulvic fractions. The non-precipitated part of the centrifuged samples was then further analysed for fulvic acid carbon (C_{FA}). The C concentrations in the supernatants were determined using the dichromate oxidation method, with the concentration of the humic acid (C_{HA}) fraction being calculated as the difference between C_{tEX} and C_{FA} . The humification ratio (HR, equation 4.1), humification index (HI, equation 4.2), percentage of humic acids (Pha, equation 4.3), and polymerization index (PI, equation 4) which are indices used for the evaluation of humification level in the vermicompost were then calculated as indicated in the equations (Sanchez-Monedero et al, 1996).

$$\text{HR} = \frac{C_{\text{tEX}}}{C} \times 100 \quad (\text{Eqn. 4.1})$$

$$\text{HI} = \frac{C_{\text{HA}}}{C} \times 100 \quad (\text{Eqn. 4.2})$$

$$\text{Pha} = \frac{C_{\text{HA}}}{C_{\text{tEX}}} \times 100 \quad (\text{Eqn.4.3})$$

$$\text{PI} = \frac{C_{\text{HA}}}{C_{\text{FA}}} \quad (\text{Eqn. 4.4})$$

4.3.9 Statistical analysis

The data were analyzed using repeated measures of analysis of variance (ANOVAR) because the sample collection was non-destructive. Where sphericity assumptions could not be met, the Greenhouse–Geisser correction of P was used. A one-way analysis of variance (ANOVA) was used to analyze humification. Mean separations were conducted using the least significant differences (LSD) at $P < 0.05$ when analysis of variance (ANOVA) indicated a significant p -value. All data analyses were done using JMP version 14.0.0 Statistical software ((SAS Institute, Inc., Cary, North Carolina, USA, 2010) whilst Microsoft Excel was used for plotting of all graphs.

4.4 RESULTS

4.4. Influence of food waste-goat manure mixtures on vermi-degradation

4.4.1 C/N ratio, pH and electrical conductivity

To evaluate the effects of how the various mixtures of goat manure and vegetable food waste influence bio-gradation during vermicomposting, we evaluated the changes in C/N ratio, humification parameters, pH and electrical conductivity. At week 0, the increase in food waste concentration resulted in an increase in C/N ratio with the 75FW:25GM manure treatment having the highest C/N ratio of 51.2, whilst the lowest was the 25FW:75GM treatment with a C/N ratio of 35.4 (Figure 4.1). After 12 weeks of vermicomposting, the final C/N ratio was 16.5; 16.6; 16.9 and 17.1 for the 75FW:25GM; control; 25FW:75GM and 50FW:50GM, respectively. However, it was noteworthy that across the 12 weeks of vermicomposting, there were no significant differences ($P \geq 0.05$) between treatments as indicated in Table 4.1, though there were significant interactions ($P \leq 0.05$) between treatments and time, indicating that the change in C/N ratio was more dependent on time.

Table 4.1: Results of repeated measures of ANOVA for changes in selected parameters following 12 weeks of vegetable food waste- goat manure mixture vermicomposting.

Parameter	Treatment		Time (weeks)		Treatments × Time	
	<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>
C/N ratio	4.64	ns	57.62	< 0.001	2.96	0.0759
pH	63.5	< 0.001	46.46	< 0.001	4.70	0.0051
EC (mS/cm)	699.5	< 0.001	63.32	< 0.001	7.14	0.0007
Olsen P (g/kg)	533.2	< 0.001	392.9	< 0.001	15.22	< 0.001
Nitrate & Nitrite (g/kg)	0.39	ns	15.91	< 0.001	2.01	ns
Ammonium (g/kg)	101.85	< 0.001	94.50	< 0.001	3.5	0.0106
Ca (g/kg)	74.46	< 0.001	17.81	0.0027	4.98	0.0308
K (g/kg)	4.623	0.0002	13.42	< 0.001	5.48	< 0.001
Mg (g/kg)	71.33	< 0.001	59.73	< 0.001	3.35	0.0047
Na (g/kg)	17.77	0.0012	28.36	≤ 0.001	4.61	< 0.001

ns = not significant at $P \leq 0.05$, $F = F$ - test, $P =$ Probability.

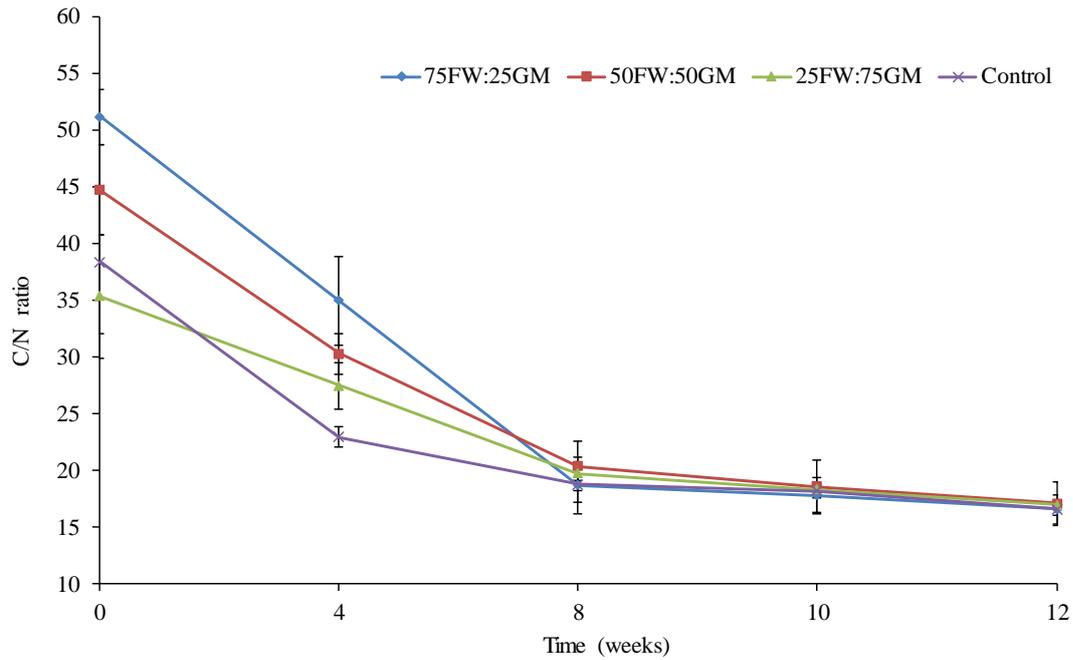


Figure 4.1 Changes in Carbon to Nitrogen ratio (C/N) during vermicomposting of vegetable food waste (FW) mixed with goat manure (GM) at different ratios. Error bars indicate standard deviation.

Across the 12 weeks of vermicomposting, the C/N ratio decreased by 208%; 161%; 108% and 131% for the 75FW:25GM; 50FW:50GM; 25FW:75GM and control treatments, respectively. A significant difference ($P \leq 0.05$) between treatments was observed on changes in pH and electrical conductivity (EC) as indicated by the repeated measures of ANOVA (Table 4.1). At the beginning of the study, pH values followed the order 75FW:25GM > 50FW:50GM > 25FW:75GM > control, with alkaline pH values of 9.6; 9.1; 8.5 and 8.3, respectively.

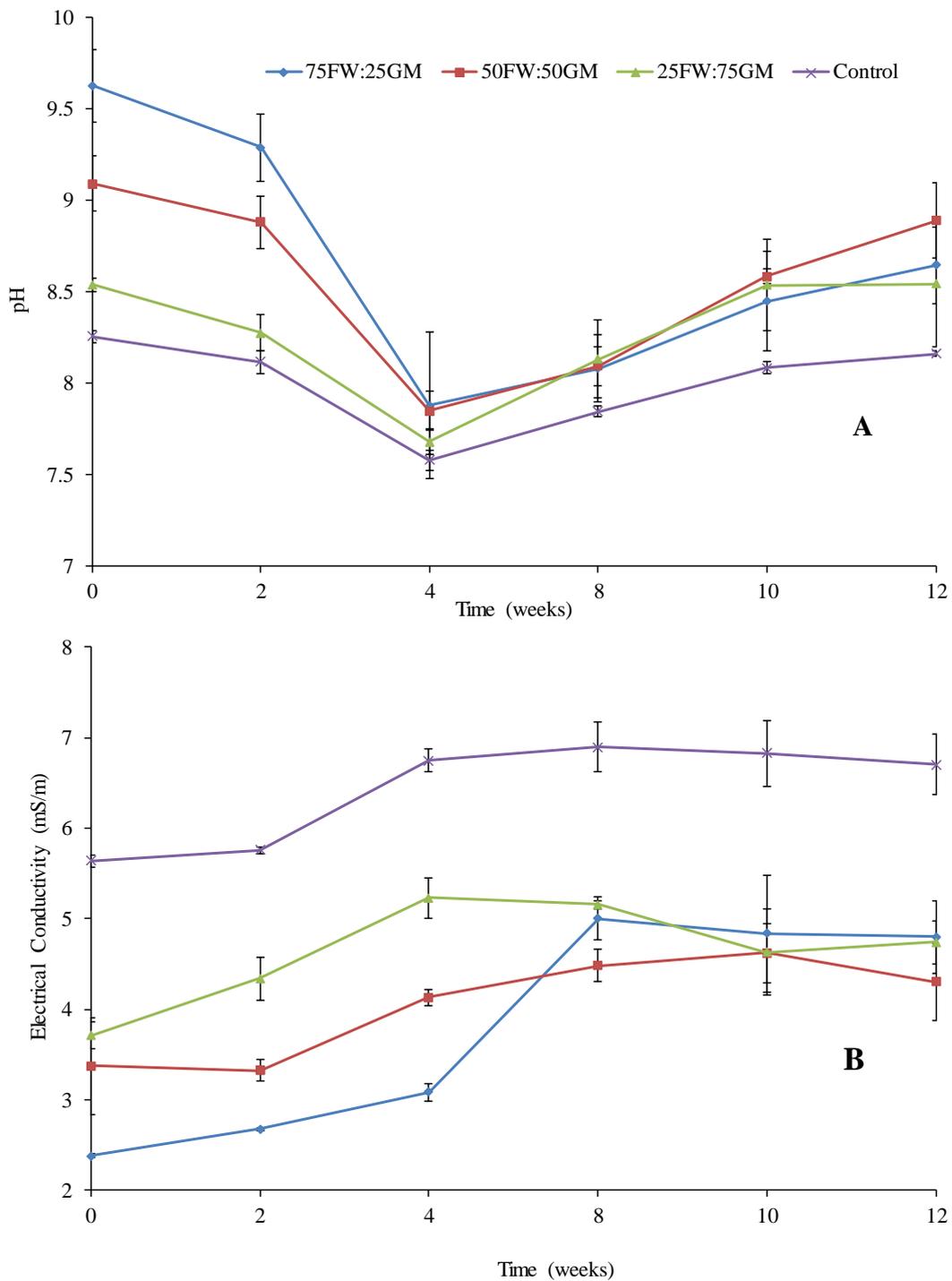


Figure 4.2 Changes in pH (A) and EC (B) during vermicomposting over 12 weeks of vegetable food waste (FW) mixed with goat manure (GM) at different ratios. Error bars indicate standard deviation.

During the 12 weeks of vermicomposting, pH in all treatments decreased from week zero to week 4 and thereafter gradually increased until week 12 (Figure 4.2 A). After 12 weeks of

vermicomposting, all treatments showed slightly alkaline values which were lower than the starting values with the 50FW:50GM treatment having the highest pH of 8.9 whilst the control had the lowest pH of 8.2. The significant changes in pH between week 0, week 4 and week 12 could explain the significant interaction observed between the treatments and time, as indicated by the repeated measures of ANOVA. Across all treatments and throughout the 12 weeks of vermicomposting, EC values generally increased resulting in significantly different final EC (mS/m) values of 4.8; 4.3; 4.7 and 6.7 for the 75FW:25GM; 50FW:50GM; 25FW:75GM and control, respectively (Figure 4.2 B). It was interesting to note that as the level of goat manure increased, the electrical conductivity also increased.

4.4.2 Humification parameters

For all four humification parameters measured, there was a significant difference ($P \leq 0.05$) observed between treatments (Figure 4.3). Apart from the humification ratio, the 75FW:25GM treatment showed the highest humification index (HI), percentage humic acids (Pha) and polymerization index (PI). For humification ratio (HR), the 25FW:75GM treatment resulted in the highest value of 17.8 whilst the lowest was the 75FW:25GM treatment with a HR of 11.8. As the level of vegetable food waste incorporation increased, it was observed that the HR decreased resulting in an 11.9%; 34.1% increase as vegetable food waste incorporation decreased from 75% to 50% and 50% to 25%, respectively. However, as the food waste content changed to 0% in the control, the HR did not continue increasing (Figure 3). Unlike the HR results, for HI, Pha and PI, as the level of vegetable food waste incorporation decreased from 75% to 25%, these parameters decreased. The decrease in vegetable food waste incorporation from 75% to 50% resulted in a 51.2%; 14.1% and 32.1% decrease in HI; Pha and PI, respectively. Similarly, the decrease in vegetable food waste incorporation from 50% to 25% resulted in a 44.1%; 110.1% and 184.6 decrease in HI, Pha and PI, respectively.

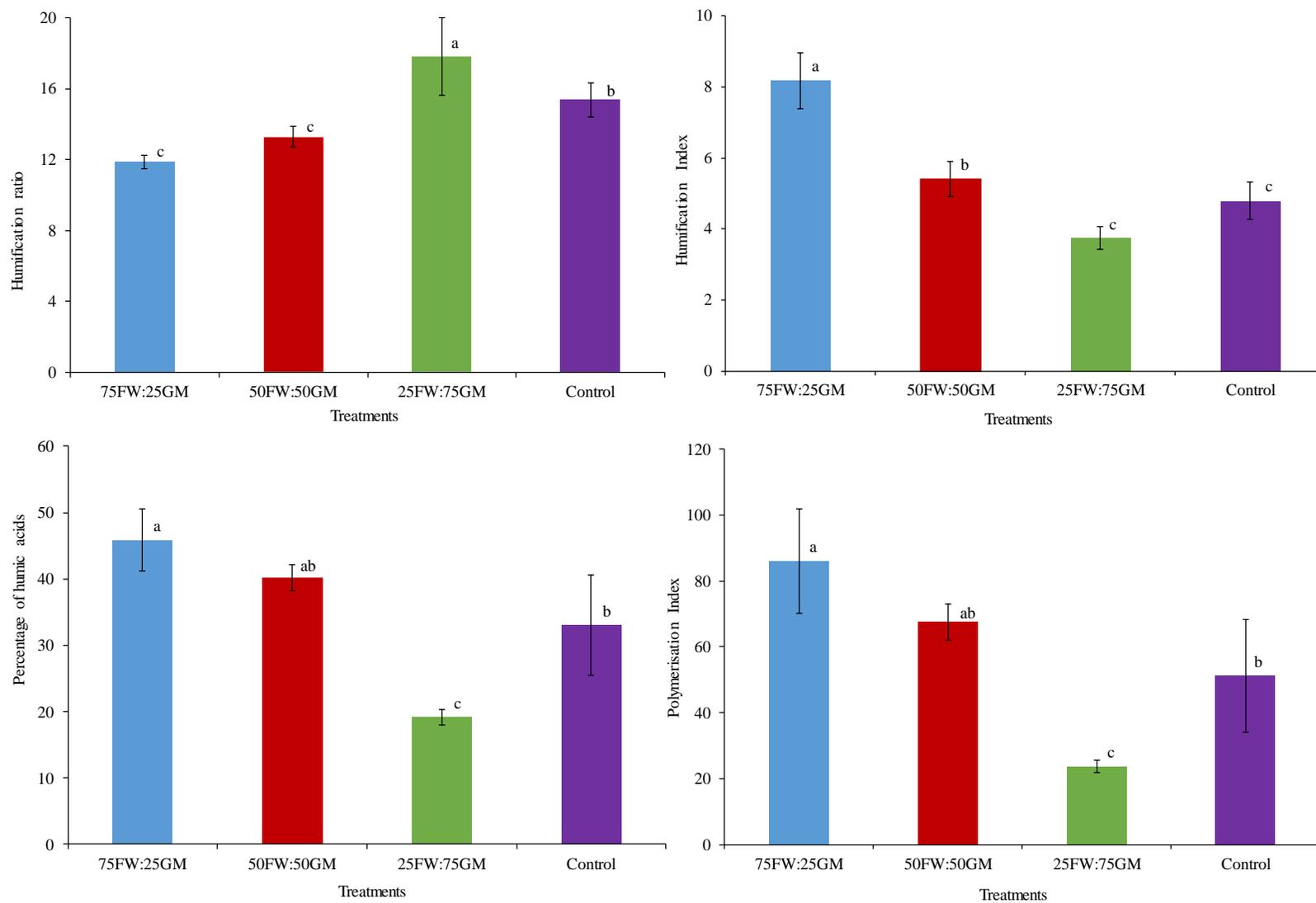


Figure 4.3 Changes in humification parameters during 12 weeks of vermicomposting of vegetable food waste (FW) mixed with goat manure (GM) at various ratios. Error bars indicate standard deviation whilst bars within each graph having different lowercase

4.4.3 Influence of food waste-goat manure mixtures on nutrient release

4.4.3.1 Olsen extractable phosphorus

Phosphorus is one of the critical elements important for plant growth and in this study, there were significant differences ($P \leq 0.05$) observed between treatments (Table 4.1). The 75FW:25GM and 50FW:50GM treatments showed the highest final concentration of Olsen phosphorus of 0.98 and 0.96 g per kg of compost whilst the control treatment had the lowest final concentration of 0.46 g per kg of compost (Figure 4.4 and Table 4.3).

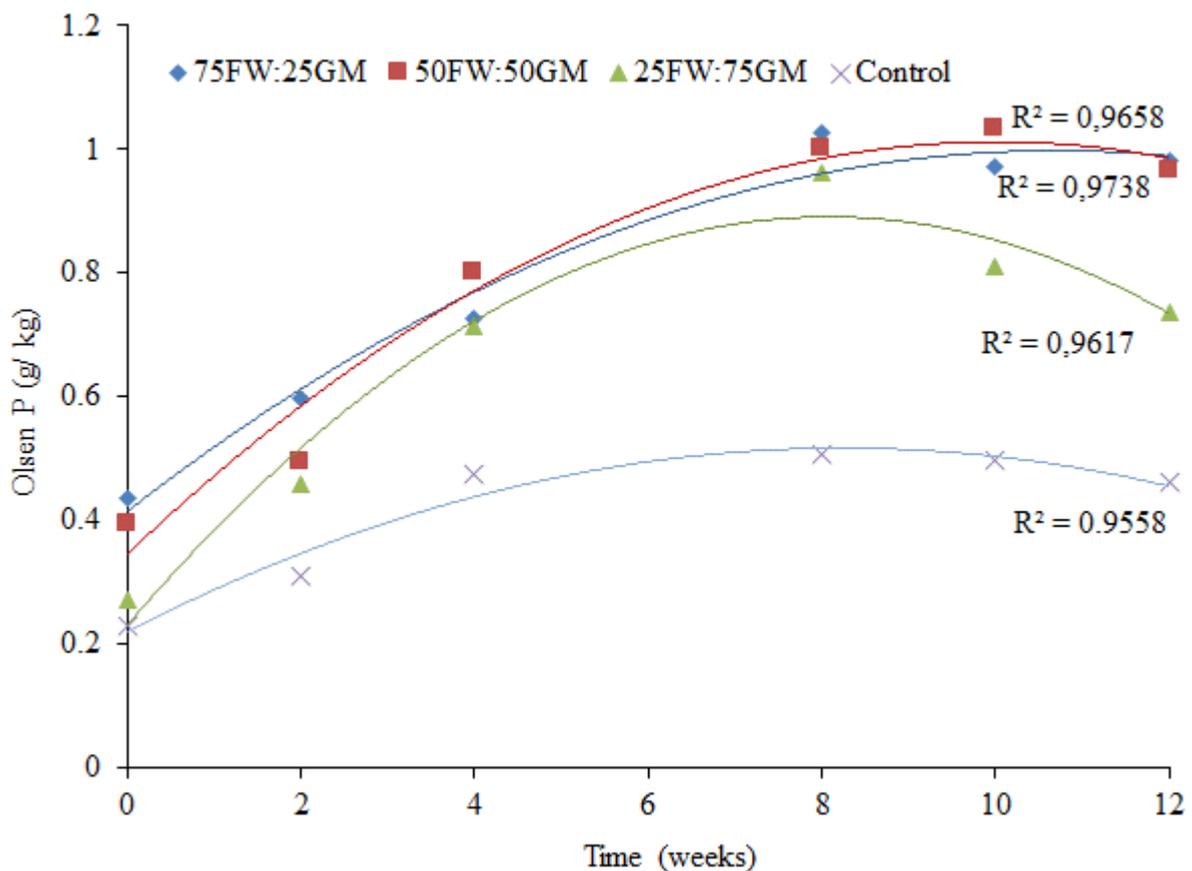


Figure 4.4 Second order polynomial line graph showing the trends of changes in Olsen P during vermicomposting of vegetable food waste (FW) mixed with goat manure (GM) at different ratios.

Table 4 .2 Influence of different vegetable food waste and goat manure mixtures during vermicomposting on Olsen extractable P.

	Regression equation	R ²	Rate of Olsen release [¶]	Predicted Olsen P at week 12 [§]	Observed Olsen P at week 12 [#]	Net Olsen P increase after 12 weeks*
Treatments			g P/kg/week	g/kg		%
Control	$-0.0044x^2 + 0.0719x + 0.2197$	0.9558	0.019c ^λ	0.4489d	0.461c	100.3c
75FW:25GM	$-0.0051x^2 + 0.1088x + 0.4148$	0.9738	0.045a	0.986b	0.982a	124.6bc
50FW:50GM	$-0.0066x^2 + 0.133x + 0.3455$	0.9658	0.048a	0.9911a	0.964a	144.5ab
25FW:75GM	$-0.0101x^2 + 0.1632x + 0.231$	0.9671	0.039b	0.735c	0.736b	172.84a

¶ Calculated as (Olsen P at 12 weeks – Olsen P at 0 weeks)/12 wk.

§ Calculated from the regression equation.

Determined as the actual Olsen P after 12 weeks of vermicomposting.

* Calculated as (Olsen P at 12 weeks – Olsen P at 0 weeks)/Olsen P at 0-week x 100.

λ Values followed by the same lowercase letter are not significantly different at $P \leq 0.05$.

Relative to the control, the treatments 75FW:25GM; 50FW:50GM and 25FW:75GM resulted in a 113%; 109% and 60% more Olsen phosphorus, respectively, after 12 weeks of vermicomposting. The final Olsen P concentration after 12 weeks of vermicomposting followed a significantly different order of 75FW: 25GM = 50GM: 50FW > 25FW:75GM > Control. It was also interesting to note that the 50FW:50GM treatment showed significantly ($P \leq 0.05$) the highest rate of Olsen P release of 48 mg P/ kg of compost per week, whilst the control had the lowest rate of 19 mg P/ kg of compost per week (Table 4.3). In the study, it was also noted that the rate of Olsen phosphorus release could be predicted with high certainty using the second order polynomial equation, as indicated by the high coefficient of determination. The equations also indicated that maximum Olsen P within vermicomposts is realized around 10 weeks, before it starts decreasing towards week 12 during food waste-goat manure vermicomposting (Figure 4.4).

4.4.4 Exchangeable inorganic nitrogen

There were no significant differences ($P \geq 0.05$) between all treatments in nitrate/nitrite concentrations across the 12 weeks of vermicomposting (Table 4.1). Generally, all treatments showed an increase in nitrate/nitrite concentrations from week 0 showing a peak concentration between week 6 and week 8 and thereafter decreased significantly until week 12 (Figure 4.5). At week 8, where peak nitrate/nitrite concentration was reached, it was observed that as the concentration of food waste increased from 25% to 75%, the concentration of nitrate/nitrite decreased. At week 9, the highest concentration of nitrate/nitrite was in the 25FW:75GM treatment with a value of 0.1 g/kg followed by 50FW:50GM > 75FW: GM and lastly the control with concentrations of 0.098; 0.068; 0.067 g/kg, respectively. Similar to nitrate/nitrite, the concentration of ammonium during the vermicomposting in all treatments showed an increasing trend until week 10 (Figure 4. 5).

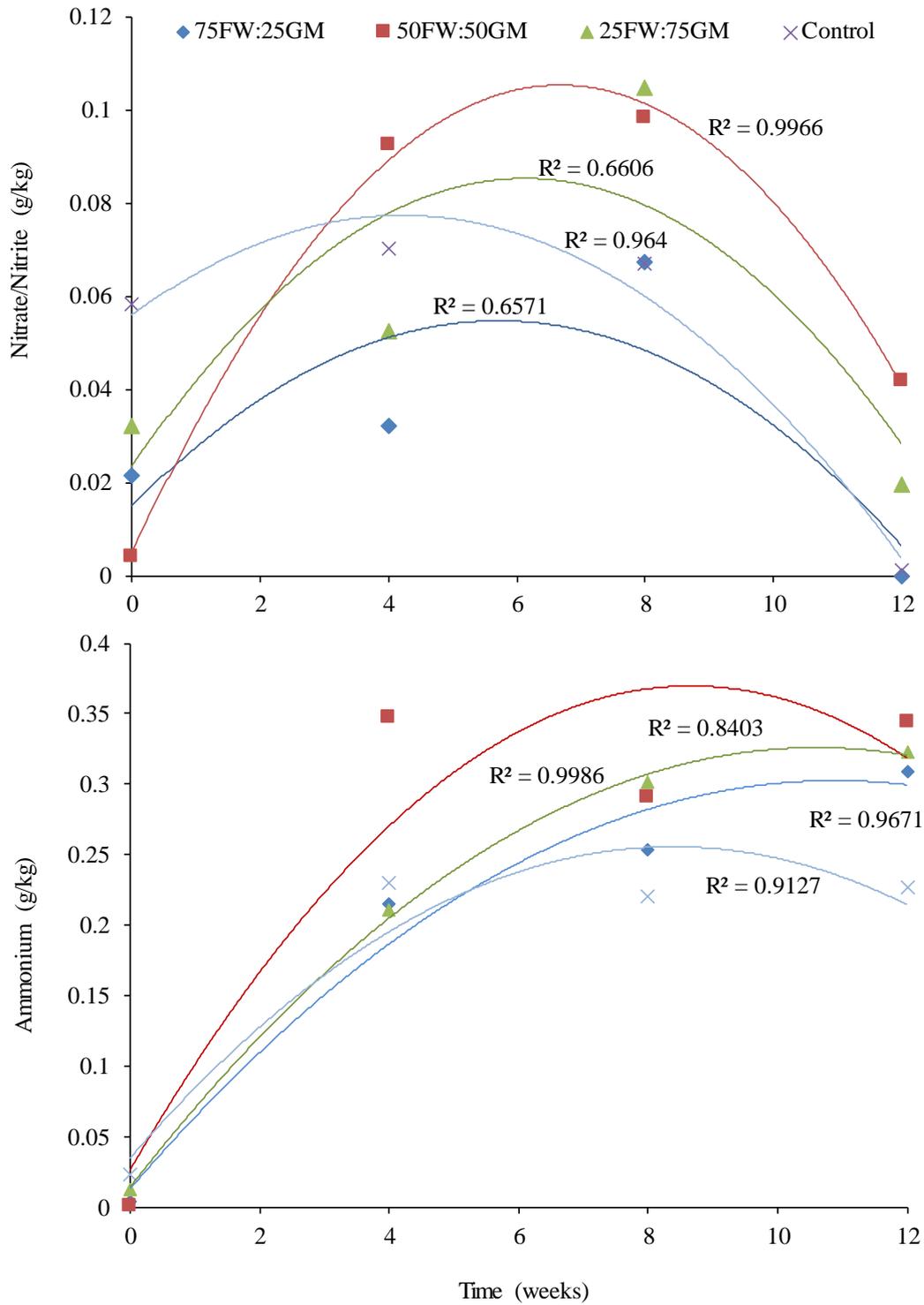


Figure 4.5 Second order polynomial line graphs showing the trends of changes in nitrate, nitrite and ammonium during vermicomposting of vegetable food waste (FW) mixed with goat manure (GM) at different ratios.

However, unlike nitrate/nitrite, there were significant differences ($P \leq 0.05$) observed between treatments (Table 4.1). The 50FW:50GM treatment resulted in consistently high concentration of ammonium, whilst the control had the lowest concentration throughout the 12 weeks of vermicomposting. It was interesting to observe that there was no consistent relationship observed between changes in the concentration of nitrate/nitrite and that of ammonium, though their concentrations during vermicomposting could be predicted using the second order polynomial equations.

4.4.5 Extractable cations

For all the cations (Ca, Mg, Na and K) determined in this study, there were significant differences ($P \leq 0.05$) observed between treatments (Table 4.1). Across the 12 weeks of vermicomposting, a general trend was observed between the treatments, where the higher the percentage of goat manure, the higher the concentrations of the cations. This resulted in the control treatment having the highest concentration of all cations during the 12 weeks of vermicomposting (Figure 4.6 A). For calcium, the concentration increased for all treatments from week 0 till week 4 and thereafter, it remained almost constant for all other treatments whilst it continued to increase for the control treatment. After 12 weeks of vermicomposting, the concentration of Ca was 6.6; 4.9; 4.8 and 4.1 g/kg for the control; 25FW:75GM; 50FW:50GM and 75FW:25GM treatments, respectively.

Potassium is among the 3 macro elements required for plant nutrition which was measured in our study. The concentration of K increased from week 0 for all treatments and then reached a maximum at either 4 or 8 weeks, and there after decreased or remained almost constant until week 12 (Figure 4. 6 B). It was interesting to note that, though the control had the highest

concentration of K at week 0, after 12 weeks of vermicomposting the concentration of K followed the order 75FW:25GM > 50FW:50GM > control > 25FW:75 GM with the concentrations ranging from 10.96 g/kg to 10.22 g/kg.

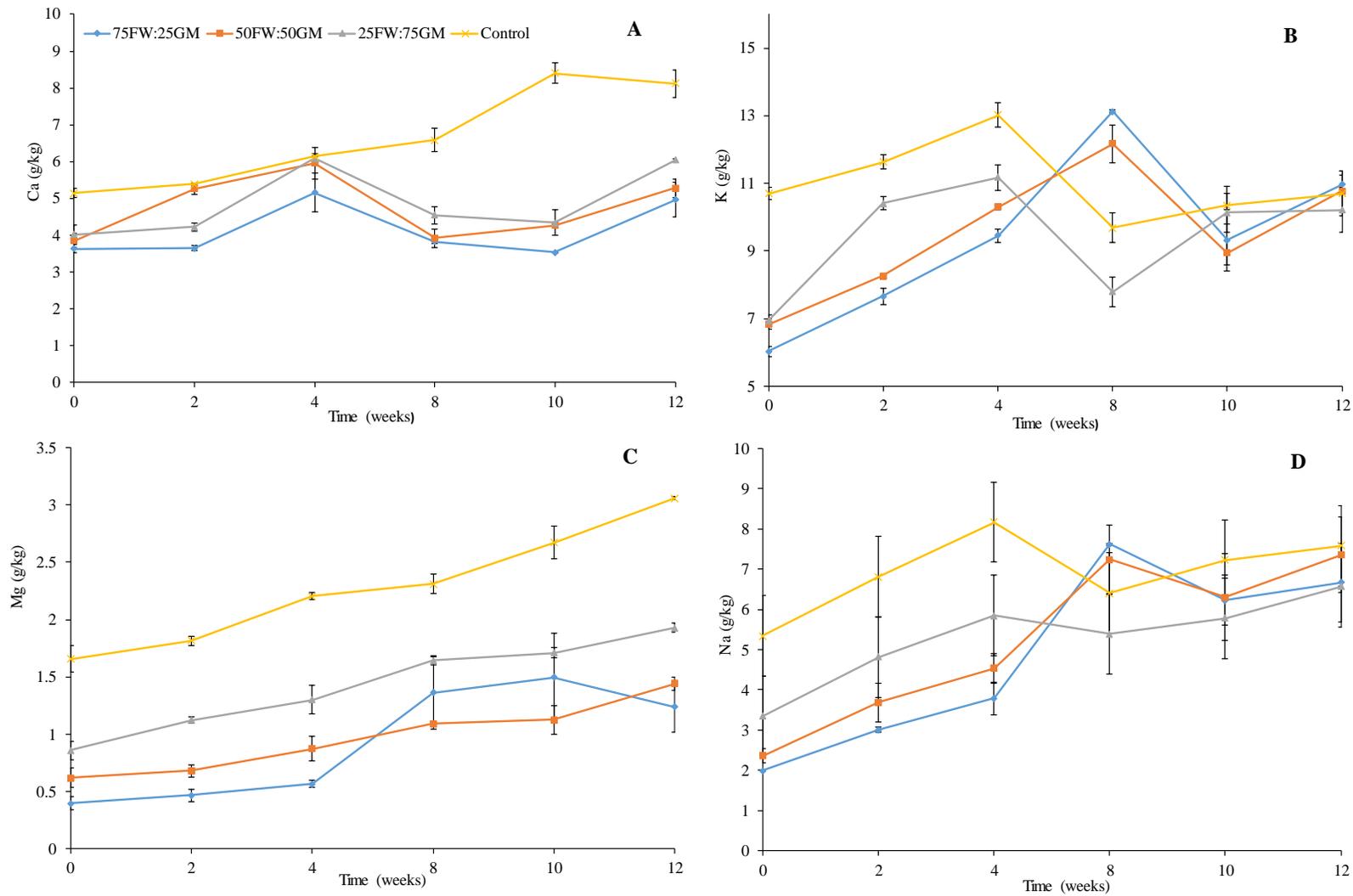


Figure 4.6 Changes in extractable Ca; K; Mg and Na indicated in graphs A; B; C and D, respectively, during 12 weeks of vermicomposting of vegetable food waste (FW) mixed with goat manure (GM) at various ratios. Error bars indicate standard deviation.

For magnesium, the concentration across all the four treatments increased from week 0 until week 12 (Figure 4.6 C). Similar to the trend of Ca, it was observed where the control had the highest concentration across all 12 weeks compared to other treatments, with the higher the inclusion level of goat manure, the higher the concentration of Mg generally. After 12 weeks of vermicomposting, final concentration of Mg was 3.1; 1.9; 1.4 and 1.2 g/kg for the control; 25FW:75GM; 50FW:50GM and 75FW:25GM treatments, respectively. The concentration of Na in composts is critical as it can induce soil dispersion when present in high concentrations. From week 0 until week 4, the control treatment showed the highest concentration of Na, with an increasing trend being observed for all treatments until week 8 (Figure 4.6 D). Though not consistent a trend, the final concentration of Na followed the order control > 50FW:50GM > 75FW:25GM > 25FW:75GM, respectively.

4.5 DISCUSSION

The combined vermicomposting of goat manure and vegetable food waste presented in our study is a novel technique that can allow the generation of nutrient rich organic soil fertilizers for use in arid countries. Though goat manure and vegetable food wastes have been reported to have considerable quantities of essential plant nutrients, these are mainly organically bound and require mineralization before they can be available for plant uptake (Cho et al. 2017; Moreno-Caselles et al. 2002). The present study thus determined the levels of biodegradation and nutrient release during the vermicomposting of different goat manure – vegetable food waste mixtures. Through the mixing of the various ratios of goat manure and vegetable food waste, the original C/N ratios of the composts were not uniform and this has been identified as critical in influencing the vermi-degradation process (Mupambwa and Mnkeni 2018). A starting C/N ratio of between 25 - 40 has been widely recommended as optimum for earthworm

growth and development during vermicomposting (Ravindran and Mnkeni; Ndegwa and Thompson 2000).

In the study, the recommended C/N ratios were not achieved for the starting mixtures. Though the original mixtures were not optimized based on recommendations, the results of the final C/N ratios in the different mixtures were quite interesting as they indicated highly mature composts indicating effective biodegradation. A C/N ratio of below 20 and preferably 15 has been reported to indicate mature compost and all treatments had values that fit this criterion (Raj and Antil 2011). The decrease in C/N ratio is an indicator of biodegradation and our results actually indicate that vermi-degradation was more effective in treatments with higher initial C/N ratio like the 75FW:25GM and 50FW:50GM treatments, contrary to what is recommended in literature.

Apart from using changes in C/N, the changes in humification parameters have also been reported to be critical in indicating the maturity of composts. The increase in humification parameters during vermicomposting indicates the effectiveness of the various treatments in transforming the easily degraded molecules into more recalcitrant molecules with higher molecular weights (Mupambwa and Mnkeni 2015).

A mature compost has been reported to have a humification ratio > 7 , a humification index > 3.5 , a polymerization index > 1 , and percentage of humic acids $> 50\%$ (Bernal et al. 2009). Although all vermicompost treatments in this study resulted in higher humification parameters, the 50FW:50GM treatment achieved superior humification properties, thus the best matured vermicompost. Higher humic fractions play an important role in the soil carbon cycling and soil nutrient mineralization, with the humic acids also acting as a source of nutrients for plants, providing a more available form of nutrients if used in the soil (Abakumor et al. 2018).

The chemistry of a vermicompost is very important as this can influence the final fertilizer value of the compost. All treatments showed consistently alkaline pH values during the vermicomposting process and this could be explained by the increased release of ammonia during biodegradation of organic compounds (Gong et al. 2018). Similar results where pH increases during vermicomposting have also been reported by Majlessi et al. (2010) and Fatehi and Shayegan (2010) attributing it also to the activity of earthworm calciferous glands secreting ammonium ions which reduce H^+ ions.

However, the pH values in all the treatments were within the values where most nutrients are available for plant uptake. Similarly, the rise in electrical conductivity reported might be due to degradation of organic matter and increased levels of soluble mineral salts in available forms during vermicomposting (Sharma and Garg et al 2019). It was however noteworthy that 50FW:50GM treatment gave the lowest EC which will not pose any salinity threats when this vermicompost is applied to soil.

The higher electrical conductivity with higher levels of goat manure can also explain the higher level of cations observed in treatments where goat manure was incorporated beyond 50%. However, the influence of the various cations, especially Na from the various vermicomposts may need to be further evaluated under field conditions to evaluate their influence on soil physical properties.

The influence of the various mixtures of goat manure and vegetable food waste was quite apparent in the transformations of phosphorus and nitrogen, with the 50FW:50GM treatment showing again superior results. The increase in Olsen extractable P observed is due to the mineralization of organic phosphorous fractions during vermicomposting process caused by phosphatase secretion from earthworms (Esmaeili et al. 2020). The other important observation during phosphorus and nitrogen release was the ability to precisely predict or model the changes in these nutrients using the second order polynomial. These equations clearly indicated

that the maximum concentrations of P and N, during goat manure-vegetable food waste mixtures, is achieved around week 10 and can be critical in development of nutrient predictable, commercially marketable organic fertilizers. Finally, the changes in nitrate/nitrite and ammonium were unlike what has been reported by many researchers where the concentration of nitrate/nitrite increases as that of ammonium decreases. Mupambwa and Mnkeni (2016) however made similar observations during fly ash vermicomposting and further research is needed to evaluate these transformations when such vermicomposts are applied to soil as a fertilizer.

4.6 Conclusions

This study focused on optimizing the vermicomposting of goat manure- food waste mixture to enhance the nutrient release and biodegradation. Though incorporation of vegetable food waste at higher levels resulted in high initial C/N ratio, the treatments between 75FW:25GM and 50FW:50GM actually resulted in optimized biodegradation which created a more superior compost in terms of maturity parameters determined. Furthermore, these same treatments (75FW:25GM and 50FW:50GM) also resulted in significantly higher release of essential macro-elements like N and P, with their peak release being observed after about 10 weeks of vermicomposting. This indicates that under arid conditions like those of Namibia, goat manure and vegetable food waste could be mixed at specific ratios of between 75FW:25GM and 50FW:50GM to generate a nutrient rich organic fertilizer. However, there is need to further evaluate the actual fertilizer value of these optimized goat manure – vegetable food waste vermicomposts in crop based studies. Supplementing such vermicomposts with materials like biochar can also be critical in reducing nutrient leaching and may need to be evaluated in future studies.

CHAPTER FIVE

5.0 SEAWEED (*GRACILARIOPSIS FUNICULARIS*) BIOCHAR INCORPORATION INTO A GOAT MANURE-FOOD WASTE VERMICOMPOST FOR OPTIMIZED VERMI-DEGRADATION AND NUTRIENT RELEASE

5.1 Abstract

Vermicomposts are organic fertilizer sources that are being promoted, however their concentration of macro nutrients such as (NPK) are very low as compared to those of inorganic fertilizers. This study evaluated the effect of biochar from seaweed (*Gracilariopsis funicularis*) incorporated into a goat manure-food waste mixture at 0% (control); 2%; 4%; 6% and 8% on compost degradation and macro-nutrient release. There were significant ($P \leq 0.05$) effects on most of the measured parameters. After 10 weeks of vermicomposting the highest pH of 9.06 was observed within the control whilst the lowest was 8.7 in the 8% treatment. The electrical conductivity showed a positive relationship with the level of biochar incorporation with the highest of 16.5 mS/cm from the 8% treatment whilst the lowest was within the control with 6 mS/cm. There were no significant differences between treatments on humification parameters, however there were significant differences on the changes of C/N ratio with the final C/N ratio of 14.4; 14.9; 16.7; 15.1 and 14.4 for the control; 2%; 4%; 6%; and 8% treatments, respectively. Higher incorporation rate resulted in the higher concentration of potassium with a value of 32.3 g/kg at week 8. The final percentage change in Olsen P was 19%; 14.2%; 7.3%; 4.1% and 3.0% for the 8%; 6%; control; 4%; and 2% treatments, respectively. An optimized level of 6 to 8 % biochar incorporation ratio can be recommended in terms of nutrition and decomposition during vermicomposting of food waste-goat manure mixture. However, incorporation of *G. funicularis* biochar does not seem to influence changes in the vermicomposting efficiency,

though it can significantly improve the macro nutrients like P, K and Mg concentrations as well as the macro element concentrations.

Keywords: Biodegradation, Compost maturity, *Eisenia fetida*, Nutrient release, Seaweed biochar.

5.2 Introduction

The advent of the green revolution saw the introduction of huge industrialization and intensification of agricultural activities which has generated huge quantities of waste materials (Chen et al. 2020). Furthermore, the use of inorganic fertilizers to increase crop yields has resulted in increased crop yield though this has also contributed to soil degradation as these fertilizers only feed the crop and not the soil which is a living ecosystem. Traditionally composts have been used as organic sources of nutrients, with their limitations being slow release of nutrients into the soil coupled with low nutrient levels and the probability of introducing potentially toxic pathogens and heavy metals into the environment (Chen et al. 2020). Recently, research has focused on improved organic fertilizers in sustainable agriculture like vermicomposts which involve the use of solid organic materials such as food waste, animal waste and sewage sludge processed using earthworms such as *Eisenia fetida* and other species (Garg and Gupta, 2009).

Though both composts and vermicomposts are being promoted as nutrient sources, they are still inferior in nutrition when compared to inorganic fertilizers. For example, urea has 46% Nitrogen, compound fertilizer like 3:2:3 (35 %) has 42.8 % Phosphorus and 28% Potassium whilst homemade compost was reported to have as low as 0.5% Nitrogen, 0.27% Phosphorus and 0.81% Potassium (Tilley 2020). This has led researchers to use various materials for amendment such as fly ash, rock phosphate and biochar as well as other methods like

phosphorus solubilising bacteria to increase nutrient content. A study by Zheng et al, (2020) reported that rock phosphate amendment increased the contents of total Phosphorus and available Phosphorus, and reduced the loss of nitrogen during sewage sludge composting. According to Lukasho et al. (2019) the inoculation of fly ash – cow dung – waste paper vermicompost with phosphate solubilising bacteria accelerated the biodegradation resulting in an improved vermicompost with low C/N ratio and high Olsen phosphorus. A study that used modified fly ash as an amendment to remediate heavy metal contamination from the soil reported that the mixture of fly ash and chicken manure reduced the concentrations of cadmium, copper and lead by 49.0%; 53.5% and 67.8 % respectively. They also observed an increase in organic matter and available NPK (Hu et al. 2020).

In another study, the incorporation of biochar made from corn cobs and wood improved soil quality as it provided higher soil organic matter and macronutrients during the production of maize (Kizito et al. 2019). Biochar as an amendment is quite interesting as it is rich in recalcitrant forms of carbon and contains elevated nutrient concentrations (Katakula et al. 2020). The use of biochar has potential to reduce the bioavailability of trace elements, increase organic matter content of composting feedstock, enhance the level of humification and increases nutrient retention during vermicomposting (Were et al. 2019). Amendment of biochar enhances plant nutrient availability in the soil, microbial activities, organic matter, water holding capacity and crop production; while lessening the fertilizer requirements, greenhouse gas productions and nutrient leaching (Wang et al, 2020).

Research has shown that amendment of 10% (w/w) plant biochar into kitchen waste – sewage sludge vermicompost increased the reproduction rate of earthworms (*Eisenia fetida*) by up to 53.9 % (Khan et al. 2019). In the same study, biochar amendments increased the concentration of macronutrients, total Nitrogen (15.8–31.0%), total phosphorus (8.6 –9.9%), total potassium (2.8–17.3%), calcium (4.1–9.9%) and magnesium (0.8–12.2%). Makini et al, (2020) reported

that amending soils with goat manure-based vermicompost enhances soil chemical properties. Among the different application rates used, the highest rate of 30 t ha⁻¹ goat manure-based vermicompost showed higher soil pH (8.00), total Nitrogen (0.606%), available Phosphorus (21.933 ppm) and exchangeable K (0.456).

Much of the research that has used biochar as an amendment has mainly used material of terrestrial origin with no research having used marine biomass. In a recent study, Katakula et al. (2020) identified that biochar derived from seaweed species such as *Laminaria pallida* and *Glacilariopsis funicularis* pyrolysed at a temperature of 400 degrees Celsius can generate nutrient rich and carbon rich biochar. Marine biomass derived biochar can be an important source of amendment for organic fertilizers in hyper arid countries such as Namibia. However, there is limited research that looked at marine biomass and converted them into biochar to be used as an amendment into vermicompost. Therefore, the objective of this study was to evaluate the seaweed biochar incorporation into food waste-goat manure vermicompost for enhanced vermicompost degradation (C/N, pH, EC) and essential plant nutrient release (Olsen P, Ca, Na, Mg, and K).

5.3 Materials and methods

5.3.1 Source of materials used in the study

The experiment was conducted at the Sam Nujoma Campus of the University of Namibia, located in Henties Bay, Erongo Region of Namibia. The optimum ratio of food waste and goat manure mixture of 50% food waste and 50% goat manure was used based on effective biodegradation and nutrient mineralization results identified in experiment 2. The seaweed biochar used in this study was identified from Katakula et al. (2020) and this was derived from *G. funicularis* which was pyrolysed at a temperature of 400 degrees C for 1 hr. Plastic buckets

of 20 L capacity were used as vermi-reactors for the process of vermicomposting, and holes were drilled at the bottom and on the lid for gaseous exchange and leachate drainage (Mupambwa 2015). The earthworms were obtained from the local wormery at the University of Namibia- Sam Nujoma Campus, where the species *Eisenia fetida* was kept feeding on mainly vegetable food waste and goat manure.

5.3.2 Treatments, experimental design and setup

Seaweed biochar (SB) was incorporated into the optimised food waste -goat manure mixture (FWGM) at 5 different levels on a dry w/w basis and this gave 5 treatment combinations which are: Control (FWGM only); 2% SB + FWGM; 4% SB + FWGM; 6% SB + FWGM; 8% SB + FWGM. The experiment was laid in a completely randomized design with 3 replications. The mixtures from the different treatments were allowed to pre compost for 2 weeks to moisten the materials and to get rid of volatile toxic gases (Mupambwa and Mnkeni 2016). After the 2-week pre-composting, earthworms (*E. fetida*) were introduced into each vermireactor at a stocking density of 25 g of worms per kg of compost following recommendations of Mupambwa and Mnkeni (2016). A total of 3 kg (dry mass basis) food waste and goat manure mixture was used for the experiment and this was kept at a moisture content of 70-80% under shade at room temperature. Sampling was done for each treatment at 0; 4; 8; 10; weeks. The collected compost samples were air dried and ground using a mechanical grinder (POLYMIX PX-MFC 90 D made in Switzerland by KINEMATICA AG) to analyse for the selected parameters as described below.

5.3.3 Electrical conductivity and pH

Electrical conductivity (EC) and pH were measured in water at a ratio of 1:10 (w/v) as described by AgriLASA (2004). Briefly, a 5g of the compost was mixed with 50 mL of deionized water and shaken with a horizontally reciprocating shaker at 120 rpm for 30 minutes

and then pH and EC were measured using a calibrated multi - meter (Lovibond Water Testing, Senso Direct 150).

5.3.4 Olsen extractable P

The Olsen method was used to determine extractable phosphorus because it has been shown to be effective for acidic materials (Schoenau and Halloran 2006). A solution of 0.5M sodium hydrogen carbonate adjusted to a pH of 8.5 using 1M of Sodium hydroxide was used for extraction. Briefly, a 2.5g of the vermicompost was shaken in 50 mL of the extracting solution for 30 minutes at 120 rpm and then filtered with Whatman Number 2 filter paper. The extracts were then analysed for P using the Ascorbic acid method as described by Kuo (1996).

5.3.5 Total C and N

Total C and N were determined using the dry combustion method employing a LECO CHN628 auto analyser (LECO Corporation, USA).

5.3.6 Extractable cations (Ca, Mg, Na, and K)

The cations were extracted using the ammonium acetate method as described by (AgriLASA 2004). Solution of 1 Mol ammonium acetate adjusted to pH 7 was used to extract the cations. To prepare this, an amount of 57 mL of glacial acetic acid was diluted with de-ionised water to a volume of 500 mL. An amount of 69 mL concentrated ammonia solution was then added to the diluted solution of acetic acid. The solution was mixed well and diluted to about 900 mL with deionised water and then pH was adjusted to 7 using either acetic acid or ammonia solution. A 5 g of compost was placed in 100 mL extraction bottle and 50 mL of ammonium acetate solution was added into the extraction bottle and the mixture was shaken horizontally on a reciprocating shaker at 180 rpm for 30 minutes. The extracts were filtered using Whatman Number 2 filter paper and the cation concentrations in the solution were determined using a

calibrated Inductively Coupled Plasma – Optical Emission Spectrometer (ICP-OES – iCAP 6000 SERIES).

5.3.7 Humification parameters

The humic and fulvic acid fractions in the composts were extracted using a method described by Sanchez-Monedero et al, (1996). A 0.1 mol L⁻¹ NaOH solution was used at an extraction ratio of 2:40 (w/v), and shaken for 4 hours with a reciprocal shaker. The extracts were then centrifuged at 4000 rpm for 15 min. After centrifugation, the supernatant was divided into two fractions, with one half stored for analysis of total extractable C fraction (C_{tEX}). The other half was acidified to pH 2 by adding drops of concentrated H₂SO₄, to form a precipitate representing the humic acid fractions (HA) whilst the liquid part represented the fulvic acid fraction. The acidified extracts were allowed to coagulate for 24 hours at 4 °C and further centrifuged at 4000 rpm to separate the humic and fulvic fractions. The non-precipitated part of the centrifuged samples was then further analysed for fulvic acid carbon (CFA). The C concentrations in the supernatants were determined using the dichromate oxidation method, with the concentration of the humic acid (CHA) fraction being calculated as the difference between C_{tEX} and CFA. The humification ratio (HR, equation 5.1), humification index (HI, equation 5.2), percentage of humic acids (Pha, equation 5.3), and polymerization index (PI, equation 5.4) which are indices used for the evaluation of humification level in the vermicompost were then calculated as indicated in the equations.

$$HR = \frac{C_{tEX}}{C} \times 100 \quad (\text{Eqn. 5.1})$$

$$HI = \frac{C_{HA}}{C} \times 100 \quad (\text{Eqn. 5.2})$$

$$\text{Pha} = \frac{C_{\text{HA}}}{C_{\text{tEX}}} \times 100 \quad (\text{Eqn.5.3})$$

$$\text{PI} = \frac{C_{\text{HA}}}{C_{\text{FA}}} \times 100 \quad (\text{Eqn. 5.4})$$

5.4 Statistical analysis

The data were analysed using repeated measures of analysis of variance (ANOVAR). Where sphericity assumptions could not be met, the Greenhouse-Geisser correction of P was used. For the humification parameters, a one-way analysis of variance (ANOVA) was done for the data collected at week 10. Mean separations were conducted using the Fishers protected least significant at $P \leq 0.05$ when analysis of variance indicated a significant P -value. All data were analysed using JMP version 14.0.0 Statistical software (SAS Institute, Inc., Cary, North Carolina, USA, 2010), whilst all the graphs were plotted using Microsoft Excel (2007).

5.5 Results

5.5. Effects of seaweed biochar incorporation during vermicomposting on compost maturity

Compost maturity was measured using parameters such as: C/N ratio and humification parameters as presented in the next section.

5.5.1 pH and Electrical conductivity (EC)

There was a significant interaction between pH and EC. A significant difference ($P \leq 0.05$) was observed between treatments on changes in pH across the 10 weeks of vermicomposting (Table 5.1). Generally, pH was alkaline throughout the vermicomposting process with pH actually increasing from the initial values across all treatments (Figure 5.1). After 10 weeks of vermicomposting, the control treatment which had no biochar incorporated into it had the highest pH of 9.06 whilst the lowest pH was 8.72 observed in the 8% biochar treatment. It was

interesting to observe that the final pH values at 10 weeks showed a strong link to the level of biochar incorporation as it followed the order 0% > 2%; 6% > 4% > 8% biochar. After 10 weeks of vermicomposting, the pH values observed were 9.06; 8.83; 8.79; 8.82; and 8.72 for the treatments 0%, 2%, 4%, 6% and 8% treatments, respectively.

Table 5. 1 Repeated measures ANOVA for changes in the selected parameters during vermicomposting of a goat manure food waste mixture amended with seaweed (*G. funicularis*) biochar.

Parameter	Treatment		Time (Weeks)		Treatments × Time	
	<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>
C/N ratio	0.67	< 0.0001	373.85	< 0.0001	1.84	ns
pH	29.45	< 0.0001	520.45	< 0.0001	8.79	< 0.0001
EC (mS/cm)	129.4	< 0.0001	397.59	< 0.0001	22.06	< 0.0001
Olsen P (g/kg)	11.12	< 0.0024	11.48	0.0010	1.42	ns
Ca (g/kg)	1.06	Ns	110.14	< 0.0001	14.15	< 0.0001
K (g/kg)	887.06	< 0.0001	42.62	< 0.0001	67.71	< 0.0001
Mg (g/kg)	3.80	0.0511	46.79	< 0.0001	3.49	0.0167
Na (g/kg)	863.63	< .0001	92.58	< 0.0001	34.24	< 0.0001

ns = not significant at $P \leq 0.05$

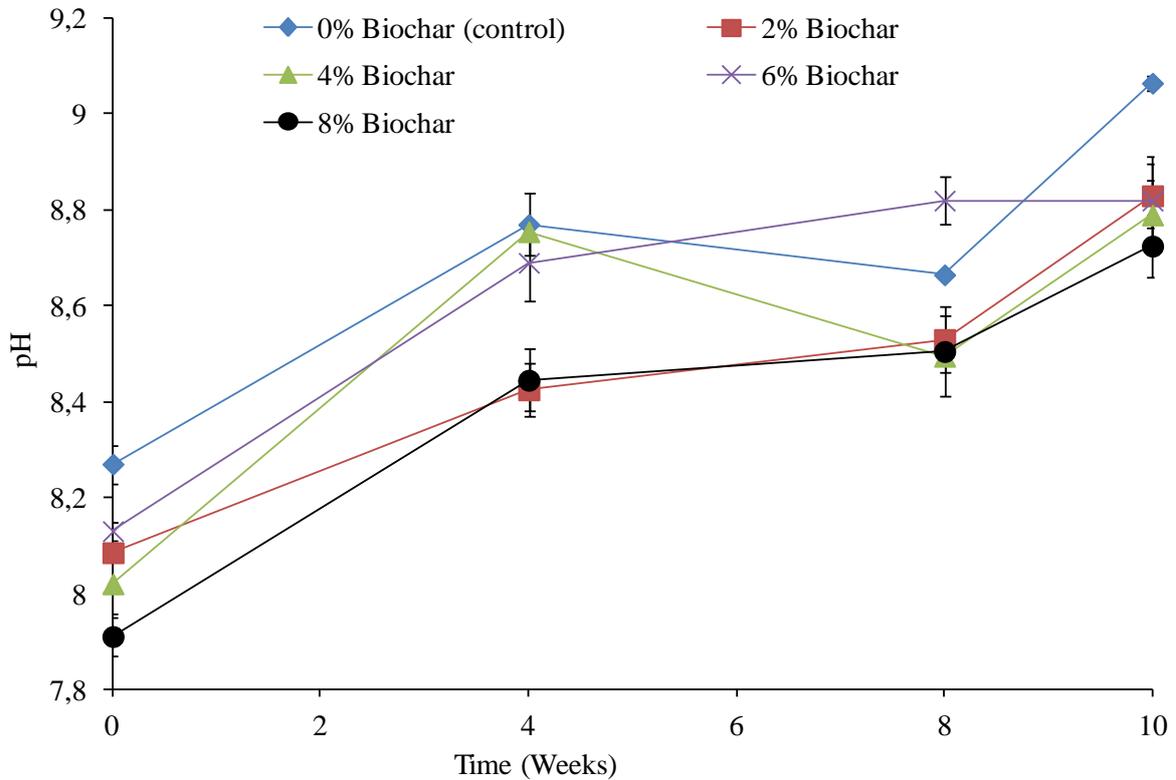


Figure 5.10 Changes in pH during vermicomposting of goat manure - food waste mixture amended with seaweed (*G. funicularis*) biochar. Error bars indicate standard deviation

Throughout the 10 weeks of vermicomposting, EC showed an almost linear increase for all treatments, with a significant difference ($P \leq 0.05$) being observed between treatments and time as indicated in Table 5.1 and Figure 5.2. Similar to the observations of pH, the higher the biochar incorporation rate, the higher the EC and also the higher the incorporation rate the higher the rate of change (Figure 5.2). Across all treatments between 0 and 10 weeks, EC increased by 28%; 42.2%; 42.3%; 54% and 67% for the 0%, 2%, 4%, 6% and 8% treatments, respectively. After the 10 weeks of vermicomposting the final EC values were 6 mS/cm; 9.2 mS/cm; 10.9 mS/cm; 12.4 mS/cm and 16.5 mS/cm for 0%; 2%; 4%; 6%; and 8% treatments, respectively.

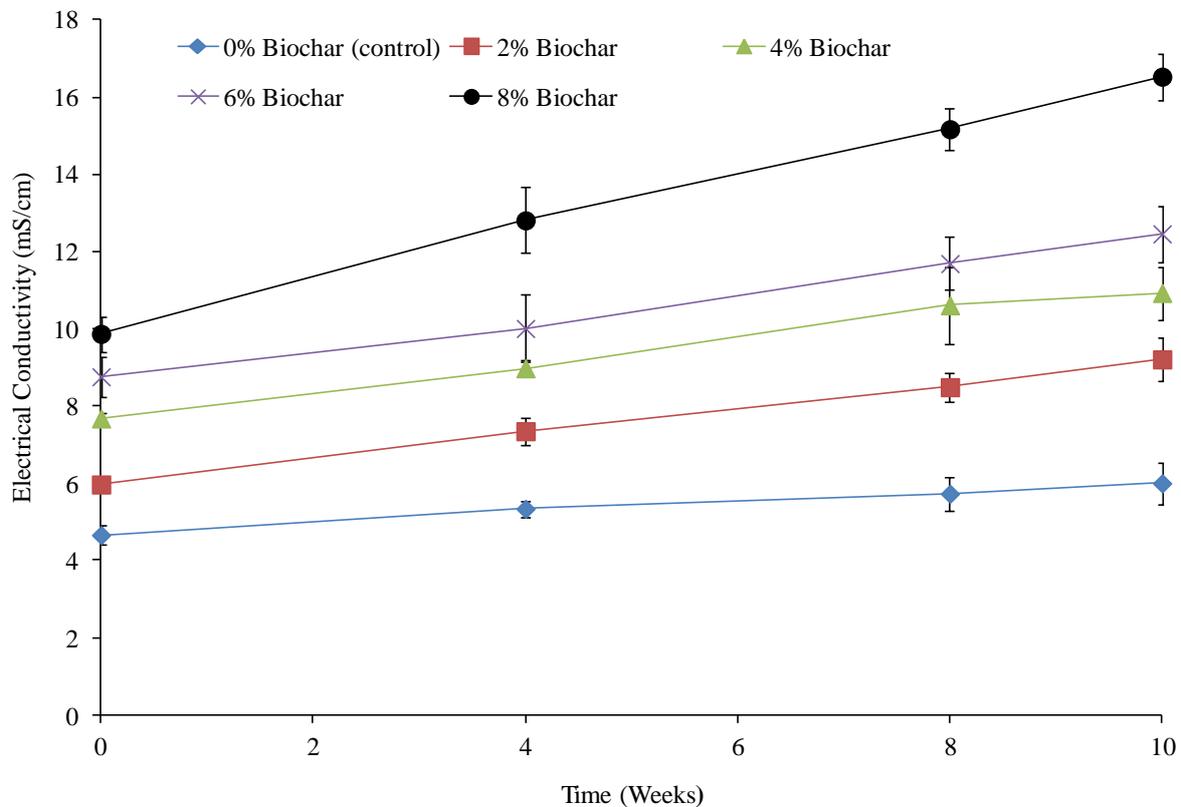


Figure 5.2 Changes in Electrical conductivity during vermicomposting of goat manure - food waste mixture amended with seaweed (*G. funicularis*) biochar. Error bars indicate standard deviation.

5.5.2 C/N ratio

Across the different treatments, there were significant differences ($P \leq 0.05$) observed on changes in C/N ratio, across the 10 weeks of vermicomposting (Table 5.1). Generally, across all the treatments the C/N decreased from an average C/N of 34:1 at week 0 to a final average of 15:1 at week 10 (Figure 5.3). After the 10 weeks of vermicomposting, relative to the starting values, the C/N ratio decreased by 97%; 116%; 155%; 128% and 134% for the 4%; 6%; 2%; 8% and control treatments, respectively, as indicated by the significant difference on time indicated in Table 1. At the end of the vermicomposting, the final C/N ratios were 14.4; 14.9;

16.7; 15.1 and 14.4 for the treatments 0%; 2%; 4%; 6% and 8%. There were no significant interactions ($P \geq 0.05$) between treatments and time (Table 5.1).

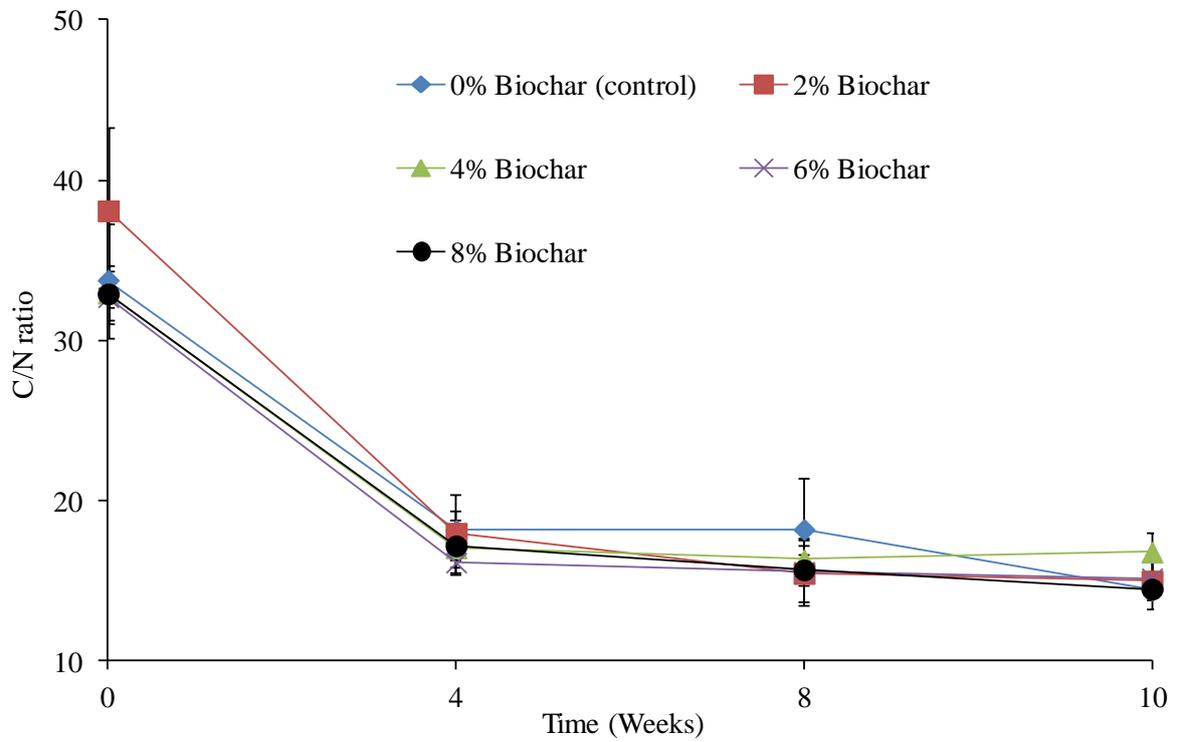


Figure 5.3 Changes in Carbon to Nitrogen ratio (C/N) during vermicomposting of a goat manure food waste mixture amended with seaweed (*G. funicularis*) biochar into. Error bars indicate standard deviation.

5.5.3 Humification parameters

For all the four humification indices measured which are humification ratio (HR), humification index (HI), percentage of humic acids (Pha), and polymerisation index (PI) there were no significant differences ($P \geq 0.05$) between all the treatments after 10 weeks of vermicomposting. The highest humification ratio was observed in the control treatment with a humification ratio of 16.1 whilst the lowest was the 4% treatment with the HR of 13.8. The HR values followed the order 0% > 8% > 2% > 6% > 4% with final values of 16.1; 16.0; 15.6; 15.3 and 13.8, for biochar treatments, respectively (Figure 5.4 A). For humification index, the control treatment had the highest value of 2.8 whilst the lowest was with the 4% treatment with a HI of 2.3. The final HI after 10 weeks of vermicomposting followed the order of 4% < 6% < 2% < 8% < Control (Figure 5.4 B).

Similarly, the control treatment also resulted in the highest value of percentage humic acids of 17.8 whilst the 6% treatment had the lowest Pha of 16.5. The Pha followed the order of 6% < 4% < 8% < 2% < control with 16.5; 16.7; 16.9; 17.1 and 17.8, respectively (Figure 5.4 C). For polymerization index, the highest value was observed at the control treatment with the PI of 21.6 whilst the lowest was 19.9 at 6% biochar treatment. The PI followed the order of 6% < 4% < 8% < 2% < control with 19.9; 20.1; 20.4; 20.6 and 21.6 (Figure 5.4 D).

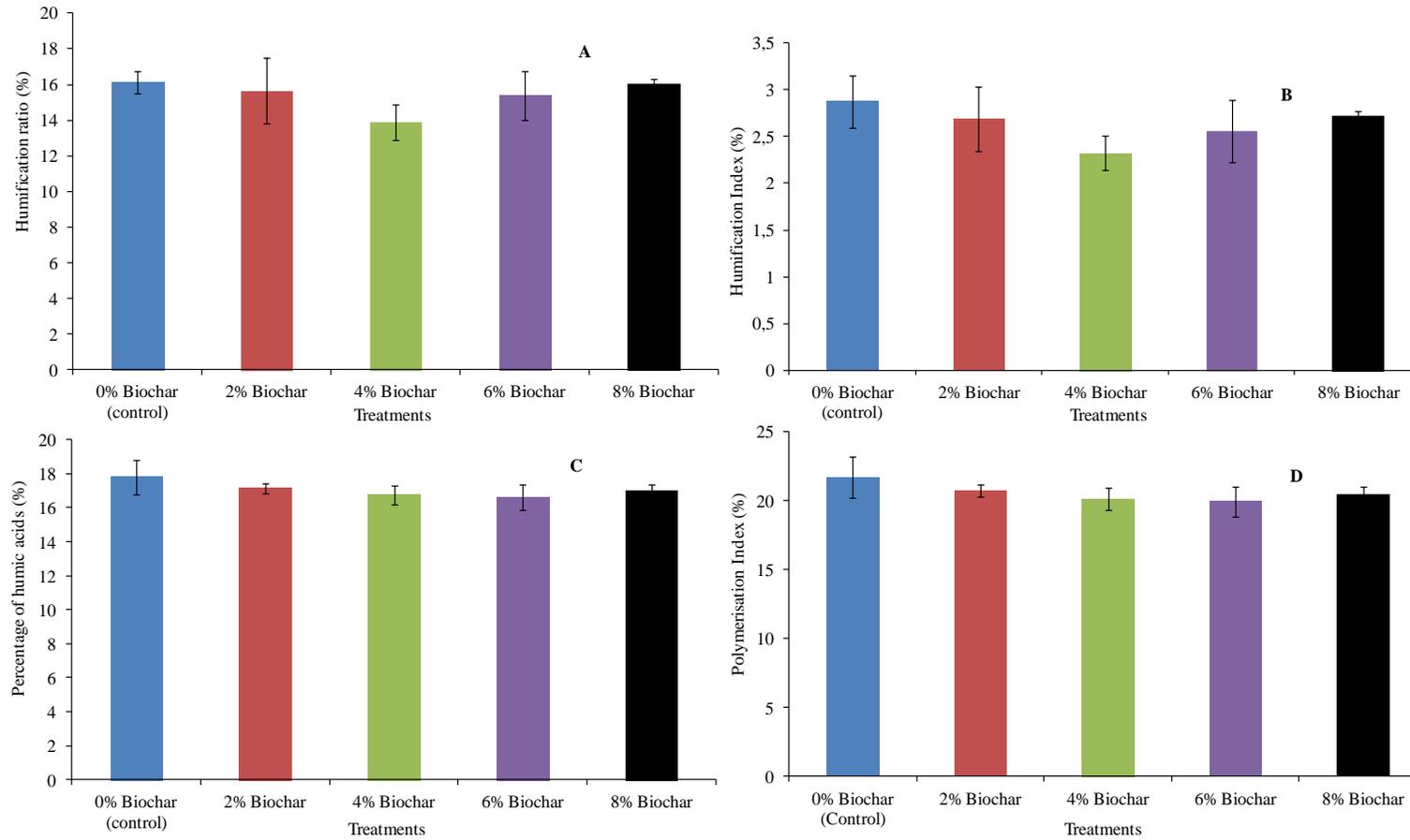


Figure 5.4 Final humification parameters after 10 weeks of vermicomposting of a goat manure food waste mixture amended with seaweed (*G. funicularis*) biochar. A (Humification ratio); B (Humification index); C (Percentage of humic acids) and D- (Polymerisation Index).

5.5.4 Effects of seaweed biochar incorporation during vermicomposting on nutritional content

Olsen extractable phosphorus

There were significant differences ($P \leq 0.05$) observed between treatments on changes in Olsen phosphorus as shown in the repeated measures ANOVA Table 5.1. The 8% treatment gave the highest final concentration of Olsen extractable phosphorus of 0.40 g per kg whilst the 2% treatment had the lowest final concentration of 0.30 g per kg of compost at ten weeks. It was interesting to observe that the changes in Olsen P during vermicomposting could be modelled using a second order polynomial equations with very high R^2 values (Figure 5.5). For all the treatments, the peak Olsen phosphorus was observed between week 6 and week 8 of vermicomposting and after 8 weeks, almost all the treatments started showing a decrease in Olsen P concentration. After 10 weeks of vermicomposting the final percentage change in Olsen P was 19%; 14.2%; 7.3%; 4.1% and 3.0% for 8%; 6%; 0%; 4%; and 2% treatments, respectively (Figure 5.5).

For the control treatment, the peak concentration of Olsen P was observed at week 4 with a concentration of 0.37 g/kg which was a 7.3% increase in Olsen P compared to the initial concentration. The peak concentration for 2% biochar treatment for Olsen P was observed at week 4 with a concentration of 0.38 g/kg with a 3.0 % increase. For the 4% treatment the peak concentration of Olsen P was observed at week 8 with a concentration of 0.40 g/kg with a 4.1 % increase in Olsen concentration. The peak concentration of 6% treatment was observed at week 4 with the Olsen concentration of 0.41 g/kg with a 14.2 % increase in Olsen P. For the 8% treatment the peak Olsen concentration was 0.42 g/kg observed at week 8 with 19% increase in Olsen P relative to the starting values (Figure 5.5).

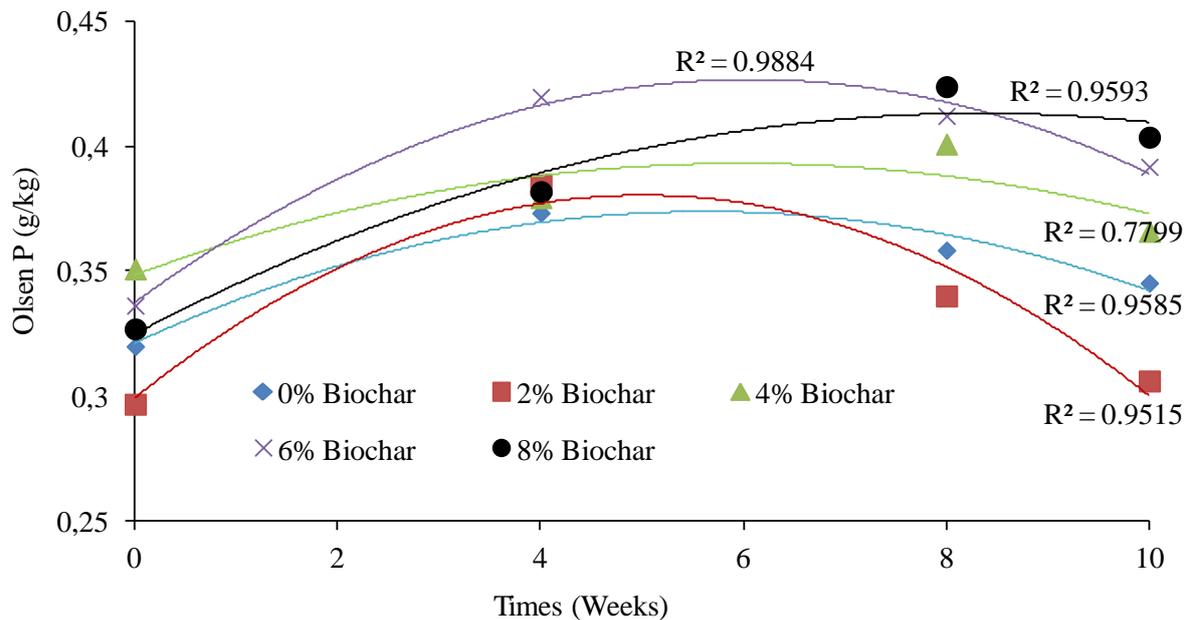


Figure 5.5 polynomial line graphs showing the trends of changes in Olsen P after ten weeks of vermicomposting of a goat manure food waste mixture incorporated with seaweed (*G. funicularis*) biochar.

5.5.5 Extractable cations (Ca, Mg, Na and K)

There were significant differences ($P \leq 0.05$) observed between treatments for Mg, K, and Na except for Ca as shown in repeated measures ANOVA (Table 5.1). For all treatments except for the control, the concentration of calcium increased from week 0 until week 8 and thereafter significantly decreased to concentrations that were below the original concentrations. However, the control treatment showed an almost constant concentration of calcium across the 10 weeks of vermicomposting. After ten weeks of vermicomposting the final concentration of calcium was observed to be high in the control followed by 2%; 4%; 8% and 6% treatment with the final concentration of 18.9 g/kg; 16.2 g/kg; 15.7 g/kg; 15.5 g/kg and 15.3 g/kg (Figure 5.6 A). It was observed that the final concentration of calcium for all treatments was lower than that of the treatment without biochar.

Similar to the changes in electrical conductivity, the higher the concentration of biochar incorporation rate the higher the concentration of potassium. For all treatments the concentration was almost consistent across the 10 weeks of vermicomposting, whilst it increased only for the 8% biochar concentration (Figure 5.6 B). The final concentration of potassium followed the order 8% > 6% > 4% > 2% > 0% with the final concentration of 27.4 g/kg; 15.8 g/kg; 15.0 g/kg; 14.1 g/kg and 6.4 g/kg after 10 weeks of vermicomposting. For all treatments the concentration of magnesium increased until week 8 and thereafter decreased (Figure 5.6 C). The final concentration of magnesium followed the order 0% > 2% > 4% > 8% > 6% with the final concentration of 0.59 g/kg; 0.58 g/kg; 0.58 g/kg; 0.55 g/kg and 0.52 g/kg at 10 weeks (Figure 5.6 C). Generally, sodium showed a small decrease for most of the treatments across the 10 weeks of vermicomposting. After 10 weeks, the higher the biochar incorporation rate the higher the concentration of sodium. The final concentration of sodium followed the order 8% > 6% > 4% > 2% > 0% with the final concentration of 5.44 g/kg; 3.45 g/kg; 3.29 g/kg; 3.2 g/kg and 2.2 g/kg at 10 weeks (Figure 5.6 D).

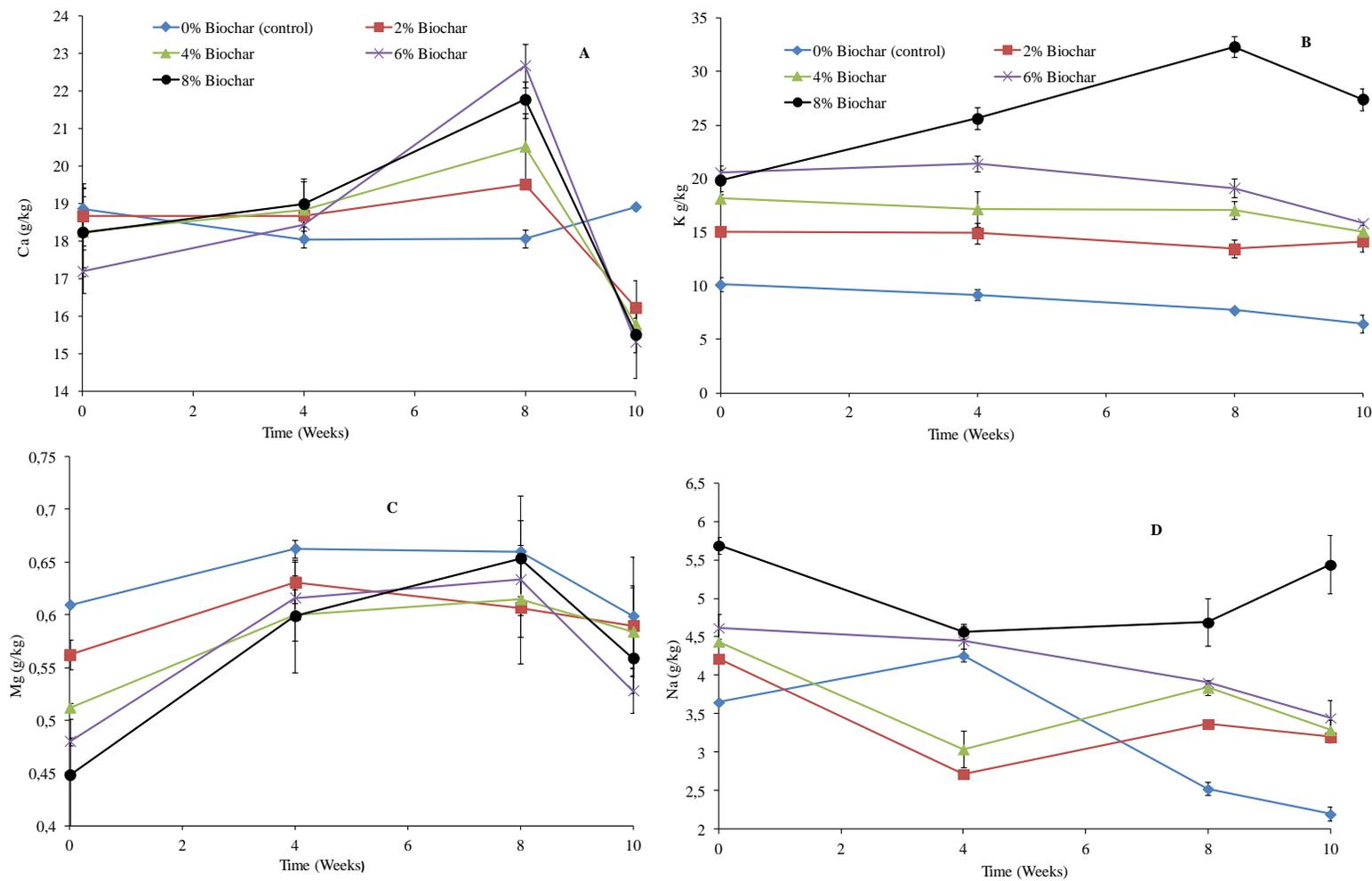


Figure 5.6 Changes in extractable A (Ca); B (K); C (Mg) and D (Na) after 10 weeks of vermicomposting of a goat manure food waste mixture amended with seaweed (*G. funicularis*) biochar. Error bars indicate standard deviation.

5.6 Discussion

5.6.0 Influence of seaweed biochar incorporation on vermicomposting

According to results in the study, the alkalinity of the pH is probably from the materials used which are goat manure and food waste which had a pH of 9 and not due to the application rate of biochar. The increase in pH across all treatments could be attributed to the release of ammonia and calcium as suggested by Karwal and Kaushik (2020) and this has been attributed to earthworm's activity. Furthermore, the pH increase has also been reported to be attributed to the increase in ash formation and mineralization of organic nitrogen as a result of microbial activities (Jain et al. 2018). The alkaline vermicompost may present an opportunity to use it as an amendment for acidic soils though there is no clear link between the level inclusion of biochar and the changes in pH. Though the pH was alkaline it was still within the region where most of the macro-nutrients needed by the plants will still be bio available as recommended by Simms and Mahato (2020). In our study, it was clear that the higher EC observed is attributed to the level of biochar incorporation. The increase in EC is attributed to the high levels of cations mainly calcium and potassium that are present in the biochar hence the higher the biochar level the higher the EC. Higher EC reflects the presence of more soluble salts, metabolites like ammonium and inorganic ions that are produced by earthworm's activities during vermicomposting (Lukashe et al. 2019). It will be critical to monitor the changes in electrical conductivity when seaweed biochar amended vermicompost is incorporated into the soil as the EC value of beyond 4 mS/m has been reported to result in soil salinity (Sarfaraz et al. 2020).

The incorporation of biochar did not influence the trend of change in C/N ratio as all the treatments followed the same trend. C/N ratio is the key indicator of biodegradation during the vermicomposting process (Karwal and Kaushik 2020) and C/N of less than 20 has been reported to represent mature compost though a C/N ratio of below 15 represent a much more

stable compost (Bernal et al. 2009). In our study it was observed that the treatments that had biochar incorporated at 6 to 8% resulted in much more stable compost though even the control resulted in mature compost with a C/N ratio of below 20. The decrease in C/N ratio may be due to the accumulation of nitrogenous compounds, the release of CO₂ by earthworm metabolism and enzyme-microbe induced decomposition of organic matter (Bhat et al. 2015; Karwal and Kaushik 2020; Alidadi et al 2016; Zhi-wei 2019). This was also observed in a study by Ravindran and Mnkeni (2016) who reported that C/N ratio decreases may be due to a higher loss of carbon accompanied by an increase in nitrogen during vermicomposting of waste paper and chicken manure. In the study the decrease in C/N may have been a result from the rapid breakdown of organic matter for microbial metabolism. When you incorporate biochar into compost you may get faster maturity relative to the control without biochar. Inclusion of biochar does not result in reduced decomposition but rather enhanced decomposition as observed in our study.

The humification parameters showed a different trend with the control having the highest humification parameters though they were not significantly different from the other treatments. As observed in our study, incorporation of biochar does not result in reduced humification parameters. Mature compost has been indicated to have a humification ratio of > 7 which was achieved in all treatments in our study (Bernal et al. 2009). However, according to (Bernal et al. 2009) the humification index, polymerisation index and percentage of humic acids were all below the recommended levels for mature compost. This may be because the humification takes place later after the initial decomposition during vermicomposting; therefore, it might be interesting to do this vermicomposting over a longer period to see if humification can be improved. It's interesting that the C/N ratio indicated mature compost unlike the humification parameters. There is still need to evaluate other parameters other than humification parameters that are critical in the evaluation of compost maturity (Li et al. 2015)

5.6.1 Influence of seaweed biochar incorporation on nutrient transformations

An incorporation rate of biochar of between 6 and 8% resulted in the highest Olsen extractable P after 10 weeks of vermicomposting. The peak concentration of Olsen P was observed between week 6 and 8 which unfortunately was when the compost was not yet matured. The decline in Olsen P may be due to leaching of the nutrients in the leachate. The higher the biochar the higher the concentrations of Olsen P observed. The higher concentrations in vermicomposts may be due to various earthworm activities during vermicomposting with the enzymes that help the release of phosphorus from feedstock (Sharma and Garg 2020). According to the food and Agricultural Organisation of the United Nations (FAO 2001), the potassium content in organic fertilizer should not be less than 1.5% while the concentration of calcium and other essential micro nutrients should be in the range from 0.01% - 0.05%. In our study the concentration of potassium was above 1.5% which means incorporation of biochar may enhance the concentration of potassium in composts. Zhang et al. (2016) found that the concentration of Na ions was lower in the composted mixture amended with biochar as compared to mixture without biochar addition. However, in our study that was not the case and this might be attributed to the origin of the material that was used to prepare the biochar which is from the marine environment where salinity is higher. It will be important to monitor the changes in soil physical properties when this seaweed biochar amended vermicompost is used as soil amendment. The increase in exchangeable calcium and magnesium be ascribed to the higher content of these nutrients in the materials used (Madiwe et al. 2020). In our study, that was not the case; the increase in Calcium and magnesium could be attributed to the effect of organic acids produced during the process of decomposition which enhances the solubility of Calcium and Magnesium. The incorporation level of biochar increased the potassium concentration and this may be due to the biochar used which had high amounts of potassium from the first experiment.

5.6.2 Conclusion

The study observed that biochar incorporation does not really influence the biodegradation process. In terms of nutrients, the seaweed biochar resulted in significantly higher Olsen extractable P levels as compared to the control. An optimized level of 6 to 8 % biochar incorporation ratio can be recommended in terms of nutrition and decomposition during vermicomposting of food waste-goat manure mixture. During vermicomposting, the peak concentration of elements such as P was observed before the vermicompost indicated maturity based on C/N ratio and humification parameter. The study recommends to evaluate the changes in soil parameters and plant growth when vermicompost with seaweed biochar incorporated between 6 to 8% is used.

CHAPTER 6

6. GENERAL DISCUSSION, CONCLUSIONS AND RECOMMENDATIONS

6.1 GENERAL DISCUSSION

The introduction of green revolution technologies has led to the use of huge quantities of inorganic fertilizers such as urea and other compound NPK fertilizers, which increased the yields among most farmers. However, over the years, this has led to soil and environmental pollution as well as degradation of the soil quality (Verma et al. 2020). These green revolution technologies tend to only feed the crop whilst ignoring to feed the soil which is living, hence soil degradation has been observed by most smallholder farmers. Recently there has been a shift towards organic agriculture as a sustainable technology that allows nutrients to be fed to the soil and then the soil can feed the crop (Tripathy and Khan 2020).

The realization of the importance of soil in crop production has seen a drive towards technologies like conservation agriculture, sustainable intensification, organic agriculture, to mention a few. However, the major challenge with the use of organic nutrient sources is the lack of the organic materials that are rich in macro nutrients such as nitrogen and phosphorus, comparable to that present in inorganic fertilizers. Though composts have been used over several years, they have been reported to lack a lot of nutrients when compared to inorganic fertilizers (Verma et al. 2020). Therefore, current research has been driving towards the identification of various materials that can be used as nutrient amendments for this organic fertilizer, such as fly ash, rock phosphate and most recently biochar (Katakula et al. 2020; Mupambwa et al. 2020).

In the study, a unique type of biochar was prepared which was developed from marine biomass as much of the research on biochar has focused on using materials of terrestrial origin (Huang

et al. 2020). The results showed highly significant differences on chemical properties between the biochar prepared from the two seaweed species (*Gracilariopsis funicularis* and *Laminaria pallida*) with alkaline pH. This may be attributed to the feedstock and pyrolysis temperature used during the biochar production (Roberts et al. 2015). The alkaline pH observed in the biochar from *Gracilariopsis funicularis* and *Laminaria pallida* can create an opportunity for the use of this biochar as a pH controller in acidic soils similar to what was recommended with alkaline fly ash by Mupambwa and Mnkeni (2015).

A pyrolysis temperature of 400°C was observed to give biochar with the best quality in terms of macro and micro nutrients, pH and total C for both seaweed species. However, it was also observed that the seaweed based biochar contained very small quantities of nitrogen, but very high phosphorus and other macro-nutrient concentrations. The heavy metals measured in our study included Zn, Cr, Cd, Ni, As and Pb, and these were not really a major challenge in terms of their concentrations with our marine biomass derived biochar; which meant that the prepared biochar may be used safely as a nutrient source in organic agriculture. However, only Cd was above the permissible limit and this might need to be monitored. Though the heavy metals in this biochar were lower than permissible limit, there is need to monitor the concentration of heavy metals if this type of biochar applied successively as a soil amendment.

The two seaweed species that were evaluated in the study showed different chemical characteristics; therefore, it will be interesting to evaluate other different seaweed species that are under-utilized in Namibia such as *Ulva* spp and *Porphyra capensis*. Furthermore, there is limited information on the quantities of biomass that is beach-cast from marine environments in Namibia. There is therefore a need for research to quantify the harvested biomass in order to evaluate the feasibility of harvesting this biomass to sustainably produce marine biomass biochar for organic soil fertility management in Namibia. In our study, we have also observed that marine biomass from the two seaweed studied can result in very good yield, high carbon

concentrations and high total concentration most of the macro nutrients (Katakula et al. 2020). However, relative to the total concentrations of most essential plant nutrients, the concentration of these bio available nutrients in the biochar was not much; this means that there is an opportunity to improve the bio available nutrient concentrations through the processes such as vermicomposting.

One of the main reasons why marine biomass can have potential in arid countries such as Namibia is the lack of terrestrial productivity because of the aridity of the environment. Due to the aridity of the Namibian environment, much of the fruit and vegetables are actually imported from various countries. This generates a lot of food waste due to transportation that causes post-harvest losses. Furthermore, most of the farmers keep small stocks such as goats and sheep which are highly adapted to the arid environment of Namibia, which generate large quantities of animal manures. Most of these wastes such as food waste and animal manure are dumped into open areas causing environmental pollution (Lukashe et al. 2019).

These wastes like animal manures and food wastes have a potential to be used as organic sources, which may be recycled and transformed into rich nutrient sources through the process of vermicomposting. Goat manure is one of the underutilized resources that a lot of farmers may use as an amendment but there is limited information on the nutrient composition. The study evaluated the optimization of food waste and goat manure mixture and we have observed that a mixture of 75% food waste and 25% goat manure as well as 50% food waste and 50% goat manure, achieved superior compost in terms of macro-nutrients and biodegradation. After 12 weeks of vermicomposting it was observed that maximum final Olsen phosphorus was 0.96 g/kg for 50FW:50GM. It was interesting that phosphorus and nitrogen were predictable using the second order polynomial equation and this can be critical in developing nutrient predictable organic fertilizers. Though food waste and goat manure materials showed some significant concentration of nutrients there is still an opportunity to use materials such as biochar to

improve the concentration of macro elements. Therefore, the next study used the biochar from experiment 1 and the best combination of food waste and goat manure with the biochar as an amendment to improve nutrients in the vermicompost.

In the last experiment, seaweed (*Gracilariopsis funicularis*) biochar produced at a temperature of 400°C from experiment 1 was incorporated into goat manure-food waste vermicompost at different levels between 0% and 8%, for optimized vermi-degradation and nutrient release. The incorporation of biochar did not seem to influence the biodegradation and maturity of the vermicompost across the various incorporation rates. In fact, based on the recommendations of Bernal et al (2009), incorporation of the biochar resulted in a vermicompost that was not mature based on the humification properties. In this study, the incorporation rate between 6% and 8% biochar resulted in an alkaline pH throughout the vermicomposting process with the high values of EC above 4 mS/cm.

The 6% and 8% had the highest quality compost based on the parameters such as C/N, humification ratio and Olsen P. High concentration of potassium of 32.3 g/kg at week 8 was reported to be due to the high incorporation rate. Therefore, 6% and 8% levels were found to be most appropriate as they gave a vermicompost with desirable maturity and nutritional properties. Marine biomass such as seaweed have a great potential to be used as alternative organic fertilizers since they are rich in essential macro and micro elements that are important to the soil and plants (Emadodin et al. 2020). In Namibia organic agriculture plays a role in a way that it contributes to mitigating the greenhouse effect and global warming through the ability to sequester carbon in the soil by promoting biochar to be used as an amendment (Katakula et al. 2020).

6.2 Conclusions

1. Biochar prepared from marine resources like seaweed has the potential in organic soil fertility management as this biochar was observed to contain significantly high concentration of essential nutrients such as P, K, Mg, Ca and Mg with reduced concentration of heavy metals. *G. funicularis* gives biochar with superior nutritional qualities compared to that of *L. pallida* and the pyrolysis temperature of 400°C which gives the best biochar quality across both seaweed species.

2. A ratio of 50% food waste and 50% goat manure yields more superior compost compared to other ratios where food waste or goat manure increased. During vermicomposting of food waste and goat manure the concentration of phosphorus and nitrogen can be modelled using the 2nd polynomial equations. Our study peak concentration of nitrogen and phosphorus were observed between weeks 8 and 10.

3. The higher inclusion of biochar levels might result in elevated electrical conductivity. In terms of nutrients, *G. funicularis* biochar resulted in significantly higher Olsen extractable P levels as compared to the control of only food waste and goat manure with no biochar. An optimized level of between 6% to 8 % *G. funicularis* biochar incorporation can be recommended in terms of nutritional and biodegradation.

6.3 Recommendations for future studies

1. Our study only collected the seaweed biomass in one sampling. There is a need to further evaluate the chemical composition of the biochar and uncharred biomass from different seaweeds abandoned along the entire Namibian coast and collected over different seasons. More studies should be done to quantify the actual amount of beach cast biomass harvested

along the coast in order to determine the quantity of beach cast produced per season in form of seaweed.

2. There is need for further evaluation of compost made from goat manure-food waste amended with biochar into actual plant based experiments for vegetable production in order to evaluate the response of the soil and crops made from the compost produced from biochar amended to food waste-goat manure. Future studies should assess the actual amount of food waste produced in various industries in order to evaluate the quantity to be used for fertilizers as vermicompost.

3. There is also need to evaluate if nutrients can be leached from compost that was amended with biochar, to produce liquid organic nutrient solutions for hydroponic crop production in arid countries such as Namibia. More available organic materials to be used from different regions to develop biochar for amendment into soil for nutrient sources.

4. The study recommend the use of organic materials for fertilizers in Namibia especially in the northern regions where the soils are poor and low in organic matter.

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The potential fertilizer value of Namibian beach-cast seaweed (*Laminaria pallida* and *Gracilariopsis funicularis*) biochar as a nutrient source in organic agriculture



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ABSTRACT

Biochar has been reported as an efficient tool for carbon sequestration and soil quality improvement, though most research has evaluated the quality of biochar from terrestrial based materials. The study evaluated the macro; micro nutrients and heavy metal concentrations of biochar from seaweeds from Namibian coast. Seaweed species *Laminaria pallida* and *Gracilariopsis funicularis* were pyrolysed at different temperatures i.e. control (uncharred); 200; 400; 600 and 800 °C in order to determine the chemical and physical properties of the biochar produced. There were significant interactions ($P < 0.05$) between temperature and seaweed species on most parameters measured. A pyrolysis temperature of 400 °C resulted in an almost 50% reduction in biochar yield, with higher temperatures resulting in even more yield loss whilst producing biochar with higher alkaline pH above 7 and high EC above 20 mS/cm. *G. funicularis* biochar resulted in the highest macro element content with total C 38.3%; N 4.3% and P 6.3 g/kg, whilst *L. pallida* biochar had the highest contents of cations of 16.2 g/kg Ca; 6.4 g/kg Mg; 151 g/kg K and 45 g/kg Na. Higher pyrolysis temperatures did not result in consistent changes in total heavy metal content, with only Cd content being above the maximum permissible limits for biochar of 3.9 mg/kg. Generally, a pyrolysis temperature of 400°C was observed to give biochar with the best quality in terms of macro elements, pH and total C. *G. funicularis* biochar showed significantly higher nutrient concentrations, therefore has great potential in soil quality improvement.

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Optimized vermicomposting of a goat manure-vegetable food waste mixture for enhanced nutrient release



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ABSTRACT

In arid countries, goats are the preferred small livestock and these generate dung that can be used as manure to improve the soil quality. Furthermore, in arid countries like Namibia, most of the fruits and vegetables are imported, and during transportation, damage and deterioration may occur, which results in losses. This study seeks to optimize the vermicomposting of goat manure and vegetable food waste to generate a nutrient rich organic fertilizer using *Eisenia fetida* earthworms. The influence of various ratios of vegetable food waste (FW) and goat manure (GM) mixtures on vermi-degradation and plant nutrient release was evaluated. There were significant differences ($P < 0.05$) on humification parameters across treatments. Biodegradation measured as changes in Carbon/Nitrogen ratio and humification properties followed the order of 75FW:25GM > 50FW:50GM > control (100GM) > 25FW:75GM. Similarly, the 75FW:25GM and 50FW:50GM treatments had the highest final concentration of Olsen phosphorus of 0.98 and 0.96 g per kg of compost, respectively. Relative to the control, the treatments with 75%; 50% and 25% FW resulted in a 113%; 109% and 60% more Olsen phosphorus, respectively, after 12 weeks of vermicomposting. There were no significant differences ($P > 0.05$) across all treatments on nitrate/nitrite concentrations following 12 weeks of vermicomposting. The changes in plant available P and N during vermicomposting were predictable using second order polynomial equations. This study concludes that a compost mixture of the ratio 75FW: 25GM or 50FW:50GM will give an optimized vermicompost for plant P nutrition after about 10 to 12 weeks of vermicomposting.

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