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ORGANIC MATTER CONTRIBUTION OF AMELIORANTS TO RECLAIMED SURFACE MINED SOIL AND CARBON STORAGE

By

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In loving memory of my father

Rudolph W. Coetzee

(8 August 1955 – 7 December 2014)

DECLARATION

I, Marc Coetzee declare that the dissertation, which I hereby submit for the degree MSc Environmental Management at the University of Pretoria, is my own work and has not previously been submitted by me for a degree at this or any other tertiary institution.

Signed……………………………………

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ABSTRACT

Coal mining in South Africa is renowned for large scale removal of topsoil and subsoil through opencast mining. Such processes lead to an enormous amount of land degradation and thus, limit the land capability after mine closure. The removal and stockpiling of topsoil leads to adverse effects not only on the fertility of the soil but also the physical properties of the soil which greatly limit the ability of the soil to sustain plant development. Resultant mined soils have a varied nutrient content range most commonly exhibiting a decreased organic carbon content and as a result, scientists today, have invested a considerable amount of time and resources into research which is focused upon the sequestration of atmospheric carbon into stable soil organic matter as a means to reduce the concentrations of carbon in the atmosphere and at the same time improve the physical and chemical properties of the soil for plant growth. However, the processes affecting the carbon cycle of the reclaimed mine soil are not well understood and are highly complex thus, requiring further investigation. One of the most practical and most efficient ways to improve soil impacted by surface coal mining is to address soil organic carbon levels by physically incorporating organic matter into the soil as a soil ameliorant together with a continuous supply of organic matter through plant growth and decay as soil organic matter forms a very important component of early soil formation and the re-establishment of ecosystem functionality on rehabilitated post-mining sites. Introducing pastures on replaced topsoil will provide a rapid method to stabilize soil, build soil organic matter though root and plant decay and prevent soil erosion, all of which are crucial during the initial stages of mine reclamation. Vegetation development physically incorporates organic matter into the soil and thereby, aids in lowering the bulk density and helping to prevent compacted conditions. This creates a habitable environment for microbial populations to proliferate and ultimately support plant life. In this investigation we evaluate the use of different organic soil amendments and the use of a cover crop to improve the soil organic carbon content as well as the physical and chemical properties of soil impacted by a South African surface coal mine.

In this investigation 40, 1 m long by 1 m wide by 1.2 m high experimental soil research bins “mini-lysimeters” which were constructed to represent a rehabilitated soil/substrate profile which is commonly created through rehabilitation practices on surface coal mines in South Africa. The bins were filled with mine spoil, subsoil and topsoil all of which came from a coal mine in Mpumalanga. The trial consisted of ten different topsoil treatments including a control treatment consisting only of the cover soil. *Avena sativa* was planted as the winter cover crop and *Eragrostis tef* was planted as the summer test pasture. Results indicate that the lucerne, manure and woodchips combination treatment (T9) was the best overall treatment used in this trial and illustrated an increase in *A. sativa* aboveground biomass of 128% and an increase in *E. tef* aboveground biomass of 44% when compared to the control. It was the only treatment to have a significant difference (p<0.05) on the soil pH, bulk density, aboveground biomass and root biomass when compared to the control. Treatment T9 also illustrated an increase in the total amount of carbon stored within the soil after the *E. tef* harvest of 17% when compared to the control. This improvement to both the physical and chemical properties of the soil can also be attributed to the value in which each individual component within the combination treatment had and thereby complemented one another in terms of improving overall plant development. Conclusions from this trial have highlighted the value of using a combination soil ameliorant, made from resources found near coal mine sites in South Africa, which can address organic soil limitations and thus, have a substantial effect on both the physical and chemical soil properties which play such a crucial role in plant development and soil reclamation.

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CHAPTER 1

A REVIEW INTO THE VALUE OF CARBON STORAGE ON REHABILITATED SURFCE MINED SOIL

1. Introduction

Coal is the world's most abundant fossil fuel and the primary energy source in South Africa. South Africa relies heavily on the coal mining industry for electricity production and will continue to mine coal for many years to come. However, the mining of coal is a destructive process with very sensitive environmental and economic implications. In 2004, Neke and Du Plessis estimated that 100 000 hectares in the Eastern Highveld of South Africa had been negatively impacted by coal mining and have estimated that this may increase to 325 081 hectares with the economically mineable land available. Coal mining is renowned for resulting in large scale removal of topsoil and subsoil through opencast mining which can lead to an enormous amount of land degradation. Resultant overburden materials are unsuitable for plant growth as they are high in gravel content, have unfavorable particle size and have relatively shallow soil profiles (Bradshaw and Chadwick, 1980). These soils have a varied nutrient content range and in order to incorporate a successful reclamation project into work, one needs to understand the production values of each of these materials in such an environment. Therefore, reclamation and re-vegetation of drastically disturbed soils together with the development of profitable and environmentally sound land uses depend on a comprehensive understanding of the chemical, physical and biological properties of the soil under these new ecosystems (Shrestha and Lal, 2006).

A popular environmental topic of interest which directly affects the mining industry is the management of carbon and carbon emissions. The management of carbon has become an energy and environmental issue in South Africa as carbon emissions continue to rise and will continue to rise through the next century. In 2013, the atmospheric concentration of carbon dioxide in the world was measured at 397 ppm (Arce *et al*., 2014) and thus driving national policies and treaties to prioritize implementing carbon mitigation measures. National Treasury in South Africa has stated that they will be implementing carbon tax in 2016 in which South African companies will be paying R120 per ton of carbon dioxide emitted over the basic tax free threshold of 60% which is limited to 90% of total carbon emissions (Department of National Treasury, 2014). This carbon tax will cover emissions resulting directly from fuel combustion and gasification, and from non-energy industrial processes.

However, while these policies loom, there are carbon management strategies which exist and will help mitigate carbon emissions. One of the four carbon management methods which is of the greatest significance in terms of mine rehabilitation is terrestrial carbon sequestration in which carbon is captured and stored in the soil and in vegetation growing on such soil. Soil carbon ultimately derives from vegetation and, therefore, must be managed directly by the incorporation of soil nutrients and indirectly through aboveground management of vegetation. Soil organic matter (SOM) forms an important component of early soil formation and re-establishment of ecosystem functionality on rehabilitated post-mining sites (Pietrzyhowski and Daniels, 2014). Many approaches to increase terrestrial carbon storage are focused upon increasing the carbon content in the vegetation and most importantly, the carbon content within the soil. Therefore, re-vegetation of reclaimed mine land presents an excellent opportunity to optimize carbon storage as the soil often has very low inherent organic matter content. This illustrates the great potential for carbon storage through soil reclamation and applying appropriate soil management techniques (Shrestha and Lal, 2006). However, the fluxes and inputs affecting carbon balance of these restorative measures are not well documented. The processes affecting the carbon cycle of the reclaimed mine soil ecosystems must therefore, be investigated further as the natural processes involved in carbon sequestration of disturbed soils are highly complex and research results are relatively scarce.

2. Soil carbon sequestration

Soil contains approximately 75% more carbon than vegetation and twice the amount at the present atmospheric concentration, making it the largest carbon reservoir of the terrestrial carbon pool (Krishan *et al.*, 2009). Biogeochemical cycling of carbon between the atmosphere, oceans, biosphere and the soil is controlled over a period of 100 000 years (Berner, 2003) while the short-term carbon cycle spans over just decades and forms the focus when evaluating soil carbon improvement strategies with regards to agricultural and mining ecosystems.

Carbon sequestration is a natural process in which carbon is removed from the atmosphere and deposited in a reservoir of which two processes exist. The first being direct carbon sequestration by inorganic chemical reactions that convert carbon dioxide (CO_2) into soil inorganic carbon compounds and the second being indirect carbon sequestration, in which carbon sequestration occurs as plants essentially transform atmospheric CO_2 into plant biomass through photosynthesis and incorporate this biomass into the soil as humus (Nair, *et al.*, 2010). This incorporation of organic biomass into the soil is the main source of carbon and nutrients for microbial and plant growth. However, soil organic carbon has different ages which can be quantified by carbon 14 (C^{14}) dating. Through the process of photosynthesis, the plant incorporates carbon from CO_2 within the atmosphere into the plant's biomass in a proportion relative to the amount of C^{14} in the atmosphere (Nair, *et al.*, 2010). When the plant decays the amount of C^{14} decreases at a fixed rate due to radioactive decay which can then be determined and thus, the age of the carbon identified (Kaiser *et al.*, 2002).

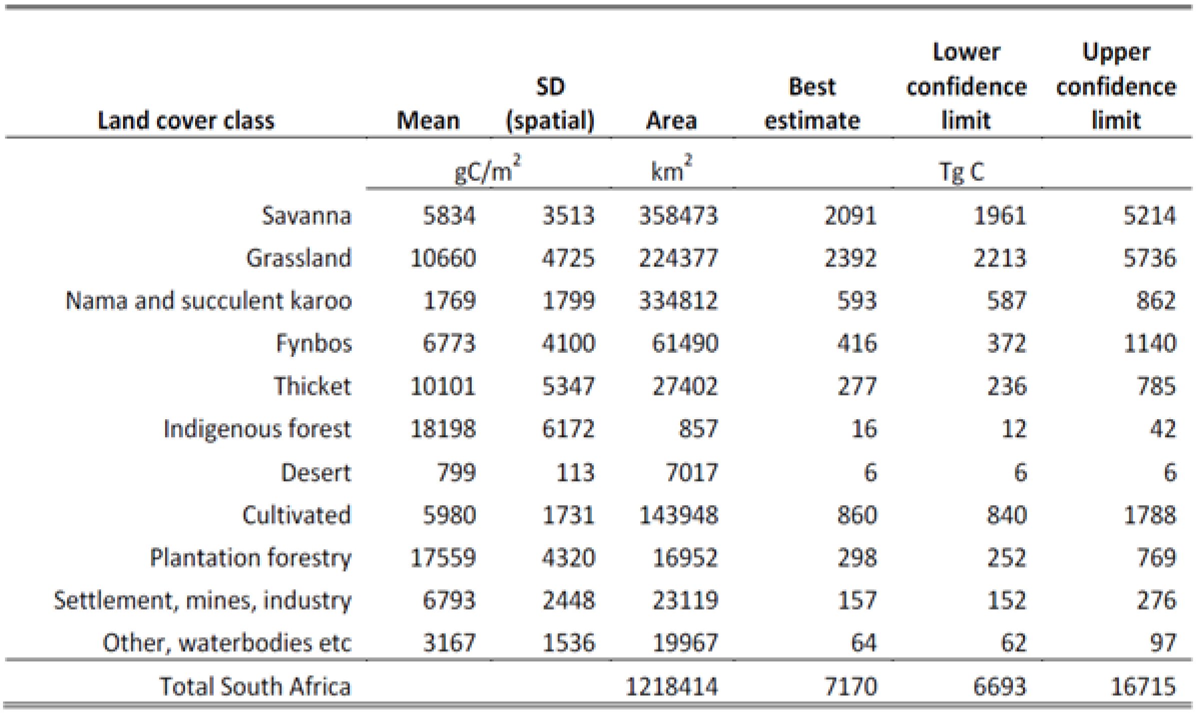
Angers and Chenu (1997) found that about one-third of the total SOM breaks down at a much slower rate compared to the rest of the organic matter and thus, can still be present in the soil one year later. Decomposition of organic matter, plant respiration and microbial respiration completes this cycle between the atmosphere and the soil (Tripathi *et al.*, 2014) and over time, and under undisturbed environmental conditions, the rates of carbon emissions and additions tend to level out, leading to a stabilization of the amount of soil organic carbon (Tripathi *et al.*, 2014). However, there are varying opinions regarding the SOM stability within the soil and the resistance of SOM to decomposition (Paustian *et al.*, 1998; Schmidt *et al.*, 2011). However, changes in land cover and/or mining have a large impact on the carbon cycle and consequently on the earth's climatic system. Hence, investigating the carbon cycle under different soil and vegetation types can play an important role towards the development of appropriate methodologies which enhance ecosystem functions and overall ecosystem health (Prescott *et al.*, 2000; Schoenholtz *et al.*, 2000; Chung *et al.*, 2012).

3. Terrestrial carbon stocks in South Africa

The global average atmospheric concentration of CO_2 has increased from 280 ppm from the industrial revolution in 1800 to a current level of approximately 397 ppm in 2013 with an annual increase of two ppm for the past decade (Arce *et al.*, 2014). This has resulted in major global and environmental changes, such as: increases in the frequency of extreme weather events, change in precipitation patterns and amounts, and global temperature increases, all of which can be attributed to burning fossil fuels, mining and drastic land use changes (Easterling *et al*, 2000). Therefore, mitigating the impact of global warming demands a reduction in atmospheric concentrations of CO_2 which has encouraged scientists to consider mitigating CO_2 concentrations in the atmosphere by facilitating and enhancing carbon sequestration in terrestrial ecosystems (Paustian *et al.*, 1998). Natural biological processes such as photosynthesis assimilates CO_2 and redistributes carbon into the plant tissue and reincorporates it into the soil where carbon can be stored for long periods of time. The long term ecosystem carbon balance is defined by the disturbances on carbon uptake by vegetation (Randerson *et al.*, 2002) and the amount of carbon from the atmosphere which is sequestered into plant biomass through photosynthesis is called Gross Primary Production (GPP), half of which is returned to the atmosphere through plant respiration (Scholes *et* al., 2014). This places emphasis on the fact that the majority of carbon is stored in the soil and thus, should be the focus of carbon storage projects. Furthermore, the conversion of CO_2 into soil organic matter helps to prevent carbon breakdown by microbial decomposition which emphasizes the importance of protecting and enhancing the natural capacity of plants and trees to facilitated carbon sequestration and reduces greenhouse gases.

In an approach to identify the total carbon stocks in South Africa, Scholes *et al*., (2014) followed a stratified-random procedure by using remote sensing and geostatistical methods to extrapolate field measurements to help estimate the average carbon stocks of each vegetation/land use type. From Figures 1 and 2 it is evident that the amount of carbon stored in the aboveground woody and herbaceous biomass increases from west to east in South Africa with the amount of carbon stored in the soil following the same pattern (Figure 3). These terrestrial carbon stocks are mainly determined by the following factors: temperature, soil conditions, plant available water and vegetation cover. It is evident from Figure 1 and Figure 2 that carbon stocks in the desert and Karoo biomes tend to be very low, while the carbon stocks found in the coastal and montane forests are much higher resulting from the large amount of trees present which have the ability to store larger amounts of carbon (Table 1). However, it is important to note that forest areas in South Africa constitute only a small proportion of the total land compared to grassland and savannah biomes. Therefore, while trees in these forests sequester larger amounts of carbon, the savannah and grassland biomes together tend to dominate the national carbon stocks with three quarters of South Africa’s total terrestrial carbon stock (Table 1). From Figure 4 and 5 it is evident that the coalfields coincide with these grassland and savannah biomes and thus, the mining of coal directly affects the carbon sequestration within these biomes. Therefore, in terms of coal mine rehabilitation, one should review restoring the carbon content of the soil as these areas play an important role in terms of sequestering large amounts of carbon dioxide.

Table 1: Total ecosystem organic carbon in South Africa (Scholes *et al.*, 2014).



Land cover class	Mean	SD (spatial)	Area	Best estimate	Lower confidence limit	Upper confidence limit
	gC/m^2		km^2		Tg C	
Savanna	5834	3513	358473	2091	1961	5214
Grassland	10660	4725	224377	2392	2213	5736
Nama and succulent karoo	1769	1799	334812	593	587	862
Fynbos	6773	4100	61490	416	372	1140
Thicket	10101	5347	27402	277	236	785
Indigenous forest	18198	6172	857	16	12	42
Desert	799	113	7017	6	6	6
Cultivated	5980	1731	143948	860	840	1788
Plantation forestry	17559	4320	16952	298	252	769
Settlement, mines, industry	6793	2448	23119	157	152	276
Other, waterbodies etc	3167	1536	19967	64	62	97
Total South Africa			1218414	7170	6693	16715

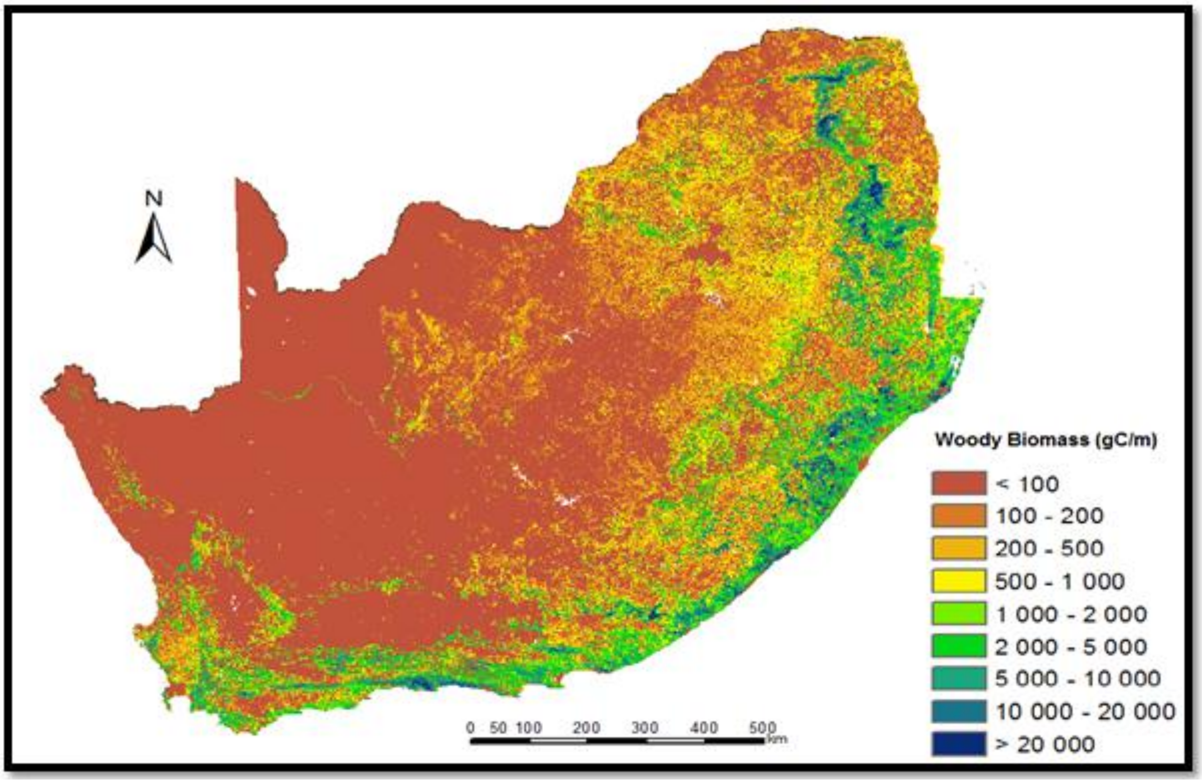


Figure 1: Map of South Africa illustrating carbon stocks in the aboveground woody biomass (Scholes *et al*., 2014).

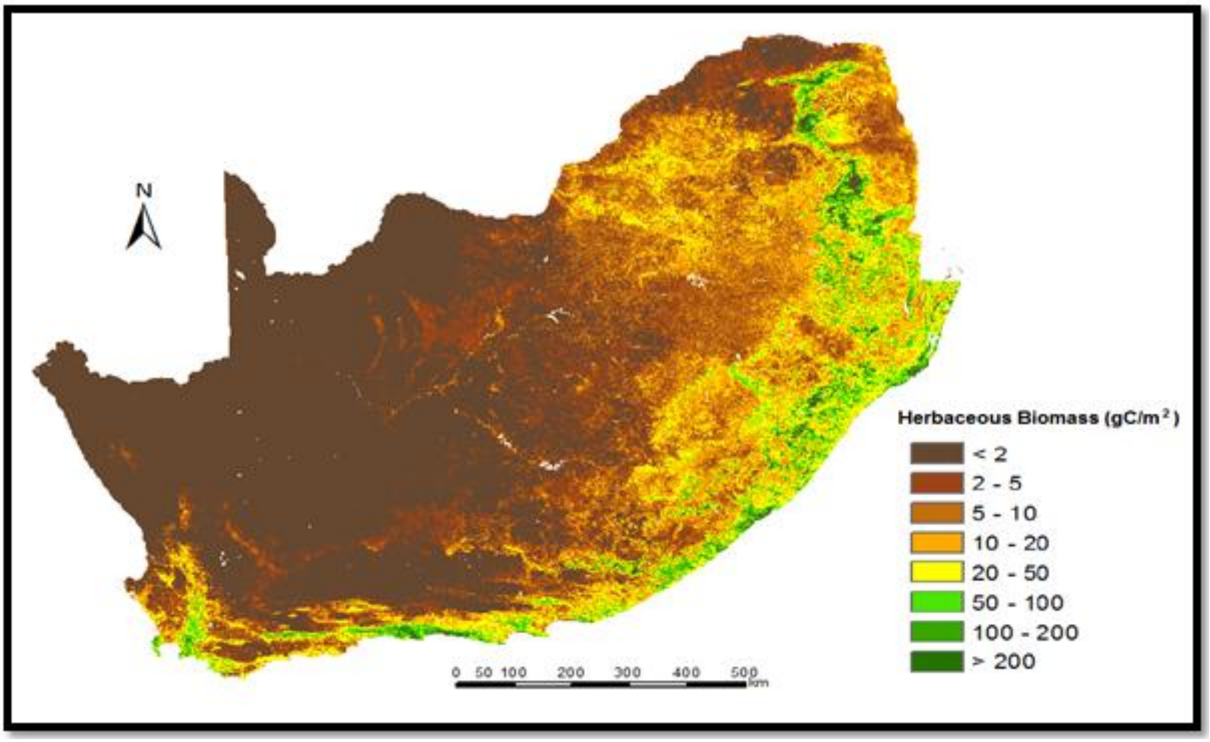


Figure 2: Map of South Africa illustrating carbon stocks in the aboveground herbaceous biomass (Scholes *et* al., 2014).

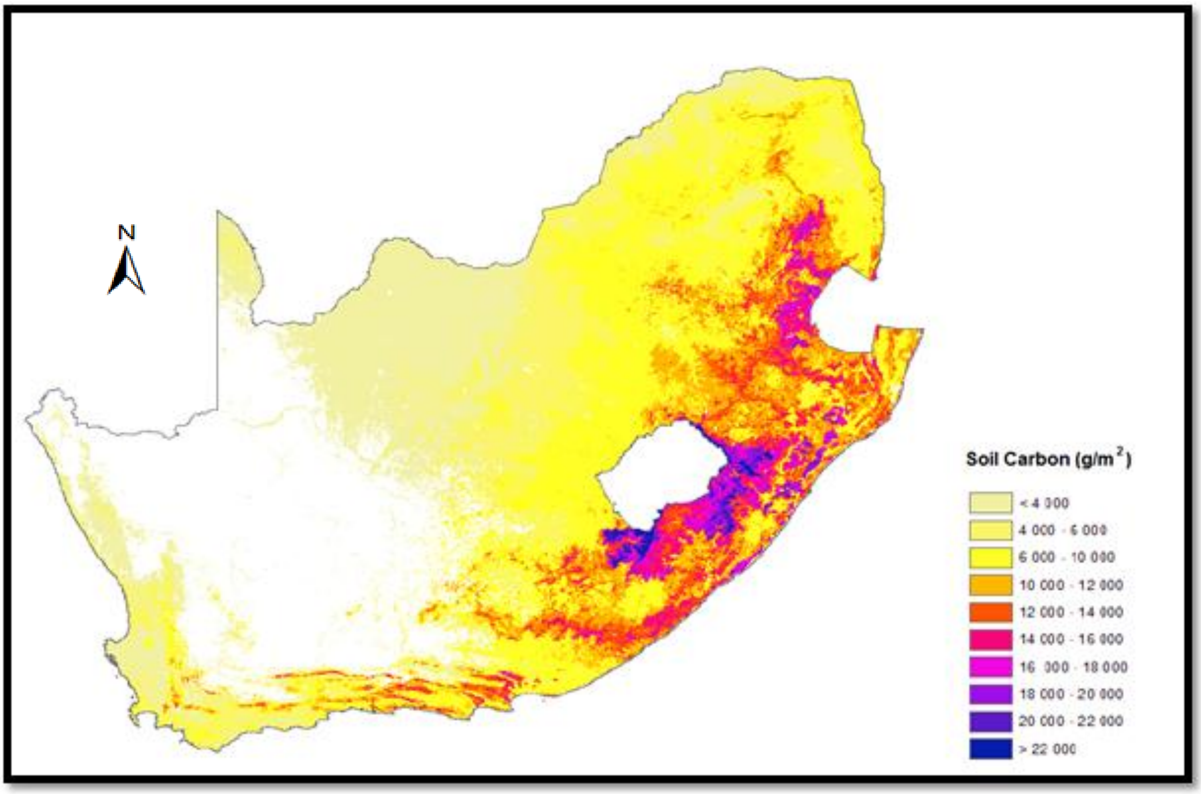


Figure 3: Map of South Africa illustrating carbon present in the soil (Scholes *et al*., 2014).

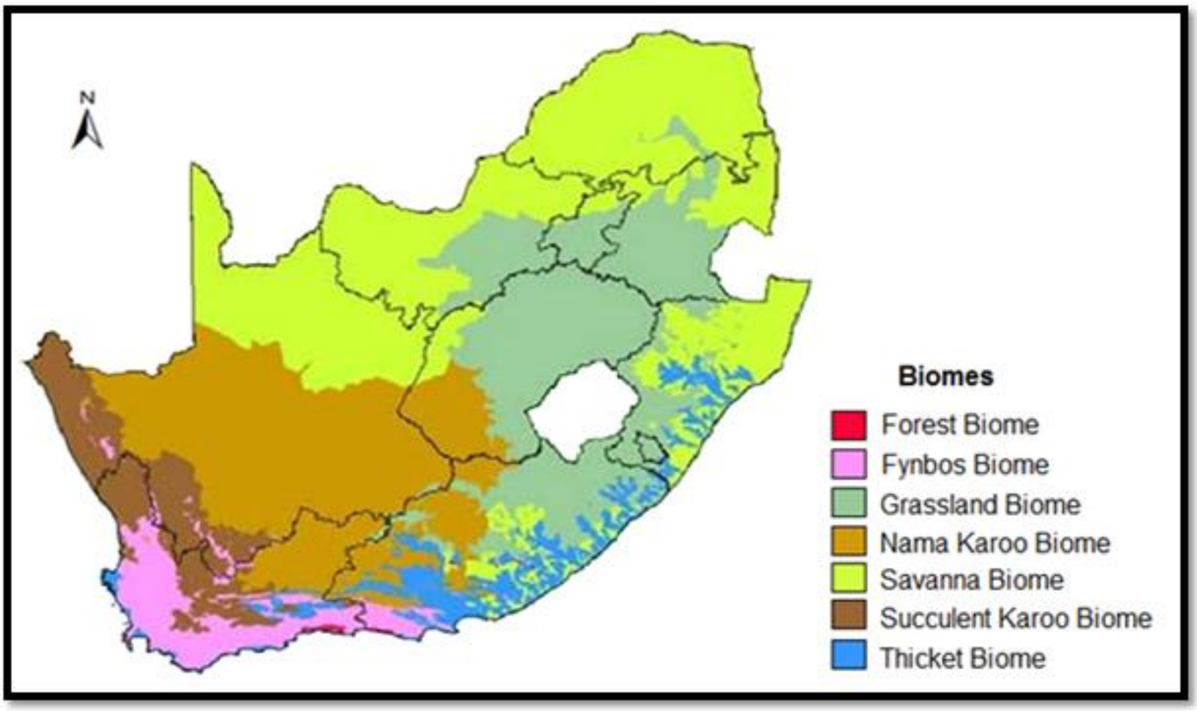


Figure 4: Map of South Africa illustrating the various vegetation biomes (Department of Environmental Affairs and Tourism, 2015).

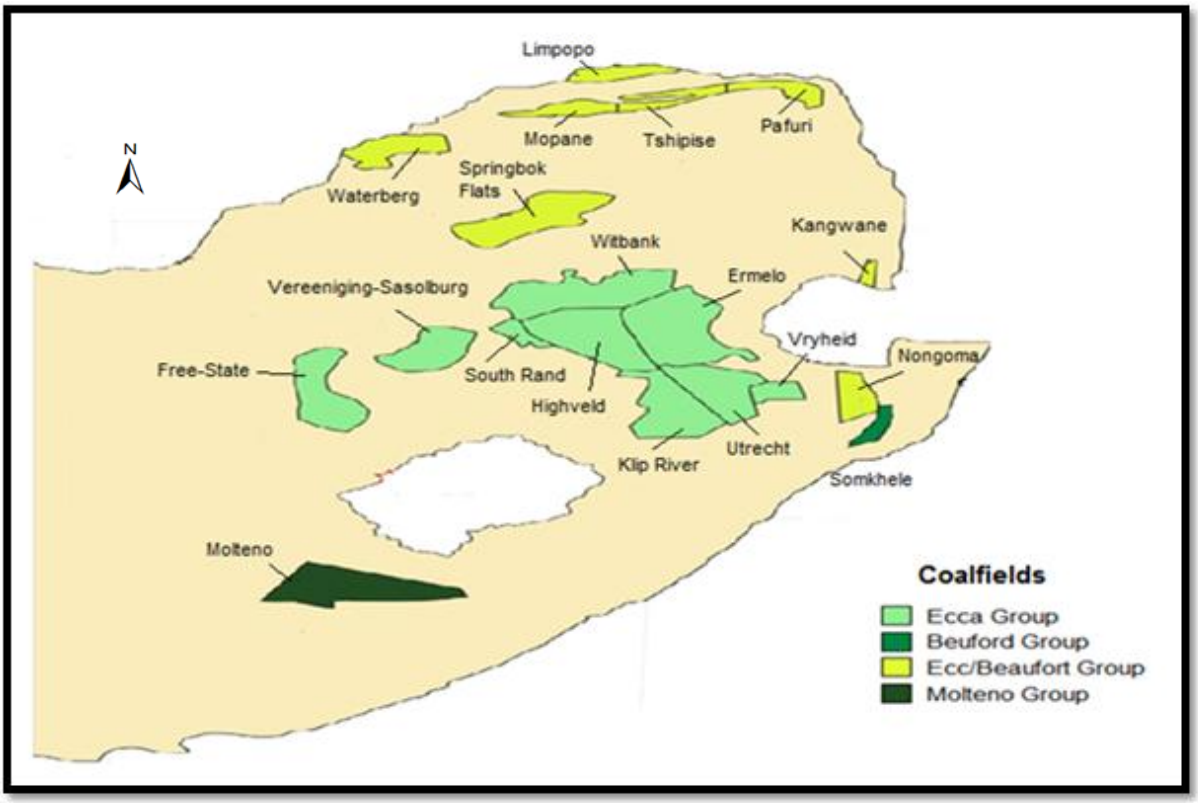


Figure 5: Map of South Africa illustrating the various coalfields (Hancox and Götz, 2014).

4. Surface mine rehabilitation in South Africa

Fairbanks *et al*., (2000) has estimated that South Africa has the following land cover percentages: 0.14% mines and quarries, 1.1% urban/built up land and 4.9% degraded land. While 175 421 ha consist of mines and quarries, a large amount of this land overlies arable soil in South Africa which encompasses 12% of the total land (GCIS, 2012). This begs the question as to what extent do mines and quarries impact the quality of arable land after mine closure and how can this be avoided or mitigated. In order to gain access to minable coal, the above lying material called overburden is excavated, removed and profiled. The highest proportion of waste produced by industrial activity is mine spoil, with billions of tons produced per year (Bell, 1998). Typically, topsoil is stripped and either placed on the profiled overburden or stockpiled. Once the topsoil has been placed, it is then leveled and prepared for seeding leading to vegetation establishment. The fertility, depth and slope of the topsoil which is placed on the overburden once mining in that area has ceased, will determine the land capability class of the soil.

Removal and stockpiling of topsoil leads to adverse effects on not only the fertility of the soil but also the bulk density of the soil and therefore, can exasperate the negative side effects on the plant production potential of the soil (Ross *et al*., 1982, 1984; Hart *et al*., 1986). The magnitude and impact of these surface mining techniques on the soil depends on the physical and chemical composition of the minerals present, the method of reclamation, surface and subsurface hydrological patterns and more importantly the skills, management and technology employed (Shrestha and Lal, 2006). Coal mine spoil is commonly characterized as containing a low amount of organic matter, having few plant available nutrients, limited soil microbes and little soil moisture as the spoil contains either a very coarse texture resulting in low water retention or a very compacted texture preventing water drainage (Jha and Singh, 1992; Singh *et al*., 1996; Tripathi and Singh, 2009; Tripathi *et al*., 2012). This negatively impacts the functionality of the ecosystem through adversely affecting the soil water and nutrient cycle (Insam and Domsch, 1988; Harris *et al*., 1993; Anderson *et al*., 2004). Taking into consideration the challenges mentioned above, rehabilitating surface mined soil which is aimed at improving the carbon content of the soil itself, poses a complicated and sophisticated task. Therefore, it is important that there exists a sound understanding of the challenges present and with concurrent mining and rehabilitation planning, one can identify the soil limitations before a reclamation plan is put into work and thereby increase the probability of reclaiming soil quality which can productively sustain vegetation.

According to the South African law, opencast mines are required to undergo rehabilitation with the end goal of producing a sustainable land use (Minerals and Petroleum Resources Act of 2002; National Environmental Management act). However, over the past few years, the South African mining industry has often witnessed poor planning with regards to addressing closure issues such as supplying sufficient time, personnel and finances to achieve satisfactory closure outcomes. This has resulted in a vast heritage of degraded land in the country. One of the many issues with regards to mine site rehabilitation is that the rehabilitation planning done by most mining companies in South Africa occurs during mining activity and during decommissioning rather than before the mining activity. This can lend itself to closure problems such as: insufficient funds left for rehabilitation and inability to address the needs of the interested and affected parties. Therefore, these mining companies should be targeting a balance between providing what the affected community of the mined land needs, providing a sustainable land use capability and minimal net loss of biodiversity, all of which can only be achieved with meticulous and well-timed mine closure planning.

5. Re-vegetation to enhance carbon storage

One of the objectives of mine rehabilitation in South Africa is to restore land to former agricultural capability by using pasture species which are adapted to the climatic region of the mine and are fertilizer responsive (Mentis, 2006). During the initial stages of surface mine soil re-vegetation, grasses are introduced to stabilize soils and reduce erosion on replaced topsoil while additionally providing a rapid method to build soil organic matter (Tripathi *et al*., 2014). The typical seed mix used on rehabilitated coal mines in South Africa comprises annual species such as *Eragrostis tef* in combination with perennial species such as *Eragrostis curvula, Cynadon dactylon, Cenchrus ciliaris, Digitaria eriantha and Medicago sativa* (Mentis, 1999). Grasses (C4) are often chosen because they offer greater drought tolerance, have a good rooting structure, require less soil nutrients and are more climate resilient. However, one must not forget that once the pasture has established, proper maintenance must follow in terms of supplying appropriate fertilizer applications and defoliation management. Without proper management, rehabilitated land will degrade and one will be forced to rehabilitate, rehabilitated land.

The introduction of a nitrogen fixing species such as a legume is often an approach in which nitrogen levels in the soil can be increased. However, in South Africa the use of legumes to naturally increase the nitrogen content in the soil during mine rehabilitation has not been properly explored. Reasons for this may be because legumes are not tolerant of acidic soils which commonly occur on surface coal mines in South Africa. Furthermore, there are very few indigenous legumes cultivated as a pasture crop and the fact that once the crop has established, one must be aware that these plants can cause bloat in ruminant animals grazing on land rehabilitated with legumes if not correctly managed. However, legumes have been found to be highly beneficial as they improve the nutrient cycle by not only increasing the nitrogen content of the soil but also producing decomposable nutrient rich litter through the turnover of fine roots and nodules (Tripathi *et al*., 2014). Additionally, it's important to note that plant roots contribute SOM through root death and exudation of organic substances during plant growth (Singh *et al*., 1991). Exudates comprise secretions, soluble compounds and lysates which have the potential for a greater contribution to the long-term soil carbon storage (Tripathi *et al*., 2014). However, different plant species produce different types and amounts of plant residues which will have an impact on the amount of SOM (Nair, *et al*., 2010). Additionally, the amount of carbon that is sequestered through photosynthesis is largely dependent on the plant species and the development of the plant itself, therefore, placing extreme importance on sustaining healthy soil conditions for plant growth. Hence, these species can have a direct impact on the biogeochemical carbon cycle and provide a pathway for carbon movement into deeper soil horizons (Tripathi *et al*., 2014).

6. Factors affecting ecosystem productivity and carbon storage

Soil compaction and hard setting is one of the most common and devastating problems found on open cast mine rehabilitation sites in South Africa (Nell and Steenekamp, 1998). This is often a result of intense trafficking of machinery on topsoil and the pressure created when topsoil is stockpiled (Bradshaw, 1997). Compaction limits root establishment and promotes soil erosion which makes proper development of grass species next to impossible (Jha and Singh, 1992; Singh *et al*, 1996). Compaction of stockpiled topsoil deteriorates the chemical properties of the soil as the oxygen within the soil becomes limited and an anaerobic environment results. Once such an environment is created, nitrogen can be lost through de-nitrification to the atmosphere as N_2 or N_2O (Davies *et al*., 1995). This further impacts the habitable conditions of the soil for organisms to proliferate (Haigh, 2000). Additionally the impact of compaction on rooting alters water drainage patterns and the water holding capacity of the soil in the rooting zone (Bell *et al*, 1994), which reduces overall grass development and thereby, having an impact on the amount of carbon stored within the soil. Failure of vegetation to properly develop on such soil results in lower inputs of organic matter in both the upper and lower regions of the soil (Tripathi *et al.*, 2014) further reducing the physical protection of the organic carbon through intra-aggregate and organo-mineral complexes (Post and Kwon, 2000). It is, therefore, extremely important that compacted mine soil undergoes rehabilitation in order to create a depth and moisture content which is favorable for plant development as the ecosystem productivity depends largely on the ability of the soil to support and sustain vegetation (Tripathi *et al.*, 2014). Vegetation development will physically incorporate organic matter into the soil and thereby, introduce microbial populations back into the soil which will aid in lowering the bulk density and help prevent compacted conditions (Bradshaw, 1997). Vegetation can also potentially improve the quality of the soil through the increase in water storage and infiltration, reduced loss of top soil through vegetation stabilization, improve nitrogen fixation through the introduction of nitrogen fixing plant species and improved the soil physical structure (Singh *et al*, 2004). Thus, the impact in which developing vegetation has on soil properties is profound.

6.1 Soil amendments

In South Africa, soil impacted by surface coal mining is commonly rehabilitated with inorganic fertilizers such as limestone ammonium nitrate, diammoium phosphate and potassium which promotes vegetation growth and thereby, aids in the reduction of soil compaction issues. While using inorganic amendments are useful and effective, they can be very costly, which is why resourceful organic amendments have gained substantial interest for use in increasing plant development and alleviating soil compaction. The use of organic amendments has also been found to not only address soil chemical deficiencies but also promote soil carbon storage (Nair, *et al.*, 2010). Nonetheless, soils devoid of sufficient phosphorus, magnesium, calcium and potassium are effectively permanent unless these nutrients are locked up in unweathered minerals and are released through natural weathering which can take many years (Williams and Walker, 1969).

In many cases carbon sequestration has been enhanced by the incorporation of a mixture of amendments, such as the by-products from coal combustion (fly ash) and mulch or compost (Matsi and Keramidas, 1999; Hearing *et al.*, 2000). The positive effects on the soil from the mixture mentioned above can be attributed to: fly ash enhancing flocculation between soil particles, thereby stabilizing soil structure and organic matter from compost adhering to mineral surfaces, thereby allowing for more reactive interactions between the plant roots, soil nutrients and water within the soil (Palumbo *et al.*, 2004). However, it is important to remember that certain organic amendments such as cattle manure are less likely to be retained in the soil over long periods of time and, therefore, must be made aware of when used alone or in combination with inorganic amendments as the manure will contribute little to long-term carbon sequestration (Palumbo *et al*., 2004). Conversely, woody biomass amendments which are recalcitrant and lignin-rich are known to degraded over longer periods of time and therefore, have a higher affinity for binding to soil particles and ultimately increasing long-term carbon storage (Palumbo *et al*., 2004). Studies have shown that certain soil amendments are capable of sequestering as much as 3 PgC yr^{-1} (Lal *et al*., 1998). Thus, it becomes important that focus be placed on enhancing the natural biological processes that assimilate CO_2 and direct the carbon into plant tissues and into microbial resistant pools of soil organic matter to achieve greater carbon sequestration (Palumbo *et al*., 2004). While the central role of carbon sequestration management is to enhance the conditions and the capacity of plants and soil to capture carbon, there is however, very little known about the interactions and overall functionality of carbon storage in terrestrial ecosystems (Palumbo *et al*., 2004).

6.2 Green manure

Cover crops are crops usually grown in the winter period and ploughed into the soil as a green manure (Poeplau and Don, 2015) of which there are four categories that exist, main crops, companion crops, catch crops and winter cover crops (Pieters, 2006). These crops are all broadly defined as crops grown for the physical and chemical benefit of the soil. Green manure crops have shown the following soil advantages: increasing soil organic matter content, increased biological activity within soil, increased soil pH, improved soil structure, suppressed water evaporation from the soil surface, reduced erosion and increased nutrient supply within the soil (Harris and Megharaj, 2001; Pieters, 2006). Furthermore, it is important to note that different green manuring strategies must be selected depending on the purpose in which they are grown. In the case of improving soil properties of impacted coal mine soils, winter cover crops are of the most significant use, as growing vegetation continuously incorporates organic matter into the soil and thereby, improving soil conditions during the winter period which usually has very little active plant development. Additionally, the value of a legume species grown as a winter crop cannot be emphasized quite enough as legumes naturally increase the nitrogen content of the soil (Bradshaw, 1997). Keeping this in mind, the choice of a legume species as a winter cover crop has, however, has been overlooked within the mining industries of South Africa.

As expected, green manuring increases the carbon content in soils of which a large part of this carbon is immediately lost as CO² during plant decay, leaving only a small quantity of carbon left in the form of stable SOM (Yadav *et al.*, 2000). Potter and Snyder (1916) found that an addition of one ton of dry oats into the soil resulted in 163.5 kilograms was lost in the form of CO². Yet, the loss of CO² in the soil through decay has been found to increase the solubility of phosphates and therefore, improve the nutrient content of the soil (Pieters, 2006). Nevertheless, a large part of carbon introduced into the soil through root decomposition has been found to contribute more effectively to a relatively stable carbon pool (Kätterer *et al.*, 2011). There is, however, no doubt that enrichment of soil with the green manure crops will increase the SOM content and help to compensate for anthropogenic greenhouse gas emissions. The challenge is, how to accurately quantifying the amount of carbon in soil incorporated with green manure (Lal, 2004; Harris and Megharaj, 2001).

6.3 Microbial biomass

Decomposition of organic biomass by microbial activities is an important component of healthy soil and determines the availability of nutrients for plants. Thus, the understanding of soil quality is interlinked with microbial decomposition as the microbial population within the soil determines, to an extent, the fertility of the soil. The rate of plant decomposition by microbes in the soil are dependent on a number of factors such as; soil pH, moisture content, temperature, presence of toxins, availability of nutrients and soil aeration (Piao *et al.*, 2001). Therefore, one can gain an idea of the fertility and quality of the soil by measuring the productivity of the microbial community (Edgerton *et al.*, 1995). Microorganisms are highly adaptable to adverse soil conditions such as poor nutrient content and adverse chemical concentrations commonly found in surface mined soil (Tripathi *et al.*, 2014). Conversely, poor water holding capacity and coarse textured soils create a completely inhabitable environment for not only soil microbial populations but also vegetation (Insam and Domsch, 1988; Ŝourková *et* al., 2005). Therefore, improving the chemical and physical soil properties will result in an improvement in microbial activity. Additionally, soil structure becomes extremely important in terms of not only the soil health but also the physical protection of soil organic matter by balancing microbial access to the soil organic material and the food web interactions which can diminish carbon pools (Elliott and Coleman, 1988; van Veen and Kuikman, 1990).

Furthermore, aboveground vegetation and soil-based carbon sequestration strategies play an important role in reducing CO_2 emissions (Shrestha and Lal, 2006) as a large proportion of carbon is lost in the form of CO_2 through microbial decomposition. In cases where soils are devoid of actively growing vegetation, no added input of organic matter results and existing microbial populations will continue to metabolise the remaining organic material into CO_2 until all material has been decomposed. This highlights the importance of introducing a cover crop on rehabilitated land in winter rather than leaving soil bare and thereby, preventing the total decomposition of plant nutrients which would result in more nutrients left over for summer pastures to establish. However, one needs to also be aware of the fact that the introduction of a cover crop into the soil as organic matter will stimulate denitrification by microbial activities under wet conditions (Mosier, *et al*., 2004) which highlights the fact that while green manuring can increase carbon sequestered in the soil it also stimulates the increase in nitrate fluxes into the atmosphere.

6.4 Root biomass

Roots have a large impact on the biogeochemical carbon cycle in terms of the amount of carbon contributed through root death and root exudation (Singh e*t al*., 1991). Root exudates are composed of soluble sugars, amino acids and organic acids which are readily decomposed by bacteria (Jones, 2008), while more recalcitrant organic materials require much longer periods of time to be decomposed (Boer *et al*., 2005). Furthermore, allowing vegetation to develop on bare soil plays an important role in terms of stabilising the soil surface and minimising soil erosion as the soil surface structure can be enhanced by root polysaccharides and labile mucilages produced by root development (Ruiz-Colmenero *et al*., 2013). Gyssels *et al.*, (2005) found that plant roots reduce soil erosion by penetrating the soil macrospores and thereby, reducing surface runoff and enhancing water infiltration. Roots have also been found to transport carbon into deeper soil horizons which is often controlled by: root turnover rates, root exudation, mycorrhizal colonization, soil characteristics and plant species (Tripathi *et al*., 2014).

Haynes and Beare (1997) found that plants with higher root densities resulted in a greater amount of soil aggregate stability, microbial biomass and soil organic carbon as the lysates, soluble secretions and dead fine roots help with the physical and chemical binding of microaggregates into larger macroaggregates. In another study by Tisdall and Oades (1982) the amount of soil organic carbon was found to be much higher in macroaggregates compared to microaggreagtes which was due to the presence of decomposing roots and hyphae holding macroaggregates together. Furthermore, carbon from root decomposition has also been identified as a more stable form of carbon than from shoot decomposition owing to larger amounts of chemical recalcitrance within the plant roots (Rasse *et al*., 2005).

7. Soil carbon protection

There are two main processes in which soil organic matter is protected; biochemical recalcitrance and physical protection (Christensen, 1996; von Luetzow *et al*., 2008). The protection process of biochemical recalcitrance occurs when aromatic polymers such as lignin and other structures which are difficult for microbes to break down form the chemical makeup of SOM (Christensen, 1996). Physical protection involves the physical binding of SOM in soil aggregates, limiting microbial access to the SOM and thereby, preventing decomposition (Christensen, 1996).

Soil aggregates are classified into different size classes according to their diameter with the microaggregates being the smallest, 53 µm in diameter and the macroaggregates being greater than 250 µm in diameter and often formed by smaller microaggregates which are bonded together by fine roots, hyphae and organic materials (Oades and Waters, 1991; Tisdall and Oades, 1982). The size of the soil aggregate is known to play an important role in terms of carbon retention in the soil (Six *et al*., 2004). Macroaggregates are the least stable while the microaggregates are the most stable (Tisdall and Oades, 1982). The amount and age of the carbon present differs in both the microaggregates and the macroaggregates with the lowest amount of carbon present in the microaggregates but on average having the oldest form of carbon and the highest amount of carbon in the macroaggregates with the youngest form of carbon (Nair, *et al*., 2010). Furthermore, plant residues which have a recognizable cell structure, are known as coarse intra-aggregate particulate organic matter (iPOM) and form the basis of aggregate formation (Kogel-Knabner *et al*., 2008). These aggregates physically protect the SOM by creating a physical boundary between the interactions within a food web, microorganisms and the rate at which microorganisms turn over SOM (Six *et al*., 2000). Hence, plants species with high root densities result in greater aggregate stability (Haynes and Beare 1997).

Another form of physical protection, which takes a longer period of time to develop, is the formation of organomineral complexes (Six *et al*., 2000). These complexes bind SOM through microbial activity and abiotic factors and remain in the soil for longer periods of time (Nair, *et al*., 2010). Through organomineral complexes, carbon can be stored within the soil for longer periods of time as it leads to larger amounts of recalcitrant microaggregates within macroaggregates and thereby, increasing the amount of carbon sequestered (Nair, *et al*., 2010).

8. Problem statement, aim, objectives and hypotheses

Soil impacted by surface coal mining has a whole host of negative impacts which affect the chemical and physical properties of the soil and therefore, the overall plant production potential. In this project soil carbon improvement strategies are investigated by evaluating the application of organic amendments to the soil impacted by surface coal mining. The associated effects on plant growth of various plant species and the amount of soil carbon stored as a result of the application of various organic soil amendments will be investigated in this project. The aim will be to synthesize information with regards to the assessment of soil carbon storage through effective reclamation practices and investigate the possible measures to reclaim soils for agricultural purposes by addressing pH, bulk density, plant yield and soil organic carbon. Therefore, the objectives of this study will be to compare the effects of each soil amendment on soil pH, soil bulk density, aboveground plant biomass, root biomass, soil fertility and soil organic carbon content and to test the hypotheses that:

- Improved plant yield will be associated with improved soil fertility
- Improved soil fertility will increase soil organic carbon storage

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CHAPTER 2

EFFECTS OF ORGANIC SOIL AMENDMENTS ON THE PROPERTIES OF SOIL IMPACTED BY SURFACE COAL MINING

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Abstract

In South Africa, open cast mines are required by law to undergo rehabilitation with the end goal of producing a sustainable land use after mine closure. While soil rehabilitative practice involves the removal and stockpiling of topsoil to conserve a healthy soil condition, this process, however, often leads to severe soil compaction and soil nutrient loss. Yet, allowing damaged mine soils to undergo natural processes in terms of restoration involves a period of 50-100 years before any acceptable vegetation cover is able to develop. Therefore, mine rehabilitation and reclamation requires additional soil amendments to decrease the time needed before acceptable vegetation can properly develop. In this investigation we evaluated the use of a different organic soil amendments and the use of a cover crop to improve the physical and chemical properties of soil impacted by surface coal mining. The investigation involved 40, 1 m long by 1 m wide by 1.2 m high experimental soil research bins "mini-lysimeters" which were constructed to represent a rehabilitated soil/substrate profile commonly created through rehabilitation practices on surface coal mines in South Africa. The bins were filled with mine spoil, subsoil and topsoil all of which came from a coal mine in Mpumalanga. The trial consisted of ten different topsoil treatments with four replicates per treatment including a control treatment consisting only of the cover soil. *Avena sativa* was planted as the winter cover crop and *Eragrostis tef* was planted as the summer test pasture. Results indicate that the combination treatment (T9) containing chopped lucerne, composted woodchips and cattle manure illustrated the greatest increase in aboveground and root biomass, and was the only treatment to show significant differences ($p<0.05$) in all parameters measured in this trial (pH, bulk density, aboveground biomass and root biomass) when compared to the control.

This emphasizes the different advantages one can expect by using a combination treatment which can improve different soil properties simultaneously to result in an overall positive impact on plant development.

1. Introduction

The mining of coal in South Africa is responsible for massive amounts of land degradation through the removal of large quantities of topsoil and subsoil in the process of open cast and sub-surface mining (Bradshaw, 1997). These mining operations are known to result in a wide variety of waste materials with many different characteristics which are related to the geology of the associated strata, the type of minerals being mined and the mining procedure (Bradshaw, 1997). Such processes have been shown to negatively impact the fertility of the soil (Ross *et al*., 1982; 1984), the plant production potential (Hart *et al*., 1986) and the physical properties of the soil (Cook *et al*., 1986). Furthermore, the removal and stockpiling of topsoil leads to severe soil compaction which further impacts the fertility of the soil and as a result, the overall plant development (Hart *et al*., 1986; Ross *et al*., 1982, 1984;). Resultant soil will undoubtedly be detrimental to the long-term ability of the land to sustain biomass production on an economic basis (Hart *et al.*, 1999) and will ultimately lead to the loss of biodiversity (Bradshaw, 1993). Therefore, many countries have come forth with legislation which serves to protect and conserve surface soils.

In South Africa open cast mines are required by law to undergo rehabilitation with the end goal of producing a sustainable land use (South Africa, Minerals and Petroleum Resources Act of 2002; National Environmental Management Act). However, before such legislation, mining companies were not bound to implement rehabilitation programmes and have thus resulted in a heritage of vast amounts of degraded land. Industries which have created this heritage have often discontinued and left no funds to restore the land which in most cases needed to be achieved as cheaply and effectively as possible (Bradshaw, 1997). Furthermore, allowing damaged mine soils to undergo natural processes in terms of restoration involves a period of 50-100 years before any acceptable vegetation cover is able to develop (Bradshaw, 1997). Such progressions are slow and therefore, require additional soil amendments in order to speed up the process of soil reclamation. Thus, the value of organic soil amendments have recently captured the interests of mining companies as these types of amendments physically incorporate additional organic matter into the soil which increases the overall soil organic matter content which in turn has positive soil remediation effects, such as: increased plant nutrients, soil stabilization through aggregate formation, increased porosity, increase water infiltration, increase water retention, decreased bulk density and increased microbial activity (Singh *et al*, 2004). However, one should always be reminded of the end land use and therefore, proper care should be taken in order to strike a balance between, providing what the affected community of the mined land wants, restoration of previous land use capability and minimal net loss of biodiversity, which can only be achieved with meticulous and well-timed mine closure planning.

One of the most prolific problems which occurs on coal mine rehabilitation sites in South Africa is the issue of topsoil compaction. During the process of mine rehabilitation, topsoil is typically stripped and either placed on profiled overburden or stockpiled. The compaction of the soil in most cases in South Africa is due to intense trafficking of machinery which the physically compacts topsoil during replacement and severely impacts the ability of vegetation to properly establish. This removal and stockpiling of topsoil leads to adverse effects on the fertility of the soil as a result of soil compaction and thus having a detrimental effect on root establishment which further promotes soil erosion making re-vegetation extremely difficult (Jha and Singh, 1992; Singh *et al*, 1996). Rehabilitation must, therefore, alleviate compaction issues by creating a depth and moisture content which is favourable for plant development as ecosystem productivity depends largely on the ability of the soil to support and sustain vegetation (Tripathi *et al*., 2014).

Re-vegetation is one of the most commonly used mine spoil stabilization techniques with phyto-remedation proving to aid in decreasing bulk density, minimizing erosion, minimizing pollution and improving the aesthetic quality of the environment (Wong, 2003). Re-vegetation physically incorporates organic matter into the soil and introduces microbial populations back into the soil which helps lower the bulk density and helps prevent compacted conditions (Bradshaw, 1997). Restoration projects in New Zealand have indicated that the most successful methods of land restoration focus on restoring soils in order to supply sufficient plant available nutrients and to insure soils are physically suited for root growth (Hart and Mine, 1986; Hart *et al*., 1990). Zobel (2005) states that root system dynamics form an integral part to the maintenance of biological and chemical equilibrium within the soil and are heavily involved in contributing to soil quality changes. Growing roots provide a gum-like material which cements soil particles into aggregates and thus, improving soil aggregation which itself controls the biological and hydrological properties of the soil (Melillo and Gosz, 1983; Tresder *et al*., 2005). Exudates, therefore, have an influence on hydrological properties, microbial activity and the subsequent metabolism of root detritus which can ultimately improve soil fertility. Additionally, Bradshaw (1997) makes mention of the fact there is no need to provide large amounts of nitrogen to degraded soils if there is a healthy level of nitrogen-fixing bacteria present, which can reintroduce productive amounts of nitrogen. This emphasizes the value of re-vegetating soils with nitrogen-fixing species. Additionally, green manuring with roots alone has the potential to improve the soil organic matter content and contribute to an improved nitrogen cycle and increases in microbial activity (Sainju *et al*., 2005).

Thus, the evaluation of the soil properties, plant growth characteristics, soil amendments and biomass production on soils impacted by surface coal mining is extremely important as it helps to assess the suitability of the soil to sustain vegetation and thereby, provide insight into the success of soil reclamation. In this study the application of different organic soil ameliorants on soil impacted by surface coal mining will be evaluated with regards to the effect in which these ameliorants have on, soil fertility, soil compaction and plant production compared to untreated control.

2. Material and methods

2.1 Experimental site

A field trial was conducted at the Hatfield Experimental Farm of the University of Pretoria, Pretoria, South Africa (25°45°S, 28°16°E) 1327 m above sea level in 2014 and 2015. The trial involved 40, 1 m long by 1 m wide by 1.2 m high experimental soil research bins "mini-lysimeters" (Figure 1).

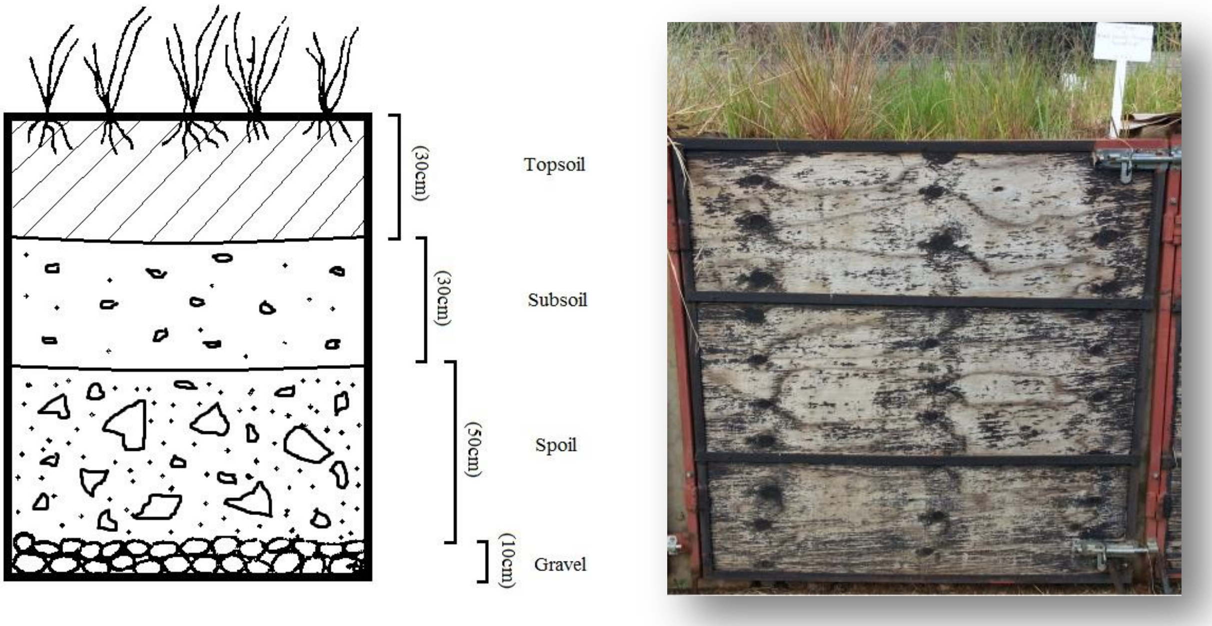

Figure 1 - Soil research bin "mini-lysimiter'' illustrating the different substrate levels.

The research bins were first filled with a 10 cm layer of gravel (to allow for free water drainage), followed by 50 cm layer of mine spoil followed by 30 cm layer of subsoil (Figure 2) and lastly 30 cm layer of mine top soil which was treated with different organic soil amendments (Table 1). The mine spoil, subsoil and top soil (Hutton, sandy loam) were all acquired from a surface coal mine in Mpumalanga, South Africa.

Figure 2 - Images of the mine spoil (left) and subsoil (right).

The treatments used in this trial have been specifically selected with regards to addressing possible concerns as to the, quantity available, access and proximity. The cut grass (*Digitaria eriantha*) was selected to be used as a soil treatment because this species is commonly grown on already rehabilitated mine sites in South Africa (Mentis, 1999) and can additionally be used as a soil ameliorant which physically introduces organic matter into the soil on land which still needs to be rehabilitated. The chopped lucerne (*Medicago sativa*) was selected because this species has a high inherent nitrogen content and therefore, when incorporated into the soil it can greatly improve the soil nitrogen status. Furthermore, because of the symbiotic relationship with nitrogen fixing bacteria, lucerne can be planted on already rehabilitated mine sites to improve the soil nitrogen status and additionally be used as a chopped hay lucerne soil ameliorant on other areas of the mine which still needs to undergo soil amelioration.

The black wattle (*Acacia mearnsii)* composted woodchips treatment was selected because black wattle is a highly invasive Australian species commonly found in large numbers on mine sites in South Africa. Therefore, eradicating this species while simultaneously creating a highly nutritious composted soil treatment from resources found on sight. Lastly the choice of composted cattle manure and fly ash were selected as these two treatments have proven very useful in their ability to ameliorate drastically disturbed soils (Shrestha and Lal, 2006) and can easily be sourced in large quantities from surrounding cattle farms and fly ash from nearby coal power stations.

Application rates of soil ameliorants for rehabilitation of mined areas differ according to the soil analysis, purpose of rehabilitation and the soil ameliorant used. In this trial an application rate of 40 tons per hectare for the all treatments except those containing fly ash were chosen. The application rates for the fly ash and manure treatment and the fly ash and woodchips treatment were based on alleviating the pH and the physical properties of the topsoil used in this trial. Previous research conducted by Fail (1987) reported fly ash application rates of 70 tons per hectare for stripped mine soil while Twardowska (1990) reported fly ash application rates of 366 tons per hectare for the amelioration of coal mine spoil. Therefore, in this trial it was decided the an application rate of 132 tons per hectare (66 tons of fly ash and 66 tons of manure or woodchips) for the fly ash and manure treatment (T1) and the fly ash and woodchips treatment (T3) would be used.

The design of the soil research bins were to simulate and represent a rehabilitated soil/substrate profile which is commonly created through rehabilitation practices on a surface coal mines in South Africa. Prior to the treatment application, the topsoil, subsoil and the different soil treatment were analysed for pH and total mineral content.

Table 1: List of the soil treatments applied (n = 4). Where combinations of treatments exist, the total application rate is equally divided among each treatment.

Soil Treatments	Treatment numbers	Origin of Treatment
Fly ash (class F) and composted cattle manure[b]	T1	A + B
Composted cattle manure[a]	T2	B
Fly ash and black wattle composted woodchips *(Acacia mearnsii)*[b]	T3	A + B
Black wattle composted woodchips *(Acacia mearnsii)*[a]	T4	C
Cut grass (*Digitaria eriantha*)[a]	T5	D
Cut grass (*Digitaria eriantha*) and composted cattle manure[a]	T6	D + B
Cut grass (*Digitaria eriantha*) and black wattle composted woodchips *(Acacia mearnsii)*[a]	T7	D + B
Lucerne hay (*Medicago sativa)*[a]	T8	D
Lucerne hay (*Medicago sativa)* and composted cattle manure and black wattle composted woodchips *(Acacia mearnsii)*[a]	T9	D + B + B
Top soil only (control)	T10	E

[a] Applied at 40 tons per hectare

[b] Applied at 132 tons per hectare

A = Fly ash from Ash Resources (Pty) limited, South Africa
B = Conradie Organics, Pretoria, South Africa
C = Jacklin Organics, Pretoria, South Africa
D = Hatfield experimental farm, Pretoria, South Africa
E = Surface coal mine in Mpumalanga, South Africa

2.2 Experimental layout and treatment application

The treatment applications for each research bin were arranged in a complete randomized block design and consisted of nine different top soil treatments applied at either 40 or 132 tons per hectare (Table 1) with four replicates per treatment including a control treatment containing no soil ameliorant. The topsoil was placed into a concrete mixer together with the respective soil treatment to insure that the treatment was thoroughly blended with the top soil. Once the top soil was thoroughly mixed it was offloaded into the research bins to make a 30 cm amended top soil layer. After the amended soil had been placed in the research bins it was left to settle for four weeks. Once the four week period had passed the soil had subsided due to heavy rainfall during March of 2014 (Table 2) and therefore, an additional application of amended soil was applied to fill the research bins to the 30 cm surface mark once more. *Avena sativa* L. (Overberg cultivar) was selected as the winter cover crop test species. This particular species was chosen because it is one of the strongest winter growing annuals in South Africa which can tolerate low fertility soils. Furthermore, this species was used strictly as a cover crop to improve the physical and chemical properties of the soil such as: increasing soil organic matter content, increasing microbial activity, improving soil nitrogen status through nitrogen fixation, increasing soil pH, improving soil structure, reducing erosion and increasing nutrient supply within the soil (Harris and Megharaj, 2001; Pieters, 2006). Additionally, the *A. sativa* winter cover crop was used to evaluate the treatments applied to impacted surface mined soil and their effects on plant development.

Avena sativa seeds were planted in rows at an application rate of 40 kg per hectare on 1 April 2014 as the winter test pasture. These plants were then watered until seedlings emerged after which they were left without irrigation and, were therefore, rain fed for the remaining days. After the 90 day growth period, aboveground plant material was harvested by sampling from a 0.09 m^2 area from each of the 40 research bins at a height of 30 mm above the soil surface and maintaining a 15 cm buffer strip from the boarders of the research bins. The aboveground material was placed in paper bags and put into an oven at 60 ºC for 48 hours (Goering and Van Soest, 1970) after which the samples were weighed. After harvest, the soil research bins were equally irrigated and left for three days before soil penetrometer readings were taken with the Geotron Hand Penetrometer Model P5 to determine the effect that each soil treatment had on soil compaction. Once compaction readings were taken, an auger was used to take soil samples at a depth of 25 cm from each research bin. The pH (KCL) was then determined and the macro nutrients of all soil samples were then analysed by ammonium acetate extraction.

Eragrostis tef (Zucc.), (SA Brown cultivar) was selected as the summer pasture because it is one of the strongest growing summer annuals and is an excellent nurse crop. This particular species has a very hardy nature (C4) and is commonly used on coal mine rehabilitation sites in South Africa (Mentis, 1999). On the 16 of October 2014, *Eragrostis tef* seeds were broadcasted at an application rate of 10 kg per hectare, irrigated until seedlings emerged after which the plants were rain fed until final harvest on the 9 of April 2015. The aboveground biomass and root samples where then harvested by sampling from a 0.09 m^2 area after which the root samples were then rinsed with tap water to insure all debris was removed before drying. Both the aboveground biomass and the root biomass were dried using the same method mentioned above, after which the samples were weighed and recorded. Penetrometer readings were taken following the same method mentioned above, after which soil samples were collected and analysed to once again determine the effect that each treatment had on soil fertility.

Figure 3 - Different soil treatments applied.

Table 2: Chemical composition of ameliorants and soil substrates.

Treatments	**P%**	**K%**	**Na%**	**Ca%**	**Mg%**	**N%**	**pH**
Fly ash	0.08	0.02	0.12	2.68	0.47	0.02	8.99
Composted cattle manure	0.83	2.74	0.41	1.80	0.81	2.50	9.06
Black wattle composted woodchips	0.32	1.05	0.16	0.92	0.36	1.71	5.60
Cut grass	0.13	1.27	0.08	0.24	0.09	0.94	5.50
Lucerne	0.18	0.69	0.03	0.81	0.26	3.18	5.61
Substrates			mg kg^{-1}			pH	
Topsoil	21	59	13	343	78	5.18	
Subsoil	1	76	14	295	283	4.28	

Figure 4 - Experimental trial at the Hatfield Experimental Farm, Pretoria, South Africa (May 2014).

2.3 Weather

All weather data was collected on Hatfield experimental farm using an automatic weather station which consisted of: one LI 200X pyranometer (LiCor, Lincoln, Nebraska, USA) measuring solar radiation, an electronic cup anemometer (MET ONE, Inc. USA) measuring wind speed, an electronic rain gauge (RIMCO, R/TBR and tipping bucket rain gauge, Rauchfuss Instruments Division, Australia) measuring rainfall, a temperature and relative humidity sensor, and a CR 10X data logger (Campbell Scientific Inc., USA) which recorded the data at 10 second intervals.

Table 3: The mean maximum and minimum temperatures in addition to the total rainfall recorded per month for years 2014 and 2015.

	Maximum Temperature (°C)	Minimum Temperature (°C)	Average Rainfall (mm)
		2014	
March	25.68	13.65	253.30
April	24.43	13.81	11.70
May	24.47	7.21	0.50
June	21.57	3.46	0.90
July	20.55	3.51	0.00
August	23.32	7.35	13.70
September	28.95	11.93	0.70
October	29.05	12.90	30.70
November	26.95	14.29	103.70
December	28.39	16.25	265.40
		2015	
January	30.17	16.86	85.20
February	31.28	16.63	22.10
March	29.41	15.62	57.80
April	26.42	11.84	29.70

3. Statistical analysis

Ten different soil treatments including a control treatment were replicated four times. Treatments were allocated to research bins in a complete randomised block design while all statistical analysis was conducted using IBM SPSS statistical software version 22© (IBM Corp released, 2013). Normal distribution of the mean values were confirmed by examining both the Normal Q-Q plot and the Detrended Normal Q-Q plot along with the Shapiro-Wilk test of normality (P>0.05). The homogeneity of variance was tested by Levene’s test for equality of variances (P>0.05). Significant differences were tested on the mean values using the One-Way Analysis of Variance (ANOVA) with Fisher’s Least Significant Difference (LSD) post hoc test at P<0.05 conducted on the soil bulk density and Tukey’s highest significant difference post-hoc test conducted on the pH and dry matter yield at P<0.05. Lastly, the non-parametric Kruskal-Wallis test was conducted to establish significant differences in soil fertility results as the data was the only resultant data from this trial which was not normally distributed (Shapiro-Wilk) together with a violation of the homogeneity of variance (Levene’s test).

4. Results and discussion

Soil impacted by surface mining is often considered to have a low organic matter content, low microbial activity, low pH, low nutrient status, low water holding capacity and a high bulk density (Boerner *et al*., 1998; Hearing *et al*., 2000; Indorante *et al*., 1981; Palumbo *et al*., 2004; Seybold *et al*., 2004; Sinclair *et al*., 2004). Restoration of such soil therefore, requires additional soil amendments to decrease the time needed before acceptable vegetation can develop. All data collected in this study was used to illustrate to what extent the different soil treatments affected the soil properties and the associated plant biomass. The aboveground dry biomass and the root dry biomass were extrapolated from the 0.09 m^2 area to 1 m^2. Data was constantly reported in comparison to the control and the results indicate that the application of these different organic soil ameliorants have a substantial effect on the physical properties of the soil, the fertility of soil and the associated plant development.

4.1 Soil pH

Generally, low pH of soil impacted by surface coal mining has proven to create a very adverse soil environment for microbial and plant growth (Tripathi *et al*., 2014). This toxicity often arises due to oxidation of sulphide minerals from the underlying coal discard which impacts cover soils through capillary action of the resultant acidic water (Truter, 2007). Hence, soil amelioration is desperately needed in order to raise the pH to create an environment in which plants can utilize soluble nutrients within the soil. Luthe and Peterson (1997) suggest that the most favourable soil pH for *A. sativa* development is between 5.5 and 7 (KCL), while *E.tef* is known to tolerate a wide range of soil acidities.

The statistical analysis on the resultant pH of the soil after the *A. sativa* harvest shows that the soil pH was significantly ($p<0.05$) influenced by treatment applications. From Figure 5 it is evident that the only soil treatments which had a significant difference ($p<0.05$) in the mean pH compared to the control after the *A. sativa* harvest were from treatments T1 and T6. Treatment T1 and T6 showed an increase of 17.86% and 13.69% respectively when compared to the control but showed no significant difference when compared to each other. From Table 2 it is evident that the fly ash treatment is alkaline in nature with a pH of 8.99 and thus was the reason for the increase the soil pH.

The cut grass treatment (T5) measured, effectively the same pH as the control which may be due to the low pH content of the treatment itself (Table 2). The composted cattle manure treatment (T2) is a common soil ameliorant used for rehabilitation of degraded soils as it is alkaline in nature (Table 2) and is a great source of organic matter with an abundant supply of essential plant nutrients (Cooperband, 2002). Applying this source of organic matter to the soil, moderates changes in the soil pH and as a result, creates a suitable soil environment for plant growth (Cooperband, 2002). However, it is important to note that the manure is less likely to be retained within the soil over a long period compared to a treatment such as woodchips, and the manure will therefore, only have an influence on soil pH over a shorter timeframe (Palumbo *et al.*, 2004). The woodchips treatment (T4) showed a mean increase of 5.75% compared to the control. However, this treatment is a recalcitrant and lignin-rich treatment which is known to degrade over a much longer time period (Palumbo *et al.*, 2004) compared to the other soil ameliorants used in this trial. It therefore, will only start to have a greater impact on the soil pH once the treatment has decomposed to a greater extent. The lucerne treatment (T8) showed an increase in pH of 5.21% compared to the control which may be due to the fact that lucerne treatment itself has a moderate pH, moderate calcium content and a high nitrogen content (Table 2) which together facilitates greater plant growth, ultimately resulting in an improvement on the soil alkalinity. Comparing the fly ash and manure treatment (T1) with the manure treatment (T2) and comparing the fly ash and woodchips treatment (T3) with the woodchips treatment (T4) after the *A. sativa* harvest revealed no significant difference in mean pH (p<0.05). This suggests that while the fly ash and manure treatment (T1) and fly ash and woodchips treatment (T3) were applied at 132 t ha^{-1} and the manure treatment (T2) and the woodchips treatment (T4) were applied at 40 t ha^{-1}, this difference in application rate, revealed no significant difference (p<0.05) on the mean pH of the soil.

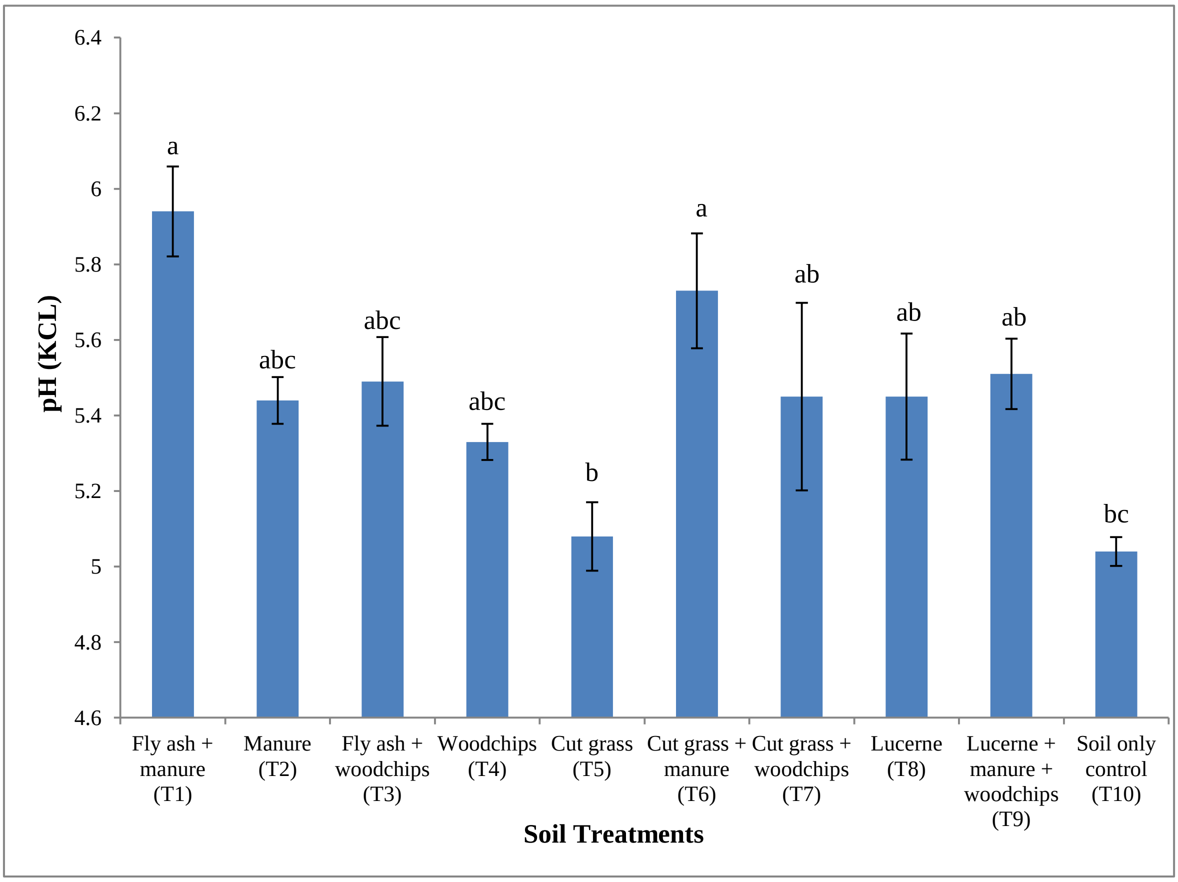

Figure 5 - Mean soil pH measured after *A. sativa* harvest as a result of the different soil treatments applied. Bars with the same letter have no significant difference in the mean pH at (*p*<0.05). Vertical lines represent the standard error of the mean.

The pH of the soil after the *E. tef* harvest was also significantly (p<0.05) influenced by the treatment applications. However, treatments T5, T6, T7, and T8 showed no significant difference (p<0.05) in mean pH compared to the control after the *E. tef* harvest. The cut grass treatment (T5), once again, measured effectively the same pH as the control as a result of the treatment itself being acidic and containing very low amounts of plant nutrients (Table 2) which play an important role in improving plant growth and thus, soil pH. The woodchips component within treatment T7 proved to increase the pH but not to the same extent as the manure did within treatment T6.

Comparing the fly ash and manure treatment (T1) with the manure treatment (T2) and comparing the fly ash and woodchips treatment (T3) with the woodchips treatment (T4) after the *E. tef* harvest revealed no significant difference amongst each other in mean pH (*p*<0.05). This again suggests that the larger application rate for treatments T1 and T3 had no significant difference (*p*<0.05) in mean pH of the soil compared to treatments T2 and T4. However, fly ash is known to play an important role in improving the soil pH and the physical properties of soil as describe in a pot trial conducted by Truter (2007), which can be seen for treatment T1 and T3 in this trial as compared to other treatments that have no fly ash component. However, the greatest increase in mean pH measured after the *E. tef* harvest compared to the control was measured from soil incorporated with treatments T1, T2, T3, T4 and T9 which showed increases in mean pH of 12.31%, 13.68%, 16.63%, 9.00% and 9.25% respectively. Furthermore, these treatment all showed a significant difference ($p<0.05$) in mean pH compared to the control. Yet, it is evident that a decrease in mean pH was measured after the *E. tef* harvest compared to the mean pH measured after the *A. sativa* harvest in all amended soils except in treatments T2, T4, T7 and T9 which interestingly all contained composted woodchips component. This may due to the neutralizing effect in which the woodchips had on the soil pH as the woodchips at this stage of the trial had decomposed to a greater extent than compared to the extent of decomposition after the *A.sativa* harvest and thereby, having a greater influence on soil pH. Nonetheless, as a collective, the mean pH of all treatments measured after the *A. sativa* harvest was recorded at 5.44 while the mean pH of all the treatments measured after the *E. tef* harvest was recorded at 5.29, therefore illustrating a general decrease in pH measured after the second harvest (*E. tef*). This may be due the high rainfall during the summer season (Table 3) resulting in carbonic acid, causing a slight acidic effect on the soil. Furthermore, Boone (1994), Haider *et al*. (1993), Norby and Cortrufo (1998) and Sanchez *et al*., (2002) state that plant roots have a greater influence on the amount of soil organic matter contribution compared to the aboveground plant biomass. Therefore, the organic matter contribution from the roots of the *A. sativa* species may also have been the reason why we see this change in pH as this species produced a larger root biomass compared to *E. tef* species, (Figure 11 and 12) thus resulting in a larger soil organic matter contribution which in turn improved the buffering capacity of the soil and limited large changes in soil acidification.

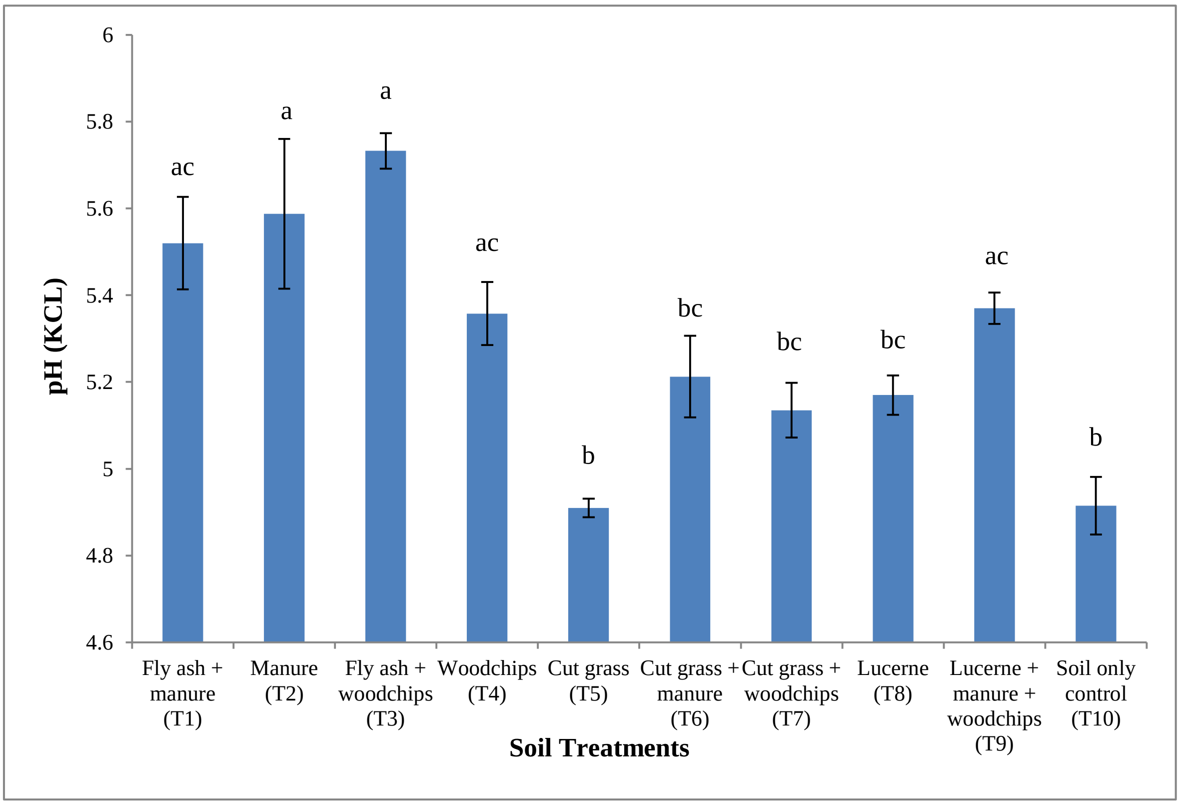

Figure 6 - Mean soil pH measured after *E. tef* harvest as a result of the different soil treatments applied. Bars with the same letter have no significant difference in the mean pH at (p<0.05). Vertical lines represent the standard error of the mean.

4.2 Penetrometer resistance and bulk density

The penetrometer measures pressure in kilopascals exerted from the soil on the cone base of the penetrometer which gets vertically driven into the soil. As the cone enters the soil, measurements of soil resistance are recorded at one centimetre intervals until maximum pressure is reached. With this particular piece of equipment a pressure can be recorded between a range of 0 - 5400 kPa before the maximum capacity of the penetrometer is reached. Using the pressure recorded from the penetrometer per centimetre, bulk density can be calculated by using the equation formulated by Hernanz *et al.*,(2000) granted that the moisture within the soil exceeds 8 (%w/w) and the cone based used is bigger than 98mm².

$$Bulk\ Density = 0.013PR^{0.096}d^{-0.061} \qquad \text{[Equation 1]}$$

Where PR is the penetration resistance (kPa) and d is the depth (cm).

Initial soil stockpiling, topsoil replacement and the process in which topsoil is transported and handled by heavy machinery on coal mines leads to soil compaction and an unfavourable soil bulk density for plants to properly develop. While these heavily compacted soils are known to negatively impact root development and microbial proliferation (Haigh, 2000), the addition of soil organic matter from soil ameliorants is known to decrease the bulk density of the soil and create a more favourable soil environment for plants to develop (Stock *et al*., 2007). From Figure 7 it is evident that the only soil treatment having a significant difference (p<0.05) on the mean bulk density compared to the control was from the manure treatment (T2) which illustrated a mean decrease of 3.51% in comparison to the control. Treatment T8 showed no significant difference (p<0.05) in mean bulk density compared to any other treatment but measured the second greatest decrease in mean bulk density with mean decrease 2.57% compared to the control.

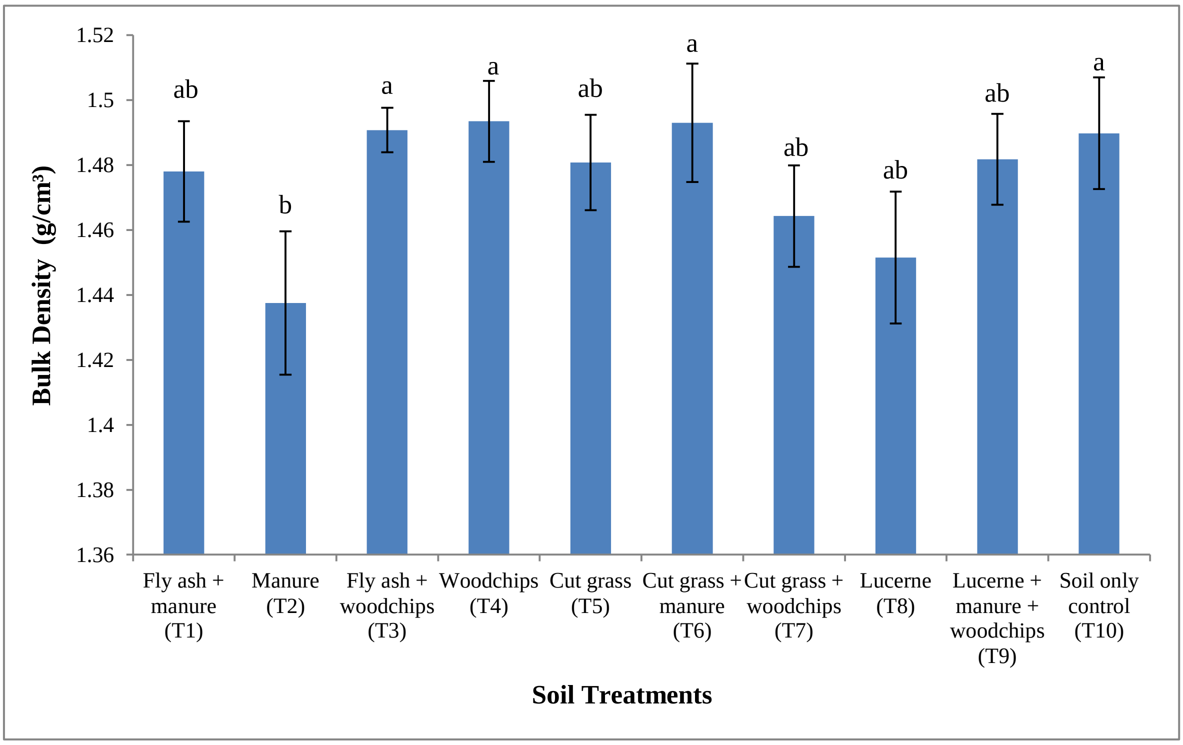

Figure 7 - Mean bulk density calculated to a depth of 25cm after *A. sativa* harvest as a result of the different soil treatments applied. Bars with the same letter have no significant difference in the mean bulk density at (*p*<0.05). Vertical lines represent the standard error of the mean.

From Figure 8 it is evident that all the treatments had a significant difference (p<0.05) on the mean bulk density compared to the control except treatment T6. Treatment T8 had the greatest significant difference (p<0.05) in mean bulk density with a mean decrease of

5.87% compared to the control. This may be due to the fact that the lucerne treatment was in the form of a chopped hay soil ameliorant (Figure 3) which helped to aerate the soil by physically pushing the soil particles away from one another. Treatment T8 also substantially increase the root production (Figure 8 and 9) compared to the rest of the soil ameliorants which in turn, increase the organic matter within the soil from the process of root decay and root exudation, thereby decreasing the bulk density (Bradshaw,1997). When considering the cut grass treatment (T5), which was also a chopped hay product, a decrease in mean bulk density of 3.74% was calculated in comparison to the control after the *E. tef* harvest. A possible reason why the cut grass treatment (T5) may not have shown a decrease in bulk density quite to the same extent to which the lucerne treatment did, even though the T5 is also in the form of chopped hay soil ameliorant, may be because the treatment T8 had the additional benefit of supplying more nutrients (Table 4 and 5) for plant growth and, therefore, resulting in more root development (Figure 11 and 12) and subsequent root decomposition, both of which play an important role in decreasing bulk density. In a study conducted by Haynes and Beare (1997) it was found that plants with higher root densities resulted in a greater amount of soil aggregate stability, microbial biomass and soil organic carbon, which illustrates the importance of sustaining a healthy root system within soil to provide support for the formation of a good soil structure. Additionally, roots secrete exudates which contain lysates and soluble secretions which can help with the physical and chemical binding of micro aggregates into larger macro aggregates again, highlighting the importance in which developing vegetation has on improving soil physical properties (Nair *et al*., 2010). Furthermore, Osmont *et al*., (2007) describes how soil compaction can be significantly decreased through root growth of grass species as their roots are able to penetrate into tiny pore spaces and radially expand, thereby increasing the pore space and ultimately decreasing bulk density. This may also be the reason for the decrease in bulk density calculated as a result of the application of treatment T9 which resulted in a large root biomass.

Treatments T1, T2, T3, T4 all had a significant difference ($p<0.05$) in mean bulk density compared to the control but showed no significant difference ($p<0.05$) compared to one another. This suggest that the application of 132 t ha^{-1} for treatment T1 and T3 had no significant difference ($p<0.05$) on bulk density compared to an application of 40 t ha^{-1} for treatments T2 and T4. The overall mean bulk density of each treatment calculated after the *E. tef* harvest showed a mean decrease of 3.50% compared to the overall mean bulk density of each of the treatments calculated after the *A. sativa* harvest, illustrating overall compaction alleviation over time.

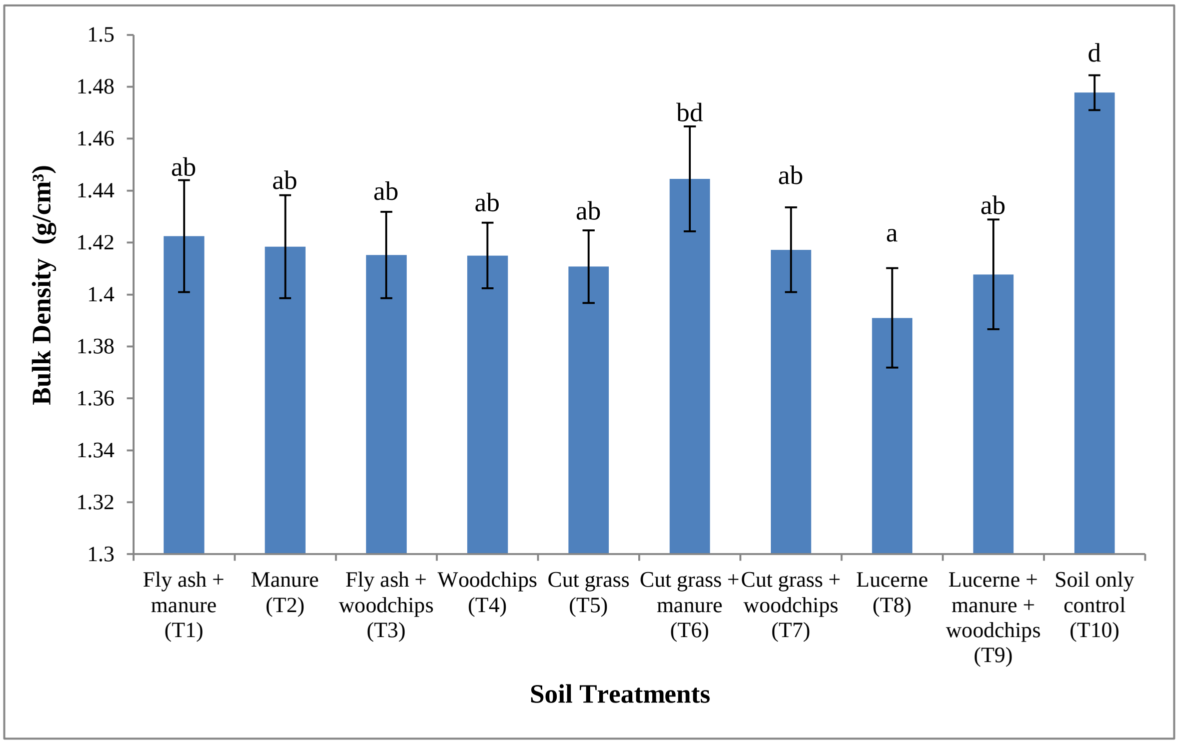

Figure 8 - Mean bulk density calculated to a depth of 25cm after *E. tef* harvest as a result of the different soil treatments applied. Bars with the same letter have no significant difference in the mean bulk density at (p<0.05).Vertical lines represent the standard error of the mean.

4.3 Dry matter yield

4.3.1 Aboveground dry matter yield of *A. sativa* and *E. tef*

The application of soil ameliorants have three basic functions on improving soil conditions for plant growth, a physical soil function, a chemical soil function and a microbial soil function. The physical function focuses on addressing the structural properties of the soil such as the bulk density, porosity, water holding potential and the texture. The chemical function involves improving the nutrients available to the plant as well as locking up toxins and improving the soil pH. Lastly, the microbial function comprises the ability in which the ameliorant creates a soil environment which supports microbial growth and indirectly, plant available nutrients. All these functions determine to what extent vegetation will develop on soil which has been ameliorated. Therefore, one can relate the plant development to the success in which the soil ameliorant addresses soil deficiencies within those three areas.

From Figure 9 it is evident that the only treatment which had a significant difference ($p<0.05$) on mean dry matter yield compared to the control was treatments T9 ($p<0.05$), with a mean increases in yield of 128.08% when compared to the control. When looking at the analysis of the treatments (Table 2) one can see that some treatments are plentiful in the amount of certain marco nutrients but scarce in others. By examining the analysis of the lucerne, manure and woodchips treatment (T9), one can see how each individual component within the combination treatment, complemented one another. The lucerne and manure components provided the majority of the nitrogen, while the woodchips and manure components provided the majority of the phosphorous, potassium and calcium. From Table 4 it is evident that the lucerne, manure and woodchips treatment (T9) provided vast improvements in the amount of phosphorus, potassium, sodium and magnesium within the soil after the *A. sativa* harvest compared treatments T3, T4, T5, T7, T8 and T10.

Furthermore, it is important to note, that while manure is known to rapidly improve the soil nutrient status over the first few years with readily available nutrients as it is decomposed quickly, it however will not provide a sustained amount of nutrients for plants and microbes (Cooperband, 2002). Composted woodchips on the other hand is a more recalcitrant treatment and will decompose at a much slower rate owing to the higher lignin content and, therefore, releasing nutrients over a longer time period (Palumbo *et al*., 2004). This places emphasis on the use of a soil treatment which harbours different components which complement each other in respect to addressing inherent deficiencies that may exist in each component's function. One must not forget that treatment T9 was also applied at 40 $t.ha^{-1}$ with each individual component applied at 13.3 $t\ ha^{-1}$. Therefore, illustrating the value of diversifying a soil ameliorant without increasing the application rate of each individual component. Furthermore, evidence from Davis and Whiting (2012), suggests that woody soil ameliorants tie up nitrogen, which is later released once the wood has decomposed and therefore, illustrates that the use of a soil ameliorant which is comprised of both a quick and slow decomposing materials, having both fast and long term effects on soil quality.

The analysis of the fly ash treatment showed poor levels of important plant macro nutrients (phosphorous, potassium and nitrogen) but much higher levels of calcium which highlights the fact that this treatment may have a larger impact on the physical structure of the soil than compared to improving the nutritional quality of the soil for plant development. Furthermore, it enhances the flocculation between soil particles (Appendix A1) and thereby stabilizing soil structure (Palumbo *et al*., 2004) and improving soil water holding capacity and plant available water (Park *et al*., 2014). Additionally, because of the alkaline nature of the fly ash, studies show that fly ash can improve the pH of acidic mine soils (Martens and Frankenberger, 1992) and in this trial, all treatment which contained fly ash showed a substantial increases in the mean soil pH compared to the control.

A possible reason as to why the fly ash and manure treatment (T1) showed greater aboveground biomass (Figure 9 and 10) as compared to most of the other treatments may be due to the fact that this treatment contributed an abundant amount of calcium from the fly ash which helped to physically ameliorate the soil structure and chemically improve the soil pH (Figure 5), while at the same time addressing nutritional deficiencies with the application of the manure which contributed abundant levels of nitrogen, potassium and phosphorous (Table 2 and 3). According to Bradshaw (1997), in order for vegetation to properly develop on soil impacted by mining, soil must have a healthy amount of nitrogen available to plants. This is supported in this trial as the highest nitrogen containing treatment, the lucerne treatment (T8), resulted in substantial amount of aboveground biomass for both the *A. sativa* and the *E. tef* pastures (Figure 9 and 10).

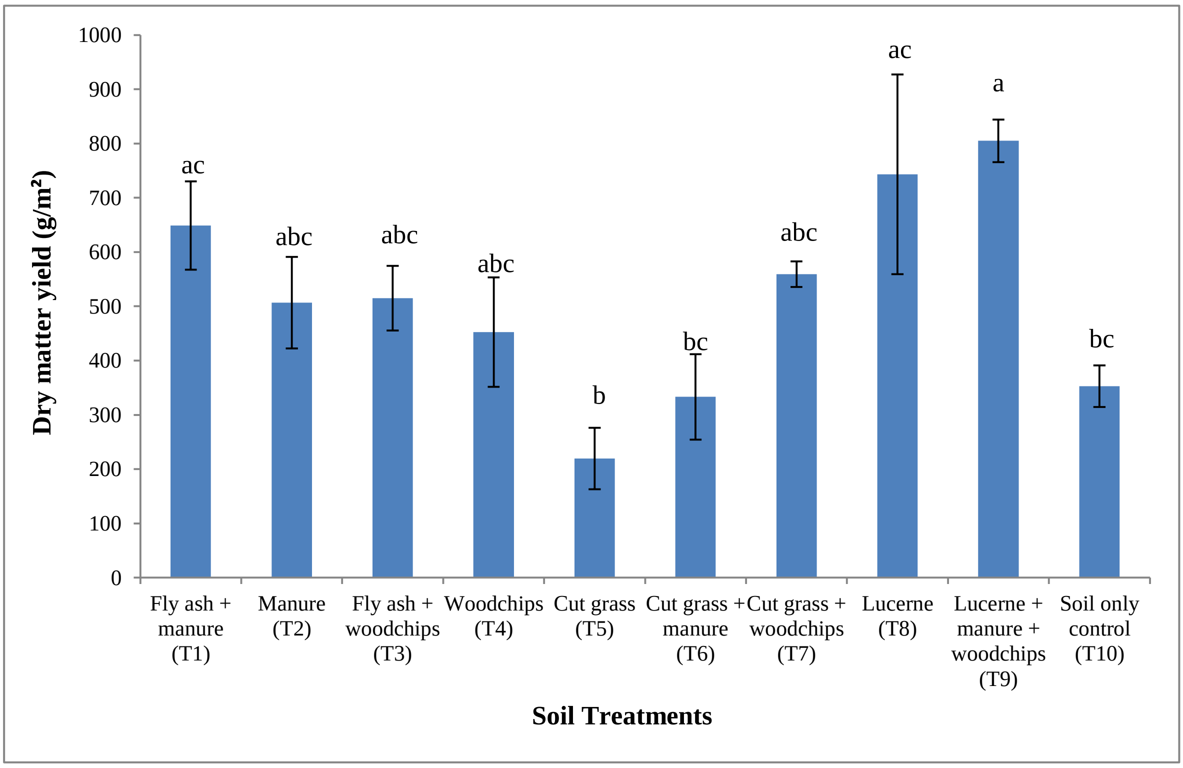

Figure 9 - Mean *A. sativa* aboveground dry matter yield from 1 m² as a result of the different soil treatments applied. Bars with the same letter have no significant difference in the mean dry matter yield at (*p*<0.05). Vertical lines represent the standard error of the mean.

The *E. tef* aboveground biomass showed no significant difference ($p<0.05$) as a result of the different treatment applications. However, the *E. tef* species proved to be the more pronounced species in terms of total aboveground yield when compared to the *A. sativa* species. From Figure 10 it is evident that the lucerne, manure and woodchips treatment (T9), the woodchips treatment (T4) and the fly ash and manure treatment (T1) had the greatest increase in mean aboveground *E. tef* biomass with increases of 44.18%, 30.08% and 20.13% respectively when compared to the control. Treatment T9 was the best soil treatment in terms of resultant aboveground biomass which can again be attributed to the contribution of nutrients made by each individual component within the soil treatment. Furthermore, the lower bulk density calculated after the *E. tef* harvest (Figure 8) as a result of the lucerne, manure and woodchips treatment (T9) application brings forth the fact that this treatment further supports vegetation development by decreasing the bulk density and therefore, having a positive effect on the water infiltration and water holding capacity (Martens and Frankenberger, 1992; Chaulya *et al*., 2000a, 2000b) and as a result, creating a soil environment which further improves plant development.

Treatment T4 showed the second greatest increase in mean aboveground biomass with an increase of 30.08% in comparison to the control. From Table 4 and 5, it is evident that all treatments which contained woodchips T3, T4, T7, and T9 showed substantial increase in the amount of potassium measured within the soil after the *E. tef* harvest. This increase in potassium is possibly caused by the greater extent of the woodchip decomposition which resulted in the release of more potassium as a nutrient. As stated earlier, soil ameliorants which contain wood products have a higher lignin content which prevents rapid decomposition and promotes a delayed nutrient release (Palumbo *et al*., 2004). Potassium is an essential macro nutrient for plant development as it plays a crucial role in photosynthesis, starch formation, protein synthesis and the translocation of sugars (Brady and Weil, 2008) and therefore, contributes to improved plant development.

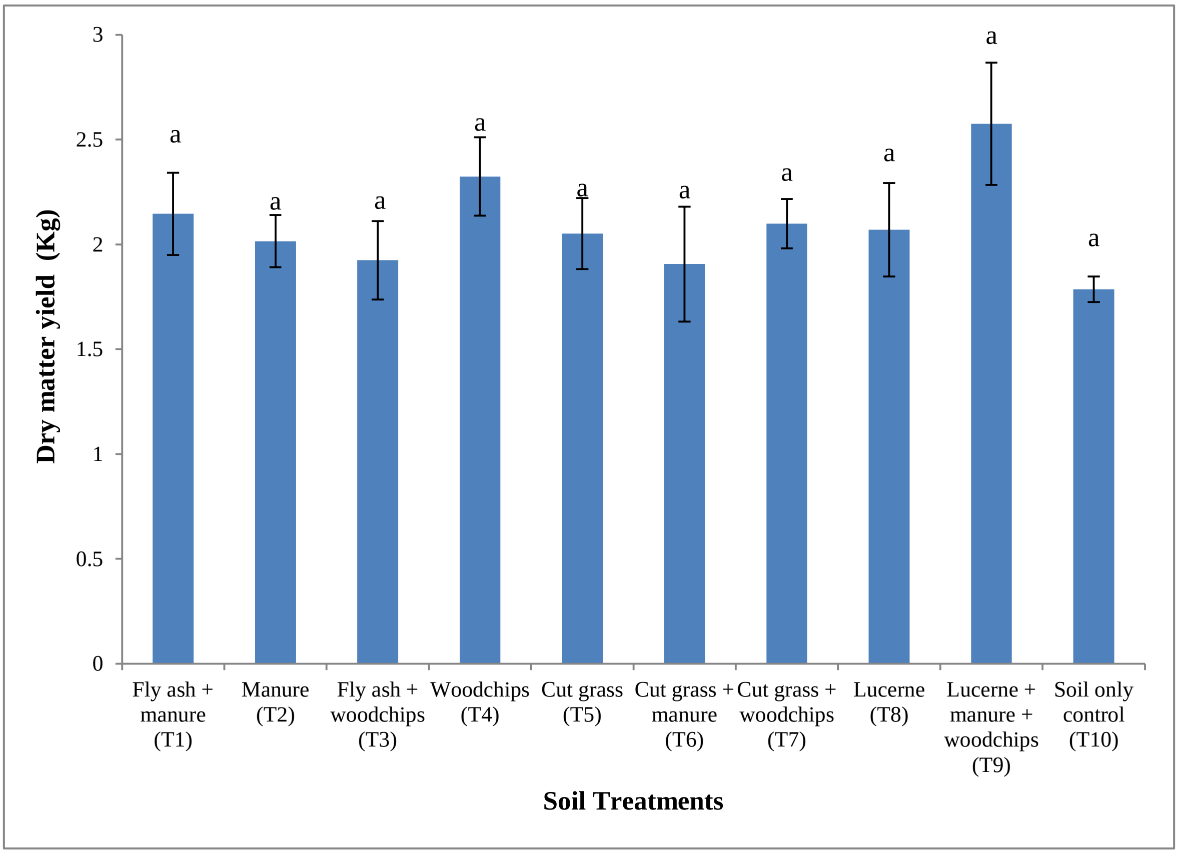

Figure 10 - Mean *E. tef* aboveground dry matter yield from 1 m^2 as a result of the different soil treatments applied. Bars with the same letter have no significant difference in the mean dry matter yield at (*p*<0.05).Vertical lines represent the standard error of the mean.

4.3.2 Root dry matter yield of *A. sativa* and *E. tef.*

The influence in which the soil environment impacts root development ultimately determines overall plant growth as the plant roots supply the entire plant with nutrients and water for plant growth. The soil physical and chemical properties influence root growth and therefore, an indirectly assessment of soil reclamation can be done by evaluating resultant root yield.

The mean root dry matter yield for *A.sativa* was calculated using the mean shoot to root response (3.89) measured after a 90 day emergence pot trial was conducted on *A.sativa* by Amanullah *et al.*, (2015). The *A. sativa* root biomass showed a significant difference (p<0.05) as a result of the different soil treatments applied. From Figure 11 and 12 it is evident that the root yield follows the same trend as the aboveground yield as the soil within the research bins which supported greater aboveground biomass also supported greater root biomass. This could be attributed to the fact that a larger crop canopy photosynthesizes more sunlight and therefore, provides more nutrients to root development granted that the soil conditions do not limit root growth. The treatment which showed the greatest significant difference ($p<0.05$) in mean *A. sativa* root biomass was treatment T9 with and a mean increase of 128.03% compared to the control. Treatment T9 helped to create soil environment conducive to root development by improving the physical and chemical properties of the soil. A substantial decrease in bulk density was measured as a result of the lucerne, manure and woodchips treatment (T9) which has been shown to help to increase the pore space and the increase in water content range within which root development is less likely to be constrained and thereby, allowing for greater root proliferation (da Silva and Kay, 1997). Additionally, the soil analysis after the harvest of both pasture species (Table 4 and 5) indicated that the application of treatment T9 helped increase the amount of nutrients within the soil which in turn resulted in greater aboveground and belowground plant growth. This however, was not the case when considering resultant root yield from the application of treatment T5 which showed very poor levels of macro nutrients within the soil and thus, poor root yield. Furthermore from Table 2 it is evident that the cut grass treatment (T5) has a high carbon content but a low nitrogen content which according to Hoorman and Islam (2010), will result in a lock up of soil nitrogen form microbial action and thereby, deplete the plant available nitrogen status within the soil.

Additionally, drastic changes in soil pH can detrimentally affect the microbial and root symbiotic relationships that exist within the soil and thus, negatively impacting the nutrient absorption within the soil (Shrestha and Lal, 2006). While the pH measured as a result of the different applications of organic treatments are not within extreme levels of acidity, the treatments which increased the soil pH may have enhanced root symbiotic relationships and thereby, improved nutrient absorption and root development.

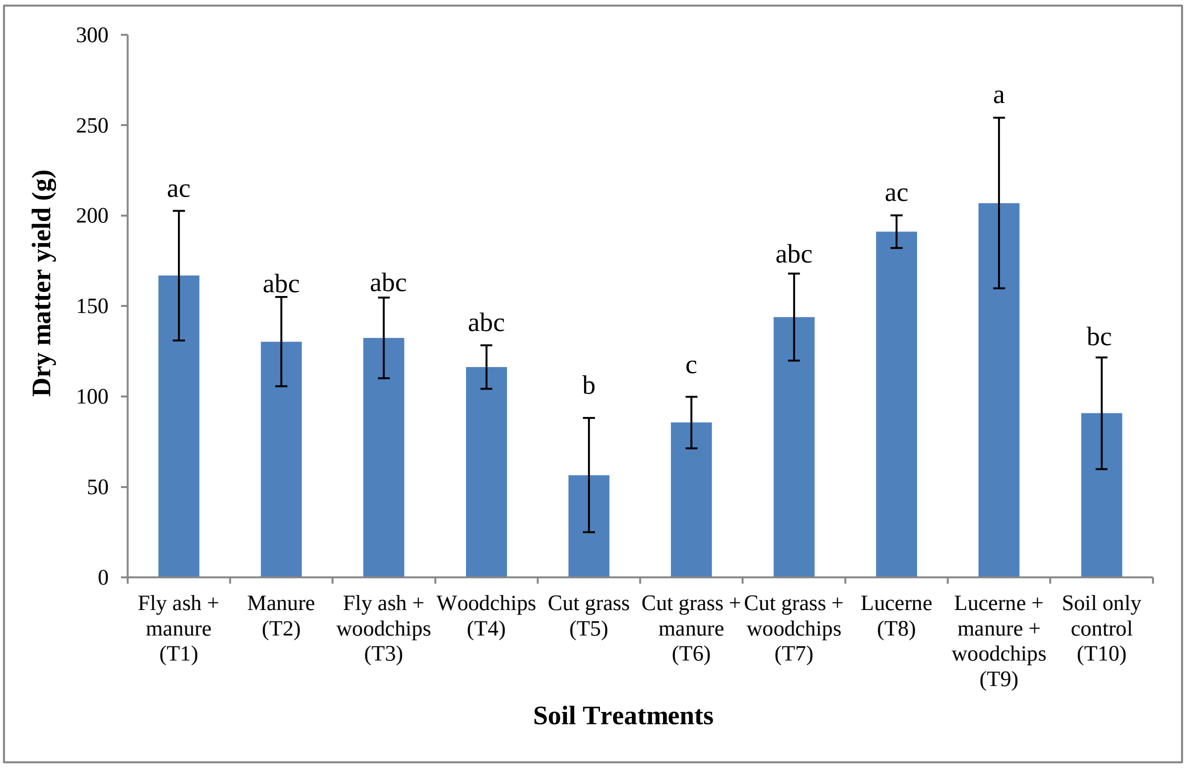

Figure 11 - Mean root dry matter yield of *A. sativa* from 1 m² as a result of the different soil treatments applied. Bars with the same letter have no significant difference in the mean dry matter yield at (*p*<0.05). Vertical lines represent the standard error of the mean.

The mean root dry matter yield of *E. tef* was significantly influenced by treatment applications at ($p<0.05$). Treatment T9 showed the greatest significant difference ($p<0.05$) in mean root biomass with a mean increase in root biomass of 453.41% compare to the control. Treatment T3 measured the second greatest root yield with a mean increase of 257.31% compare to the control. Both T9 and T3 showed significant differences ($p<0.05$) in mean bulk density compared to the control after the *E. tef* harvest. These decreases in bulk density may have helped to create a soil environment which, again, is more conducive to root development as a decrease in bulk density is known to increase the water content range within which root development is less likely to be inhibited (da Silva and Kay, 1997). As mentioned before, the treatments which measured higher levels of soil macro nutrients after the harvest expressed greater overall root yields. Lastly, it is evident from Figure 11 and 12 that *A. sativa* has a more pronounced rooting system compared to the *E. tef* species as a result of the species difference and thus, has shown a larger biomass yield for each of the different soil treatments.

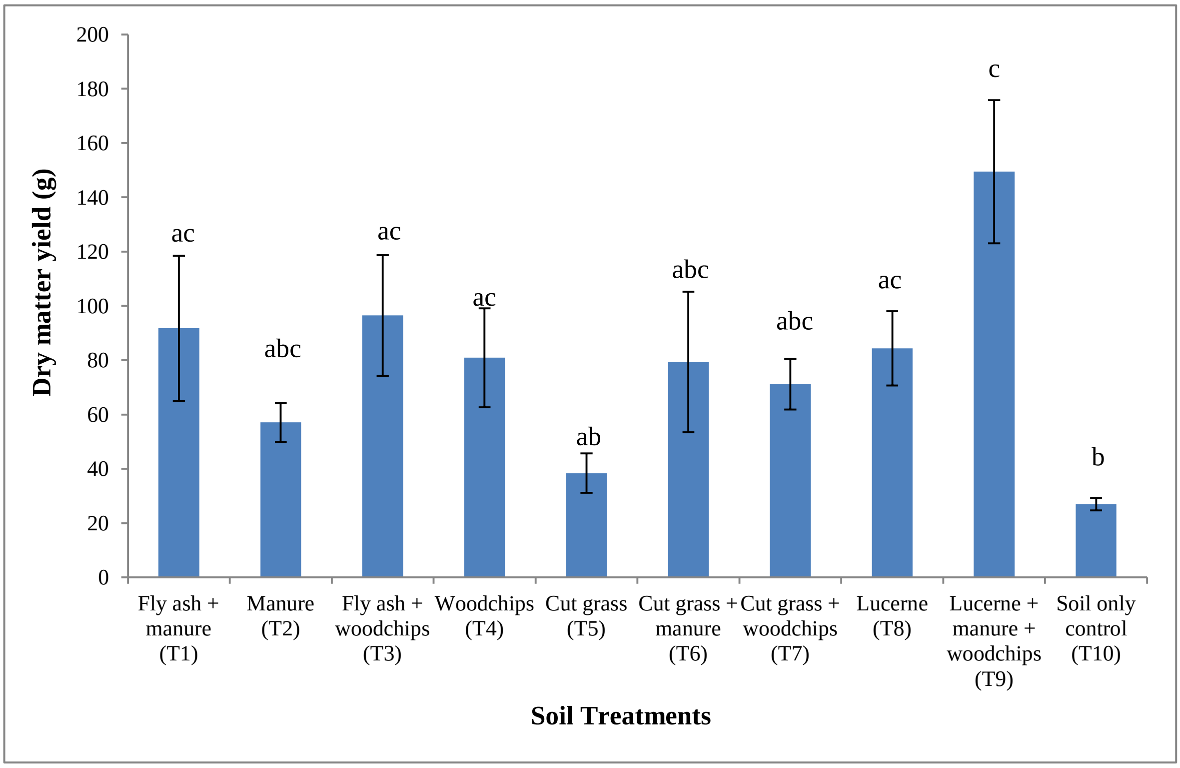

Figure 12 - Mean root dry matter yield of *E. tef* extrapolated from 0.09 m² to 1 m² as a result of the different soil treatments applied. Bars with the same letter have no significant difference in the mean dry matter yield at (*p*<0.05). Vertical lines represent the standard error of the mean.

4.4 Soil fertility measured after *A. sativa* and *E. tef* harvest

The Kruskal-Wallis test showed a significant difference (p<0.01) between the means of all the different soil nutrients after the *A. sativa* harvest (Table 4) as a result of the different treatments applied (Appendix B1). However, the Kruskal-Wallis test showed no significant difference at (p<0.05) for the mean levels of sodium measured as a result of the different treatments applied but significant differences (p<0.05) of the rest of the soil nutrients measured after the *E. tef* harvest (Appendix B2).

The topsoil used in this trial showed a deficiency in potassium but reasonably good soil nutrient levels for the rest of the plant macro nutrients (Table 2). From Table 4 it is evident that the cut grass treatment (T5) and the control (T10) resulted in soil recorded values which fall below the recommended healthy range given in FSSA (2007) when considering the amounts of phosphorus, potassium, and magnesium. The effects of treatments T3, T4, and T8 showed recorded soil values which fall within the recommended healthy range given in FSSA (2007) for all soil nutrients shown in Table 4 while the effects of treatment T9 measured excess amounts of phosphorus within the soil. Lastly, the effects of treatments T1, T2, and T6 measured excess amounts of phosphorus and potassium within the soil with regards to the recommended healthy range given in FSSA (2007). By examining the analysis of the treatments themselves, (Table 2) one can see that fly ash contains poor amounts of macro nutrients except for the amount of calcium and as a result, all soils amended with fly ash showed an increase in calcium within the soil as compared to the rest of the soils. Soils amended with cattle manure showed large increases in potassium, sodium and magnesium due to the high inherent content of these nutrients within the treatment. Alternatively, analysis of the cut grass treatment showed poor amounts of macro nutrients except for the relatively good amounts of potassium while the lucerne treatment showed very high amounts of nitrogen. As expected, soils amended with treatment T9 showed a good balance of all macro nutrients measured after both harvests.

From Table 5 it is evident that the effects of treatments T3, T7 and T9 within the soil showed values which fall within the recommended healthy range given in FSSA (2007) for all soil nutrients, while the effects of treatments T1, T2, T6 showed excess amounts of phosphorus and potassium within the soil. The effects of treatments T5 and T10 recorded values which fall below the recommended healthy range given in FSSA (2007) when considering the amounts of phosphorus, potassium, and magnesium measured within the soil. Furthermore, Table 5 shows that the amount of nutrients within all the amended soils generally decreases after the *E. tef* harvest except for the soils amended with woodchips. Possible reasons for the decrease in nutrients may be as a result of nutrient removal after harvesting, leaching, plant uptake, or immobilization of nutrients (Truter, 2007). As stated earlier, an increase in potassium was measured in all soils amended with woodchips which may have been due to the delayed release of potassium from the woodchips as a result of the greater extent of decomposition after the *E. tef* harvest as compared to the *A. sativa* harvest.

Table 4: Soil macro nutrients recorded after *A. sativa* harvest with the general recommended soil nutrient amounts represented in square brackets according to The Fertilizer Society of South Africa Handbook (2007). The standard error of the mean is given in round brackets.

Soil Treatments	P Bray 1	Ammonium acetate extraction			
	Mean P [15-30]	Mean K [80-160]	Mean Na	Mean Ca [300-2000]	Mean Mg [80-300]
	mg.kg^{-1}				
Fly ash + manure (T1)	64 (±13.32)	283 (±66.29)	31 (±6.97)	567 (±52.99)	143 (±16.56)
Manure (T2)	47 (±12.06)	214 (±60.50)	26 (±6.22)	430 (±37.88)	113 (±16.01)
Fly ash + woodchips (T3)	21 (±3.47)	87 (±22.24)	17 (±2.96)	458 (±45.85)	85 (±12.56)
Woodchips (T4)	27 (±4.87)	97 (±9.84)	16 (±1.70)	411 (±7.32)	92 (±4.53)
Cut grass (T5)	11 (±0.41)	79 (±6.73)	13 (±1.75)	358 (±15.42)	76 (±4.77)
Cut grass + manure (T6)	54 (±13.66)	273 (±104.81)	27 (±9.14)	424 (±46.44)	117 (±19.55)
Cut grass + woodchips (T7)	19 (±2.21)	82 (±6.09)	12 (±0.48)	348 (±17.79)	74 (±4.78)
Lucerne (T8)	18 (±2.49)	131 (±12.06)	14 (±1.03)	414 (±14.20)	87 (±3.69)
Lucerne + manure + woodchips (T9)	36 (±2.36)	154 (±10.20)	19 (±0.65)	418 (±13.35)	103 (±3.18)
Soil only control After (T10)	13 (±0.48)	48 (±1.66)	11 (±0.41)	339 (±15.19)	66 (±3.14)

Above recommended anmount – 🟦
Below recommended amount – 🟥
Within recommended amount – ■

Table 5: Soil macro nutrients recorded after *E. tef* harvest with the general recommended soil nutrient amounts represented in square brackets according to The Fertilizer Society of South Africa Handbook (2007). The standard error of the mean is given in round brackets.

Soil Treatments	P Bray 1	Ammonium acetate extraction			
	Mean P [15-30]	Mean K [80-160]	Mean Na	Mean Ca [300-2000]	Mean Mg [80-300]
	mg.kg^{-1}				
Fly ash + manure (T1)	46 (±12.38)	198 (±17.03)	15 (±2.59)	475 (±52.87)	108 (±14.27)
Manure (T2)	37 (±12.31)	206 (±22.33)	12 (±0.95)	352 (±14.91)	92 (±11.21)
Fly ash + woodchips (T3)	22 (±2.68)	123 (±21.62)	14 (±1.55)	496 (±35.29)	91 (±9.69)
Woodchips (T4)	13 (±1.35)	117 (±10.35)	13 (±1.04)	353 (±30.66)	78 (±6.96)
Cut grass (T5)	8 (±0.25)	79 (±10.14)	13 (±0.71)	336 (±14.09)	68 (±4.19)
Cut grass + manure (T6)	32 (±3.63)	222 (±33.13)	13 (±0.75)	389 (±18.21)	101 (±5.02)
Cut grass + woodchips (T7)	16 (±5.55)	120 (±23.64)	13 (±1.08)	353 (±38.12)	81 (±14.03)
Lucerne (T8)	11 (±0.82)	135 (±13.79)	14 (±0.41)	369 (±8.59)	73 (±3.11)
Lucerne + manure + woodchips (T9)	20 (±1.65)	171 (±10.59)	15 (±0.95)	428 (±14.13)	98 (±4.73)
Soil only control (T10)	9 (±0.5)	66 (±5.02)	12 (±1.79)	322 (±10.26)	63 (±3.64)

Above recommended amount – 🟦
Below recommended amount – 🟥
Within recommended amount – ■

5. Conclusion

It is evident from this trial that the application of the different soil treatments significantly affected the plant yield and the soil physical and chemical properties to extents which differ substantially. The cut grass treatment (T5) proved to be the worst treatment as it showed no significant difference ($p<0.05$) in the soil pH, aboveground biomass and root biomass compared to the control. The lucerne, manure and woodchips treatment, (T9) however, showed significant differences ($p<0.05$) in all the parameters measured and thus, proved to be the best treatment used in this trial. Treatment T9 proved to be excellent in decreasing the bulk density in just one season and therefore, demonstrating its value in compaction alleviation and its overall positive impact on the physical properties of the soil. The lucerne, manure and woodchips treatment also measured the greatest increase in aboveground and root dry matter yield for both pasture species which highlights the advantages in which each individual component within treatment T9 (manure, woodchips and lucerne) had on the pH and the nutrient availability to the plant. This emphasizes the different advantages one can expect by using a combination treatment which can improve different soil properties simultaneously to result in on overall positive impact on plant development.

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For clarity reasons tables do not follow the format guidelines for the journal of The Southern African Institute of Mining and Metallurgy.

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CHAPTER 3

ORGANIC MATTER CONTRIBUTION OF SOIL AMELIORANTS TO CARBON STORAGE OF SURFACE MINED SOIL

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Abstract

Soil impacted by topsoil and vegetation stripping caused by surface coal mining activities substantially decreases the amount of organic carbon within the soil and thus, the ability of the soil to properly sustain vegetation. As a result, a considerable amount of research has been focused on the sequestration of atmospheric carbon into stable soil organic matter as a means to reduce concentrations of carbon in the atmosphere and at the same time improve soil fertility for plant growth. In this investigation we evaluate the significance of soil carbon improvement strategies on soil impacted by surface coal mining in South Africa. The investigation involved 40, 1 m long by 1 m wide by 1.2 m high experimental soil research bins “mini-lysimeters” which were constructed to represent a rehabilitated soil/substrate profile which is commonly created through rehabilitation practices on surface coal mines in South Africa. The bins were filled with mine spoil, subsoil and topsoil from a coal mine in Mpumalanga. The trial consisted of ten different topsoil treatments with four replicates per treatment including a control treatment consisting only of the cover soil. *Avena sativa* and *Eragrostis tef* were planted as the winter and summer test pasture species. Results from this trial indicate that the soil amendment which provided the greatest increase in yield and the greatest overall increase in the resultant carbon captured was the combination treatment containing chopped lucerne, composted woodchips and cattle manure (T9). Additionally, this treatment illustrated a drastic decrease in bulk density and a substantial improvement in the soil pH and plant available nutrients. When data is extrapolated to per hectare, this treatment resulted in an overall increase of 33.1 $tCO^2/ha/annum$ more than the control which illustrates the amount of carbon dioxide which can be sequestered if proper soil management techniques are applied in conjunction with the use of soil ameliorants. Furthermore, it highlights the importance of soil organic carbon

improvement strategies which provide both a soil rehabilitation function as well as an improved carbon storage function.

1. Introduction

Soil impacted by the topsoil and vegetation stripping caused by surface coal mining activities substantially decreases the amount of organic carbon within the soil with some cases illustrating a 80% loss of soil organic carbon (Akala and Lal, 2001). Stripping and stockpiling topsoil negatively impacts the ability of the soil to support plant development as the chemical, physical and biological properties are drastically altered. The compaction of soil is also known to drastically limit root establishment and increase erosion which makes the development of grass species next to impossible (Jha and Singh, 1992; Singh *et al*, 1996). Additionally, the loss of soil organic matter (SOM) as a result of poor management practices related to the stockpiling of soil leads to the dilution of organic carbon through the mixing of subsoil and topsoil, erosion losses, exposure of SOM to oxygen and the reduction in plant growth (Ripley *et al.*, 1996; Stahl *et al.*, 2002). As a result, a focus has recently been placed on research with an emphasis on the sequestration of atmospheric carbon into stable SOM on rehabilitation sites as a means to reduce concentrations of carbon in the atmosphere (Ganjehunte, *et al.*, 2009).

The sequestration of carbon by plants is a natural process as plants remove carbon dioxide from the atmosphere and incorporate it within plant biomass or within the soil. With regards to surface mine rehabilitation, carbon sequestration improves the organic matter content of the soil as plants essentially transform atmospheric CO_2 into plant biomass through photosynthesis and incorporate this biomass into the soil, ultimately forming humus and thus, increasing the carbon content of the soil and supplying a source of carbon and nutrients for plant and microbial growth (Nair, *et al.*, 2010). Plant roots contribute a substantial amount SOM through root death and exudation of organic substances during plant growth (Singh *et al.*, 1991). Boone (1994), Haider *et al.* (1993), Norby and Cortrufo (1998) and Sanchez *et al.*, (2002) state that roots have a greater influence on soil organic carbon and nitrogen levels than the aboveground plant biomass. The organic input from plant roots to the surrounding soil environment is the principal support of biological activity and abundance of organisms (Cheng *et al.*, 1994; Kirchner *et al.*, 1993) and therefore, rehabilitation with a focus towards improving soil conditions for the re-establishment of plant roots can have a direct impact on the biogeochemical carbon cycle and additionally, introduce carbon into deeper soil horizons (Tripathi *et al.*, 2014). Hence, the value of vegetation establishment and development on topsoil plays an extremely important role towards the success of rehabilitation and soil organic carbon improvement.

The potential to increase the carbon content in soil impacted by surface mining appears to offer much promise as theses soils have a low inherent organic carbon content and thus, have large room for improvement (Follet *et al*., 2000; Ingram *et al*., 2008; Lal *et al*., 1998). This poses as an excellent opportunity to increase the carbon content in soil through appropriate carbon sequestration methodologies and thereby, an opportunity to help mitigate the carbon dioxide concentration in the atmosphere while still improving the fertility of the soil for proper plant development. Furthermore, landscape changes due to the impact of surface mining results in a significant impact on the carbon cycle and consequently on the earth's climatic system. With the average global atmospheric concentration increasing at 2 ppm per year and standing at 397 ppm in 2013, the prospect of soil carbon storage displays an attractive and promising alternative to carbon mitigation (Acre *et al*., 2014). Hence, the assessment of the carbon dynamics under different soil and vegetation types provide valuable information towards understanding carbon storage methodologies and ecosystem functionality (Chung *et al*., 2012; Prescott *et al*., 2000; Schoenholtz *et al*., 2000).

National Treasury in South Africa has stated that they will be implementing carbon tax in 2016, in which South Africans will be paying R120 per ton of carbon dioxide emitted over the basic tax free threshold of 60% (Department of National Treasury, 2014). This emphasizes the value of implementing soil organic carbon improvement strategies on surface mined soil to help mitigate carbon dioxide release. However, with this being said, rehabilitating surface mined soil with a focus on improving the organic carbon content of soil is a very sophisticated and challenging task. While the value of carbon sequestration projects have shown success on rehabilitated surface coal mine areas in countries outside of South Africa, the value and potential of carbon sequestration and carbon focused rehabilitation projects in South Africa have yet to be explored. In this study, the impact in which the application of different soil organic ameliorants have on soil impacted by surface coal mining were evaluated by examining the associated impact on soil properties and soil carbon storage.

2. Material and methods

2.1 Experimental site

A field trial was conducted at the Hatfield Experimental Farm of the University of Pretoria, Pretoria, South Africa (25°45°S, 28°16°E) 1327m above sea level in 2014 and 2015. The trial involved 40, 1 m long by 1 m wide by 1.2 m high experimental soil research bins "mini-lysimeters". The research bins were first filled with a 10 cm layer of gravel (to allow for free water drainage), followed by 50 cm layer of mine spoil followed by 30 cm layer of subsoil and lastly a 30 cm layer of mine top soil which was treated with different organic soil amendments (Table 1). The mine spoil, subsoil and top soil (Hutton, sandy loam) were all acquired from a surface coal mine in Mpumalanga, South Africa. The design of the soil research bins were to simulate and represent a rehabilitated soil/substrate profile which is commonly created through rehabilitation practices on a surface coal mines in South Africa. Prior to the treatment application, the topsoil, subsoil and the different soil treatments were analysed for pH, carbon content and macro nutrient content.

2.2 Experimental layout and treatment application

The treatment applications for each research bin were arranged in a complete randomised block design and consisted of nine different top soil treatments applied at either 40 or 132 tons per hectare (Table 1) with four replicates per treatment including a control treatment containing no soil ameliorant. The topsoil was placed into a concrete mixer together with the respective soil treatment to insure that the treatment was thoroughly blended with the top soil. Once the top soil was thoroughly mixed it was offloaded into the research bins to make a 30 cm amended top soil layer. After the amended soil had been placed in the research bins it was left to settle for four weeks. Once the four week period had passed the soil had subsided due to heavy rainfall during March of 2014 (Table 2) and therefore, an additional application of amended soil was applied to fill the research bins to the 30 cm surface mark once more. *Avena sativa* L. (Overberg cultivar) was selected as the winter cover crop test species. This particular species was chosen because it is one of the strongest winter growing annuals in South Africa which can tolerate low fertility soils. Furthermore, this species was used strictly as a cover crop to improve the physical and chemical properties of the soil such as: increasing soil organic matter content, increasing microbial activity, improving soil nitrogen status through nitrogen fixation, increasing soil pH, improving soil structure, reducing erosion and increasing nutrient supply within the soil (Harris and Megharaj, 2001; Pieters, 2006). Additionally, the *A. sativa* winter cover crop was used to evaluate the treatments applied to impacted surface mined soil and their effects on plant development.

Avena sativa seeds were planted in rows at an application rate of 40 kg per hectare on 1 April 2014 as the winter test pasture. These plants were then watered until seedlings emerged after which they were left without irrigation and, were therefore, rain fed for the remaining days. After the 90 day growth period, aboveground plant material was harvested by sampling from a 0.09 m² area from each of the 40 research bins at a height of 30 mm above the soil surface and maintaining a 15 cm buffer strip from the boarders of the research bins. The aboveground material was placed in paper bags and put into an oven at 60 ºC for 48 hours (Goering and Van Soest, 1970) after which the samples were weighed and analysed for total organic carbon content by combustion analysis at 600 °C in which the carbon content is then calculated from the corresponding carbon dioxide which gets released (AOAC, 1995). Root samples were then harvested from the same 0.09 m² area and rinsed with tap water to insure all debris was removed before drying. The root biomass was then analysed to determine the total organic carbon content by combustion analysis. After harvest, the soil research bins were equally irrigated and left for three days before soil penetrometer readings were taken with the Geotron Hand Penetrometer Model P5 to determine the effect that each soil treatment had on soil compaction. Using the penetrometer readings, bulk density was calculated using the equation formulated by Hernanz *et al*., (2000). Once compaction readings were taken, an auger was used to take soil samples at a depth of 25 cm from each research bin. The pH (KCL) was then determined and the macro nutrients of all soil samples were then analysed by ammonium acetate extraction. Lastly the soil carbon was determined by using the Walkley and Black (1934) method and corrected for underestimates by multiplying the carbon percentage values by 1.33 (Schumacher, 2002).

Eragrostis tef (Zucc.), (SA Brown cultivar) was selected as the summer pasture because it is one of the strongest growing summer annuals and is an excellent nurse crop. This particular species has a very hardy nature (C4) and is commonly used on coal mine rehabilitation sites in South Africa (Mentis, 1999). On the 16 of October 2014, *Eragrostis tef* seeds were broadcasted at an application rate of 10 kg per hectare, irrigated until seedlings emerged after which the plants were rain fed until final harvest on the 9 of April 2015. The aboveground biomass and root samples where then harvested by sampling from a 0.09 m² area after which the root samples were then rinsed with tap water to insure all debris was removed before drying. Both the aboveground biomass and the root biomass were dried using the same method mentioned above, after which the samples were weighed and analysed to determine the total organic carbon content by combustion analysis. Penetrometer readings were taken following the same method mentioned above, after which soil samples were taken at a depth of 25 cm from each research bin. Lastly using the same methods described earlier, all soils were analysed for pH, carbon content (Walkley Black), and macro nutrient content.

Table 1: List of the soil treatments applied (n = 4). Where combinations of treatments exist, the total application rate is equally divided among each treatment.

Soil Treatments	Treatment numbers	Origin of Treatment
Fly ash (class F) and composted cattle manure[b]	T1	A + B
Composted cattle manure[a]	T2	B
Fly ash and black wattle composted woodchips *(Acacia mearnsii*)[b]	T3	A + B
Black wattle composted woodchips *(Acacia mearnsii*)[a]	T4	C
Cut grass (*Digitaria eriantha*)[a]	T5	D
Cut grass (*Digitaria eriantha*) and composted cattle manure[a]	T6	D + B
Cut grass (*Digitaria eriantha*) and black wattle composted woodchips *(Acacia mearnsii*)[a]	T7	D + B
Lucerne hay (*Medicago sativa)*[a]	T8	D
Lucerne hay (*Medicago sativa)* and composted cattle manure and black wattle composted woodchips *(Acacia mearnsii*)[a]	T9	D + B + B
Top soil only (control)	T10	E

[a] Applied at 40 tons per hectare

[b] Applied at 132 tons per hectare

A = Fly ash from Ash Resources (Pty) limited, South Africa
B = Conradie Organics, Pretoria, South Africa
C = Jacklin Organics, Pretoria, South Africa
D = Hatfield experimental farm, Pretoria, South Africa
E = Surface coal mine in Mpumalanga, South Africa

Table 2: Chemical composition of ameliorants and soil substrates

Treatments	P%	K%	Na%	Ca%	Mg%	N%	C%
Fly ash	0.08	0.02	0.12	2.68	0.47	0.02	0
Composted cattle manure	0.83	2.74	0.41	1.80	0.81	2.50	32.85
Black wattle composted woodchips	0.32	1.05	0.16	0.92	0.36	1.71	21.71
Cut grass	0.13	1.27	0.08	0.24	0.09	0.94	55.92
Lucerne	0.18	0.69	0.03	0.81	0.26	3.18	54.67
Substrates			mg kg^{-1}			pH	
Topsoil	21	59	13	343	78	5.18	0.96
Subsoil	1	76	14	295	283	4.28	0.27

2.3 Weather

All weather data was collected on Hatfield experimental farm using an automatic weather station which consisted of: one LI 200X pyranometer (LiCor, Lincoln, Nebraska, USA) measuring solar radiation, an electronic cup anemometer (MET ONE, Inc. USA) measuring wind speed, an electronic rain gauge (RIMCO, R/TBR and tipping bucket rain gauge, Rauchfuss Instruments Division, Australia) measuring rainfall, a temperature and relative humidity sensor, and a CR 10X data logger (Campbell Scientific Inc., USA) which recorded the data at 10 second intervals.

Table 3: The mean maximum and minimum temperatures in addition to the total rainfall recorded per month for years 2014 and 2015.

	Maximum Temperature (°C)	**Minimum Temperature (°C)**	**Average Rainfall (mm)**
		2014	
March	25.68	13.65	253.30
April	24.43	13.81	11.70
May	24.47	7.21	0.50
June	21.57	3.46	0.90
July	20.55	3.51	0.00
August	23.32	7.35	13.70
September	28.95	11.93	0.70
October	29.05	12.90	30.70
November	26.95	14.29	103.70
December	28.39	16.25	265.40
		2015	
January	30.17	16.86	85.20
February	31.28	16.63	22.10
March	29.41	15.62	57.80
April	26.42	11.84	29.70

3. Statistical analysis

Ten different soil treatments including a control treatment were replicated four times. Treatments were allocated to research bins in a complete randomised block design while all statistical analysis was conducted using IBM SPSS statistical software version 22© (IBM Corp released, 2013). Normal distribution of the mean values were confirmed by examining both the Normal Q-Q plot and the Detrended Normal Q-Q plot along with the Shapiro-Wilk test of normality ($P>0.05$). The homogeneity of variance was tested by Levene’s test for equality of variances ($P>0.05$). One-Way Analysis of Variance (ANOVA) and Tukey’s Highest Significant Difference ($P<0.05$) post-hoc comparisons of the means test was conducted to test significant differences in all the carbon values.

4. Results and discussion

Rehabilitating soil impacted by surface coal mining with soil organic amendments helps to increase the inherent low levels of organic matter within the soil and helps to improve the chemical, physical and biological properties of the soil, thereby, simultaneously helping to mitigate the carbon dioxide concentration in the atmosphere. All data collected in this study was used to illustrate to what extent the different organic soil treatments affected the soil organic carbon content, the soil properties and the associated plant biomass. The aboveground dry biomass, the root dry biomass and the organic carbon within such biomass was extrapolated from the 0.09 m^2 area to 1 m^2. Data was constantly reported in comparison to the control and the results indicate that the application of these different organic soil ameliorants have a substantial effect on the organic carbon content of the soil, the soil fertility and associated plant biomass.

4.1 Aboveground dry matter yield and organic carbon content of *A. sativa* and *E. tef*

The amount of carbon stored within the aboveground biomass was determined by multiplying the aboveground dry matter yield by the carbon content in percentage (determined by combustion analysis) within the corresponding aboveground biomass (Appendix A). From Figure 1 and 2 it is evident that the amount of carbon in grams within the aboveground biomass of both species is directly related to the amount of dry biomass produced. The difference in the resultant aboveground biomass of each soil treatment follows the same pattern as the difference in the total carbon stored within the biomass. Therefore, as the respective soil treatments have varied effects on the amount of aboveground dry biomass produced, they to, have a varied effects on the amount of carbon stored within the aboveground biomass. Hence, the soil treatments which resulted in the greatest amount of aboveground dry biomass also resulted in the greatest amount of carbon stored. Thus, the resultant amount of carbon stored is a function of soil properties as the soil properties affect the amount of aboveground biomass. Therefore, the soil treatment which best improves the soil properties, best improves the amount of carbon stored within the resultant aboveground biomass.

From Figure 1 it is evident that treatment T9 was the only treatment which showed a significant difference ($p<0.05$) in mean carbon stored compared to the control. Treatment T9 showed the greatest increase in mean aboveground carbon stored compared to the control with a mean increase of 122.83% while T8 and T1 showed the second and third greatest aboveground carbon stored with mean increases of 107.48% and 77.01% respectively when compared to the control. The large differences in the amount of carbon stored within the aboveground biomass can be attributed to the soil treatments (T9, T8 and T1) which showed a drastic decrease in soil bulk density, and additionally increased the soil pH and the level of soil macro nutrients in comparison to the control (Appendix B). Ultimately, these improvements in soil properties helped create a soil environment which is more conducive to greater plant development and therefore, overall aboveground carbon storage.

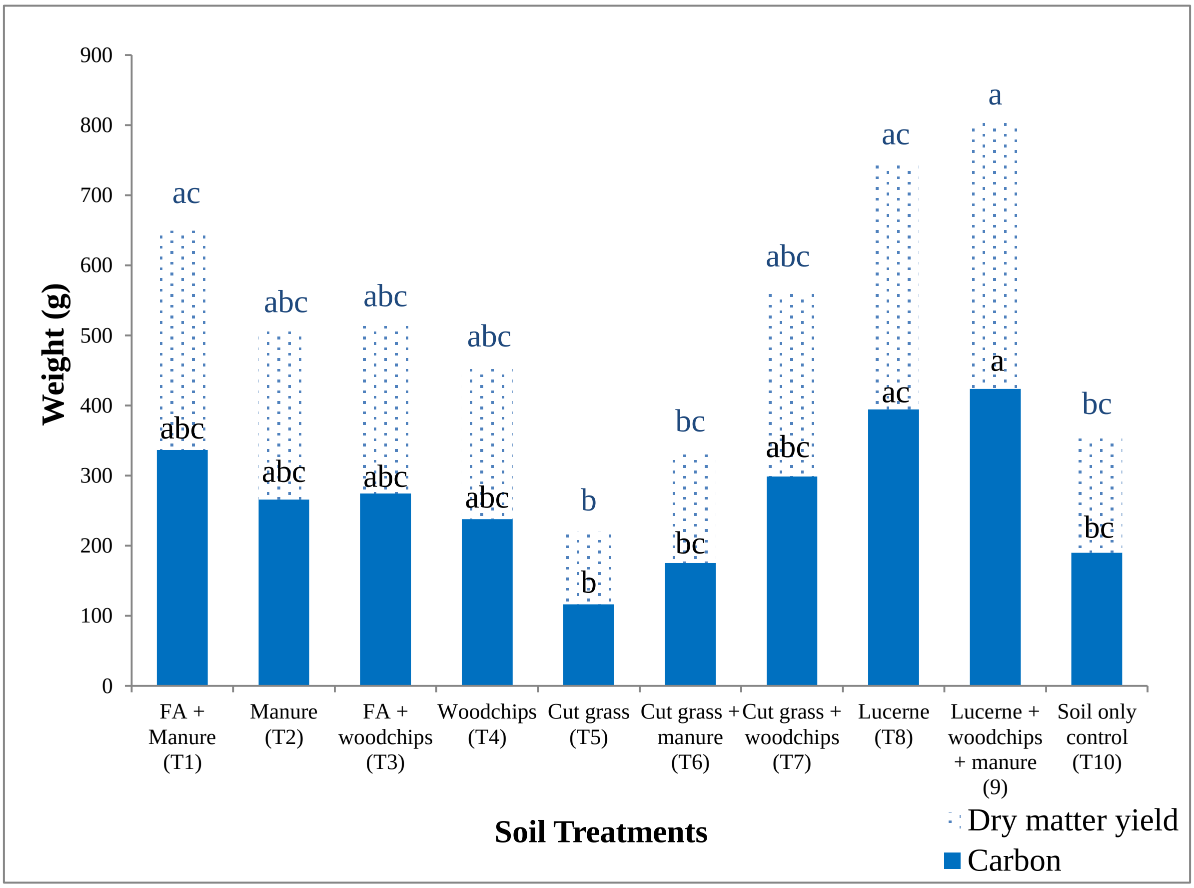

Figure 1 - Mean *A. sativa* aboveground dry matter yield and carbon content within the aboveground biomass from 1 m² as a result of the different soil treatments applied. Bars with the same respective lettering have no significant difference in the mean aboveground biomass or carbon content at (*p*<0.05).

From Figure 2 it is evident that the *E.tef* species resulted in larger amounts of aboveground carbon stored than compared to *A. sativa* due to the larger biomass produced as result of species difference. Yet, no significant difference ($p<0.05$) in mean aboveground carbon stored, was measured as a result of any of the soil treatments applied when compared to each other or to the control. However, treatments T9 and T4 showed the largest increases with a mean increase in aboveground carbon stored of 41.85% and 29.32% respectively when compared to the control. The soil analysis results from Appendix B illustrate that treatment T9 and T4 showed considerable increases in soil macro nutrient content compared to the control. These increases in plant nutrients improved the soil nutrient status and thus, had positive impact on aboveground biomass and therefore, a positive impact on the amount carbon stored.

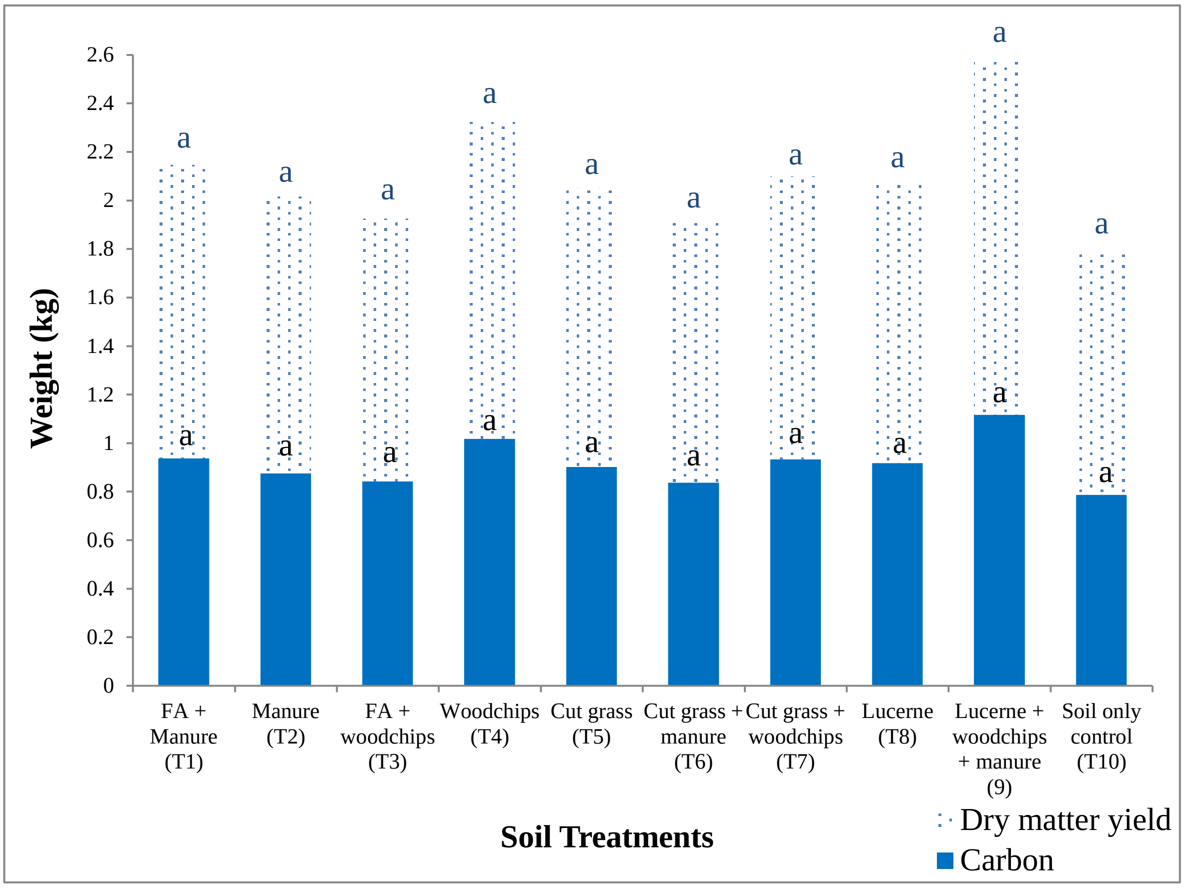

Figure 2 - Mean *E. tef* aboveground dry matter yield and carbon content within the aboveground biomass from 1 m² as a result of the different soil treatments applied. Bars with the same respective lettering have no significant difference in the mean aboveground biomass or carbon content at (*p*<0.05).

4.2 Root dry matter yield and organic carbon content of *A. sativa* and *E. tef.*

The mean root dry matter yield for *A.sativa* was calculated using the mean shoot to root response (3.89) measured after a 90 day emergence pot trial was conducted on *A.sativa* by Amanullah *et al*., (2015). The amount of carbon (in grams) stored within the root biomass was determined by the same method used for the aboveground biomass in which the dry root matter yield was multiplied by the carbon content in percentage within the corresponding root biomass (Appendix A). As was mentioned above, the amount of carbon in grams within the root biomass of both species is related to the amount of dry biomass produced. Therefore, the soil treatments which greatly improve the root biomass, greatly improve the amount of carbon stored and thus, the amount of total carbon stored within the root biomass is a function of the soil properties. However, the relationship between the amount of carbon stored within the root biomass does not follow the same pattern as the amount of carbon stored within the aboveground biomass. This is due to carbon percentage differences measured within the roots of both these pasture species (Appendix A) which drastically affects the total carbon stored. Additionally the stored carbon within the roots, measured on average, less than the stored carbon within the aboveground biomass (Appendix A).

From Figure 3 it is evident that there were no significant differences ($p<0.05$) in the mean root carbon stored as a result of the different soil treatments applied when compared to the control. However, significant differences ($p<0.05$) exist when comparing the treatment results amongst each other. Figure 3 illustrates the positive effect in which the lucerne treatment (T8) and the lucerne, manure and woodchips treatment (T9) had on the root carbon stored with mean increases of 120.37% and 118.21% respectively when compared to the control. Once again this can be attributed to the positive effects in which these treatments have on the soil properties (Appendix B) which in turn had an improvement on root yield and thus improved carbon storage within the plant roots.

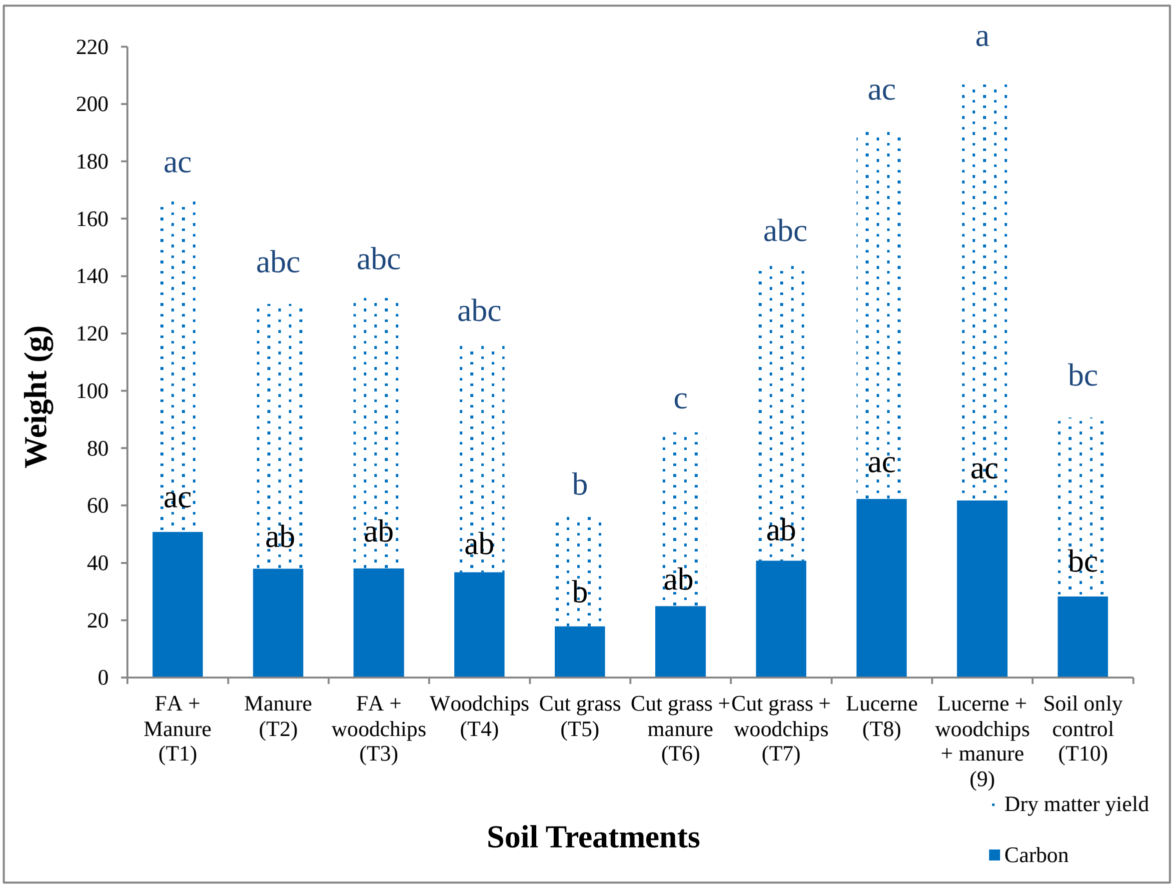

Figure 3 - Mean *A. sativa* root dry matter yield and carbon content within the root biomass from 1 m^2 as a result of the different soil treatments applied. Bars with the same respective lettering have no significant difference in the mean root biomass or carbon content at (p<0.05).

The carbon stored within the *E. tef* root biomass was significantly influenced (p<0.05) by the different treatment applications. Again, treatment T9 illustrating the largest increase in root carbon stored. This treatment showed a mean increase of 418.91% and a significant difference (p<0.05) when compared to the control. Treatment T3 was the only other treatment to show a significant difference (p<0.05) in mean root carbon stored when compared to the control with an increase of 247.78%. Both treatments showed a substantial decrease in soil bulk density and considerable improvements in pH and the levels of macro nutrients (Appendix B). Therefore, providing a soil environment which is more conducive to root development and thus, carbon storage.

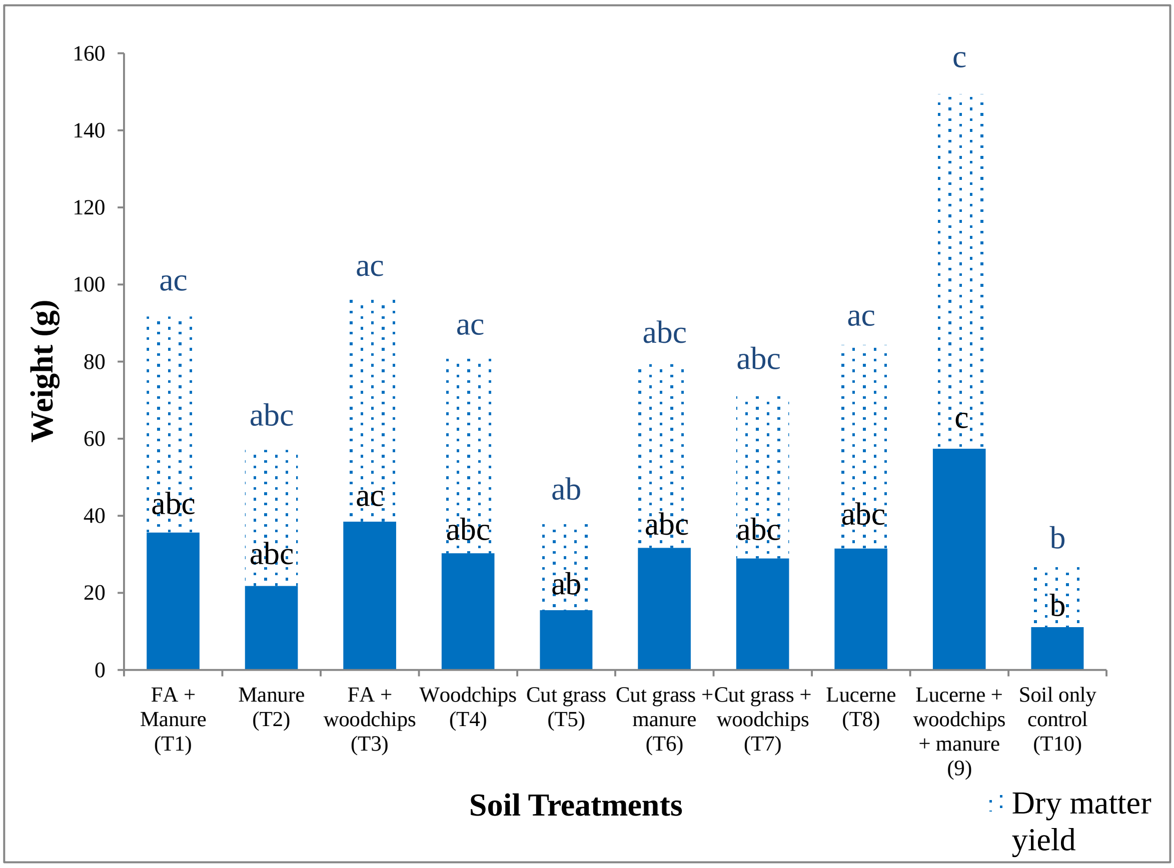

Figure 4 - Mean *E. tef* root dry matter yield and carbon content within the root biomass from 1 m^2 as a result of the different soil treatments applied. Bars with the same respective lettering have no significant difference in the mean root biomass or carbon content at (p<0.05).

4.3 Organic carbon captured

List of organic carbon equations (Carbon captured over the 265 day growth period)

- Soil organic matter (%)

Soil organic carbon (%) x 1.72 (Bowen and Rovira, 1999)

[Equation 1]

- Soil organic carbon content (kgC m^{-2})

[Soil organic carbon (%) x soil bulk density (g.cm^3) x sampling depth (cm)] / 10 (Chan, 2008a)

[Equation 2]

- Total organic carbon sequestered (kgC m^{-2})

Soil organic carbon content (kgC m^{-2}) + aboveground biomass carbon content (kgC)

+ root biomass carbon content (kgC)

[Equation 3]

- Total carbon dioxide sequestered ($kgCO^2$ m^{-2})

Total organic carbon sequestered (kgC m^{-2}) x 3.667 (Chan, 2008b)

[Equation 4]

4.3.1 Soil organic carbon (%)

Soil impacted by surface coal mining generally results in a very low soil organic carbon content (Ojeda, *et* al., 2015). In this trial the organic carbon content of the topsoil before the trial, was measured at 0.96% (Table 2) while a healthy soil organic carbon content of topsoil is generally 3.5% and more. This illustrates the large potential for soil carbon storage. In order to increase the organic carbon within the soil one needs to re-introduce a source of carbon or recapture carbon to restore the soil organic matter content. The best way to do this is to have a constant supply of organic matter through continuous vegetation development. Improving the organic matter within the soil through the application of soil ameliorants and plant growth will in turn improve the soil structure and the nutrients within the soil (Nair, *et al.*, 2010). Additionally, soil amendments themselves physically add organic matter once incorporated into the soil and improve the soil environment for plant development, thereby, supporting continuous vegetation development and thus improving the soil organic carbon content through a constant source of organic carbon.

The soil organic carbon measured after the *A. sativa* harvest showed no significant difference ($p<0.05$) in the mean carbon percentage. From Table 4 it is evident that woodchips treatment (T4) showed a mean increase in soil organic carbon of 33.58% when compared to the control. Additionally, all treatments which contained woodchips (T3, T4, T7 and T9) showed an increase in soil organic carbon when compared to the control. The woodchip treatment itself, showed a carbon content of 21.71% (Table 2). However, soil organic carbon is comprised of all sources of organic carbon not only from the soil treatment but also from rapidly decomposable organic matter such as plant and animal matter which is “living” or “dead” and more resistant organic matter such as humus and lignin rich materials which is “very dead” and therefore, more stable (Hoorman and Islam,

2010) (Figure 5). This may be the reason why a greater soil organic carbon content was measured as a result of the incorporation of woodchips than compared to the incorporation of cut grass, in which the treatment itself, measured a carbon content of 55.92% (Table 2). The organic carbon stored within the soil after the woodchips treatment (T4) not only came from the carbon content of the treatment itself but also from decaying roots and the associated root exudates which are comprise of soluble compounds, lysates, secretions and dead fine roots which also contribute to long-term soil carbon storage (Tripathi *et al*., 2014). Furthermore, all research bins incorporated with woodchips showed a substantial increase in root biomass compared to the cut grass (Table 3) and therefore, resulting in larger amounts of soil organic inputs. As stated by Boone (1994), Haider *et al*. (1993), Norby and Cortrufo (1998) and Sanchez *et al*., (2002) roots have a greater influence on soil organic carbon levels than compared to the aboveground plant biomass.

However, microbial life plays an important role on the amount of organic carbon within the soil. Nitrogen rich soil treatments such as manure and lucerne, supply readily available nutrients (Cooperband, 2002) which are decomposed by soil microbial actions a lot faster than treatments which are more resistant to decomposition, therefore, affecting the amount of soil organic carbon over time due to carbon release through soil respiration. The woodchips treatment is a lignin rich treatment which decomposes more slowly than the other soil treatments used in this trial, which together with the high production of root biomass, may have been the reason why an increase in soil organic carbon with all treatments which contain woodchips was seen.

The soil organic carbon measured after the *E. tef* harvest showed no significant difference (p<0.05) in the mean carbon percentage. From Table 5 it is evident that all the soil treatments have resulted in an increase in soil organic carbon after the *E. tef* harvest when compared to the control, yet, none show a significant difference (p<0.05) when compared to the control or when compared to one another. The largest increases in soil organic carbon content was as a result of the application of cut grass and manure treatment (T6) and the application of the woodchips treatment (T4) with similar mean increases of 23.14% and 21.49% respectively when compared to the control. Treatment T6 resulted in a similar amount of root biomass as treatment T4 largely due to the nutrients which came from the cattle manure (Table 2). This helped to increase the soil organic carbon inputs from the plant roots while the cut grass treatment helped to increase the carbon content of the soil itself due to the relatively high carbon content of the treatment (Table 2). One may be asking oneself why treatment T6 did not show the same success after the *A. sativa* harvest.

The possible reason for this may be due to the fact that treatment T6 did not result in a large amount of root biomass after the *A. sativa* harvest (Figure 3). Therefore, these increases in soil organic carbon can be attributed to the carbon content of the treatments themselves (Table 2) and the resultant root biomass from the application of these treatments (Figure 3 and 4). Furthermore, one must not forget that treatments T5-T9 are combination treatments which were applied in equal fractions making up 40 t ha^{-1} and thus should be kept in mind when comparing results between one another.

It is important to remember that the soil organic carbon cycle is a very dynamic system which fluctuates according to organic inputs and microbial actions (Figure 5) thus, emphasizing the importance in which the contribution of root biomass plays in the amount of organic carbon stored within the soil. The *A. sativa* pasture produced a larger amount of root biomass compared to the *E. tef* pasture (Figure 3 and 4). Therefore, when the soil organic carbon was measured from the control treatment (T10) after the *A. sativa* harvest, the soil contained a larger amount of roots and all associated carbon inputs such as root exudates. When the soil organic carbon was measured from the control treatment (T10) after the *E. tef* harvest the root biomass from the *A. sativa* harvest may have been digested by soil microbes and released as carbon dioxide resulting in less soil organic carbon. Furthermore, the root contribution of the *E. tef* pasture was a lot less than the *A. sativa* pasture and therefore, resulted in fewer soil carbon inputs from the associated root exudates and root decay which may explain why a decrease in soil organic carbon content after the *E. tef* harvest was seen for the control treatment (Table 6).

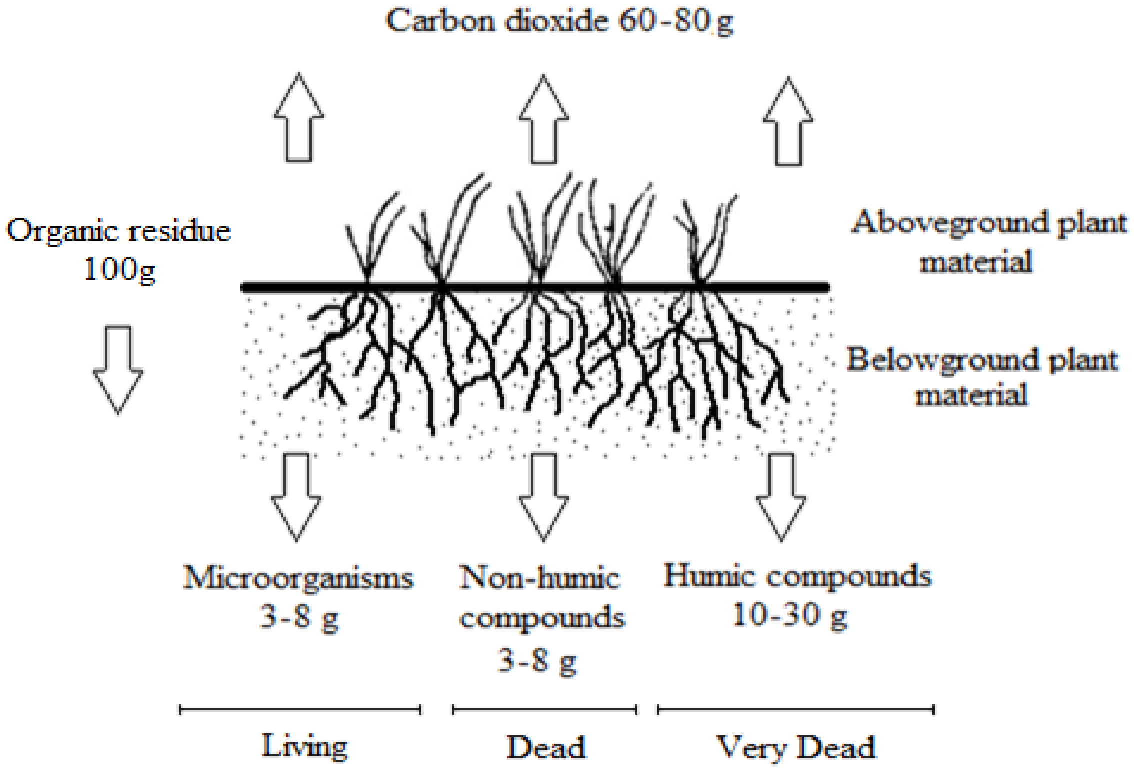

Figure 5 - Simplified illustration of the carbon fluxes from an addition of 100 grams of organic residue (Hoorman and Islam, 2010).

4.3.2 Soil organic carbon content (kg m^{-2} at depth of 25 cm)

The soil organic carbon content (kg m^{-2} at depth of 25 cm) was calculated using equation 2. The soil organic carbon measured in $kg.m^2$ after the *A. sativa* harvest showed no significant difference ($p<0.05$). From Table 4 it is evident that the woodchips treatment (T4) showed the largest increase in carbon sequestered within the soil with a mean increase of 35.56% when compared to the control. Once again all treatments which contained woodchips showed a mean increase when compared to the control (Table 4). This can again be explained by the contributions made by the resultant root biomass and carbon content of the soil treatment. Furthermore, the kg m^{-2} of soil organic carbon is largely influenced by the soil bulk density (Equation 2) therefore, soils incorporated with treatments which resulted in high bulk density will have a greater influence on the total kg m^{-2} of organic carbon. From appendix B it is evident that research bins amended with treatment T4 resulted in a high soil bulk density and thus, resulted in more kilograms of carbon stored per m^2. However, while one of the aims of this trial was to determine which treatment resulted in more kilograms of organic carbon stored per square metre, another aim was to determine which treatment showed the greatest improvement on soil properties and plant growth. Hence, it is important to note that while a higher bulk density can result in a larger amount of carbon stored within the soil (equation 2), it alternatively has a negative impact on resultant plant development and therefore, plant biomass. Furthermore, by lowering the bulk density, one creates a more favourable soil environment for plant development.

The soil organic carbon measured in kg m^{-2} after the *E. tef* harvest showed no significant difference ($p<0.05$) when compared to each other or compared to the control. From Table 5 it is evident that the largest increase in soil organic carbon stored per m^2 was measured from treatment T6. The cut grass and manure treatment (T6) showed a mean increase of 19.59% organic carbon stored when compared to the control. The high carbon content measured per square metre as a result of the application of treatment T6 is mainly due to the large root biomass (Figure 4) and the high bulk density (appendix B) measured after the *E. tef* harvest. The cut grass and manure treatment had the second largest bulk density and thus, largely influencing the soil carbon content measured per square metre.

4.3.3 Total organic carbon sequestered (kg m^{-2} at depth 25cm)

The total organic carbon sequestered was calculated using equation (3) in which the soil organic carbon content (kg m^{-2}) was added to the aboveground biomass carbon content (kg) and the root biomass carbon content (kg). The total carbon dioxide sequestered (kg m^{-2}) was calculated using equation (4) in which the total organic carbon sequestered (kg m^{-2}) was multiplied by 3.667 (Chan, 2008b). Both the total organic carbon sequestered and the total carbon dioxide sequestered showed no significant difference ($p<0.05$) after both the *A. sativa* harvest and the *E. tef* harvest.

Table 4 shows that the woodchips treatment (T4) had the largest increase in total organic carbon sequestered with an increase of 35.01% when compared to the control. Additionally, from Table 4 it is evident that treatments T1, T2, and T5 showed decreases in the total organic carbon sequestered when compared to the control after the *A. sativa* harvest. It is important to note that the amount of organic carbon sequestered within the soil (kg m^{-2}) forms the majority of the total amount of organic carbon sequestered and in the case of treatment T4, 96.13% of the total organic carbon was sequestered within the soil leaving only 3.87% sequestered within the aboveground and belowground biomass. The treatment which showed the greatest amount of organic carbon sequestered in the aboveground and root biomass was the lucerne, manure and woodchips treatment (T9) with an increase of 122.73% when compared to the control and an increase of 75% when compared to treatment T4 (Table 4). However, treatment T9 showed a decrease of 16.62% in total organic carbon sequestered when compared to treatment T4 due to treatment T4 having a higher bulk density and higher soil organic carbon content.

The total organic carbon sequestered after the *E. tef* harvest showed no significant difference (p<0.05) as a result of the application of the different soil treatments. From Table 5 it is evident that the woodchips treatment (T4) and the cut grass and woodchips treatment (T6) resulted in the same increase in total organic carbon sequestered when compared to the control with an increase of 18.18% for both treatments. In contrast to the results of the total organic carbon sequestered after the *A. sativa* harvest, the total organic carbon sequestered after the *E. tef* harvest all showed distinct increases in the mean values when compared to the control, except for treatment T5. Once again the lucerne, manure and woodchips treatment (T9) showed the greatest increase in organic carbon sequestered within the aboveground and root biomass with an increase 48.10% when compared to the control and a 10.38% increase when compared to treatment T4 and T6. From Table 6 it is evident that increases were measured in the mean total value for the organic carbon sequestered in the aboveground and root biomass for all the treatments measured after the *E. tef* harvest. These increase are largely attributed to the aboveground biomass produced from the *E. tef* species which was significantly greater in comparison to the amount produced from *A. sativa* species and thus, resulted in a greater amount of organic carbon sequestered in the total plant biomass (Table 5). While the majority of the organic carbon is stored within the soil, the aboveground and root biomass plays a significant part in the total organic carbon sequestered. Therefore, treatments which improve soil properties such as pH, bulk density and plant nutrient supply tend to increase the organic carbon sequestered as they significantly influence the amount of biomass produced.

Table 4: Resultant organic carbon sequestered 4 months after soil treatment.

	Soil organic carbon content (%)	**Soil organic matter content (%)**	**Soil organic carbon content at depth 25 cm (KgC m^{-2})**	**Organic carbon sequestered in aboveground and root biomass (KgC m^{-2})**	**Total organic carbon sequestered (KgC m^{-2})**	**Total carbon dioxide sequestered (kgCO^2 m^{-2})**
Fly ash + manure (T1)	1.14 (±0.52)	1.96 (±0.45)	4.22 (±0.99)	0.39 (±0.05) ac	4.61 (±0.98)	16.91 (±3.61)
Manure (T2)	1.34 (±0.43)	2.30 (±0.37)	4.83 (±0.83)	0.30 (±0.05) abc	5.14 (±0.80)	18.83 (±2.95)
Fly ash + woodchips (T3)	1.52 (±0.61)	2.61 (±0.53)	5.66 (±1.15)	0.31 (±0.04) abc	5.97 (±1.17)	21.89 (±4.29)
Woodchips (T4)	1.79 (±0.19)	3.09 (±0.17)	6.71 (±0.39)	0.28 (±0.06) abc	6.98 (±0.35)	25.60 (±1.29)
Cut grass (T5)	1.33(±0.20)	2.29 (±0.17)	4.94 (±0.41)	0.14 (±0.04) b	5.08 (±0.39)	18.63 (±1.43)
Cut grass + manure (T6)	1.37 (±0.33)	2.36 (±0.29)	5.12 (±0.62)	0.20 (±0.04) ab	5.32 (±0.66)	19.52 (±2.43)
Cut grass + woodchips (T7)	1.62 (±0.30)	2.79 (±0.26)	5.92 (±0.52)	0.34 (±0.01) ac	6.26 (±0.53)	22.94 (±1.95)
Lucerne (T8)	1.37 (±0.43)	2.35 (±0.37)	4.98 (±0.83)	0.46 (±0.11) ac	5.44 (±0.92)	19.94 (±3.36)
Lucerne + manure + woodchips (T9)	1.44(±0.25)	2.48 (±0.22)	5.34 (±0.48)	0.49 (±0.03) c	5.82 (±0.47)	21.35 (±1.72)
Soil only control (T10)	1.34 (±0.92)	2.31 (±0.79)	4.95 (±1.70)	0.22 (±0.02) abc	5.17 (±1.71)	18.96 (±6.26)

Means within columns with the same letter do not differ significantly ($p>0.05$), columns with not lettering have no significant difference ($p>0.05$) in mean value.

Table 5: Resultant organic carbon sequestered 12 months after soil treatment.

	Soil organic carbon content (%)	**Soil organic matter content (%)**	**Soil organic carbon content at depth 25 cm (KgC m^{-2})**	**Organic carbon sequestered in aboveground and root biomass (KgC m^{-2})**	**Total organic carbon sequestered (KgC m^{-2})**	**Total carbon dioxide sequestered (kgCO^2 m^{-2})**
Fly ash + manure (T1)	1.43 (±0.19)	2.45 (±0.33)	5.07 (±0.71)	0.97 (±0.08)	6.05 (±0.67)	22.17 (±2.45)
Manure (T2)	1.35 (±0.09)	2.32 (±0.16)	4.78 (±0.37)	0.89 (±0.05)	5.68 (±0.34)	20.82 (±1.26)
Fly ash + woodchips (T3)	1.41 (±0.11)	2.43 (±0.19)	4.98 (±0.33)	0.88 (±0.09)	5.86 (±0.39)	21.48 (±1.47)
Woodchips (T4)	1.47 (±0.08)	2.52 (±0.13)	5.19 (±0.29)	1.06 (±0.08)	6.24 (±0.35)	22.87 (±1.29)
Cut grass (T5)	1.23 (±0.70)	2.12 (±0.12)	4.34 (±0.20)	0.92 (±0.08)	5.26 (±0.26)	19.29 (±0.94)
Cut grass + manure (T6)	1.49 (±0.15)	2.56 (±0.25)	5.37 (±0.49)	0.87 (±0.13)	6.24 (±0.48)	22.87 (±1.74)
Cut grass + woodchips (T7)	1.46 (±0.12)	2.52 (±0.20)	5.19 (±0.45)	0.96 (±0.05)	6.15 (±0.48)	22.56 (±1.73)
Lucerne (T8)	1.38 (±0.03)	2.38 (±0.05)	4.81 (±0.16)	0.95 (±0.11)	5.76 (±0.18)	21.13 (±0.69)
Lucerne + manure + woodchips (T9)	1.42 (±0.04)	2.45 (±0.06)	5.01 (±0.18)	1.17 (±0.13)	6.18 (±0.22)	22.68 (±0.80)
Soil only control (T10)	1.21 (±0.07)	2.09 (±0.06)	4.49 (±0.27)	0.79 (±0.02)	5.28 (±0.27)	19.37 (±0.96)

Means within columns show no significant difference ($p>0.05$).

Table 6: Change in organic carbon sequestered measured from the winter (*A. sativa*) to the summer (*E. tef*) harvest.

	Soil organic carbon content (%)	**Soil organic matter content (%)**	**Soil organic carbon content at depth 25 cm (KgC m^{-2})**	**Organic carbon sequestered in aboveground and root biomass (KgC m^{-2})**	**Total organic carbon sequestered (KgC m^{-2})**	**Total carbon dioxide sequestered (kgCO^2 m^{-2})**
Fly ash + manure (T1)	↑0.29	↑0.49	↑0.85	↑0.58	↑1.44	↑5.26
Manure (T2)	↑0.01	↑0.02	↓0.05	↑0.59	↑0.54	↑1.99
Fly ash + woodchips (T3)	↓0.11	↓0.18	↓0.68	↑0.57	↓0.11	↓0.31
Woodchips (T4)	↓0.32	↓0.57	↓1.52	↑0.78	↓0.74	↓2.73
Cut grass (T5)	↓0.10	↓0.17	↓0.60	↑0.78	↑0.18	↑0.66
Cut grass + manure (T6)	↑0.12	↑0.20	↑0.25	↑0.67	↑0.92	↑3.35
Cut grass + woodchips (T7)	↓0.16	↓0.27	↓0.73	↑0.62	↓0.11	↓0.38
Lucerne (T8)	↑0.01	↑0.03	↓0.17	↑0.49	↑0.32	↑1.19
Lucerne + manure + woodchips (T9)	↓0.02	↓0.03	↓0.33	↑0.68	↑0.36	↑1.33
Soil only control (T10)	↓0.13	↓0.22	↓0.46	↑0.57	↑0.11	↑0.41

5 Conclusion

Carbon sequestration on rehabilitated surface coal mines in South Africa is not a well understood process and the methods used to extract coal result in soils with low organic carbon content. The results from this trial illustrate that the use of organic treatments can improve both soil conditions and the total organic carbon sequestered. In this trial the greatest increase in total organic carbon compared to the control was measured from the soil which was applied with the woodchips treatment (T4). However, when considering an organic treatment which increases the total soil organic content and greatly improved the soil properties, one must consider the lucerne, manure and woodchips treatment (T9). This treatment showed a slight decrease in total organic carbon of 0.06 kg m^{-2} when compared to the woodchips treatment (T4) after the *E. tef* harvest but showed a substantial increase in soil nutrients and plant biomass. This illustrates that the lucerne, manure and woodchips treatment (T9) resulted in better soil conditions for plant growth and, therefore, when considering a soil ameliorant which both improves soil organic carbon content and soil properties for plant growth, the lucerne, manure and woodchips treatment best fitted the task in this trial.

However, one cannot neglect the carbon lost through soil and plant respiration when determining the effective carbon sequestered. Cook and Lloyd, (2012) calculated soil and plant respiration from a one year old rehabilitated and grassed area occurring on same mine from which the topsoil, subsoil and spoil came from in this trial to be 0.38 kgCO^2/m^2/annum. Subtracting this value from the resultant carbon dioxide sequestered after the lucerne, manure and woodchips (T9) application showed an increase of 3.31 kgCO^2/m^2/annum when compared to the control. Extrapolated to per hectare, this value equates to 33.1 tCO^2/ha/annum stored more than the control. This shows that the incorporation of organic matter and the use of a winter cover crop can substantially increase the amount of carbon sequestered within the soil after just one year while simultaneously improving the soil properties and therefore, also illustrating its value as a soil ameliorant in rehabilitation.

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For clarity reasons tables do not follow the format guidelines for the journal of The Southern African Institute of Mining and Metallurgy

CHAPTER 4

GENERAL CONCLUSIONS AND RECOMMENDATIONS

Re-establishment of vegetation for the purpose of rehabilitation and reclamation on coal mines have for many years presented a challenging and complicated task. Rehabilitation has thus, necessitated the use of soil ameliorants to improve soil properties and aid in the process of proper vegetation development. The value of incorporating organic matter directly into the soil in the form of a soil ameliorant together with the use of a winter cover crop to improve overall soil carbon storage and soil fertility has yet to be properly investigated as a rehabilitative option on mine sites in South Africa. In this investigation, conclusions were based on the restorative values in which the different soil amendments impacted the physical and chemical properties of the soil for improved plant development and improved carbon storage. The results from these trials indicate that the use of these soil ameliorants as a sole treatment or as a combine treatment can have and significant improvements on the physical and chemical properties of the cover soil while at the same time improving soil carbon storage.

It is evident form the results that the lucerne, manure and woodchips combination treatment (T9) was the best overall treatment used in this trial. It was the only treatment to have a significant difference ($p<0.05$) on the soil pH, soil bulk density, aboveground biomass and root biomass. This improvement to both the physical and chemical properties of the soil can be attributed to the value in which each individual component within the combination treatment complemented one another in terms of addressing different soil deficiencies. With regards to the pH, the alkaline nature of the manure within the combination treatment helped to increase the pH to a level in which plant development is more favorable for both plant species used in this trial. Additionally, manure is rich in phosphorus, potassium, calcium and nitrogen and is rapidly decomposed and therefore, quickly improves the soil nutrient status which has an immediate impact of plant development. Evidence of this increase in plant nutrients could be seen when looking at the soil analysis after both plant harvests which revealed large increases in macro nutrients when compared to the control. However, because manure is readily decomposed, the nutrient release is short lived and, therefore, will only improve the soil nutrient status over the first few years. The composted woodchips on the other hand is recalcitrant and lignin-rich, therefore, requiring more time in order to be properly decomposed. Evidence of this was seen after the *Eragrotis tef* (*E. tef*) harvest when the chemical soil analysis illustrated increases in the level of potassium concentration in all soils amended with woodchips. This however, was not the case when examining the soil analysis after *Avena sativa* (*A. sativa*) harvest which was the first harvest four months after soil treatment. This illustrates that much more time was needed for the woodchips to be broken down in order to release the potassium. Furthermore, because there exists high potential for soil organic carbon improvement due to the low inherent carbon content of soil impacted by surface coal mining, larges increase in soil organic carbon are achievable if the proper soil carbon management tools are used. In this study, all treatments measured after the *E. tef* harvest illustrated a mean increase in total organic carbon stored when compared to the control, except for the cut grass treatment (T5). Furthermore, the analysis of the soil as a result of the application of the woodchips treatment (T4) revealed the greatest mean increase in total organic carbon of 18.18% when compared to the control. However, as mentioned before, nitrogen rich soil treatments such as manure and lucerne supply readily available nutrients which are decomposed by soil microbial actions much faster than treatments which are more resistant to decomposition such as the woodchips treatment and therefore, affect the amount of soil organic carbon present over time due to carbon release through soil respiration thus, emphasizing the value of having both a fast (nitrogen rich) and a slow (lignin rich) nutrient releasing soil ameliorant for promoting carbon storage and plant development on a long term basis.

The lucerne treatment (T8) had the most profound impact on the bulk density of the soil measured after the *E. tef* harvest with a mean decrease of 5.87% compared to the control. This can be attributed to the fact that the lucerne was in the form of a chopped hay ameliorant which helped to aerate the soil by physically pushing the soil particles away from one another and at the same time supplying abundant levels of nitrogen for root development. Furthermore, nitrogen is one of the most important soil nutrients needed for plant growth and is instrumental in promoting aboveground and belowground plant growth which was evident from the resultant aboveground and belowground plant yields from the application of lucerne treatment. The lucerne treatment alone illustrated the second greatest mean increase in aboveground yield of 110.64% when compared to the control after the *A. sativa* harvest and a mean increase in root yield of 191.21% when compared to the control after the *E. tef* harvest. The increase in root growth of the pasture species increased the amount of pores spaces occupied by growing roots. As the roots continue to develop they radially expand and thereby, increase the total pore space through eventual root decay and ultimately decreasing bulk density. Therefore, by linking the advantages in which each soil ameliorant offers into a combination treatment, one is able to greatly increase the soil organic carbon content while simultaneously having a substantial improvement to the chemical and physical properties of the soil, all of which has as positive impact on vegetation development and thus, the functionality of the ecosystem. Furthermore, improved ecosystem functionality increases the amount of carbon dioxide sequestered and ultimately has a positive role towards decreasing greenhouse gas emissions.

Recommendations

The results from this trial are only indicative of the affects in which the different soil ameliorants had on the soil properties and plant growth following one winter and summer period. Therefore, a long-term study into the soil properties of the cover soil is needed to evaluate the long term effects in which theses soil ameliorants have on soil pH, soil bulk density, aboveground biomass, root biomass, soil fertility and soil carbon storage. Long term data will help determine the effective longevity of each soil ameliorant. Furthermore, evaluating the amount of carbon captured and the overall carbon cycle over a longer term will provide more reliable data as carbon fluxes are highly variable. One can go even further and determine the stability of the organic carbon in order to determine the permanence of the captured carbon which can be used to reclaim carbon credits. Lastly, this experiment was conducted on a confined rehabilitated soil profile which was reconstructed on the Hatfield experimental farm. Evaluating these ameliorants on a replaced and profiled soil on the areas of a coal mine which needs to undergo rehabilitation could provide more reliable data which can be used in a model and extrapolated for use on other surface coal mines.

APPENDIX

A: Soil texture images.

Figure A1 - Soil texture after soil was thoroughly mixed with fly ash (top) and cut grass (bottom).

B. Summary of the statistical analysis tables chapter 2

Table B1: Summary of ANOVA table on the soil pH after the *A. sativa* harvest.

ANOVA

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	2.564	9	.285	4.311	.001
Within Groups	1.982	30	.066		
Total	4.546	39			

Table B1.1: Tukey's post hoc comparison on the mean soil pH measured after the *A. sativa* harvest.

pH

Tukey HSD[a]

Treatment	N	Subset for alpha = 0.05	
		1	2
Soil only (control) (T10)	4	5.0350	
Cut grass (T5)	4	5.0800	
Woodchips (T4)	4	5.3300	5.3300
Manure (T2)	4	5.4350	5.4350
Lucerne (T8)	4	5.4450	5.4450
Cut grass + woodchips (T7)	4	5.4500	5.4500
Fly ash + woodchips (T3)	4	5.4875	5.4875
Lucerne + manure + woodchips (T9)	4	5.5050	5.5050
Cut grass + manure (T6)	4		5.7250
Fly ash + manure (T1)	4		5.9375
Sig.		.267	.059

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 4.000.

Table B2: Summary of ANOVA table on the soil pH after the *E. tef* harvest.

ANOVA

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	2.710	9	.301	10.895	.000
Within Groups	.829	30	.028		
Total	3.540	39			

Table B2.1: Tukey's post hoc comparison on the mean pH measured after the *E. tef* harvest.

pH

Tukey HSD[a]

Treatment	N	Subset for alpha = 0.05			
		1	2	3	4
Cut grass (T5)	4	4.9100			
Soil only (control) (T10)	4	4.9150			
Cut grass + woodchips (T7)	4	5.1350	5.1350		
Lucerne (T8)	4	5.1700	5.1700		
Cut grass + manure (T6)	4	5.2125	5.2125	5.2125	
Woodchips (T4)	4		5.3575	5.3575	5.3575
Lucerne + manure + woodchips (T9)	4		5.3700	5.3700	5.3700
Fly ash + manure (T1)	4		5.5200	5.5200	5.5200
Manure (T2)	4			5.5875	5.5875
Fly ash + woodchips (T3)	4				5.7325
Sig.		.273	.068	.082	.082

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 4.000.

Table B3: Summary of ANOVA table on the soil bulk density measured after the *A. sativa* harvest.

ANOVA

Fishers Least Significant Difference

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	.013	9	.001	1.388	.237
Within Groups	.031	30	.001		
Total	.045	39			

Table B4: Summary of ANOVA table on the soil bulk density measured after the *E. tef* harvest.

ANOVA

Fishers Least Significant Difference

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	.020	9	.002	1.853	.099
Within Groups	.036	30	.001		
Total	.056	39			

Table B5: Summary of ANOVA table on the aboveground biomass of *A. sativa*.

ANOVA

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	1226636.033	9	136292.893	4.586	.001
Within Groups	891652.035	30	29721.734		
Total	2118288.067	39			

Table B5.1: Tukey's post hoc comparison on the mean aboveground biomass of *A. sativa*.

Biomass

Tukey HSD[a]

Treatment	N	Subset for alpha = 0.05		
		1	2	3
Cut grass (T5)	4	219.9444		
Cut grass + manure (T6)	4	333.1111	333.1111	
Soil only (control) (T10)	4	353.0277	353.0277	
Woodchips (T4)	4	452.3611	452.3611	452.3611
Manure(T2)	4	506.8888	506.8888	506.8888
Fly ash + woodchips (T3)	4	515.2222	515.2222	515.2222
Cut grass + woodchips (T7)	4	559.3611	559.3611	559.3611
Fly ash + manure (T1)	4		649.0555	649.0555
Lucerne(T8)	4		743.5555	743.5555
Lucerne + manure + woodchips (T9)	4			805.0555
Sig.		.187	.055	.152

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 4.000.

Table B6: Summary of ANOVA table on the root biomass of *A. sativa*.

ANOVA

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	81065.892	9	9007.321	4.587	.001
Within Groups	58912.889	30	1963.763		
Total	139978.782	39			

Table B6.1: Tukey's post hoc comparison on the mean root biomass of *A. sativa*.

Biomass

Tukey HSD[a]

Treatment	N	Subset for alpha = 0.05		
		1	2	3
Cut grass (T5)	4	56.5278		
Cut grass + manure (T6)	4	85.6111	85.6111	
Soil only (control) (T10)	4	90.7500	90.7500	
Woodchips (T4)	4	116.2778	116.2778	116.2778
Manure (T2)	4	130.3333	130.3333	130.3333
Fly ash + woodchips (T3)	4	132.4444	132.4444	132.4444
Cut grass + woodchips(T7)	4	143.8055	143.8055	143.8055
Fly ash + manure (T1)	4		166.8333	166.8333
Lucerne (T8)	4		191.1389	191.1389
Lucerne + manure + woodchips (T9)	4			206.9444
Sig.		.187	.055	.152

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 4.000.

Table B7: Summary of ANOVA table on the aboveground biomass of *E. tef*.

ANOVA

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	1.817	9	.202	1.329	.264
Within Groups	4.558	30	.152		
Total	6.374	39			

Table B8: Summary of ANOVA table on the root biomass of *E. tef*.

ANOVA

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	7.897	9	.877	5.052	.000
Within Groups	5.210	30	.174		
Total	13.106	39			

Table B8.1: Tukey's post hoc comparison on the mean root biomass of *E. tef* (log transformed).

Biomass

Tukey HSD[a]

Treatment	N	Subset for alpha = 0.05		
		1	2	3
Soil only (control) (T10)	4	3.2849		
Cut grass (T5)	4	3.6012	3.6012	
Manure (T2)	4	4.0224	4.0224	4.0224
Cut grass + manure (T6)	4	4.2076	4.2076	4.2076
Cut grass + woodchips (T7)	4	4.2417	4.2417	4.2417
Woodchips (T4)	4		4.2958	4.2958
Lucerne (T8)	4		4.3881	4.3881
Fly ash + manure (T1)	4		4.4097	4.4097
Fly ash + woochips (T3)	4		4.4987	4.4987
Lucerne + manure + woochips (T10)	4			4.9536
Sig.		.072	.111	.088

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 4.000.

Table B9: Kruskal Wallis test on the soil macro nutrients after the *A. sativa* harvest.

Test Statistics

	PBray	K	Na	Ca	Mg
Chi-Square	33.653	31.184	23.400	22.383	26.867
df	9	9	9	9	9
Asymp. Sig.	.000	.000	.005	.008	.001

Table B10: Kruskal Wallis test on the soil macro nutrients after the *E. tef* harvest.

Test Statistics

	PBray	K	Na	Ca	Mg
Chi-Square	30.912	29.242	7.413	23.107	20.050
df	9	9	9	9	9
Asymp. Sig.	.000	.001	.594	.006	.018

C. Carbon content (%) of aboveground and root biomass.

Table C1: Mean carbon content (%) of the aboveground and root carbon biomass of *A. sativa* and *E. tef*.

	A. sativa		***E. tef***	
	Mean aboveground carbon content (%)	Mean root carbon content (%)	Mean aboveground carbon content (%)	Mean root carbon content (%)
Fly ash + manure (T1)	51.91 (±0.58)	30.66 (±1.29)	43.71 (±0.51)	37.32 (±3.35)
Manure (T2)	52.29 (±0.46)	29.80 (±2.19)	43.38 (±0.11)	38.22 (±0.49)
Fly ash + woodchips (T3)	52.36 (±0.28)	28.42 (±4.06)	43.65 (±0.36)	39.61 (±0.49)
Woodchips (T4)	52.74 (±0.47)	31.84 (±1.03)	43.80 (±0.31)	37.80 (±0.88)
Cut grass (T5)	52.81 (±0.66)	31.54 (±2.30)	43.95 (±0.44)	40.39 (±0.39)
Cut grass + manure (T6)	52.50 (±0.36)	29.39 (±1.07)	43.73 (±0.46)	39.66 (±0.83)
Cut grass + woodchips (T7)	53.35 (±0.29)	28.64 (±1.61)	44.46 (±0.18)	40.39 (±0.98)
Lucerne (T8)	52.69 (±0.59)	32.57 (±1.03)	44.24 (±0.45)	36.59 (±1.63)
Lucerne + manure + woodchips (T9)	52.62 (±0.16)	29.56 (±3.23)	43.37 (±0.47)	37.44 (±2.34)
Soil only control (T10)	53.83 (±0.18)	31.57 (±1.22)	44.08 (±0.35)	41.19 (±1.29)
Mean total	52.81 (±0.15)	30.39 (±0.63)	43.84 (±0.12)	38.86 (±0.49)

D. Soil bulk density, soil pH and soil nutrients

Table D1: Soil bulk density, soil pH and soil macro nutrients recorded after *A. sativa* harvest with the general recommended soil nutrient amounts represented in square brackets according to The Fertilizer Society of South Africa Handbook (2007). The standard error of the mean is given in round brackets.

Soil Treatments	P Bray 1	Ammonium acetate extraction					KCL
	Mean P [15-30]	Mean K [80-160]	Mean Ca [300-2000]	Mean Mg [80-300]	Mean Na	Mean Bulk density	Mean pH
	mg.kg^{-1}					g.cm³	
Fly ash + manure (T1)	64 (±13.32)	283 (±66.29)	567 (±52.99)	143 (±16.56)	31 (±6.97)	1.48 (±0.02)	5.94 (±0.12)
Manure (T2)	47 (±12.06)	214 (±60.50)	430 (±37.88)	113 (±16.01)	26 (±6.22)	1.44 (±0.02)	5.44 (±0.06)
Fly ash + woodchips (T3)	21 (±3.47)	87 (±22.24)	458 (±45.85)	85 (±12.56)	17 (±2.96)	1.49 (±0.01)	5.49 (±0.12)
Woodchips (T4)	27 (±4.87)	97 (±9.84)	411 (±7.32)	92 (±4.53)	16 (±1.70)	1.49 (±0.01)	5.33 (±0.05)
Cut grass (T5)	11 (±0.41)	79 (±6.73)	358 (±15.42)	76 (±4.77)	13 (±1.75)	1.48 (±0.01)	5.08 (±0.09)
Cut grass + manure (T6)	54 (±13.66)	273 (±104.81)	424 (±46.44)	117 (±19.55)	27 (±9.14)	1.49 (±0.02)	5.73 (±0.15)
Cut grass + woodchips (T7)	19 (±2.21)	82 (±6.09)	348 (±17.79)	74 (±4.78)	12 (±0.48)	1.46 (±0.02)	5.45 (±0.25)
Lucerne (T8)	18 (±2.49)	131 (±12.06)	414 (±14.20)	87 (±3.69)	14 (±1.03)	1.45 (±0.02)	5.45 (±0.17)
Lucerne + manure + woodchips (T9)	36 (±2.36)	154 (±10.20)	418 (±13.35)	103 (±3.18)	19 (±0.65)	1.48 (±0.01)	5.51 (±0.09)
Soil only control (T10)	13 (±0.48)	48 (±1.66)	339 (±15.19)	66 (±3.14)	11 (±0.41)	1.49 (±0.02)	5.04 (±0.04)

Table D2: Soil bulk density, soil pH and soil macro nutrients recorded after *E. tef* harvest with the general recommended soil nutrient amounts represented in square brackets according to The Fertilizer Society of South Africa Handbook (2007). The standard error of the mean is given in round brackets.

Soil Treatments	P Bray 1	Ammonium acetate extraction				KCL
	Mean P [15-30]	Mean K [80-160]	Mean Ca [300-2000]	Mean Mg [80-300]	Mean Na	Mean Bulk density	Mean pH
	mg.kg^{-1}					g.cm^{3}	
Fly ash + manure (T1)	46 (±12.38)	198 (±17.03)	475 (±52.87)	108 (±14.27)	15 (±2.59)	1.42 (±0.02)	5.63 (±0.10)
Manure (T2)	37 (±12.31)	206 (±22.33)	352 (±14.91)	92 (±11.21)	12 (±0.95)	1.42 (±0.02)	5.45 (±0.15)
Fly ash + woodchips (T3)	22 (±2.68)	123 (±21.62)	496 (±35.29)	91 (±9.69)	14 (±1.55)	1.42 (±0.02)	5.70 (±0.04)
Woodchips (T4)	13 (±1.35)	117 (±10.35)	353 (±30.66)	78 (±6.96)	13 (±1.04)	1.42 (±0.01)	5.41 (±0.07)
Cut grass (T5)	8 (±0.25)	79 (±10.14)	336 (±14.09)	68 (±4.19)	13 (±0.71)	1.41 (±0.01)	4.89 (±0.02)
Cut grass + manure (T6)	32 (±3.63)	222 (±33.13)	389 (±18.21)	101 (±5.02)	13 (±0.75)	1.44 (±0.02)	5.30 (±0.05)
Cut grass + woodchips (T7)	16 (±5.55)	120 (±23.64)	353 (±38.12)	81 (±14.03)	13 (±1.08)	1.42 (±0.02)	5.06 (±0.16)
Lucerne (T8)	11 (±0.82)	135 (±13.79)	369 (±8.59)	73 (±3.11)	14 (±0.41)	1.39 (±0.02)	5.20 (±0.04)
Lucerne + manure + woodchips (T9)	20 (±1.65)	171 (±10.59)	428 (±14.13)	98 (±4.73)	15 (±0.95)	1.41 (±0.02)	5.39 (±0.03)
Soil only control (T10)	9 (±0.50)	66 (±5.02)	322 (±10.26)	63 (±3.64)	12 (±1.79)	1.48 (±0.01)	4.88 (±0.08)

E. Summaries of the statistical analysis tables chapter 3

Table E1: Summary of ANOVA table on the aboveground carbon content (g) of *A. sativa*.

ANOVA

Tukey

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	336843.911	9	37427.101	4.435	.001
Within Groups	253191.285	30	8439.710		
Total	590035.196	39			

Table E1.1: Tukey's post hoc comparison on the mean aboveground carbon content (g) of *A. sativa*.

Carbon

Tukey HSD[a]

		Subset for alpha = 0.05		
Treatment	N	1	2	3
Cut grass (T5)	4	116.5854		
Cut grass + manure (T6)	4	175.3660	175.3660	
Soil only (control) (T10)	4	190.0824	190.0824	
Woodchips (T4)	4	237.8204	237.8204	237.8204
Manure (T2)	4	265.8424	265.8424	265.8424
Fly ash + woodchips (T3)	4	274.4664	274.4664	274.4664
Cut grass + woodchips (T7)	4	298.4945	298.4945	298.4945
Fly ash + manure (T1)	4	336.4557	336.4557	336.4557
Lucerne (T8)	4		394.3836	394.3836
Lucerne + manure + woodchips (T9)	4			423.5487
Sig.		.053	.055	.162

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 4.000.

Table E2: Summary of ANOVA table on the root carbon content (g) of *A. sativa*.

ANOVA

Tukey

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	5.916	9	.657	3.639	.004
Within Groups	5.419	30	.181		
Total	11.336	39			

Table E2.2: Tukey's post hoc comparison table on the mean root carbon content (g) of *A. sativa* (log transformed).

Carbon

Tukey HSD[a]

		Subset for alpha = 0.05	
Treatment	N	1	2
Cut grass (T5)	4	2.7834	
Cut grass + manure (T6)	4	3.1107	3.1107
Soil only (control) (T10)	4	3.3348	3.3348
Woodchips (T4)	4	3.5039	3.5039
Fly ash + woodchips (T3)	4	3.5736	3.5736
Manure (T2)	4	3.6064	3.6064
Cut grass + woodchips (T7)	4	3.7103	3.7103
Fly ash + manure (T1)	4		3.9110
Lucerne (T8)	4		3.9983
Lucerne + manure + woodchips (T9)	4		4.0892
Sig.		.103	.071

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 4.000.

Table E3: Summary of ANOVA table on the aboveground carbon content (g) of *E. tef*.

ANOVA

Tukey

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	.326	9	.036	1.217	.321
Within Groups	.893	30	.030		
Total	1.219	39			

Table E4: Summary of ANOVA table on the root carbon content (g) of *E. tef*.

ANOVA

Tukey

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	6.974	9	.775	3.658	.003
Within Groups	6.356	30	.212		
Total	13.330	39			

Table E4.1 Tukey's post hoc comparison table on the mean root carbon content (g) of *E. tef* (log transformed).

Carbon

Tukey HSD[a]

Treatment	N	Subset for alpha = 0.05		
		1	2	3
Soil only (control) (T10)	4	2.3964		
Cut grass (T5)	4	2.6944	2.6944	
Manure (T2)	4	3.0603	3.0603	3.0603
Cut grass + manure (T6)	4	3.2820	3.2820	3.2820
Woodchips (T4)	4	3.3221	3.3221	3.3221
Cut grass + woodchips (T7)	4	3.3344	3.3344	3.3344
Lucerne (T8)	4	3.3796	3.3796	3.3796
Fly ash + manure (T1)	4	3.4105	3.4105	3.4105
Fly ash + woodchips (T3)	4		3.5724	3.5724
Lucerne + manure + woodchips (T9)	4			3.9650
Sig.		.096	.220	.189

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 4.000.

Table E5: Summary of ANOVA table on the soil organic carbon content (%) after the *A. sativa* harvest.

ANOVA

Tukey

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	1.184	9	.132	.594	.792
Within Groups	6.645	30	.222		
Total	7.829	39			

Table E6: Summary of ANOVA table on the soil organic matter content (%) after the *A. sativa* harvest.

ANOVA

Tukey

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	3.521	9	.391	.596	.790
Within Groups	19.694	30	.656		
Total	23.215	39			

Table E7: Summary of ANOVA table on the soil organic carbon content (kg.m²) after the *A. sativa* harvest.

ANOVA

Tukey

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	16.939	9	1.882	.604	.783
Within Groups	93.439	30	3.115		
Total	110.378	39			

Table E8: Summary of ANOVA table on the organic carbon sequestered within the aboveground and root biomass (kg.m²) of *A. sativa.*

ANOVA

Tukey

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	6.011	9	.668	4.303	.001
Within Groups	4.657	30	.155		
Total	10.668	39			

Table E8.1: Tukey's post hoc comparison table on the mean aboveground and root carbon content (g) of *E. tef* (log transformed).

Carbon

Tukey HSD[a]

Treatment	N	Subset for alpha = 0.05		
		1	2	3
Cut grass (T5)	4	4.8081		
Cut grass + manure (T6)	4	5.2146	5.2146	
Soil only (control) (T10)	4	5.3696	5.3696	5.3696
Woodchips (T4)	4	5.5126	5.5126	5.5126
Manure (T2)	4	5.6718	5.6718	5.6718
Fly ash + woodchips (T3)	4	5.7238	5.7238	5.7238
Cut grass + woodchips (T7)	4		5.8249	5.8249
Fly ash + manure (T1)	4		5.9397	5.9397
Lucerne (T8)	4		5.9864	5.9864
Lucerne + manure + woodchips (T9)	4			6.1797
Sig.		.066	.192	.147

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 4.000.

Table E9: Summary of ANOVA table on the total organic matter content (kg.m²) after the *A. sativa* harvest.

ANOVA

Tukey

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	17.084	9	1.898	.597	.789
Within Groups	95.421	30	3.181		
Total	112.505	39			

Table E10: Summary of ANOVA table on the total carbon dioxide sequestered (kg.m²) after the *A. sativa* harvest.

ANOVA

Tukey

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	229.731	9	25.526	.597	.789
Within Groups	1283.114	30	42.770		
Total	1512.845	39			

Table E11: Summary of ANOVA table on the soil organic carbon content (%) after the *E. tef* harvest.

ANOVA

Tukey

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	.325	9	.036	.814	.608
Within Groups	1.331	30	.044		
Total	1.655	39			

Table E12: Summary of ANOVA table on the soil organic matter content (%) after the *E. tef* harvest.

ANOVA

Tukey

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	.949	9	.105	.797	.621
Within Groups	3.966	30	.132		
Total	4.915	39			

Table E13: Summary of ANOVA table on the soil organic carbon content (kg.m²) after the *E. tef* harvest.

ANOVA

Tukey

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	.148	9	.016	.743	.668
Within Groups	.664	30	.022		
Total	.812	39			

Table E14: Summary of ANOVA table on the organic carbon sequestered within the aboveground and root biomass (kg.m²) of *E. tef.*

ANOVA

Tukey

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	.409	9	.045	1.304	.276
Within Groups	1.045	30	.035		
Total	1.454	39			

Table E15: Summary of ANOVA table on the total organic matter content (kg.m²) after the *E. tef* harvest.

ANOVA

Tukey

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	4.976	9	.553	.910	.529
Within Groups	18.223	30	.607		
Total	23.199	39			

Table E16: Summary of ANOVA table on the total carbon dioxide sequestered (kg.m²) after the *E. tef* harvest.

ANOVA

Tukey

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	66.906	9	7.434	.910	.529
Within Groups	245.049	30	8.168		
Total	311.955	39			