APPLYING SPENT COFFEE GROUND AS AN ORGANIC SOIL AMELIORANT IN THE LIMPOPO PROVINCE, SOUTH AFRICA

By

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DECLARATION

I declare that the mini-dissertation titled ‘APPLYING SPENT COFFEE GROUND AS AN ORGANIC SOIL AMELIORANT IN THE LIMPOPO PROVINCE, SOUTH AFRICA’ hereby submitted to the University of Limpopo, for degree Master of Science in Soil science has not been previously submitted for any other degree at any institution, is my work in design and in execution, and that all sources used have been acknowledged by use of references.

Motlanthi M	10/10/2022
Surname, initials	Date

ABSTRACT

The constant growth experienced by the coffee industry has led to the high-volume production of coffee waste worldwide. One of the main coffee wastes is spent coffee ground (SCG), a residue obtained after the ground coffee beans are treated under pressure. The present study was aimed to investigate the utilization of SCG to amend soil physicochemical properties. This study was conducted at Greenhouse Biotechnologies Research Centre of Excellence, University of Limpopo, South Africa, where the effect of various rates of SCG concentration in volume percentage (vol%) was tested for a period of nine months. The spent coffee ground residue was collected from four restaurants at Haenertsburg, and the application rates were 0, 5, 10, 20, 30, 50 vol%. To evaluate the change in soil physicochemical properties overtime, the incubation period was divided into four test periods namely T1 was after a month, T3 after 3 months, T6 after 6 months, and T9 after 9 months.

Physicochemical properties including nitrogen (N), phosphorus (P), potassium (K), magnesium (Mg), calcium (Ca), total organic carbon (TOC), cadmium (Cd), copper (Cu), nickel (Ni), zinc (Zn), and lead (Pb), pH, electrical conductivity (EC), C:N ratio, large macroaggregates (LM), small macroaggregates (sM), microaggregates (m), unaggregated silt and clay (s+c), mean weight diameter (MWD) and soil moisture content (SMC) were quantified at the end of each test period.

Results revealed that the interaction between incubation periods and various SCG application rates significantly ($p<0.05$) increased pH_w, EC, MWD, LM, base cations and significantly decreased TOC, heavy metals, SMC, m, and sM. Spent coffee ground increased pH_w and EC of the soil at all application rates and reached a maximum of 7.8 units at T6 in treatment SCG-5 and 202.30 S/cm at T9 in treatment SCG-50 above the control respectively. Total organic carbon increased by 548% above control in the highest treatment (SCG-50) at T1, but, however, started declining from T3 in all treatments across the incubation period.

SCG's highest application rates (SCG-20 to SCG-50) reduced the soil Cd toxicity (threshold of >2 mg/kg), but however, also reduced the availability of micronutrients (Cu and Zn) during the incubation period. At T9, Mg, Ca, K, and P increased from mean values of 55.9 to 77.9, 40.9 to 62.2, 77.4 to 112, and 22.0 to 30.0 mg/Kg above control in treatments with high application rates. LM increased whilst sM, and m decreased across the incubation period in all treatments. MWD increased by 46% at T1 and reached its maximum of 56% at T6 in treatment SCG-50 above control. Additionally, there was a positive relationship between LM and MWD. Soil moisture content however increased to 60.26% at T1 in treatment SCG-50 and decreased from T3 across the incubation period.

Spent coffee ground has the potential to be used as a liming material, a chelating agent, and for water management in semi-arid areas. It retains and cycles nutrients and improves soil structure through aggregation. However, research should be done in field conditions to access the effectiveness of this residue.

Keywords: Spent coffee ground, bio-waste, incubation period, soil amelioration, soil fertility, sandy loam soil, heavy metal toxicity, soil fraction and stability, soil moisture content.

DEDICATION

This mini-dissertation is dedicated to my family and friends, thank you for being my source of strength.

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CHAPTER 1

INTRODUCTION

1.1. Background

In the production of espresso beverages or soluble coffee, the ground coffee bean is treated with hot water and the residue that remains is the spent coffee ground (SCG) (Cruz *et al*., 2012). Globally, coffee consumption has increased by 7% in the past 5 years from 155 million 60 kg bags in 2016/17 to 166 million 60 kg bags in 2020/21 (ICO, 2021). Consequently, such high consumption generates approximately 6 million tons of SCG residue annually and if not properly disposed of is detrimental to the environment (McNutt and He, 2019). South Africans generate more than 2 million tons of SCG annually, of which 93% end up in landfills around the country (Insight Survey, 2022). With lifestyles constantly changing according to global trends and researchers linking coffee consumption with some health benefits, the demand and trade of coffee are expected to increase strongly, so is the generation of SCG (ICO, 2021). Recent studies have shown innovative attempts to utilize this low-cost residue as a green source in agricultural production, either composting SCG with other organic material or applying it directly to the soil for vegetable growth (Liu and Price, 2011; Cruz and Cordovil, 2015).

The disposal of this organic residue in landfills from the environmental and economic standpoint is not necessary because value can be added to this residue by utilizing it as a soil ameliorant (Cervera-Mata *et al*., 2017). It contains organic components such as polysaccharides, acids and phenols, lipids, nitrogen compounds, and minerals that have beneficial effects on soil physicochemical properties and the microbial population in the soil (Ballesteros *et al*., 2014). Numerous researchers have reported that when SCG is applied in sufficient amounts, it replenishes essential plant nutrients, encourages soil aggregation, increases water holding capacity, and increases nutrient cycling microbial population (Cruz and Cordovil, 2015; Hardgrove and Livesley, 2016; Cervera-Mata *et al*., 2017). However, little has been documented on the effects of various SCG application rates on improving the soil physicochemical properties of Molopo soil form.

1.2. Problem statement

The constant growth experienced by the coffee industry has led to the high-volume production of coffee waste worldwide. According to the ICO (2021), global consumption of coffee bean for the period of 2020/21 has reached 166 million 60 kg bags. South Africa produces about 3500 thousand 60 kg bags and have been importing about 520 000 thousand 60 kg bags each year from 2011 to 2021 (ARC, 2014; ICO, 2021). With coffee being the most consumed and second traded commodity, these figures are only expected to increase (McNutt and He, 2019). It is estimated on average that a kg of coffee bean makes 2kg of wet SCG (Pfluger, 1975). These could only increase the disposal of SCG in landfills and sewage systems. The disposal of this residue in landfills produce methane and carbon dioxide, greenhouse gases that contribute to global warming. This study will gather insights on how interaction of SCG application rates and incubation period affects soil chemical and properties.

1.3. Rationale

Taking into account the adverse effects associated with chemical fertilizers on the environment, the development of sustainable management practices to restore degraded arable soil could be an innovation to ensure that soil ecosystems are brought to balance for productivity. The introduction of cheap organic fertilizers could be an intervention to reduce the impact of fertilizers on the soil. Spent coffee ground (SCG) is a bio-residue obtained from the treatment of ground coffee bean with hot water (extraction process) in the production of soluble coffee or espresso beverage (Mussatto *et al*., 2011).

Globally, approximately six million tons of SCG are dumped in landfills annually and it might be detrimental to the environment (McNutt and He, 2019). From the environmental and economic standpoint, dumping of SCG is unnecessary because the extraction process only extracts a small number of components, SCG remains enriched in components such as polysaccharides (52%), lignin (24%), protein (17%), fat (2%) and minerals (1%) in dry mass basis hence it’s a highly valuable resource that can be obtained at a cheap cost (McNutt and He, 2019; Mussatto *et al*., 2011).

Different from chemical fertilizers, SCG does not only replenish depleted nutrients in the soil but also serves other benefits such as adding organic matter to the soil, which increases the soil organic carbon, encourages soil aggregation, and further increases the microbial diversity and functionality (Hardgrove and Livesley, 2016; Cervera-Mata *et al*., 2017; Vela-Cano *et al*., 2019). However, when SCG is applied at greater application rates, it becomes detrimental to plant growth and soil microorganisms (Cruz and Cordovil, 2015; Vela-Cano *et al*., 2019). Improved understanding of which SCG application rate is suitable for improving soil properties can help in the development of sustainable management practices to restore exploited agricultural soil and improve production.

1.4. Purpose of the study

1.4.1. Aim

The study aimed to investigate the utilization of SCG as a soil ameliorant

1.4.2. Objectives:

i. To determine the effects of SCG's application rates and incubation periods on selected chemical properties (N, P, K, C, Mg, Ca, TOC, C:N ratio, EC, and pH) and heavy metals.
ii. To analyze the effects of SCG's application rates and incubation periods on soil aggregate fraction and stability, and soil moisture content.

1.4.3. Hypotheses:

i. Increased SCG application rates and incubation period will increase the concentration of N, P, K, C, Mg, Ca, TOC, C:N ratio, EC, and pH in the soil and reduce heavy metals concentration.
ii. Increased SCG application rates and incubation period will promote soil aggregate fraction and stability and increase soil moisture content.

CHAPTER 2

LITERATURE REVIEW

2.1. Work done on the problem statement.

2.1.1. Introduction

Coffee, regarded as the most popular beverage and ranked second most traded commodity after crude oil worldwide, is a tropical evergreen perennial plant that is native to Africa and widely produced for its stimulating and antioxidant cherry beans (Mussatto *et al*., 2011). With over 80 species classified in the Coffea genus, presently, the principal cultivated species in about 80 producing countries are *Coffea arabica* (arabica) and *Coffea canephora* (robusta) (ICO, 2021). Coffee production, from cultivation to beverage preparation, is a long and complex technological process. Therefore, several coffee waste residues are recovered (Fig 2.1) (Murthy and Naidu, 2012). The recovered residues are divided into two classes: the processing countries coffee waste consisting of pulp and/or husk and the consuming countries coffee waste consisting of silver skin and spent coffee ground (Cruz *et al*., 2012; Murthy and Naidu, 2012).

The present study focuses on SCG residue which represents the flux of organic waste generated from soluble coffee manufacturing industries and coffee brewing cafés in coffee-consuming countries (Mussatto *et al*., 2011). The world generates roughly six million tons of SCG residue, of which nations like South Africa and Australia reuse less than 10% annually while the remaining residue enters landfills (Mussatto *et al*., 2011; Cameron and O’Malley, 2016). A study conducted by Cruz *et al*. (2012) quantified the total soluble solids (TSS) in the coffee brew for both the popular *Coffea* sp., and findings indicated that TSS extractable in robusta and arabica were 29-37% and 26-32% respectively, illustrating that SCG is a valuable source of unextractable compounds. Hence, innovations to repurpose this residue are being carried out across disciplines and amongst them is the application in agriculture.

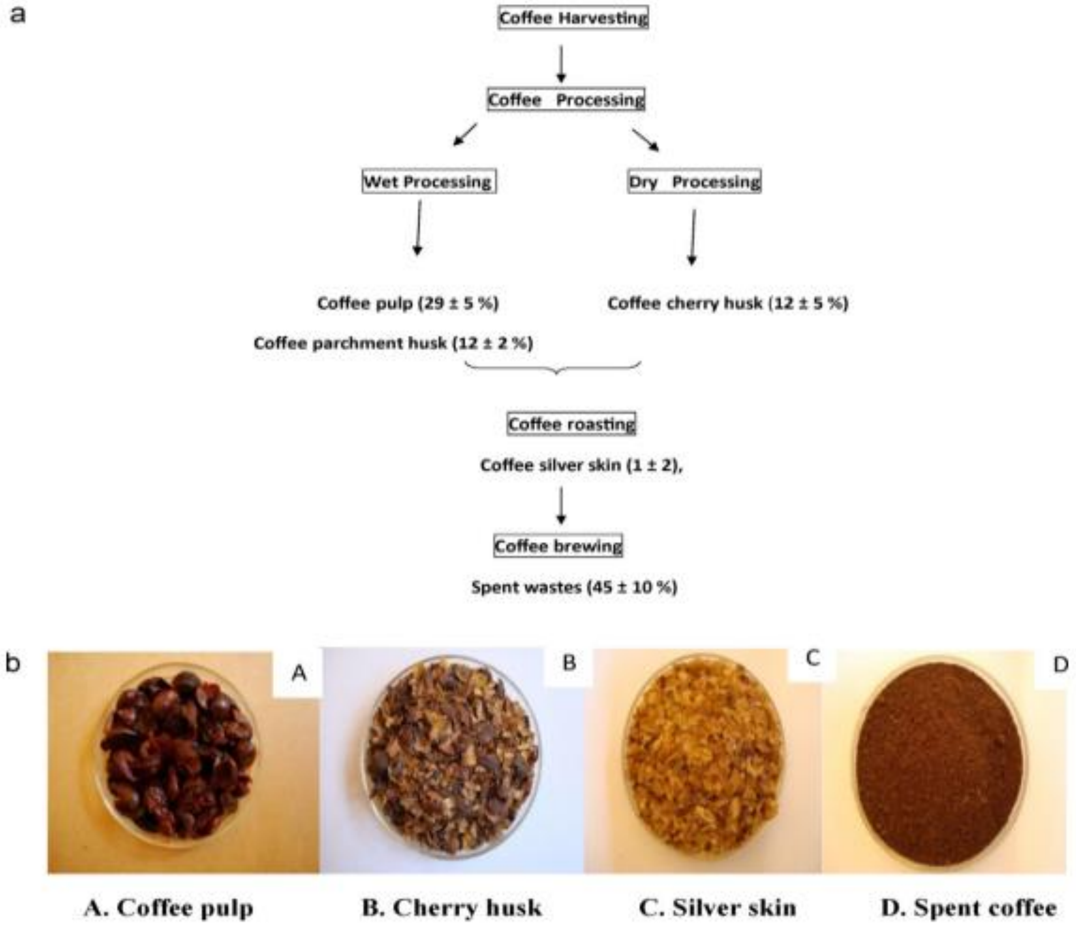

Figure 2.1: (a) Sketch of the production of various by-products from the coffee industry. (b) Coffee by-products obtained during coffee processing. Source: Murthy and Naidu, 2012

2.1.2. Spent coffee ground feasibility as an organic ameliorant

Incorporation of organic waste into the soil to improve soil properties is not a new concept in sustainable agriculture. It’s as ancient as time, the difference is the utilized organic waste. However, concerns arise when negative biological and phytotoxic responses are observed post organic waste application (Hardgrove and Livesley, 2016). Furthermore, environmental implications, feasibility, and slow nutrient release characteristics of organic waste should be evaluated for viability and efficacy of this waste. Cervera-Mata *et al*. (2017) and Cervera-Mata *et al*. (2019) brought to attention that when SCG is incorporated into the soil it contributed to improving soil chemical and physical properties while reutilizing millions of tons of SCG produced annually and mitigating climate change by incorporating carbon into soils. Supporting findings by Yamane *et al*. (2014) deduced that SCG had the potential to capture and store carbon dioxide geologically due to high levels of total carbon being recorded in the soil two years post-SCG application. A study by Santos *et al*. (2016) measured minimal greenhouse gases emitted from compost piles amended with SCG into the atmosphere. Considering the observations from these studies, it shows that utilization of SCG as a soil amendment incorporates carbon into the soil.

The outcomes of utilizing SCG are not only limited to carbon sequestration. A study conducted by Morikawa and Saigusa (2008), which utilized SCG in alkaline soil, indicated it increased plant-available iron and zinc. In addition, Kim *et al*. (2014), and da Silva Correia *et al*. (2018) indicated that utilization of SCG in soil and water contaminated with heavy metals (Pb, Ni, Cu, and As) decreased their bioavailability and accessibility thus illustrating their ability to adsorb heavy metals. Similar effects were evaluated for a herbicide, demonstrating that leaching of triazine herbicide residues through the soil was reduced post-application of SCG (Fenoll *et al*., 2014). Therefore, spent coffee ground is illustrated to be a potential chelating agent when it is incorporated into the soil. Additionally, incorporation of SCG into the soil was recorded to increase micronutrients (Cu, Fe, and Zn) for plant absorption (Cervera-Mata *et al*., 2017).

Despite efforts to reutilize this integrative resource, there are limiting factors. The chemical composition of SCG may limit the feasibility of a particular application. There are bioactive compounds that are known to be pollution hazards, induce phytotoxicity effect, or are of ecotoxicological concern (Janissen and Huynh, 2018; Cruz *et al*., 2012). For instance, Hardgrove and Livesley (2016) recorded both suppressions of weeds and crops post direct soil amelioration with fresh SCG and suggested composting SCG before application. Similar contributions were made by Cruz and Cordovil (2015), observing the reduction of plant germination and growth. Cruz *et al*. (2014) utilized fresh and composted SCG residue, and the finding on utilization of fresh SCG agreed with Hardgrove and Livesley (2016), whilst composted SCG indicated increments of essential macronutrients in crops. Moreover, the utilized compost contained a low rate of SCG.

Cruz *et al*. (2015) explored a different approach from early study and composted SCG in the soil before planting. The study illustrated that at 5 concentrations SCG had a positive impact on the crop's physical and nutritional characteristics. The study by Yamane *et al*. (2014) showed that SCG has a positive effect on plant production in the long term. Findings on other soil properties showed a reduction in soil microorganisms and increased mobility of arsenic heavy metal in soil due to high application rates of SCG (Kim *et al*., 2014; Vela-Cano *et al*., 2019). It is apparent from the contrasting results that the effects of SCG are dependent on the application form of SCG, application rates, and incubation period.

2.1.3. Spent coffee ground chemical and functionality properties

The potential use of SCG as an organic ameliorant lies in its chemical composition. Cultivation (geographic location, climate, soil conditions, plant species, and plant age) and beverage preparation (fruit cherry processing method, roasting and brewing procedure, and extraction efficiency of the industries and cafés) attribute to factors that subsequently influence the composition and quality of SCG (Mussatto *et al*., 2011; Cruz *et al*., 2012; Murthy and Naidu, 2012). There are a considerable number of publications on SCG chemical composition each presenting quantitatively varying quantities of unextracted compounds.

Ballesteros *et al*. (2014) investigated the chemical and functional properties of a mixture of robusta and arabica SCG generated from a café and findings indicated that it was chemically composed of 51.5% of polysaccharides (12.4% of cellulose and 39.1% of hemicellulose), 23.9% of lignin, 17.44% of protein, 2.29% lipids, nitrogen 2.79% and 1.30% ashes (minerals) in decreasing order on a dry mass basis (w/w). Pujol *et al*. (2013) recorded the lowest polysaccharides (22-24% w/w) as compared to the study by Mussatto *et al*., (2011) and Ballesteros *et al*. (2014), with lignin amounting to 26.51% w/w. Mussatto *et al*. (2011) recorded a protein content of 13.6% w/w not bound to amino acids and potassium was the highest of all other nutrients. This was supported in a study by Cruz *et al*. (2012), and Ballesteros *et al*. (2014). However, it disagreed with a study by Pujol *et al*. (2013), who reported that calcium was the dominant nutrient.

Components such as lignin constitute caffeine and chlorogenic acids which are polyphenolic compounds (Ballesteros *et al*., 2014). The presence of polyphenols was reported in numerous studies either being labelled antioxidant compounds for humans or bioactive toxic compounds for plants and microorganisms when leached into the environment (Cruz *et al*., 2012; Campos-Vega *et al*., 2015; Janissen and Huynh, 2018). Polyphenols like caffeine were reported to have a negative effect on plant, fungal and bacterial growth whereas tannins were marked to have a slow decomposing rate hence remaining in the environment for a longer period if applied at higher application rates (Janissen and Huynh, 2018). Moreover, tannins can adsorb heavy metals due to the polyhydroxy polyphenol functional group (Kim *et al*., 2014). These polyphenols were recorded to be a major contributor of nitrogen (46% of total nitrogen) upon decomposition (Campos-Vega *et al*., 2015). Vela-Cano *et al*. (2019) evaluated polyphenol and the findings agreed that SCG's highest application rate increased total phenol acids at initial application; however, mineralization of SCG decreased phenols concentration hence decreasing phytotoxicity. These studies demonstrated that understanding the composition and functionality of SCG compounds would result in its efficient application.

2.1.4. Effects of SCG on selected soil chemical properties

Spent coffee ground has been recorded in recent publications to supply the soil with basic nutrients required for plant growth and either showing liming or gypsum effects. However, factors such as SCG application rate, incubation period, and soil type have proven to have a major effect on the dynamics of pH and nutrients in the soil. For instance, a study conducted on alkaline soil (Andisols) in a pot experiment showed an addition of SCG decreased soil pH (became less alkaline) within a two-month period (Morikawa and Saigusa, 2008). In three months, Cervera-Mata *et al*. (2017), conducted a study on an alkaline soil pot experiment, which showed an initial pH decrease post SCG addition; however, it was followed by an increase in pH in line with the incubation period. In acidic soils, pH showed a similar trend of decreasing pH post addition and increasing in line with the incubation period; however, in a field trial there was a decrease in pH with an increasing incubation period (Hardgrove and Livesley, 2016).

The addition of the highest SCG application rate increased organic carbon (TOC), total N, C:N ratio, EC, K, and P. However, as the incubation period progressed, N increased whilst OC, C:N ratio, EC, K, and P decreased (Cervera-Mata *et al*. 2017). A study by Yamane et al. (2014) demonstrated that the highest SCG application rate caused a significant increment in soil TOC and N, but a decreased C:N ratio. A study conducted by Hardgrove and Livesley (2016) found nitrate immobilization after direct application of SCG. This was supported in a study by Cruz and Cordovil (2015), who found immobilization of nitrogen in all their incubations with SCG. The lowest concentration of bases (K, Ca, Mg) was recorded in a study by Morikawa and Saigusa (2008). Liu and Price (2011) indicated that the phosphorus increase was insignificant. Similarities in terms of phosphorus were recorded in a study by Cruz and Cordovil (2015), which detected the immobilization of phosphorus in all the incubations.

2.1.5. Effect of SCG on heavy metal abundance in the soil.

Soils can become polluted as a result of the high concentration of heavy metals in agriculture from pesticides, synthetic fertilizers, manure, and wastewater irrigation (Haider *et al*., 2021). Heavy metals are described as any metals that have a relatively high density, are poisonous and toxic at low concentrations, and are non-biodegradable (Zulfiqar *et al*., 2019; Haider *et al*., 2021). However, these metals are recognized as being essential (Zn, Cu, Fe, Ni) and non-essential (As, Cd, Pb, Hg).

Recent studies are aimed to remove heavy metals from aqueous solutions using SCG and its effectiveness is compared with recently used method such as activated carbon, Zeolite. A study by Davila-Guzman *et al*. (2016) studied the adsorption of heavy metals onto SCG and the findings indicated maximum absorbance of Cd^{2+}, $Cu^{2+,}$ and Pb^{2+} as compared to activated carbon. Similarly, Kim and Kim (2020) reported that Cd adsorption by SCG showed the highest Cd adsorption compared to zeolite. A study on adsorption of heavy metals in the soil by Kim *et al*. (2014) revealed that SCG decreased heavy metals (As, Cd, Cu, Pb, Ni, and Zn) concentration in the soil. In a study on the response of Italian ryegrass, SCG significantly reduced the absorption of Cu, Fe, Mn, and Zn due to immobilization of these metals in the soil (Kasongo *et al*., 2013). However, there are maximum permissible element concentration and plant deficiency level to consider when working with agricultural soils (Table 2.1).

Table 2.1: South African regional guideline for maximum permissible element concentration and plant deficiency level in agricultural soil

Elements	Permissible element Concentration (mg/Kg)	Plant deficiency level (mg/Kg)
Cd	2	-
Cu	6.6	1
Cr	80	-
Ni	50	-
Zn	46.5	3
Pb	6.6	-
Co	20	0.5

Cd = cadmium, Cu = copper, Cr = chromium, Ni = nickel, Zn = zinc, Pb = lead, Co = cobalt. (Source: Herselmana *et al*., 2005).

2.1.6. Effects of SCG on soil aggregate fraction and stability

A series of recent studies have indicated that amendment with SCG has a significant positive effect on some physical properties of the soil since it increases organic matter which improves soil structure (Murthy and Naidu, 2012; Kasongo *et al*., 2013; Hardgrove and Livesley, 2016). A study by Cervera-Mata *et al*. (2019) revealed that a SCG higher application rate and incubation period increased the proportion of macroaggregates and structural stability, while simultaneously decreasing bulk density and the proportion of meso- and microaggregates in a short term. The study further highlighted that the effect on the structural stability of aggregates for two months was like other organic amendments (compost, manure, and vegetable waste) for 12 years. This is of great significance in areas dominated by sandy soils which are susceptible to soil and water erosion.

2.1.7. Effects of SCG on soil water content

Soil water content indicates the quantity of water present in the soil, and it greatly depends upon soil texture and structure (Civeira, 2019). In sandy soil, the water content may vary from 3 to 10% from wilting point to field capacity and in clay soil from 20 to 40%, thus the optimum level of storage might be 20% of 1m of soil (Brandt, 2017). Practices such as the addition of organic material can help in retaining soil moisture thus assisting water management. A study by Kasongo *et al*. (2013), showed that increases in SCG amendment rates increased water retention, and this meant soils with higher SCG amendment retained most of the water. Spent coffee ground addition increased water content at -33kPa (field capacity) and -1500 kPa (permanent wilting point) compared to a soil not treated with SCG (Cervera-Mata *et al*., 2019). This was also supported in a study by Hardgrove and Livesley (2016), who observed a significant increase in soil moisture.

However, in waterlogging conditions (lower drainage) more water can be held (Brandt, 2017). Turek *et al*. (2019) revealed increased soil moisture, readily available plant water, reduced drainable porosity, and poor crop development post-application of SCG on sandy soil. Understanding the behaviour of the soil and how SCG holds and distributes water can bring about improved water management in semi-arid regions where water scarcity is a major problem.

2.2 Work not done on the problem statement.

Existing studies conducted on the effect of SCG amendment are soil-plant system and endpoint, thus don’t evaluate that the soil variables change overtime (Cervera-Mata *et al*., 2017). Hardgrove and Livesley (2016) conducted their study for 3 months which showed evidence of nitrate immobilization. More similar studies by various publications conducted for two months showed that soil parameters such as organic carbon, total N, and available K and P increased with an increasing incubation period (Cruz and Cordovil, 2015; Vela-Cano *et al*., 2019; Cervera-Mata *et al*., 2017). The results of their studies were limited to the growing season of the crop, therefore neglecting how soil parameters change over time. Diacona and Montemurro (2010) highlighted that the value of organic residue as soil ameliorants can be devalued as they tend to release nutrients in the next growing season. The present study evaluates SCG as an organic ameliorant for nine months on the agricultural utilized Molopo soil collected from Syferkuil farm in a pot experiment.

CHAPTER 3

METHODOLOGY AND ANALYTICAL PROCEDURES

3.1. Description of study location

The study was conducted at the Greenhouse Biotechnologies Research Centre of Excellence (23°53’10” S, 29°44’15” E), University of Limpopo, South Africa (Fig. 3.1). The maximum temperatures in the greenhouse are being controlled using thermostatically activated fans. The temperature averages 28°C during the day and drop to 21°C at night.

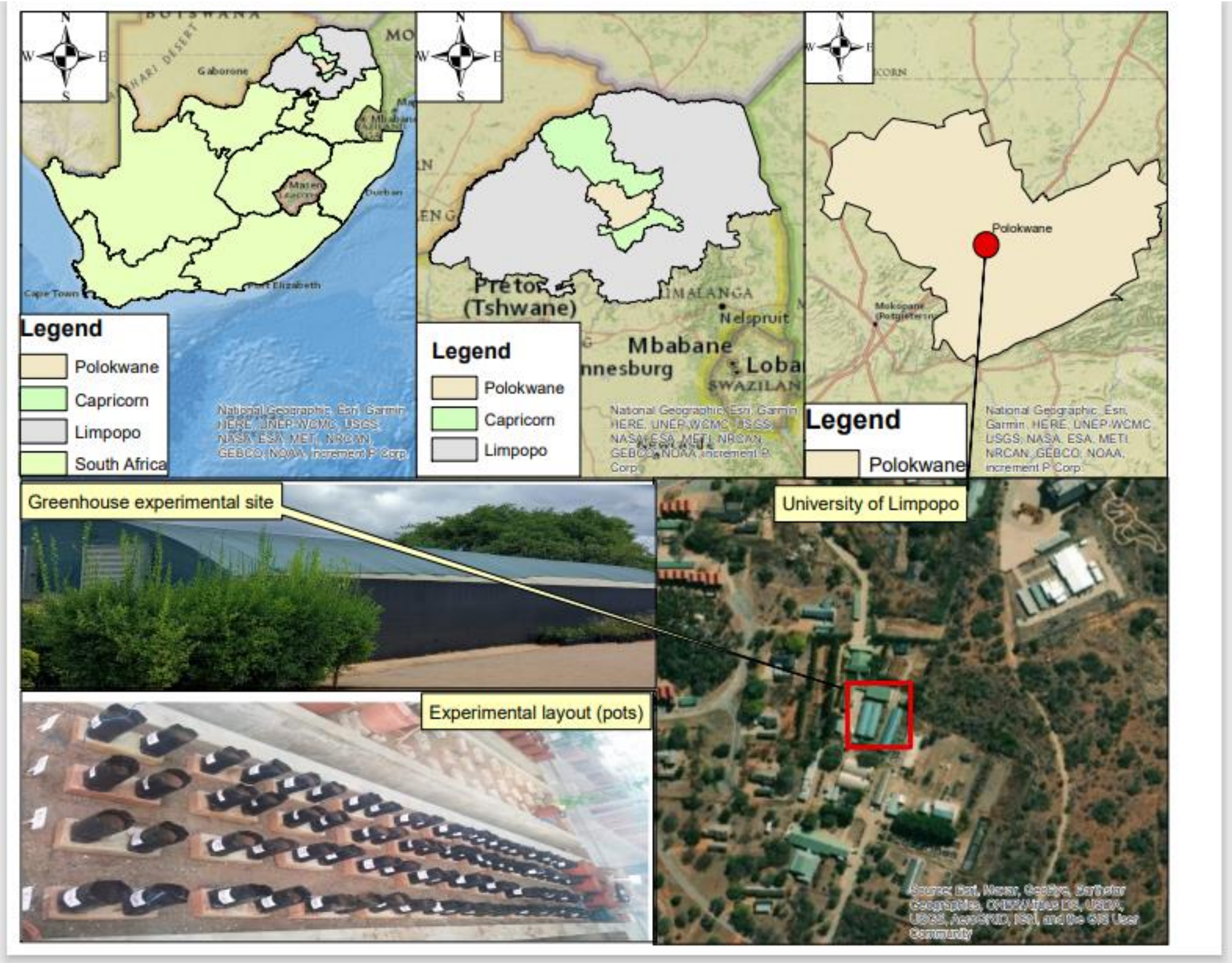

Figure 3.1: Location of the study site at the University of Limpopo Greenhouse Biotechnologies Research Centre of Excellence, Limpopo, South Africa. Shown are images of the experimental layout in a factorial (incubation period x application rate) design.

3.2. Experimental design

This study tested the effects of various rates of SCG concentration in volume percentage (vol%) for nine months. The SCG application rates were 0%, 5%, 10%, 20%, 30%, and 50% by volume. The incubation period was divided into four test periods namely T1 was after a month, T3 after 3 months, T6 after 6 months, and T9 after 9 months, hence 24 (6 SCG concentrations rates x 4 incubation periods) treatments. The soil amended treatments were 3000 cm^3 of soil plus various SCG concentration rates:

i. C – control (soil)
ii. SCG 5 – soil + 5% SCG
iii. SCG 10 – soil + 10% SCG
iv. SCG 20 – soil + 20% SCG
v. SCG 30 – soil + 30% SCG
vi. SCG 50 – soil + 50% SCG

Each treatment was thoroughly mixed and evenly distributed to the volume of 202.5 cm^3 (6 SCG treatments*4 incubation periods*4 replications) plastic bags and irrigated regularly to avoid moisture loss. To ensure successful mineralization of SCG the treatments were incubated in a controlled environment (greenhouse) with temperatures ranging from 28°C to 21°C.

3.3. Laboratory Analysis

Approximately 16 000 cm^3 of SCG was collected from The Eatery, Caffe' Villa Trattoria and The Red Plate restaurants at Haenertsburg and air-dried. The dry SCG was passed through a 2-mm sieve. Soil samples were obtained at Syferkuil Experimental Farm, under a fallowed land previously utilized for crop production experimental trails. Only the topsoil (0-15 cm) was sampled and combined to form a representative sample for the entire area. The composite sample was subjected to chemical (N, P, K, Mg, Ca, TOC, Cd, Cu, Ni, Zn, and Pb) and physical (aggregate fraction and stability, particle size distribution-hydrometer method by Bouyoucos (1962), and soil moisture content analysis. Additionally, at the end of each incubation period, the samples were dried and analyzed for chemical and physical analysis.

Achieving objective 1:

Chemical properties were determined with specific methods: soil pH was determined in a 1:2.5 soil to deionized water and soil to 1M KCl suspension ratio using pH meter (electrode method). The soil electrical conductivity (EC) was measured in a 1:5 solution ratio of deionized water using the Hanna EC meter. Nitrogen was measured using TruSpec–Leco instrument which uses the combustion method. Organic carbon was quantified using the ignition method (Davies, 1974) in which a sample ignited slowly in a muffle furnace to a final temperature of 550^{o}C for 8 hours. Calculations to compute for OC:

Ash percentage = $\frac{w3-W1}{W2-W1}$x100

Organic matter % = 100 - Ash %

$$\text{OC} = \frac{\text{Organic matter }\%}{1.72}$$

Where W1 represented the weight of the empty crucible, W2 was the weight of the crucible containing sample before ignition and W3 was the weight of crucible containing sample after ignition. C:N ratio computed from N and OC.

Exchangeable Mg, Ca, and K, available P and, heavy metals (Cd, Cu, Ni, Zn, and Pb) were analyzed using ICPE 900 instrument (Lin and Coleman, 1960). A 3.5 g sample of soil was extracted in acid (70 % HNO_3) with a PerkinElmer Titan MPS (microwave digestion), the extracts were then analyzed using ICPE 900 instrument.

Achieving objective 2:

Soil aggregate fraction and stability was determined using the wet sieving method (Elliott, 1986). The aggregate fractions were: i) >2000 µm (large macroaggregates), ii) 212-2000 µm (small macroaggregates), iii) 50-212 µm (micro-aggregates), and iv) <50 µm (silt and clay). Mean weight diameter (MWD), a measure of soil aggregate stability for each treatment was calculated using the following equation:

MWD = (2 x LM) + (1.106 x sM) + (0.131 x m) + (0.025 x (s + c))

Where MWD = aggregate stability, LM = percentage of large macroaggregates, sM = percentage of small macroaggregates, m = percentage of microaggregates, and (s + c) = percentage of unaggregated silt and clay in each soil sample.

Soil moisture content was measured using the gravimetric method (Black, 1965).

$$\text{Soil moisture content} = \frac{mass\ of\ moist\ soil - mass\ of\ oven\ dried\ soil}{mass\ of\ oven\ dry\ soil} * 100$$

3.4. Statistical analysis

Factorial (incubation period x application rate) analysis of variance (ANOVA) was undertaken using Statistix10 software to test the effect of various application rates of SCG on physicochemical properties across incubation periods at the significance level of 95% ($p < 0.05$). Where the F-values from treatments effect were found significant, means were separated using the Tukey HSD test at the significance level of 95% ($\alpha = 0.05$). Pearson correlation was used to determine the relationship amongst variables at 95% significance level using Statistix10. Regression analyses (R^2) was performed to determine the relationship between the incubation period and various SCG concentration on TOC, N and C:N ratio.

CHAPTER 4

RESULTS AND DISCUSSION

4.1. Results

4.1.1. Soil and SCG analyses

Spent coffee ground had the highest average values of EC, OM, TOC, N, C:N ratio, Cu and SMC of 872.00 S/cm, 98.00%, 56.99%, 1.81%, 32, 1.04% and 98.60% compared to the soil respectively (Table 4.1). The soil had the highest average values of Cd, Zn, Ni, Pb, Mg, K, Ca and P of 20.98, 4.07, 7.76, 5.07, 73.85, 63.77, 38.38 and 47.68 mg/Kg compared to SCG. The soil texture proportion distribution mean value of clay 2%, silt 10%, and sand 88% and the soil aggregate stability (MWD) of 0.56mm. The soil pH_W and pH_{KCl} was higher than in SCG with values 6.90 and 6.42 units.

4.1.2. Effect of spent coffee ground and incubation period on soil acidity

Soil pH_w

The interaction between different incubation periods and various SCG application rates had a significant effect ($P<0.05$) on pH_w (Table 4.2). Amelioration with SCG in the first test period (T1) reduced pH_w in all treatments compared to control (mean value of 7.5) with a drastic decline being observed in SCG-30 (mean value of 6.3) (Fig. 4.1). However, as the incubation period progressed to (T6), SCG-5 reached its maximum unit of 7.8 relative to control. The effect observed was that treatments with the highest application rates (SCG-50, SCG-30, and SCG-20) experienced the highest pH decline although they progressively and steadily increased across the incubation period. Whereas treatments with the lowest application rates (SCG-5 and SCG-10) raised the pH of the soil. Spent coffee ground's application rates negatively correlated with pH ($r = -0.645$, $p<0.05$) (Table 4.5), implying that increase in SCG application rate decreased pH_W.

Table 4.1: Physicochemical properties of the soil and SCG

	Soil	incubation	SCG
	Before	After	
Property		Values	
pH_w	6.90 ± 0.18	7.16 ± 0.10	5.26 ± 0.02
pH_{KCl}	6.42 ± 0.08	6.35 ± 0.02	4.77 ± 0.08
EC (S/cm)	60.8 ± 2	87.98 ± 1.52	872 ± 110
OM (%)	4.04 ± 0.92	4.60 ± 0.49	98.04 ± 0.19
TOC %	2.35 ± 0.53	2.67 ± 0.28	56.99 ± 0.11
N %	0.34 ± 0.07	0.33 ± 0.04	1.81 ± 0.31
C:N ratio	7 ± 2	8 ± 0.09	32 ± 3
P (mg/Kg)	14.40 ± 0.63	47.68 ± 0.73	4.37 ± 0.10
Mg (mg/kg)	41.93 ± 0.47	73.85 ± 0.38	5.51 ± 0.03
Ca (mg/kg)	38.38 ± 0.57	36.20 ± 0.10	3.81 ± 0.02
K (mg/Kg)	29.30 ± 0.40	63.77 ± 3.34	5.86 ± 0.46
Texture			
Clay (%)	2	2	-
Silt (%)	10	8	-
Sand (%)	88	90	-
MWD (mm)	0.56 ± 0.10	0.66 ± 0.01	-
Heavy metals			
Cd (mg/Kg)	8.70 ± 0.30	20,98 ± 0.19	0.52 ± 0.66
Cu (mg/ Kg)	0.46	0.69	1.04
Ni (mg/ Kg)	2.45 ± 0.05	7.76 ± 0.08	0.83 ± 0.03
Zn (mg/ Kg)	0.68 ± 0.07	4.07	1.74 ± 0.07
Pb (mg/Kg)	2.30 ± 0.08	5.07 ± 0.08	0.42 ± 0.02
SMC (%)	7.22 ± 0.23	5.52 ±0.55	98.06 ± 2.84

Values are means ± SE (n = 4 replications). – represent no results. SCG = spent coffee ground, EC = electrical conductivity, OM = organic matter, TOC = total organic carbon, N = nitrogen, P = phosphorus, Mg = magnesium, Ca = calcium, K = potassium, MWD = mean weight diameter, Cd = cadmium, Cu = copper, Ni = nickel, Zn = zinc, Pb = lead, and SMC = soil moisture content.

Soil pH_{KCl}

The interaction between different incubation periods and various SCG application rates had no significant effect on pH_{KCl} (Table 4.2). However, the main effect for various SCG application rates showed significant effect on pH_{KCl} suggesting that the lowest incubation period was enough to maximize the exchangeable acidity (Appendix 4.2). Treatment SCG-5, and SCG-10 significantly increased relative to control (Fig. 4.2). Treatment SCG-20, SCG-30, and SCG-50 declined relative to control, indicating that the highest application rates induced higher exchangeable acidity as compared to the lowest application rates. However, pH_{KCl} had no correlation with incubation period ($r = -0.18$; $p<0.05$) (Table 4.5).

Soil EC

The interaction between different incubation periods and various SCG application rates had a significant effect ($P<0.05$) on EC (Table 4.2). The highest values of EC were observed amongst the highest application rates treatments with SCG-50 measuring 202.30 S/cm at T9 above control (87.85 S/cm) (Fig. 4.3). The lowest EC was amongst the lowest application rates SCG-5 at T1 and T3 with 63.23 and 64.40 S/cm below control (69.25 and 79.1 S/cm), respectively. Treatments showed a trend of soil EC increasing with increased SCG application rate across different incubation periods.

Table 4.2: Interactive effect of incubation period and various SCG application rates on selected soil chemical properties.

T*SCG-AR (%)	**pH$_{W}$**	**pH$_{KCl}$**	**EC (S/cm)**	**N (%)**	**TOC (%)**	**C:N ratio**
1*0	7.46 abc	6.48 abc	69.25 fg	0.29 g	2.05 g	7.20 c
1*5	7.31 abcd	6.58 ab	63.23 g	0.33 fg	3.16 fg	11.87 abc
1*10	6.83 cdefgh	6.51 ab	67.35 fg	0.35 efg	3.34 fg	10.34 abc
1*20	6.83 cdefgh	6.25 abcd	82.93 defg	0.36 efg	4.99 def	17.12 abc
1*30	6.33 h	5.86 bcd	108.50 bcdefg	0.44 efg	7.27 bcd	19.39 ab
1*50	6.59 efgh	5.89 bcd	124.10 bcde	0.69 abcd	13.29 a	19.47 a
3*0	7.64 ab	6.07 abcd	79.10 efg	0.30 g	2.11 g	7.56 c
3*5	7.19 abcdef	6.55 ab	64.40 g	0.33 fg	2.82 fg	8.83 abc
3*10	7.05 bcdefg	6.35 abc	66.63 fg	0.38 efg	2.87 fg	7.77 bc
3*20	7.13 abcdef	6.09 abcd	72.07 fg	0.41 efg	4.76 defg	11.95 abc
3*30	6.85 cdefgh	6.01 abcd	96.15 cdefg	0.57 bcdef	8.26 bc	15.09 abc
3*50	6.72 defgh	5.85 bcd	128.70 bcd	0.73 abc	9.85 b	13.82 abc
6*0	7.26 abcde	6.22 abcd	111.73 bcdefg	0.31 g	2.34 fg	8.28 abc
6*5	7.78 a	6.41 abc	96.82 cdefg	0.33 fg	2.60 fg	7.78 abc
6*10	7.14 abcdef	6.43 abc	70.05 fg	0.38 efg	3.27 fg	8.53 abc
6*20	6.70 defgh	6.07 abcd	89.30 cdefg	0.45 defg	4.08 efg	9.21 abc
6*30	6.51 fgh	5.81 bcd	110.08 bcdefg	0.59 bcde	6.56 cde	11.31 abc
6*50	6.53 fgh	5.72 cd	150.73 b	0.75 ab	10.08 b	13.97 abc
9*0	7.16 abcdef	6.35 abc	87.85 defg	0.33 fg	2.67 fg	8.07 abc
9*5	7.51 abc	6.69 a	87.98 defg	0.35 efg	2.85 fg	8.43 abc
9*10	6.96 bcdefgh	6.28 abc	84.50 defg	0.39 efg	2.73 fg	7.11 c
9*20	6.38 gh	5.47 d	115.5 bcdef	0.49 cdefg	3.99 efg	8.17 abc
9*30	6.60 efgh	5.82 bcd	137.05 bc	0.59 bcde	6.49 cde	11.06 abc
9*50	6.70 defgh	5.71 cd	202.30 a	0.85 a	8.72 bc	10.54 abc
F values						
T	0.02 *	0.09 *	0.00 *	0.01 *	0.00 *	0.00 *
SCG-AR	0.00 *	0.00 *	0.00 *	0.00 *	0.00 *	0.00 *
T*SCG-AR	0.00 *	0.21	0.01 *	0.95	0.00 *	0.72

Values are means (n = 4). Means with different letters are statistically significant. *Significant at 0.05 level, T = incubation period, SCG = spent coffee ground, SCG-AR = SCG-Application rates, T*SCG-AR = incubation period* SCG-Application rates, EC = electrical conductivity, OM = organic matter, TOC = total organic carbon, N = nitrogen, P = phosphorus, Mg = magnesium, Ca = calcium, K = potassium.

Table 4.2 cont’d

T*SCG-AR (%)	Mg (mg/Kg)	Ca (mg/Kg)	K (mg/Kg)	P (mg/Kg)
1*0	58.88 g	35.20 gh	49.2 hij	45.28 c
1*5	36.40 n	17.25 m	32.05 klm	13.93 k
1*10	46.33 kl	24.13 j	45.70 ijk	27.65 fghi
1*20	70.58 d	37.25 fg	29.23 lm	51.68 b
1*30	55.58 h	28.78 i	26.75 lmn	42.98 cd
1*50	82.13 a	43.55 d	60.62 efgh	61.83 a
3*0	49.98 j	29.53 i	60.03 fghi	29.05 fgh
3*5	50.75 j	27.58 i	40.70 jkl	13.80 k
3*10	50.18 j	21.10 l	68.55 def	23.20 hij
3*20	27.68 o	11.48 n	28.48 lm	2.63 l
3*30	52.40 ij	34.03 h	13.20 n	37.53 de
3*50	47.03 k	39.63 ef	49.28 ghij	20.63 j
6*0	43.58 m	21.40 kl	67.0 def	33.48 ef
6*5	64.88 e	52.53 b	24.90 mn	37.98 de
6*10	46.90 k	39.68 ef	50.55 ghij	27.08 ghi
6*20	53.98 hi	42.95 d	28.63 lm	3.04 l
6*30	44.13 lm	22.85 jkl	75.0 cde	-4.6 m
6*50	60.63 fg	37.10 fg	77.43 cd	24.63 ghij
9*0	73.85 c	36.20 gh	63.77 defg	47.68 bc
9*5	51.88 ij	23.83 jk	59.70 fghi	23.90 ghij
9*10	43.43 m	34.63 gh	86.88 bc	11.30 k
9*20	55.93 h	40.93 de	102.25 a	21.975 ij
9*30	62.60 ef	48.28 c	101 ab	23.55 hij
9*50	77.85 b	62.15 a	112 a	29.95 fg
F values				
T	0.00 *	0.00 *	0.00 *	0.00 *
SCG-AR	0.00 *	0.00 *	0.00 *	0.00 *
T*SCG-AR	0.00 *	0.00 *	0.00 *	0.00 *

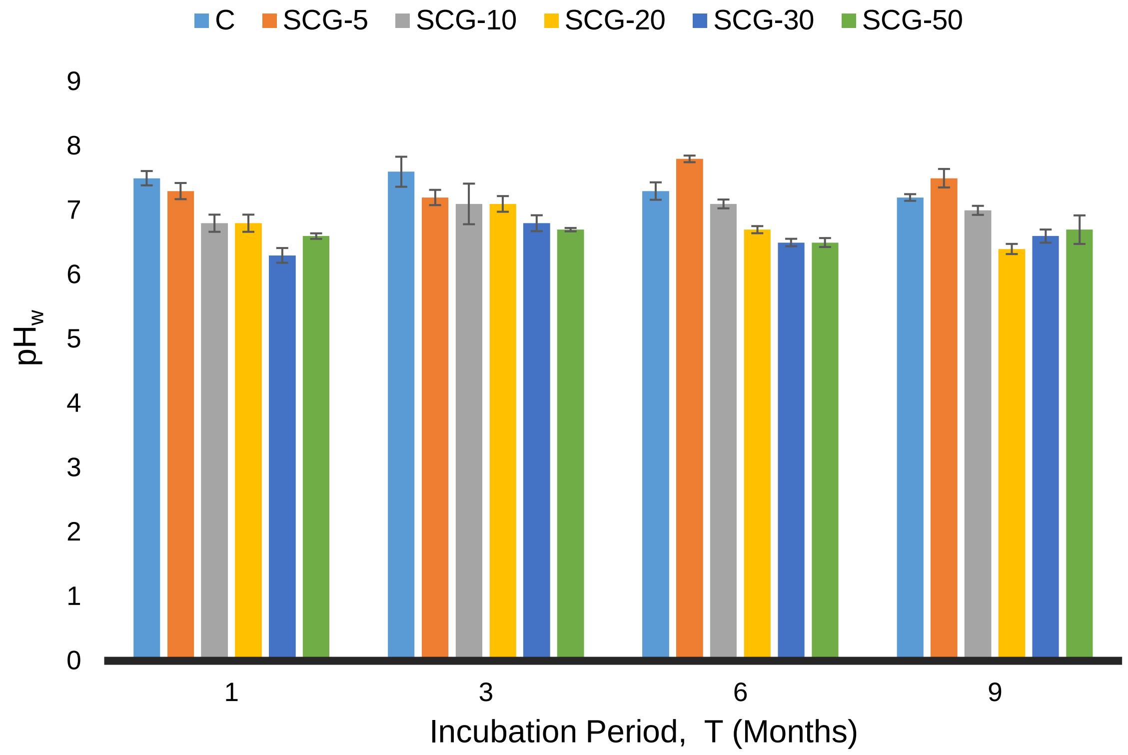

Figure 4.1: Response of pH_w to various SCG application rates across different incubation periods in means ± SE values (n = 4 replications)

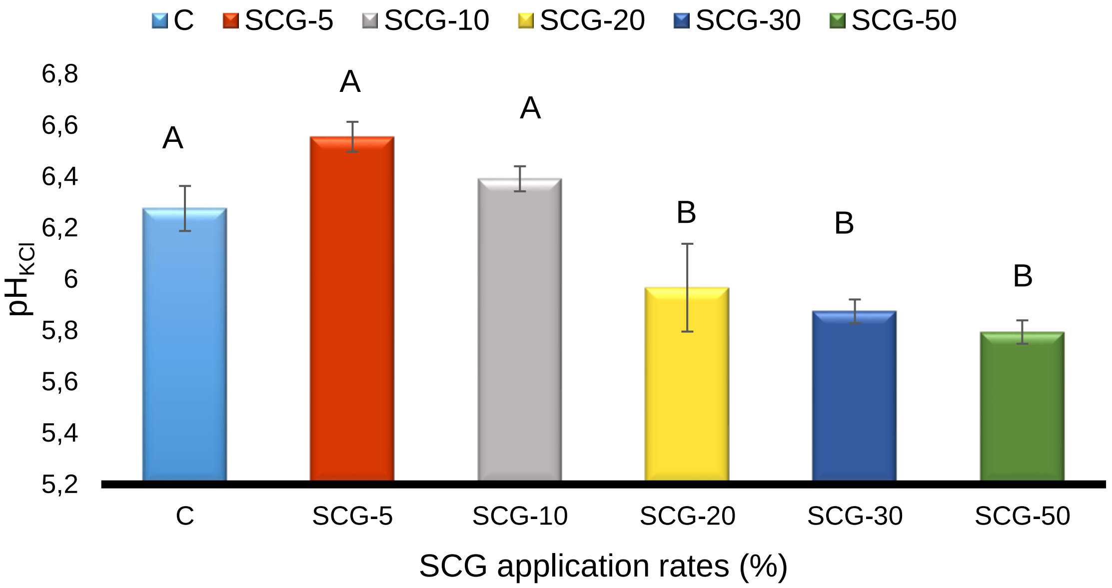

Figure 4.2: Response of pH_{KCl} to various SCG application rates in means ± SE values (n = 4 replications). Means with different letters are statistically significant.

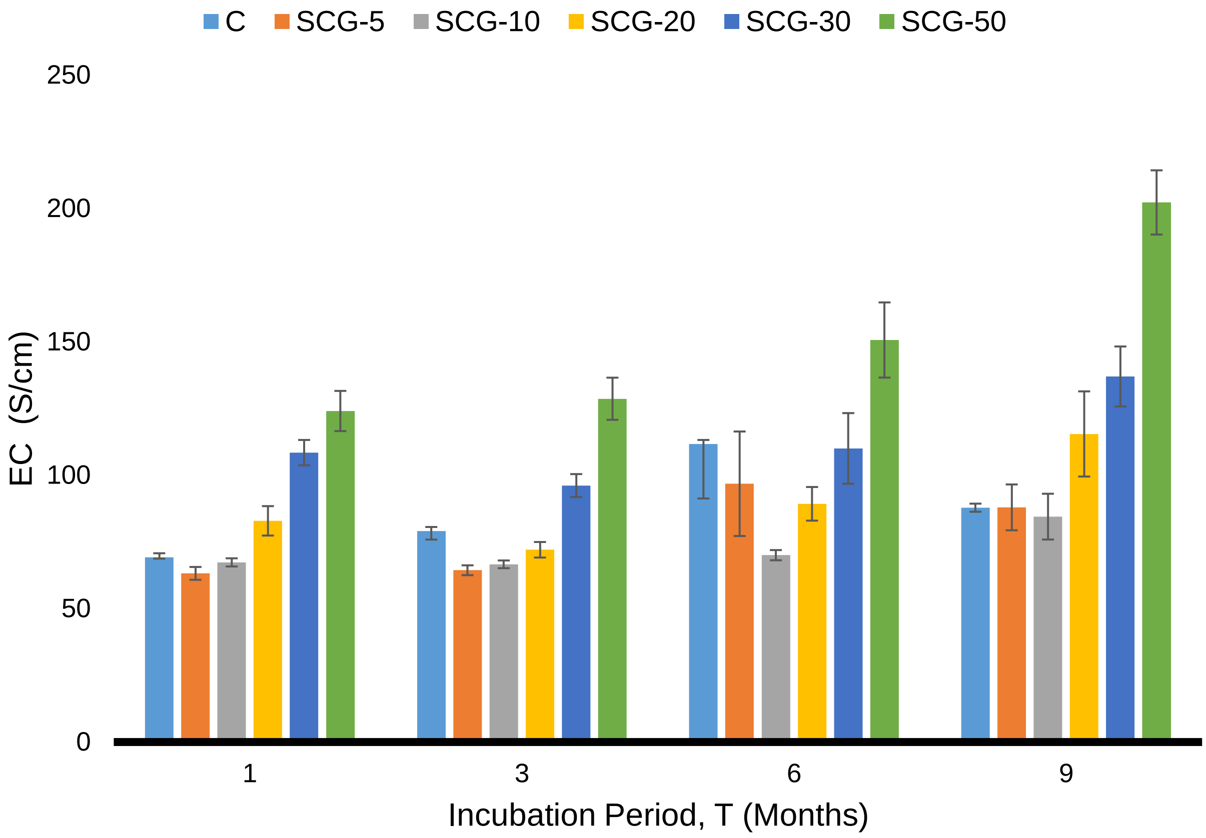

Figure 4.3: Response of soil EC to various application rates of SCG across different incubation periods in means ± SE values (n = 4 replications).

4.1.3. Effect of spent coffee ground and incubation period on nutrients supply

TOC, N and C:N ratio

Interaction between incubation periods and various SCG application rates significantly influenced TOC ($P<0.05$) and insignificantly had an effect on N and C:N ratio. However, the incubation period and various SCG concentrations levels individually showed a significant difference in N and C:N ratio without interaction (Appendix 4.5 and 4.6). Moreover, based on the regression analysis (R^2), this indicates over 60% overall variation in N and C:N ratio which can be explained by incubation period of 9 months across the various SCG application rates (Fig. 4.4).

Amelioration with SCG in the first test period of incubation increased the TOC level of the soil ranging from 3.16 to 13.29 % in all treatments compared to control. The drastic increase was seen in SCG-50 which in the first test period increased by 548% from 2.05% control to 13.29% SCG-50. A decline in TOC was observed from the 2nd test period and across the incubation period in all treatments. A drastic decline was observed in SCG-50 decreasing by 34.39 % from 13.29% at T1 test period to 8.72% at T9. However, TOC levels were still higher in SCG-50 as compared to control. Moreover, decrease in pH increased TOC ($r=-0.51$, $p<0.05$) (Table 4.5).

Nitrogen was higher during the T6 and T9 periods at a relatively high SCG application rate. Figure 4.4 shows a trend of increasing N across the incubation period for all treatments, with a maximum value of 0.85% in SCG-50 at T9. Nitrogen positively correlated with EC, TOC, MWD, LM, and SMC ($r = 0.61$, $r = 0.75$, $r = 0.72$, 0.72, 0.74; $p<0.05$) and negatively correlated with sM, M and s+c ($r = -0.56$, $r = -0.70$, -0.56) respectively (Table 4.5). Increased TOC and N post addition of SCG increased C:N ratio above control in T1, with SCG-50 reaching a maximum point of 25 units (Fig. 4.4). However, as incubation progressed, C:N ratio decreased across the incubation period and positively correlated with TOC (Table 4.5).

Base cations (Mg, Ca and K), and available P

Various SCG application rates had a significant effect ($p<0.05$) on Mg, Ca, K and P across different incubation periods (Table 4.2). There were three noticeable trends with exception to potassium (Figure 4.5): firstly, amendment with SCG in the first test period (T1) increased relatively high application rate treatments above control, Mg, Ca and P in treatment SCG-50 increased by 23.3, 8.4, and 16,6 mg/Kg above control (58.9, 35.2 and 45.3 mg/Kg) respectively. Secondly, treatment SCG-50 peaked at the 3rd test period (T6). Mg, Ca and P in treatment SCG-5 peaked by 33%, 59%, and 12% at the 3rd test period respectively. Lastly, a release of nutrients at the 4th test period (T9) from treatments with the highest application rate was found and an epic decline for treatments with the lowest application rate. At T9, Mg, Ca and P increased from mean values of 55.9 to 77.9, 40.9 to 62.2, and 22.0 to 30.0 mg/Kg above control in treatments with high application rates and declined below control from 51.9 to 43.4, 34.6 to 23.8 and 23.9 to 11.1 mg/Kg in treatments with low application rates respectively.

Potassium showed a different behaviour: firstly, treatments with a high application rate (excluding SCG-50) fell below control. Treatment SCG-5 through to SCG-30 fell within range of mean values of 45.7 to 26.8 mg/Kg below control of 49.2 mg/Kg. Secondly, with monthly progression, at the 3rd test period (T6) through to the 4th test period (T9), the high application rate treatments reached a maximum above control. High application rate treatments, SCG-20, SCG-30, and SCG-50, across T6 and T9, increased from 28.6 to 102.3 mg/Kg, 75 to 101 mg/Kg, and 77.4 to 112 mg/Kg respectively. Lastly, low application rate treatments peaked at the 2nd and 4th test period above control. Mg, Ca and K positively correlated to EC ($r = 0.46$, $r = 0.57$, $r = 0.50$; $p<0.05$).

4.1.4. Effect of spent coffee ground and incubation period on heavy metal abundance in the soil

Interaction between different incubation periods and various SCG application rates had a significant effect on heavy metals concentration (Table 4.3), respectively. Heavy metals were ranked Cd>Ni>Pb>Zn>Cu from highest to lowest (Fig. 4.6). Application of SCG on Cd, Cu Ni, Zn, and Pb showed that SCG-5 increased with increasing incubation period relative to control, whereas SCG application rate SCG-10 through to   decreased with an increasing incubation period below control.

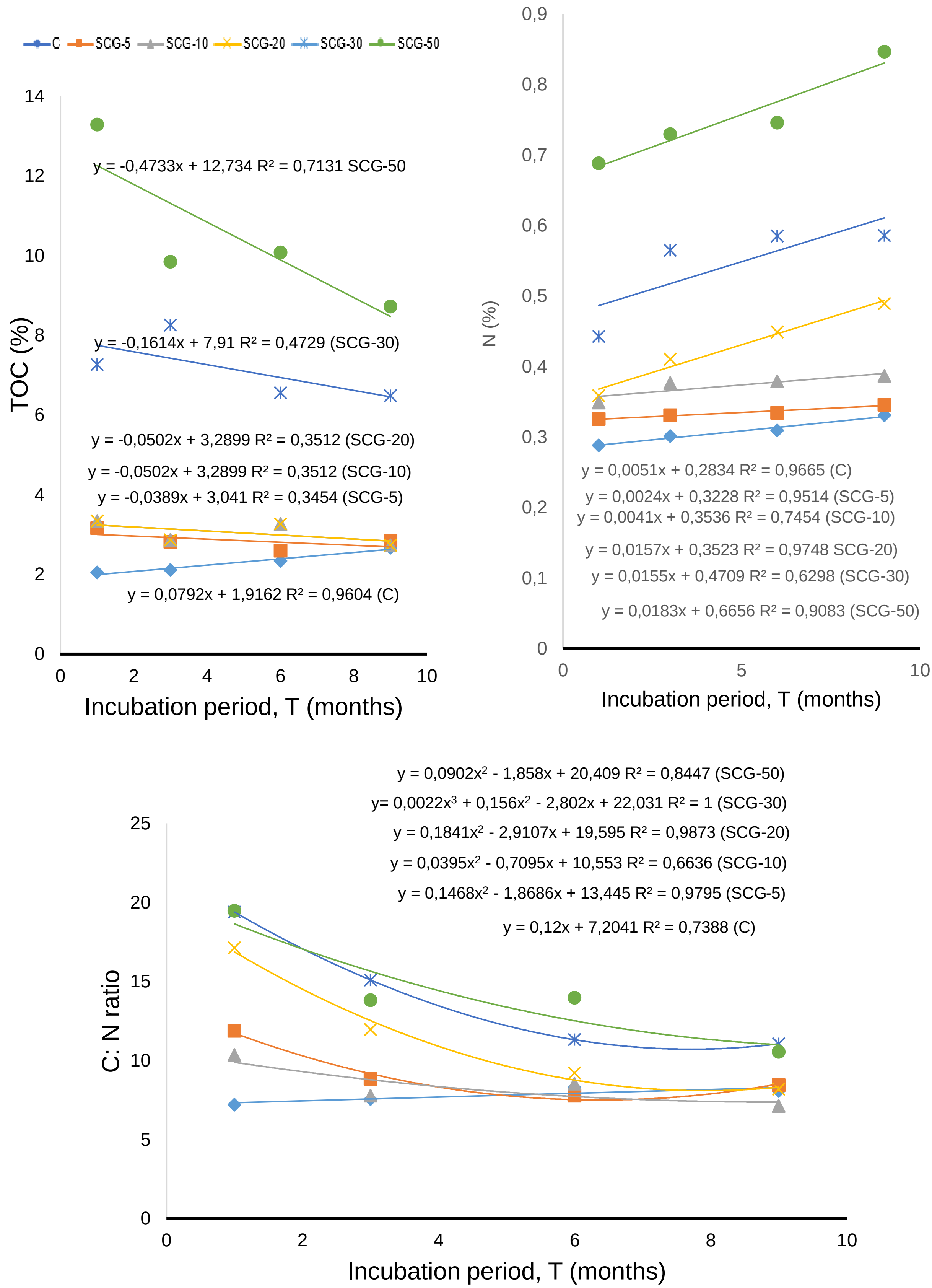

Figure 4.4: Response of TOC, N, and C:N ratio to the interactive effect of various SCG application rates and different incubation periods. Values are means (n = 4 replications). TOC = Total organic carbon, N = Nitrogen, and C:N ratio = carbon to nitrogen Ratio

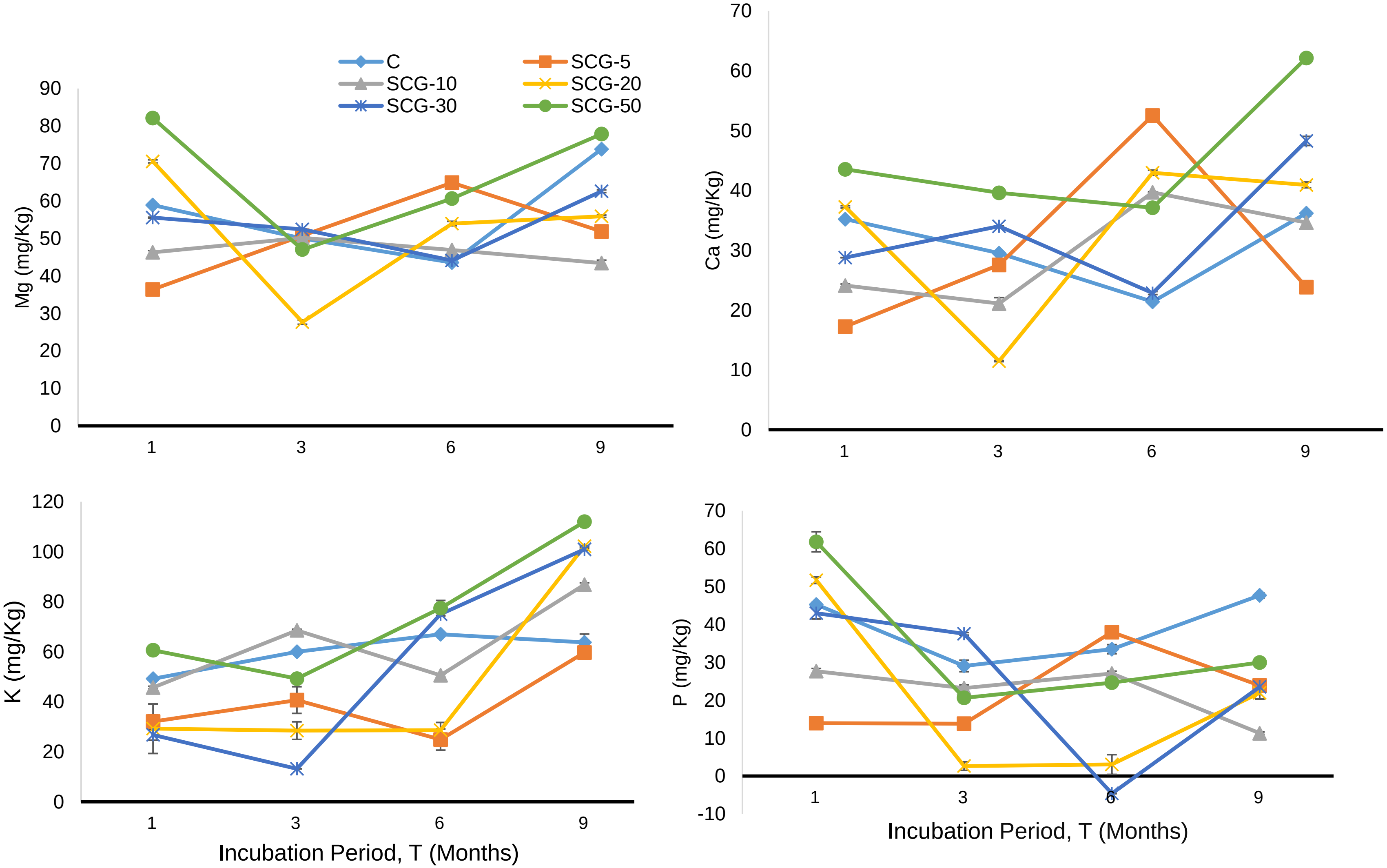

Figure 4.5: Response of selected nutrients to interactive effect of various SCG application rate and different incubation periods. Values are means ± SE (n = 4 replications). Mg = magnesium, Ca = calcium, K = potassium, and P = phosphorus.

Table 4.3: Interactive effect of incubation period and various SCG application rates on heavy metals concentration in the soil.

T*SCG-AR (%)	Cd (mg/Kg)	Cu (mg/Kg)	Ni (mg/Kg)	Zn (mg/Kg)	Pb (mg/Kg)
1*0	17,98 d	0,21 def	6,82 d	3,08 c	4,78 cd
1*5	5,65 n	0,47 b	2,82 pq	0,65 k	0,79 p
1*10	14,8o fg	0,09 ghijk	5,62 f	2,21 d	3,78 g
1*20	22,60 b	0,47 b	8,51 b	4,08 b	5,65 b
1*30	16,90 de	0,05 ijk	6,25 e	2,99 c	4,22 ef
1*50	24,28 a	0,72 a	9,28 a	4,61 a	6,07 a
3*0	13,08 ij	0,09 ghijk	4,52 ij	2,18 d	2,95 ijk
3*5	11.00 kl	0,05 ijk	4,24 jk	1,29 ghi	2,15 no
3*10	10,39 klm	0,07 hijk	4,39 ij	1,59 efg	1,88 o
3*20	1,97 o	0,66 a	1,55 r	0,16 m	0,58 p
3*30	14,63 fgh	0,07 hijk	5,28 fg	2,49 d	3,88 fg
3*50	9,82 lm	0,24 cde	2,85 pq	1,15 hi	2,84 jkl
6*0	14,13 ghi	0,16 efg	5,01 gh	2,25 d	3,28 hi
6*5	15,78 ef	0,26 cd	4,66 hi	2,33 d	4,44 de
6*10	11,73 jk	0,15 efgh	3,11 nop	1,40 efgh	3,19 hij
6*20	11,63 k	0,02 k	3,61 lm	0,79 jk	3,36 h
6*30	5,07 n	0,03 jk	3,25 mno	0,61 kl	0,98 p
6*50	11,60 k	0,29 cd	4,34 ij	1,74 e	2,52 lmn
9*0	20,98 c	0,69 a	7,76 c	4,07 b	5,08 c
9*5	10,13 lm	0,03 jk	3,92 kl	1,46 efgh	2,02 o
9*10	4,89 n	0,13 fghi	1,42 r	0,28 lm	0,96 p
9*20	9,07 m	0,12 fghj	2,58 q	1,05 ij	2,22 mno
9*30	10,49 klm	0,30 c	3,03 op	1,38 fghi	2,59 klm
9*50	13,20 hi	0,53 b	3,46 mn	1,70 ef	3,54 gh
F values					
T	0.00*	0.00*	0.00*	0.00*	0.00*
SCG-AR	0.00*	0.00*	0.00*	0.00*	0.00*
T*SCG-AR	0.00*	0.00*	0.00*	0.00*	0.00*

Values are means (n = 4). Means with different letters are statistically significant. *Significant at 0.05 level, T = incubation period, SCG = spent coffee ground, SCG-AR = SCG-Application rates, T*SCG-AR = incubation period* SCG-Application rates, Cd = cadmium, Cu = copper, Ni = nickel, Zn = zinc, Pb = lead.

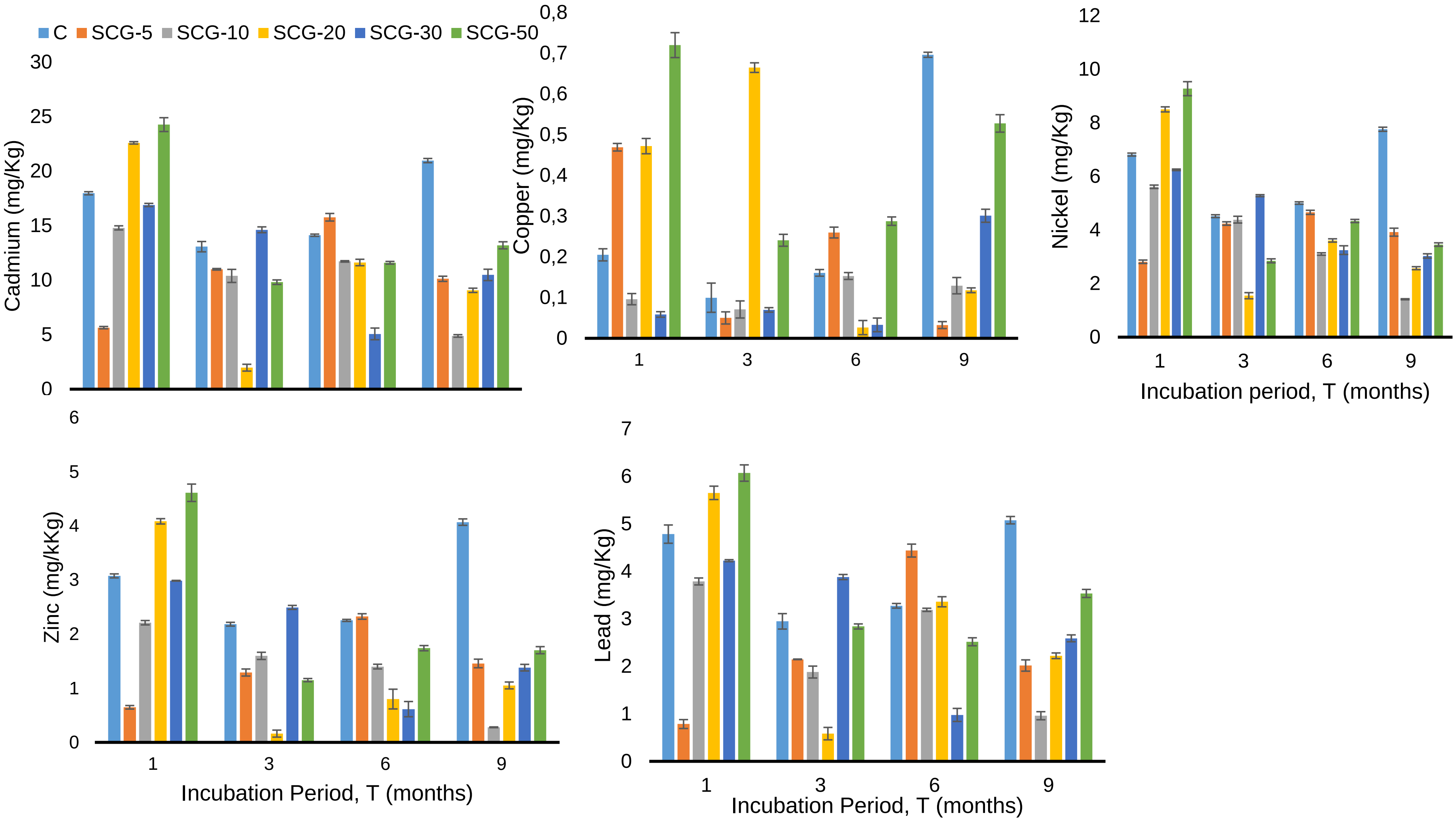

Figure 4.6: Response of selected heavy metals to interactive effect of various SCG application rates and different incubation periods. Values are means ± SE (n = 4 replications).

4.1.5. Effect of spent coffee ground and incubation period on soil aggregate fraction and stability

The interaction between incubation period and SCG various application rates had a significant effect (P<0.05) on LM, sM, and m fraction and MWD and showed no significant effect on s+c (Table 4.4). At test period T1, there were high presence of unaggregated s+c fraction in all treatments like control (Fig. 4.7). At test period T3, there were highest amounts of sM and m fraction in all treatments compared to control. At test period T6, there were highest amounts of LM fraction in all treatments compared to control.

Enrichment with SCG in the first test period (T1) post application increased MWD, with the drastic increase by 46% from control of 0,56 mm to 1,03 mm SCG-50. At the 3rd test period (T6), SCG-50 treatment reached a maximum point of 160 mm above control (0.71 mm) increasing by 56%. A steady decrease was observed after the 3rd test period. Various SCG application rates positively correlated MWD (r=0.77, p<0.05) respectively. TOC was positively correlated with MWD and LM (r = 0.60, p<0.05; r=0,98 p<0.05) and negatively correlated with sM, m and s+c (r=-0.50, p<0.05; r=-0.57, p<0.05; r=-0.45, p<0.05), respectively. pH positively correlated with sM, m and s+c (r = 0.41, p<0.05; r = 0.62, p<0.05; r = 0.48, p<0.05) and negatively correlated with LM and MWD (r = -0.55, p<0.05; r = -0.55, p<0.05). Soil moisture content was positively correlated with MWD and LM (r = 0.57, p<0.05; r = 0.63, p<0.05) and negatively correlated with sM, m and s+c, respectively.

4.1.6. Effect of spent coffee ground and incubation period on SMC

Interaction between different incubation periods and various SCG application rates had a significant effect (p<0.05) on SMC. The maximum value of 60.26 w% was observed in the first test period (T1) and the highest SCG application rate (SCG-50). In the first test period, SMC increased by 798 w% from an average of 6.71 w% control and 60.26 w% in SCG-50 (Fig.4.7). SMC was positively correlated with EC and TOC (r = 0.62, p<0.05; 0.89, p<0.05) and negatively with pHw (r = -0.44, p<0.05).

Table 4.4: Interactive effect of incubation period and various SCG application rates on soil physical properties

T*SCG-AR	LM (mm)	sM (mm)	M (mm)	s+c (mm)	MWD (mm)	SMC (w%)
1*0	0.04 f	0.39 cdefg	0.29 ab	0.08 a	0.56 i	6,71 g
1*5	0.06 f	0.48 abcde	0.27 abcd	0.05 abc	0.68 hi	6,76 g
1*10	0.05 f	0.49 abcde	0.29 abc	0.04 abcd	0.68 hi	9,55 efg
1*20	0.16 def	0.40 bcdefg	0.23 bcde	0.03 bcd	0.80 ghi	13,06 defg
1*30	0.14 ef	0.51 abcd	0.16 def	0.04 abcd	0.87 fghi	18,19 d
1*50	0.35 cde	0.28 fgh	0.15 efg	0.02 cd	1.04 defg	60,26 a
3*0	0.06 f	0.53 abc	0.37 a	0.07 ab	0.67 hi	5,54 g
3*5	0.06 f	0.53 abc	0.33 ab	0.04 abcd	0.74 ghi	5,79 g
3*10	0.06 f	0.54 abc	0.32 ab	0.04 abcd	0.77 ghi	8,51 fg
3*20	0.14 ef	0.52 abcd	0.22 bcde	0.03 bcd	0.88 efgh	11,39 defg
3*30	0.37 cd	0.39 cdefg	0.11 efg	0.02 cd	1.20 cde	15,92 def
3*50	0.44 bc	0.34 defg	0.10 efg	0.03 bcd	1.28 bcd	36,08 c
6*0	0.08 f	0.45 abcdef	0.38 a	0.05 abc	0.71 hi	5,29 g
6*5	0.10 f	0.50 abcd	0.33 ab	0.04 abcd	0.80 ghi	6,39 g
6*10	0.07 f	0.59 a	0.28 abcd	0.04 bcd	0.83 ghi	8,18 fg
6*20	0.45 bc	0.39 cdefg	0.12 efg	0.02 cd	1.34 abcd	8,73 fg
6*30	0.67 a	0.25 gh	0.06 fg	0.01 d	1.64 a	17,36 de
6*50	0.72 a	0.15 h	0.03 g	0.00 d	1.61 a	44,86 b
9*0	0.05 f	0.46 abcde	0.36 a	0.07 ab	0.66 hi	5.52g
9*5	0.05 f	0.54 abc	0.33 ab	0.05 abcd	0.75 ghi	5.79 g
9*10	0.09 f	0.57 ab	0.27 abcd	0.03 bcd	0.85 ghi	7.93 fg
9*20	0.33 cde	0.46 abcdef	0.17 cdef	0.02 cd	1.18 cdef	8.35 fg
9*30	0.54 abc	0.34 defg	0.07 fg	0.02 cd	1.46 abc	17.92 d
9*50	0.61 ab	0.3 efgh	0.06 fg	0.01 d	1.56 ab	40.76 bc
F values						
T	0.00*	0.00*	0.01*	0.00*	0.00*	0.00*
SCG-AR	0.00*	0.00*	0.00*	0.00*	0.00*	0.00*
T*SCG-AR	0.00*	0.00*	0.00*	0.97	0.00*	0.00*

Values are means (n = 4). Means with different letters are statistically significant. *Significant at 0.05 level, T = incubation period, SCG = spent coffee ground, SCG-AR = SCG-Application rates, T*SCG-AR = incubation period* SCG-Application rates, LM = large macroaggregates, sM = small macroaggregates, m = microaggregates, s+c = unaggregated silt and clay, MWD =aggregate stability, SMC = soil moisture content

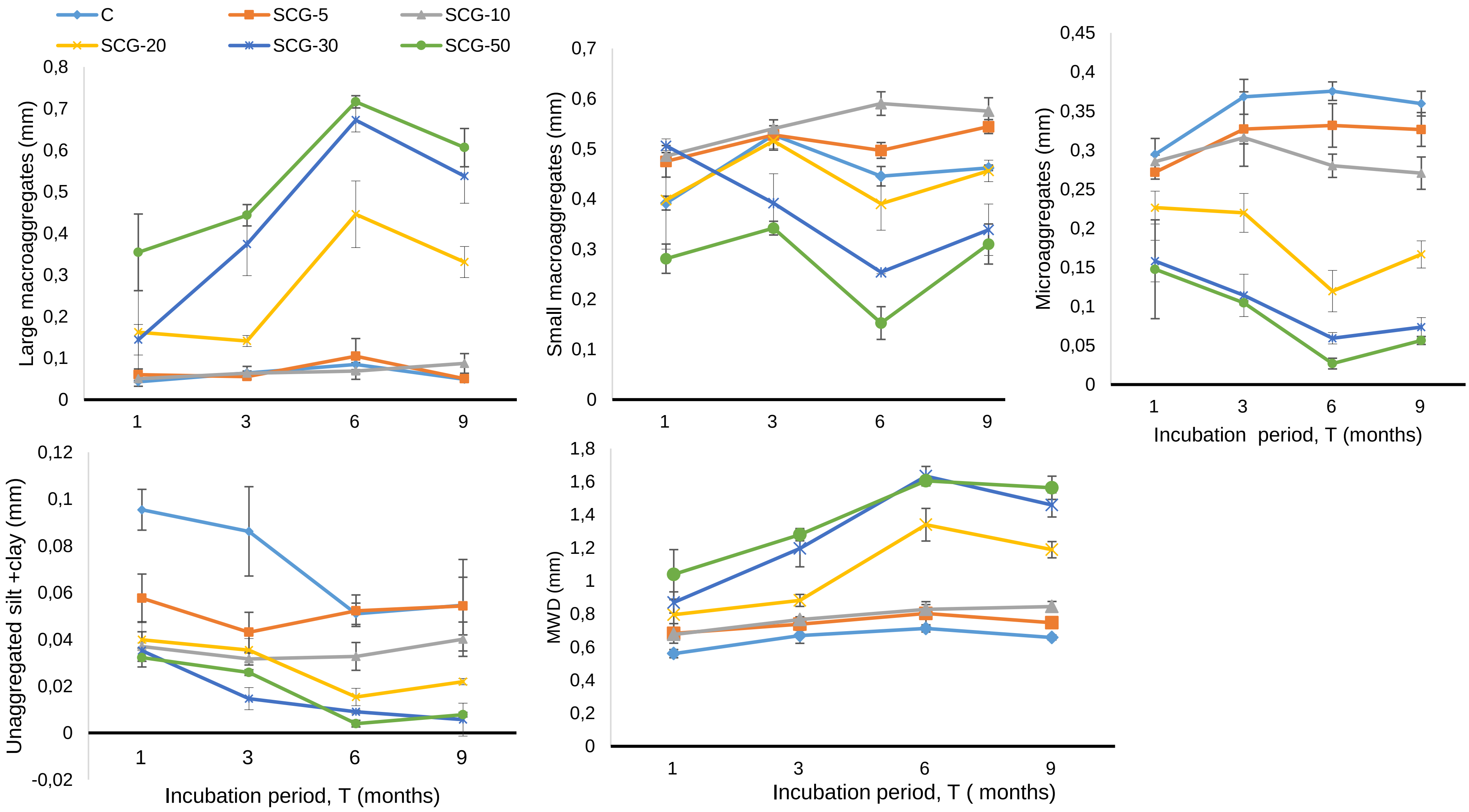

Figure 4.7: Response of aggregate fraction and stability to interactive effect of various SCG application rates and different incubation periods. Values are means ± SE (n = 4 replications). MWD = mean weight diameter

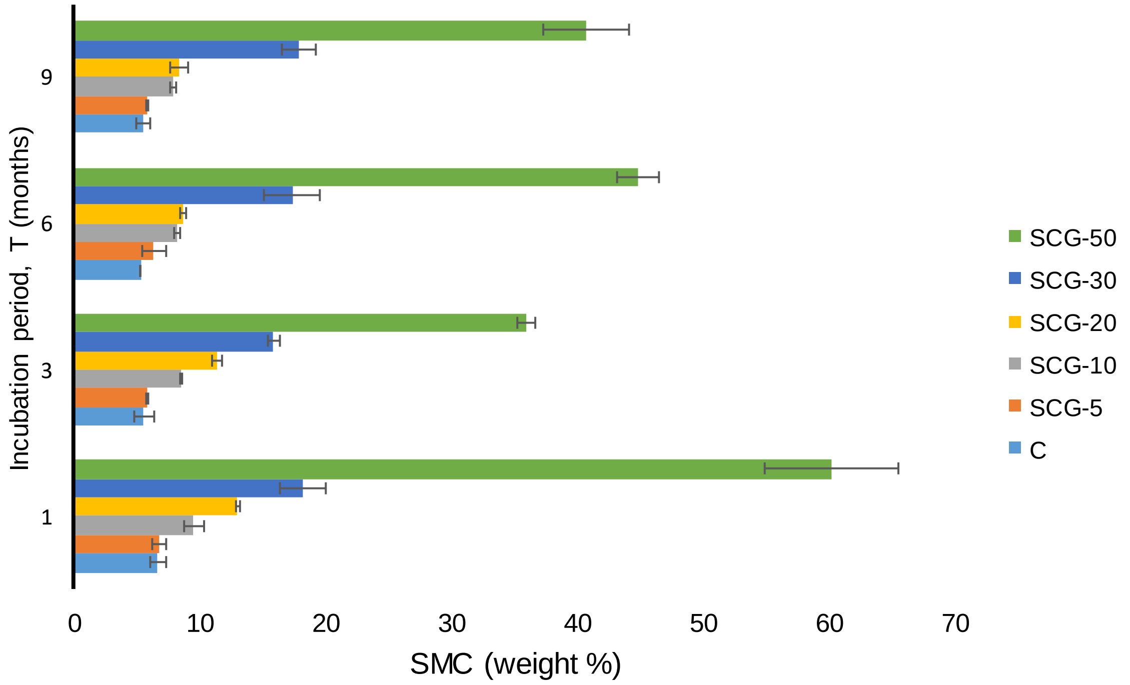

Figure 4.8: Response of SMC to the interactive effect of various SCG concentration levels across different incubation periods. Values are mean ± SE (n = 4 replications). SMC = soil moisture content.

Table 4.5: Relationships amongst selected soil chemical and physical properties

EC = electrical conductivity, OM = organic matter, TOC = total organic carbon, N = nitrogen, P = phosphorus, Mg = magnesium, Ca = calcium, K = potassium,

	pH_{KCl}	pH_{W}	EC	N	TOC	C:N ratio	P	K	Mg	Ca	Cd	Cu
pH_{KCl}	1											
pH_{W}	0.64	1										
EC	-0.47	-0.39	1									
N	-0.55	-0.51	0.67	1								
TOC	-0.51	-0.56	0.60	0.75	1							
C:N ratio	-0.17	-0.41	0.21	0.05	0.62	1						
P	0.02	0.04	0.10	-0.03	0.21	0.27	1					
K	-0.35	-0.26	0.50	0.36	0.11	-0.20	-0.11	1				
Mg	-0.20	-0.19	0.50	0.33	0.39	0.19	0.71	0.26	1			
Ca	-0.31	-0.19	0.57	0.45	0.34	0.03	0.35	0.34	0.75	1		
Cd	0.04	-0.02	0.08	-0.03	0.20	0.26	0.92	-0.18	0.78	0.40	1	
Cu	-0.05	0.00	0.20	0.20	0.32	0.24	0.37	0.06	0.40	0.17	0.29	1
Ni	0.10	-0.02	-0.04	-0.10	0.19	0.31	0.85	-0.26	0.66	0.13	0.94	0.31
Zn	0.04	0.01	0.04	-0.06	0.21	0.30	0.94	-0.18	0.73	0.25	0.96	0.37
Pb	-0.01	-0.04	0.12	0.01	0.23	0.27	0.88	-0.21	0.77	0.50	0.97	0.29
MWD	-0.56	-0.56	0.63	0.72	0.60	0.21	-0.31	0.40	0.20	0.43	-0.23	-0.00
LM	-0.55	-0.55	0.64	0.72	0.64	0.27	-0.21	0.38	0.27	0.43	-0.13	0.06
sM	0.40	0.41	-0.50	-0.56	-0.60	-0.35	-0.08	-0.25	-0.38	-0.32	-0.16	-0.23
m	0.53	0.62	-0.57	-0.70	-0.69	-0.39	0.16	-0.23	-0.21	-0.38	0.12	-0.08
s+c	0.44	0.48	-0.45	-0.56	-0.52	-0.25	0.27	-0.21	-0.10	-0.30	0.22	0.02
SMC	-0.45	-0.46	0.62	0.74	0.90	0.46	0.27	0.26	0.50	0.42	0.26	0.42
T	-0.18	-0.05	0.38	0.17	-0.13	-0.35	-0.27	0.66	0.16	0.40	-0.28	-0.05
SCG-AR	-0.59	-0.64	0.69	0.85	0.91	0.47	0.05	0.24	0.35	0.43	0.05	0.26

Cd = cadmium, Cu = copper, Ni = nickel, Zn = zinc, Pb = lead, LM = large macroaggregates, sM = small macroaggregates, m = microaggregates, s+c = unaggregated silt and clay, MWD =aggregate stability, SMC = soil moisture content, SCG-AR = SCG-Application rates, T = incubation period.

Table 4.5 cont’d

	Ni	Zn	Pb	MWD	LM	sM	m	s+c	SMC	T	SCG-AR
Ni	1										
Zn	0.97	1									
Pb	0.88	0.92	1								
MWD	-0.28	-0.27	-0.15	1							
LM	-0.16	-0.16	-0.06	0.98	1						
sM	-0.18	-0.16	-0.18	-0.69	-0.83	1					
m	0.15	0.15	0.04	-0.90	-0.90	0.67	1				
s+c	0.26	0.26	0.15	-0.74	-0.69	0.46	0.67	1			
SMC	0.23	0.26	0.27	0.57	0.63	-0.64	-0.64	-0.46	1		
T	-0.43	-0.33	-0.26	0.36	0.27	-0.02	-0.11	-0.22	-0.09	1	
SCG-AR	0.01	0.03	0.11	0.77	0.7	-0.66	-0.84	-0.65	0.89	0	1

4.2. Discussion

4.2.1. Soil and SCG analyse

Spent coffee ground used in this study had the highest average values of EC, OM, TOC, N, C:N ratio, Cu and SMC as compared to the soil. This was due to the constituents of the organic material. It was reported by McNutt and He (2019) that polysaccharides and lignin are rich in OM and are high in C:N ratio. The texture of the soil was sandy loam with a large proportion of sand (89%) which partly contributed to the low organic carbon (2.35%) and MWD (0.56 mm). According to Weil and Brady (2016) soil dominated by sand particles generally has low water holding capacity (large pores that exist between sand particles) and has a weak structure due to the low organic matter and clay content. On the other hand, the soil had the highest average values of Cd, Zn, Ni, Pb, Mg, K, P, and Ca compared to SCG. Spent coffee ground was found to have 1.30% minerals on a dry mass basis (w/w), (Ballesteros *et al*., 2014) resulting in the low nutrients in this study.

4.2.2. Effect of spent coffee ground and incubation period on soil acidity

Soil pH_w

Application of SCG at the first test period (T1) reduced pH_w in all treatments as compared to control. This was because freshly decomposing organic material (labile) lowers soil pH_w by releasing hydrogen ions that were associated with organic anions into the soil solution (Weil and Brady, 2016). Furthermore, the coffee beans are naturally acidic; this acidity constitutes 11% of the beans' original mass (Cruz *et al*., 2012). Since SCG is the by-product of coffee beans, it inherits these organic acids. A lower pH_w decline was observed in treatment SCG-5 at the first test period (T1); this was due to the low application rate of SCG.

These findings were in accordance with a study by Cruz *et al*. (2015), who found that the pH of all treatments was lower than in control and a strong negative relationship between SCG application rates with pH ($r = -0.957$, $p<0.001$). A similar strong negative relationship was observed in the present study suggesting increasing various SCG application rates decreased pH_w of the soil. Moreover, there was a noticeable increase in pH in all treatments across the incubation period with only SCG-5 increasing relative to the initial soil pH recording and control. The increase in pH_W during the incubation period in the present study was caused by buffering capacity and high CEC caused by more resistant organic matter (Weil and Brady, 2016). Overall, the difference in soil pH_w at T1 amongst treatments might be the reason for the different SCG assimilations observed across the incubation period. That is as T1 treatments with higher pH_w (SCG-5) assimilation rates were greater across the incubation period (Vela-Cano *et al*., 2019). Although pH_w of the soil increased in different treatments, the actual values fell within the optimum range of 5.5 to 7.0 which promote plant-available nutrients (Weil and Brady, 2016)

Soil pH_{KCl}

Exchangeable acidity increased with increasing SCG application rate but did not change across the incubation period. EC had a negative relationship with organic matter SCG application rates suggesting that and its various application rates increase exchangeable acidity. However, this acidity didn't increase over time due to the buffering and liming effect of the SCG (Weil and Brady, 2016). Moreover, pH_{KCl} of the soil increased in different treatments but the actual values did not cross the critical range of 5.2 to 6.5 increasing aluminium toxicity in the soil (Angelova *et al*., 2013).

Soil EC

Incorporation of SCG into soil increased EC with increasing SCG application rate and across incubation periods, especially in treatments with high application rates because EC was higher in SCG as compared to the soil (Table 4.1). The findings were in contrast with a study by Cruz *et al.* (2015) and Cervera-Mata *et al*. (2017) who found EC decreased with cultivation period and they attributed the decrease in EC to plants' absorption activity and increased cation exchange capacity caused by the transformation of the organic residue hence the greater retention of ions. However, a study by Angelova *et al*. (2013) on a different residue (compost and vermicompost) found similar findings as to the present study and they singled out that the EC depends on the raw materials and their ion concentration. Moreover, although the EC of the soil increased in different treatments the actual values did not cross the critical limit of 4000 S cm^{-1} and improved to ideal levels in SCG-50 in the last test period (T9) (Angelova *et al*., 2013).

4.2.3. Effect of spent coffee ground and incubation period on nutrients supply

Soil TOC, N, and C:N ratio

Amelioration with SCG in the first test period increased the soil TOC, N, and C:N ratio across the incubation period. It increased TOC, N, and C:N ratio by 549, 138, and 140% in the highest application rate treatments (SCG-50) and increased by 54, 12, and 65% in the lowest application rate treatments (SCG-5) as compared to control. The variability in percentage increase in the treatments was due to the difference in SCG application rates. The findings were in line with the study by Cervera-Mata *et al*. (2017) and Vela-Cano *et al*. (2019) who found a greater TOC, N, and C:N ratio increase in higher application rates as compared to lower rates.

Furthermore, enrichment of soil with SCG increased nitrogen and decreased carbon and C:N ratio across the incubation period. However, the effect differed with the SCG application rates. The trend of decreasing TOC and increasing N was due to the mineralization of SCG during the incubation period. Additionally, sandy soil tends to lose more organic matter as compared to other soils; this is because of the organic matter unprotected by soil particles resulting in rapid decomposition from microbial attack (Weil and Brady, 2016). Moreover, a negative relationship existed between pH_w and TOC (N and C:N ratio) implying pH increased with decreasing TOC. Similar findings were observed by Cervera-Mata *et al*. (2017), of increasing pH_w over time which may have allowed the mineralization of SCG. There were contrasting findings by Kasongo *et al*. (2011) and Yamane *et al*. (2014), who found an increase in organic carbon due to the carbon being incorporated with soil aggregates.

Cruz and Cordovil (2015) attributed low mineralization at the end of the incubation period (56-112 days) to the wide C:N ratio of the residue and the lack of mineralization due to amounts of caffeine as they act as nitrogen storing molecules. A study by Lui and Price (2011) revealed immobilization of nitrogen, however, and as the incubation period progressed net mineralization occurred. This was because the easily decomposable nitrogen was removed by coffee extraction leaving the insoluble N which was later attributed to the high rate of total N mineralization (Yamane *et al*. 2014). Contrasting data was found by Hardgrove and Livesley (2016), who observed immobilization of N across an incubation period of 3 months.

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Base cations (Mg, Ca, K) and available P

Amendment with SCG in the first test period increased Mg, Ca, and P above control and decreased K. However, the SCG nutrient content was lower in SCG as compared to the soil (Table 4.1). As the incubation period progressed, there were different stages of decomposition hence different SCG assimilation over time. Lower application rates were quicker to release nutrients and then followed by a decline, whereas in higher application rates nutrients were first immobilized by microorganisms followed by mineralization at the end of the incubation period.

Kasongo *et al*. (2011) observed an increase in K with a greater application rate after a year of application with coffee waste (coffee pulp and SCG); however, there was a small change in Ca and Mg concentration. They attributed this small change in these basic cations to a lack of sufficient water to promote dissolution to release the nutrients from coffee material. Phosphorus increase was contributed by the liming effect and P content of coffee waste (Kasongo *et al*., 2011). Diacona and Montemurro (2010) attributed the increase in P to the decreased P adsorption capacity of the soil due to pH or blockage of adsorption sites on soil colloids by organic molecules during manure decomposition. However, in the present study there was no correlation (0.04, $p<0.05$) between pH_w and P, therefore attributing the increase in P to blockage of adsorption sites by organic molecules.

Cervera-Mata *et al*. (2017), found that available K was related to SCG application rates and soil type, and the addition of SCG increased the amount of K in the soil due to large amounts of K in SCG. The incubation period slightly decreased the K levels, and it was attributed to K absorption by plants. However, Kasongo *et al*. (2011) observed an increase in K over the incubation period in a study without plants and attributed this to a higher application rate of the coffee waste. Vela-Cano *et al*. (2019) observed a decrease in K and P over time and attributed P to precipitation in carbonate-rich soil and K to the incorporation of this element in the illites interlayer. Cruz and Cordovil (2015) observed the same effect for SCG; however, they attributed P to immobilization by soil microorganisms. In the present study EC had a positive relationship with Mg, Ca, and K, suggesting an increase in EC increased these macronutrients.

4.2.4. Effect of spent coffee ground and incubation period on heavy metals

In this study, the application of SCG decreased the bioavailability of heavy metals across the incubation period, with the highest decrease being observed in SCG high application rate treatments. The findings agree with those by da Silva Correia *et al*. (2018) and Kim *et al*. (2014), who indicated that utilization of SCG in soil and water contaminated with heavy metals (Pb, Ni, Cu, Cd, and As) decreased their bioavailability and accessibility thus illustrating their ability to adsorb heavy metals. Kim *et al*. (2014) further elaborated that the adsorption of heavy metals to the surface of SCG might have been because of electrostatic forces by their anionic organic functional group. Cations with higher electronegativity tend to be adsorbed more (Inyang *et al*., 2012).

Even though SCG decreased the values of Cd below control, Cd values were above the maximum permissible element concentration of 2 mg/Kg (Herselmana *et al*., 2005). Spent coffee ground as well decreased the values of Cu, Ni, Zn, and Pb, however, they were never above the maximum permissible element concentration level. Additionally, Cu and Zn were deficient for plant absorption since the values were below the plant availability level (Herselmana *et al*., 2005). This was in contradiction with studies by Morikawa and Saigusa (2008) and Cervera-Mata *et al*. (2017) who found that incorporation of SCG into the soil increased micronutrient such as Cu, Fe, and Zn for plant absorption.

4.2.5. Effect of spent coffee ground on soil aggregate fraction and stability

In this study, the application of SCG significantly increased MWD under SCG application rate SCG-5 and more, across the incubation period. However, the highest increase of MWD was observed in high application rate treatments (SCG-20 through to SCG-50). The findings were in line with a study by Cervera-Mata *et al*. (2019), who found that structural stability of aggregates increased with both SCG application rate and time. Greater MWD was associated with high addition of organic matter.

During the incubation period, SCG increased the LM fraction while reducing sM, m, and s+c in all treatments as compared to control. Additionally, TOC had a positive relationship with LM, and was negatively related to sM, m, and s+c, suggesting that TOC facilitated the formation of LM fraction in the soil. That is, TOC played a role in binding primary particles (s+c) together in the fraction of microaggregates (m) and acting as a nucleus where formed microaggregates accumulate around it to form macroaggregates (sM then LM) (Bronick and Lal, 2005). Moreover, Weil and Brady (2016) indicated that organic matter joins with mineral particles of the soil, forming first the microaggregates. The fraction of microaggregates was facilitated by the labile SOC pool whereas macroaggregates were formed by the resistant pool which was more difficult to be broken down by microorganisms (Bronick and Lal, 2005). These findings agreed with a study by Cervera-Mata *et al*. (2019), who observed an increased percentage of macroaggregates and decreased percentage of meso- and microaggregates post amendment with SCG.

A positive relationship existed between LM and SMC, suggesting an increase in LM fraction increased SMC. pH_W had a negative relationship with LM, and positively related with sM, m, and s+c, suggesting decrease in pHw increased LM. There was a strong relationship between LM and MWD which suggested increase in LM increased MWD of the soil. Similar findings were reported by Lawal *et al*. (2012), they suggested that the proportion of LM fraction in the soil dictated more than 50% of the MWD value.

4.2.6. Effect of spent coffee ground and incubation period on SMC

Amelioration with SCG-50 in the 1st test period increased SMC by 798% above control. The increase in SMC was greatly attributed to SCG particle size of 20 µm. The micro-sized particles of the residue result in a high specific area of 7500 m^2/kg to hold more water (Turek *et al*., 2019). Voroney (2019) additionally indicated that organic matter increases the adhesion forces thus more water is retained in the soil. However, at test period T9, SMC had decreased to 638% compared with control.

The levels of SMC in SCG-50 were too greater than field capacity as it was above optimum storage level of 20% of 1m soil (Brandt, 2019). A study by Turek *et al*. (2019) revealed SCG's high application rate increased soil moisture, and readily available plant water, however it reduced drainable porosity and promoted poor crop development.

CHAPTER 5

CONCLUSIONS AND RECOMMENDATIONS

This study quantified the effect of various rates of SCG on physicochemical properties of the soil across the incubation period and explored the relationships between variables. Interaction between incubation periods and various SCG application rates significantly increased pHw and EC. Spent coffee ground low application rate raised pHw above control at T6 and T9 test period showing some liming capabilities. Although SCG raised pHw across the incubation period for all treatments, this increment fell within the 5.5 to 6.5 optimal range for plant growth. Spent coffee ground significantly increased the EC of the soil and improved the supply of N, P, Ca, Mg and K. Heavy metals significantly decreased across the incubation period, this also reduced essential nutrients (Zn and Cu). TOC and C:N ratio were increased at the first test period, but drastically decreased across the incubation period. C:N ratio at T9 test period fell within the 10-14 optimum range.

In this study, the application of SCG significantly increased MWD under SCG application rate SCG-5 and more, across the incubation period. During the incubation period, SCG increased the formation of large macroaggregates (LM) with the drastic increase observed in SCG-50. TOC and pHw showed a positive relationship with LM fraction suggesting an increase in TOC or pHw increased LM formation. Large macroaggregates had a positive correlation with MWD, suggesting when LM increase, MWD also increased. Soil moisture content decreased across the incubation period; however, it was still 638% higher than control. The use of SCG may help in water management in water scarcity areas; findings show MWD was positively correlated with SMC.

Various SCG application rates and different incubation periods played a role in nutrient cycling. All treatments showed potential if employed in different management strategies. For instance, SCG lower application rates released nutrients in the 2nd test period (T3) whereas with higher application rates nutrients were first immobilized and then released at T6 and T9. This trend then gives variable ways to utilize the different application rates. However, research should be done in field conditions to access the effectiveness of this residue.

REFERENCES

Angelova, V. R. Akova, V. I. Artinova N. S and Ivanov, K. I. 2013. The Effect of Organic Amendments on Soil Chemical Characteristics. *Bulgarian Journal of Agricultural Science*. 19: 958-971.

Agricultural research council (ARC). 2014. coffee. [ONLINE] Available at: http://www.arc.agric.za/arc-itsc/Pages/Coffee-Information.aspx. [Accessed 7 May 2019].

Pfluger, R.A. 1975. *Soluble coffee processing. In: Mantell, C.L. (Ed.), Solid Wastes, Origin, Collection, Processing, and Disposal*. Wiley, New York.

Ballesteros, L.F. Teixeira, J.A. and Mussatto S.I. 2014. Chemical, Functional and Structural Properties of SCGs and Coffee Silverskin. *Food Bioprocess Technology*. 7:3493–3503.

Black, C.A. (1965) Methods of Soil Analysis: Part I, Physical and Mineralogical Properties. *American Society of Agronomy*. Madison, Wiscosin, USA.

Bouyoucos, G.J. 1962. Hydrometer method improved for making particle size analysis of soils. *Agronomy Journal*. 54:464-465.

Brandt, M.J. Johnson, K. M. Elphinston, A.J. and Ratnayaka, D.D. 2017. *Twort's Water Supply: Hydrology and Surface Supplies*. Amsterdam: Elsevier Ltd

Bronick C.J. and Lal R., 2005. Soil structure and management: A review. *Geoderma*. 124:3-22.

Cameron, A and O'Malley. S. 2016. *Coffee Ground Recovery Program: Summary Report Sydney report*. Available at: https://vdocuments.mx/coffee-ground-recovery-program-summary-report.html (Accessed March 3, 2022),

Campos-Vega, R. Loarca-Pina, G. Vergara-Castaneda, H.A. and Oomah B.D. 2015. Spent coffee grounds: A review on current research and prospects. *Trends in Food Science and Technology*. 45:24-36.

Cervera-Mata, A. Martín-García, J.M. Delgado, R. Párraga, J. Sánchez-Marañón, M. and Delgado, G. 2019. Short-term effects of spent coffee grounds on the physical properties of two Mediterranean agricultural soils. *International Agrophysics*. 33:205-216.

Cervera-Mata, A. Pastoriza, S. Rufián-Henares, J. Á. Párraga, J. Martín-García, J.M. and Delgado, G. 2017. Impact of Spent Coffee Grounds as Organic Amendment on Soil Fertility and Lettuce Growth in Two Mediterranean Agricultural Soils. *Archives of Agronomy and Soil Science*. 64:1-15.

Civeira, G. 2019. *Introductory Chapter: Soil Moisture*. London, Intechopen.

Cruz, R. Cardoso, M. M. Fernandes, L. Oliveira, M. Mendes, E. Baptista, P. Morais, S. and Casal, S. 2012. Espresso coffee residues: a valuable source of unextracted compounds. *Journal of Agricultural and Food Chemistry*. 60:7777 - 7784.

Cruz, R. Mendes, E. Torrinha, Á. Morais, S. Pereira, J.A. Baptista, P. and Casal, S. 2015. Revalorization of spent coffee residues by a direct agronomic approach. *Food Resource International.* 73: 190-196

Cruz, R. Morais, S. Mendes, E. Pereira, J.A. Baptista, P. and Casal, S. 2014. Improvement of vegetables elemental quality by espresso coffee residues. *Food Chemistry*. 148:294-299.

Cruz, S. and Cordovil, C. MdS. 2015. Espresso coffee residues as a nitrogen amendment for small-scale vegetable production. *Journal of the Science of Food and Agriculture*. 95:3059-3066.

da Silva Correia, I.K. Santos, P.F. Santana, C.S. Neris, J.B. Luzardo, F.H.M. and Velasco, F.G. 2018. Application of coconut shell, banana peel, spent coffee grounds, eucalyptus bark, piassava (*Attalea funifera*) and water hyacinth (*Eichornia crassipes*) in the adsorption of Pb^{2+} and Ni^{2+} ions in water. *Journal of Environmental Chemical Engineering*. 6: 2319-2334.

Davies, D.E. 1974. Loss on ignition as an estimate of organic matter*. Soil Science Society of American Production.* 38: 150

Davila-Guzman, N.E. Cerino-Córdova, F.J. Loredo-Cancino, M. Rangel-Mendez, J.R. Gómez-González, R. Soto- Regalado, E. 2016. Studies of Adsorption of Heavy Metals onto Spent Coffee Ground: Equilibrium, Regeneration, and Dynamic Performance in a Fixed-Bed Column. *International Journal of Chemical Engineering.* 2016:11.

Diacono, M. and Montemurro, F. 2010. Long-term effects of organic amendments on soil fertility. A review. *Agronomy for Sustainable Development.* 30:401-422.

Elliott, E.T. 1986. Aggregate structure and carbon, nitrogen and phosphorus in native and cultivated soils. *Soil Science Society of America Journal*. 50:627-633.

Fenoll, J. Vela, N. Navarro, G. Pérez-Lucas, G. and Navarro, S. 2014. Assessment of agro-industrial and composted organic wastes for reducing the potential leaching of triazine herbicide residues through the soil. *Science of the Total Environment.* 493:124-132.

Haider, F.U. Liqun, C. Coulter, J.A. Cheema, S.A. Wu, J. Zhang, R. Wenjun, M. and Farooq, M. 2021. Cadmium toxicity in plants: Impacts and remediation strategies. *Ecotoxicology and Environmental Safety*. 211:

Hardgrove, S.J. and Livesley, S.J. 2016. Applying Spent Coffee Grounds Directly to Urban Agriculture Soils Greatly Reduces Plant Growth. *Urban Forestry and Urban Greening*. 18:1–8.

Herselmana, J.E. Steynb C.E. and Fey, M.V. 2005. Baseline concentration of Cd, Co, Cr, Cu, Pb, Ni and Zn in surface soils of South Africa. *South African Journal of Science* 101:509-512.

Insight Survey, 2022. *South African Industry Landscape Report. Johannesburg: B2B market research*. Available at: https://www.bizcommunity.com/Article/196/78/201839.html (accessed March 4, 2021).

International Coffee Organization (ICO), 2021. *Trade table Statistics: World Coffee Consumption*. Available at: http://www.ico.org/prices/new-consumption-table.pdf (Accessed March 4, 2022).

Inyang, M. Gao, B. Yao, Y. Xue, Y. Zimmerman, A.R. Pullammanappallil, P. and Cao, X. 2012. Removal of heavy metals from aqueous solution by biochars derived from anaerobically digested biomass. *Bioresource technology*. 110:50-6.

Janissen, B. and Huynh, T. 2018. Chemical composition and value-adding applications of coffee industry by-products: a review. *Resource, Conservation and Recycling*. 128: 110–117.

Kasongo, R.K. Verdoodt, A. Kanyankagote, P. Baert, G. and Van Ranst, E. 2011. Coffee waste as an alternative fertilizer with soil improving properties for sandy soils in humid tropical environments. *Soil Use Management*. 27: 94–102.

Kasongo, R.K. Verdoodt, A. Kanyankagote, P. Baert, G. and Van Ranst, E. 2013. Response of Italian ryegrass (Lolium multiflorum Lam.) to coffee waste application on a humid tropical sandy soil. *Soil Use and Management.* 29: 22-29

Kim, M.S. and Kim, J.G. 2020. Adsorption Characteristics of Spent Coffee Grounds as an Alternative Adsorbent for Cadmium in Solution. *Environments*. 7:24.

Kim. M.S. Min, H.G. Koo, N.M. Park, J.S. Lee, S.H. Bak, G.I. and Kim, J.G. 2014. The effectiveness of spent coffee ground and its biochar on the amelioration of heavy metals contaminated water and soil using chemical and biological assessments. *Journal of Environmental Management*. 146:124130.

Lawal, M.H. Ogunwole, O.J. and Uyovbisere, E.O. 2012. Reciprocal relationships between aggregate stability and organic carbon characteristics in a forested ecosystem of northern Nigeria. *Tropical and Subtropical Agroecosystems.* 15:481-488.

Lin, C and Coleman, N.T. 1960. The measurement of ex-changeable aluminum in the soil and clay. *Soil Science Society of America Proceedings.* 24: 444-446

Liu, K. and G.W. Price. 2011. Evaluation of three composting systems for the management of spent coffee. *Bioresource Technology.* 102:7966-7974.

McNutt, J. and He, Q.S. 2019. Spent coffee grounds: A review on Current Utilization. *Journal of Industrial and Engineering Chemistry*. 71:76-88.

Morikawa. C.K. and M. Saigusa. 2008. Recycling coffee grounds and tea leaf wastes to increase plant available Fe in alkaline soils. *Plant and Soil*. 304:249-255.

Murthy, P. S. and Naidu, M. M. 2012. Sustainable Management of Coffee Industry By-Products and Value Addition - A review. *Resources Conservation and Recycling*. 66:45-58.

Mussatto, S. I. Machado, E. M. S. Martins, S. and Teixeira, J. A. 2011. Production, Composition, and Application of Coffee and its Industrial Residues. *Food and Bioprocess Technology*. 4:661-672.

Pujol, D. Liu, C. Gominho, J. Olivella, M.À. Fiol, N. Villaescusa, I. and Pereira, H. 2013. The chemical composition of exhausted coffee waste. *Industrial Crops and Products*. 50:423-429.

Santos, C. Fonseca, J. Aires, A. Coutinho, J. and Trindade, H. 2016. Effect of different rates of spent coffee grounds (SCG) on composting process, gaseous emissions and quality of end-product. *Waste Management*. 59:37-47.

Turek, M.E. Freitas, K.S. and Armindo, R.A. 2019. Spent Coffee Grounds as Organic Amendment Modify Hydraulic Properties in a Sandy Loam Brazilian Soil. *Agricultural Water Management*. 222: 313-321.

Vela-Cano, M. Cervera-Matac, A. Purswania, J. Pozoa, C. Delgadoc, G. and González-López. J. 2019. Bacterial Community Structure of two Mediterranean Agricultural Soils Amended with Spent Coffee Grounds. *Applied Soil Ecology*. 137:12–20.

Voroney P. 2019. *Soils for Horse Pasture management*. Amsterdam: Elsevier Inc

Weil, R.R. and Brady, N.C. 2016. *The Nature and Properties of Soils*. New York: Person Education.

Yamane, K. Kono, M. Fukunaga, T. Iwai, K. and Sekine, R. 2014. Field Evaluation of Coffee Grounds Application for Crop Growth Enhancement, Weed Control, and Soil Improvement. *Plant Production Science*. 17: 93-102.

Zulfiqar, U. Farooq, M. Hussain, S. Maqsood, M. Hussain, M. Ishfaq, M. Ahmad, M. and Anjum, M.Z. 2019. Lead toxicity in plants: impacts and remediation. *Journals of Environment Management.* 250: 109557.

APPENDICES

Appendix 4.1: Factorial analysis of variance (ANOVA) for PH_W to different incubation periods and SCG various application rates effect.

Source	DF	SS	MS	F	P
rep	3	0,1539	0,0513		
T	3	0,7177	0,23924	3,47	0,0206
SCG-AR	5	11,4431	2,28862	33,22	0
T* SCG-AR	15	2,6698	0,17798	2,58	0,004
Error	69	4,7532	0,06889		
Total	95	19,7377			

Appendix 4.2: Factorial ANOVA for PH_{KCl} to different incubation periods and SCG various application rates effect.

Source	DF	SS	MS	F	P
rep	3	0,0984	0,03281		
T	3	0,5584	0,18612	2,19	0,0973
SCG-AR	5	7,5859	1,51717	17,83	0
T* SCG-AR	15	1,6989	0,11326	1,33	0,2082
Error	69	5,8707	0,08508		
Total	95	15,812			

Appendix 4.3: Factorial ANOVA for electrical conductivity to different incubation periods and SCG various application rates effect.

Source	DF	SS	MS	F	P
rep	3	2264	754,8		
T	3	19740	6580	19,61	0
SCG-AR	5	69279	13855,8	41,3	0
T* SCG-AR	15	11549	769,9	2,29	0,0105
Error	69	23148	335,5		
Total	95	125980			

Appendix 4.4: Factorial ANOVA for total organic carbon to different incubation periods and SCG various application rates effect.

Source	DF	SS	MS	F	P
rep	3	1,995	0,665		
T	3	16,339	5,446	4,8	0,0043
SCG-AR	5	811,185	162,237	142,87	0
T* SCG-AR	15	43,54	2,903	2,56	0,0044
Error	69	78,354	1,136		
Total	95	951,413			

Appendix 4.5: Factorial ANOVA for nitrogen to different incubation periods and SCG various application rates effect.

Source	DF	SS	MS	F	P
rep	3	0,06792	0,02264		
T	3	0,09832	0,03277	3,92	0,0121
SCG-AR	5	2,24886	0,44977	53,82	0
T* SCG-AR	15	0,05755	0,00384	0,46	0,9529
Error	69	0,57667	0,00836		
Total	95	3,04931			

Appendix 4.6: Factorial ANOVA for C:N ratio to different incubation periods and SCG various application rates effect.

Source	DF	SS	MS	F	P
rep	3	28,45	9,482		
T	3	389,03	129,677	6,79	0,0004
SCG-AR	5	682,92	136,585	7,15	0
T* SCG-AR	15	217,04	14,469	0,76	0,7179
Error	69	1317,86	19,099		
Total	95	2635,3			

Appendix 4.7: Factorial ANOVA for magnesium to different incubation periods and SCG various application rates effect.

Source	DF	SS	MS	F	P
rep	3	25,9	8,63		
T	3	3050,3	1016,76	1007,13	0
SCG-AR	5	3809,7	761,93	754,72	0
T* SCG-AR	15	8405,6	560,38	555,07	0
Error	69	69,7	1,01		
Total	95	15361,2			

Appendix 4.8: Factorial ANOVA for calcium to different incubation periods and SCG various application rates effect.

Source	DF	SS	MS	F	P
rep	3	9,2	3,05		
T	3	2592,9	864,284	842,71	0
SCG-AR	5	2846,7	569,332	555,12	0
T* SCG-AR	15	7263,4	484,228	472,14	0
Error	69	70,8	1,026		
Total	95	12782,8			

Appendix 4.9: Factorial ANOVA for potassium to different incubation periods and SCG various application rates effect.

Source	DF	SS	MS	F	P
rep	3	449,1	149,7		
T	3	33582,4	11194,1	381,02	0
SCG-AR	5	12444,6	2488,9	84,72	0
T* SCG-AR	15	19468,2	1297,9	44,18	0
Error	69	2027,2	29,4		
Total	95	67971,5			

Appendix 4.10: Factorial ANOVA for phosphorus to different incubation periods and SCG various application rates effect.

Source	DF	SS	MS	F	P
rep	3	94	31,32		
T	3	6330,7	2110,22	407,47	0
SCG-AR	5	4682,4	936,48	180,83	0
T* SCG-AR	15	12948,4	863,23	166,68	0
Error	69	357,3	5,18		
Total	95	24412,8			

Appendix 4.11: Factorial ANOVA for cadmium to different incubation periods and SCG various application rates effect.

Source	DF	SS	MS	F	P
rep	3	8,09	2,697		
T	3	668,77	222,922	766,26	0
SCG-AR	5	492,96	98,592	338,89	0
T* SCG-AR	15	1614,7	107,647	370,02	0
Error	69	20,07	0,291		
Total	95	2804,59			

Appendix 4.12: Factorial ANOVA for copper to different incubation periods and SCG various application rates effect.

Source	DF	SS	MS	F	P
rep	3	0,00032	0,00011		
T	3	0,52781	0,17594	157,98	0
SCG-AR	5	1,335	0,267	239,75	0
T* SCG-AR	15	2,7527	0,18351	164,78	0
Error	69	0,07684	0,00111		
Total	95	4,69267			

Appendix 4.13: Factorial ANOVA for nickel to different incubation periods and SCG various application rates effect.

Source	DF	SS	MS	F	P
rep	3	1,087	0,3622		
T	3	133,81	44,6033	2014,09	0
SCG-AR	5	61,774	12,3547	557,89	0
T* SCG-AR	15	186,633	12,4422	561,84	0
Error	69	1,528	0,0221		
Total	95	384,832			

Appendix 4.14: Factorial ANOVA for zinc to different incubation periods and SCG various application rates effect.

Source	DF	SS	MS	F	P
rep	3	0,448	0,1493		
T	3	34,825	11,6082	716,83	0
SCG-AR	5	28,669	5,7339	354,08	0
T* SCG-AR	15	67,727	4,5151	278,82	0
Error	69	1,117	0,0162		
Total	95	132,786			

Appendix 4.15: Factorial ANOVA for lead to different incubation periods and SCG various application rates effect.

Source	DF	SS	MS	F	P
rep	3	1,636	0,5454		
T	3	46,036	15,3453	667,25	0
SCG-AR	5	36,681	7,3361	318,99	0
T* SCG-AR	15	126,495	8,433	366,69	0
Error	69	1,587	0,023		
Total	95	212,435			

Appendix 4.16: Factorial ANOVA for large macroaggregates to different incubation periods and SCG various application rates effect.

Source	DF	SS	MS	F	P
rep	3	0,06469	0,02156		
T	3	0,63649	0,21216	30,8	0
SCG-AR	5	3,41792	0,68358	99,24	0
T* SCG-AR	15	0,56466	0,03764	5,47	0
Error	69	0,47529	0,00689		
Total	95	5,15905			

Appendix 4.17: Factorial ANOVA for small macroaggregates to different incubation periods and SCG various application rates effect.

Source	DF	SS	MS	F	P
rep	3	0,03432	0,01144		
T	3	0,09632	0,03211	7,27	0,0003
SCG-AR	5	0,79374	0,15875	35,94	0
T* SCG-AR	15	0,23555	0,0157	3,55	0,0002
Error	69	0,30481	0,00442		
Total	95	1,46474			

Appendix 4.18: Factorial ANOVA for microaggregates to different incubation periods and SCG various application rates effect.

Source	DF	SS	MS	F	P
rep	3	0,01733	0,00578		
T	3	0,02768	0,00923	4,41	0,0067
SCG-AR	5	1,02795	0,20559	98,37	0
T* SCG-AR	15	0,09096	0,00606	2,9	0,0014
Error	69	0,14421	0,00209		
Total	95	1,30813			

Appendix 4.19: Factorial ANOVA for unaggregated silt and clay to different incubation periods and SCG various application rates effect.

Source	DF	SS	MS	F	P
rep	3	0,00055	1,85E-04		
T	3	0,00511	1,71E-03	6,46	0,0006
SCG-AR	5	0,03005	6,01E-03	22,78	0
T* SCG-AR	15	0,00165	1,10E-04	0,42	0,9697
Error	69	0,01821	2,64E-04		
Total	95	0,05558			

Appendix 4.20: Factorial ANOVA for mean weight diameter to different incubation periods and SCG various application rates effect.

Source	DF	SS	MS	F	P
rep	3	0,1448	0,04827		
T	3	2,0804	0,69346	49,48	0
SCG-AR	5	7,3401	1,46802	104,74	0
T* SCG-AR	15	1,0107	0,06738	4,81	0
Error	69	0,9671	0,01402		
Total	95	11,543			

Appendix 4.21: Factorial ANOVA for soil moisture content to different incubation periods and SCG various application rates effect.

Source	DF	SS	MS	F	P
rep	3	27,3	9,1		
T	3	404,7	134,91	13,87	0
SCG-AR	5	18540,9	3708,18	381,21	0
T* SCG-AR	15	999,2	66,61	6,85	0
Error	69	671,2	9,73		
Total	95	20643,3			