The evaluation of coated and uncoated seed mixtures for the rehabilitation of gold and platinum mine tailings

CA Kruger

orcid.org/ 0000-0001-5716-6456

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Supervisor: Prof K Kellner

Co-supervisor: Mr PW van Deventer

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Disclaimer

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Abstract

The establishment of vegetation on platinum and gold Tailing storage facilities (TSFs) to stabilise the TSF surface against erosion (phytostabilisation) are met with many challenges; acid and alkaline pH levels of tailings, the lack of organic matter and nutrients, salinity, strained microbial populations and drought conditions. Coated or enhanced seed also known as “Agricote®” supplied by AGT Foods Africa, incorporates growth stimulants, nutrients, rhizobia inoculants with pesticides and fungicides within lime and a protective polymer to enhance seed germination and seedling growth. Throughout the mine rehabilitation industry, commercial seed mixtures containing perennial grass species in combination with nursing crops (E.g. *Eragrostis tef* [*E. tef*] & *Sorghum bicolor* [*S. bicolor*]) and perennial legumes (e.g. *Medicago sativa* [*M. sativa*]) are used to establish vegetation cover on gold and platinum TSFs. However, due to weight differences of coated and uncoated seed, the amount of seed per unit weight effectively sown is less for coated seed. Grass species also exhibit different behaviours regarding seed dormancy, competition strategies and growth vigour that may influence species establishment within a heterogeneous seed mixture. This study aims to provide insights toward understanding the community dynamics of such an established grass community on gold and platinum TSFs by comparing the emergence and establishment of coated and uncoated seed treatments with adjusted seeding rates and evaluating the use of bio-stimulants to improve vegetation establishment. The hypothesis was that increases in species seeding rates; that take into account the weight of the Agricote® coating formula for selected species used in the seed mixture and the application of bio-stimulants, would increase the success of vegetation establishment. Field trial plots at two gold TSFs were ameliorated with lime, compost and fertiliser and platinum tailings were placed in 1 m x 1m x 0,25 m bulk bags and ameliorated with compost and fertiliser. Seed selected and sown together within experimental seed treatments at the trial sites after soil amelioration includes coated and uncoated seed of *Eragrostis curvula* (*E. curvula*), *Digitaria eriantha* (*D. eriantha*) and *Cynodon dactylon* (*C. dactylon*). *Sorghum bicolor* and *E tef* were sown in seed treatments as annual nurse crops and *M sativa* was sown as a legume. The emergence and growth of *E. curvula*, *D*. *eriantha* and *C. dactylon* from coated and uncoated seed and *E. tef* from uncoated seed were evaluated separately in gold and platinum tailings in supporting pot trials to verify results of field trials. Results indicated no difference between the emergence densities of coated and uncoated seed treatments after two months. An increase in seeding rate of coated seed to sow the same number of seed as uncoated seed of the same species within treatments did not result in a more efficient

increased seedling emergence density. At both the gold TSF trial sites, the initial plant composition changed from being dominated by *E. tef* and *E. curvula* in June 2016 to *E. curvula and C. dactylon* being the dominant species from November 2016 until the end of the trials in May and June 2017. In the platinum tailing trials, *E.* curvula and *C. dactylon* were the dominant species throughout the trial period (April 2016 tot May 2017) due to irrigation of the platinum tailing, the growth of *M. sativa* suppressed the emergence of other grass species in the limited growth space. *Trichoderma* as a fungal bio-stimulant has the potential to increase seedling emergence although further research is required.

Keywords: TSF, rehabilitation, re-vegetation, phytostabilisation, bio-stimulants, establishment, seed mixture, coated ratios.

Opsomming

Die vestiging van plantegroei op platina- en goudmyn slik opgaar damme om die oppervlak daarvan te stabliseer teen erosie (Phytostabiliseering) word beperk deur verskeie uitdagings soos bv, suur en alkalise pH vlakke van slik material, afwesigheid van organiese material en nutriënte, sout toestande, lae mikroorganisme populasies en droogtes. Bedekte “coated” saad ook bekend as “Agricote®” soos verskaf deur AGT Foods Africa is in staat om groei stimulante, nutriënte, rhizobium inoculante en gifstowwe in ‘n beskermende kalk polimeer te inkorporeer en die saad dan daarmee te enkapsuleer wat die vestiging en groei van saailinge moontlik kan verbeter. In die myn slikdam rehabilitatsie industrie word kommersiele saad mengsels gebruik wat meerjarige gras spesies (bv. *E. curvula* en *C. dactylon*) bevat in kombinasie met eenjarige gras spesies (*E. tef* en *S. bicolor*) en peulgewasse (bv. *M. sativa*) vir die vestiging van plantegroei. Bedekte saad is swaarder as onbekte saad en veroorsaak dat minder bedekte saad per gewig eenheid gesaai word as onbedekte saad. Gras spesies verskil in terme van saad dormansie, kompetisie stratigiëe en groei vermoë wat ‘n invloed het op die vestiging van spesies binne ‘n saadmengsel. Die doel van hierdie studie is om insig te lewer oor die dinamika van spesies gesaai op goud en platinum slikdamme deur die opkoms en vestiging van bedekte en onbedekte saad behandelings met aangepaste saai digthede te vergelyk en die gebruik van mikrobiese bio-stimulante om opkomste te verbeter te evalueer. Die hipotese was dat hoër saailing vestiging verkry sal word wanneer die saaidigteid van bedekte saad vermeerder word volgens bedekte en onbedekte saad gewig verhoudings en die behandeling van saad en proef persele met bio-stimulante sal ook tot gevolg saailing opkomste verbeter. Veld proef persele by twee goud myn slikdamme is voorberei met kalk, kompos en kunsmis volgens bemestings aanbevelings, platina slik materiaal is geplaas in een m x een m x 0,25 m vierkantige grootmaatsakke voordat dit voorberei is met kompos en kunsmis. Saad wat gebruik is en in eksperimentele saadmengsels op die verskillende proef persele gesaai is na ameliorasie sluit in, bedekte en onbedekte saad van *E. curvula*, *D. eriantha* en *C. dactylon*. *S. bicolor* en *E. tef* is saam in saad mengsels gesaai as eenjarige hulp gewasse en *M. sativa* is gesaai as ‘n peul gewas. Die opkoms en groei van bedekte en onbedekte *E. curvula*, *D. eriantha*, en *C. dactylon* saad en onbedekte *E. tef* saad is geëvalueer in platina en goud slik mediums deur pot proewe om resultate van veldproewe te verifieer. Daar was geen merkwaardige verskil tussen die opkoms digthede van bedekte saad en onbedekte saad mengsels nie. Vermeerdering van die saaidigtheid van bedekte saad om soveel saad neer te sit soos bevat in onbedekte saad mengsels het nie gelei tot ‘n merkwaardige toename in die digtheid van saailinge na twee maande nie. Dit het ook nie gelei tot ‘n hoër oorlewings digtheid van

saailinge na ses maande nie. Op altwee die goud slikdamme was die spesie versameling in Junie 2016 gedomineer deur *E. tef* en *E. curvula* maar van November 2016 was *E. curvula* en *C. dactylon* die dominante spesies tot die einde van die proef tydperk. Op die platina proewe was *E. curvula* en *C. dactylon* die dominante species deur die proef tydperk (April 2016 tot Mei 2017) omdat die platina slik proewe besproei was het *Medicago sativa* goed gegroei en die opkoms van gras spesies in die beperkte spasie onderdruk. Die fungiese bio-stimulant *Trichoderma* het die potensiaal om die opkoms van meerjarige spesies op goud slikdamme te verbeter, maar verdere navorsing is egter nodig om dit te besvestig.

Sleutel woorde: slikdam, phyto-stabiliseering, saadmengsel, vestiging, bedekte saad.

Abbreviations

ANOVA:	Analysis of Variance
C:	Coated seed
C. dactylon:	*Cynodon dactylon*
CRWN:	Crown gold mine tailings
CRWNC:	Crown gold mine tailings coated seed
CRWNUC:	Crown gold mine tailings uncoated seed
CTRL:	Control soil
CTRLC:	Control soil coated seed
CTRLUC:	Control soil uncoated seed
D. eriantha:	*Digitaria eriantha*
DHA:	Dehydrogenase Activity
DM:	Dry Matter (Biomass)
E. curvula:	*Eragrostos curvula*
E. tef:	*Eragrostis tef*
GDP:	Gross Domestic Product
INF:	Iodonitrotetrazolium formazan
ISTA:	International seed testing association
KCl:	Potassium chloride
LD19C:	Lower seed density treatment 19 kg/ha using coated seed
LD19UC:	Lower seed density treatment 19 kg/ha using uncoated seed
LD12C:	Lower seed density treatment 12 kg/ha using coated seed
LD12UC:	Lower seed density treatment 12 kg/ha using uncoated seed
LD5C:	Lower seed density treatment 5 kg/ha using coated seed
LD5UC:	Lower seed density treatment 5 kg/ha using uncoated seed
MA1:	Microbial ameliorant treatment one
MA2:	Microbial ameliorant treatment two
MA3:	Microbial ameliorant treatment three

MA4: Microbial ameliorant treatments four

M. sativa: *Medicago sativa*

NAP: Net Acid Potential

NARGT: Non-ameliorated Rooikraal gold mine tailings

NEMA: National Environmental Management Act of 1992

NWU: North-West University

PGPR: Plant growth promoting rhizobacteria

PT: Platinum tailings

PTC: Platinum tailings coated seed

PTUC: Platinum tailings uncoated seed

RK: Rooikraal gold mine tailings

RKC: Rooikraal gold mine tailings coated seed

RKUC: Rooikraal gold mine tailings uncoated seed

S. bicolor: *Sorghum bicolor*

SER: Society for Ecological Restoration

T1C: Coated seed treatment 1

T2UC: Uncoated seed treatment 2

T3C: Coated seed treatment 3

T4C: Coated seed treatment 4

T5UC: Uncoated seed treatment 5

TSF: Tailings Storage Facility

UC: Uncoated seed

Glossary

Ameliorant:	A substance added to soil to improve the growth conditions for plant roots
Coated seed:	Seed treated with inert materials such as growth stimulants, fertiliser, pesticides and fungicides in a protective polymer surrounding the seed coat (Nel, 2014).
Uncoated seed:	Seed without any seed coating material applied to the natural seed exterior.
Bio-stimulant:	Any microorganism-containing substance applied to plants with the purpose of enhancing plant growth by enhancing abiotic stress tolerance, nutrient efficiency and crop quality traits, regardless of the soil nutrient content (Du Jardin, 2015:3).
Seed treatment:	An experimental mixture of seed from different species sown together to establish a vegetation community.
TSF:	An acronym for Tailing Storage Facility, it refers to the rock milled civil dam structure that is used to store the tailings material in after minerals of value have been extracted from the host rock (Weiersbye *et al.,* 2006:103)

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Chapter 1: Introduction

1.1 Justification of study

Gold mine tailings are commonly characterised by acidic conditions with pH levels below five due to the presence and oxidation of iron sulphides (Cooke & Johnson, 2002:49). Elevated concentrations of metals such as manganese (Mn), copper (Cu), zinc (Zn), cadmium (Cd), nickel (Ni), iron (Fe), mercury (Hg) and arsenic are also commonly associated with gold mine tailings (Mulugisi *et al.*, 2009:512). Platinum tailings are characterised by increased salinity and alkalinity (Van der Walt *et al.*, 2012:103). Both Mendez and Maier (2008a:278) and Cooke and Johnson (2002:49) explain that mine tailings in general are practically void of macronutrients and organic matter. According to Cooke and Johnson (2002:49), the absence of organic matter in fine-textured soils is regularly the case with soils of mine tailings. Furthermore, mine tailings support a strained microbial community and do not have good soil structure. These soils also have the potential to cause severe soil compaction, high bulk densities, reduced water infiltration rates and waterlogging at the soil surface (Mendez and Maier, 2008a:278). Nel *et al.* (2014:97) and Cooke and Johnson (2002:49) elaborate that the adverse negative chemical and physical soil properties regularly encountered in gold and other mine tailings inhibit the colonisation of natural vegetation and increase the difficulty of successfully establishing sustainable vegetation communities during rehabilitation.

Degradation of the environment caused by TSFs commonly spreads much farther than the boundaries of the TSF by means of erosion. Wind and water erosion are increased, which lead to the pollution of the surrounding air, soils and water sources, including dams, streams, rivers and underlying sediments. Environmental aspects that are greatly influenced by the erosion of TSFs and gold mining acid mine drainage include soil nutrient cycling, regeneration of vegetation and the biogeochemical cycling of potentially toxic elements such as aluminium (Al), Cd, Hg, chromium (Cr), Cu, Fe, Mn, lead (Pb) and Zn (Weiersbye *et al*., 2006:101).

Erosion and dust control at a mining site is therefore critical and forms part of the rehabilitation process. Rehabilitation is required by legislation and has to be carried out by the most practical and cost-effective means necessary during the rehabilitation process (Weiersbye *et al*., 2006:101). The establishment of grass mixtures on TSFs is considered by the rehabilitation industry as one of the best methods to address erosion and dust problems. The use of grass mixtures to rehabilitate TSFs is also called ‘re-grassing’ or ‘phytostabilisation’ (Mendez & Maier, 008a:279; Weiersbye *et al.*, 2006:101).

Phytostabilisation entails the use of stress-tolerant plants to stabilise TSFs in the plant rhizosphere, reducing erosion and immobilising potentially toxic elements that can occur in the tailings material. Subsequently, the heterotrophic microbial community will be improved, which will likely result in improved vegetation establishment and growth on the tailings materials (Cooke & Johnson, 2002:49; Mendez & Maier, 2008a:279). Re-grassing entails the use of grass species for phytostabilisation (Weiersbye *et al.,* 2006:101). Mendez and Maier (2008a:279) state the following: ”The ultimate objective for successful phytostabilisation, is the long-term succession of the plant community in mine tailings to promote soil development processes, microbial diversity and finally, to restore soil ecosystem functions to a state of self-sustainability.”

Turner *et al.* (2006) as cited by Nel *et al.* (2014:97) have tested improved seeding technologies and elaborate that seed coating technologies are able to assist in the establishment of vegetation on mine tailings. Seed coating improves the micro-environment surrounding the seed. Tow and Lazenby (2001:1) explain that a proper understanding of plant competition, succession and management principles is required in effective rehabilitation, especially for maintaining a desired vegetation composition over the long term. Bradshaw (2002:5) supports Tow and Lazenby (2001:1) and explains that a complete understanding of ecosystem functions, processes and structure is required in effective rehabilitation.

This study focused on the establishment and growth of three perennial grasses, namely *C. dactylon*, *D. eriantha* and *E. curvula*, one annual grass, *E. tef*, a nurse crop, *S. bicolor*, and a perennial legume, *M. sativa*, to rehabilitate gold and platinum TSFs. The species were used in different seed mixtures of coated and uncoated seed. Three coated and two uncoated seed mixtures were sown in ameliorated field plots at two gold mine TSF sites and in platinum tailings. Supporting pot trials were conducted with the same growth mediums to determine the establishment and growth potential of the species in the different tailings material. The third phase of this study evaluated the use of fungal and microbial bio-stimulants on the early establishment of grasses.

1.2 Aims and objectives

1.2.1 Aims

The main aim of this study was to evaluate the establishment and growth of coated and uncoated seeds for plant species in a seed mixture for the rehabilitation of gold and platinum TSFs. Furthermore, the study aimed to test the emergence of seedlings when lower seeding densities

are used and whether bio-stimulants are able to improve seedling emergence of seed mixtures on gold TSFs for rehabilitation.

1.2.2 Objectives

Objectives for this study were to:

- Compare the use of coated and uncoated grass seed mixtures for the rehabilitation of gold and platinum TSFs in field trials.
- Determine the ratio of coated to uncoated seeds for specific species in a seed mixture used for rehabilitation.
- Evaluate the effects of fungal and microbial bio-stimulant application and lower seeding rates on the emergence of grass seed mixtures on gold TSFs.

1.3 Hypotheses

It is hypothesised that the coated seed mixtures would have better establishment than the uncoated seed mixtures. Furthermore, an increased seeding rate would result in higher plant emergence and survival, whereas a decrease in the seeding rate would result in a lower establishment of plants. Lastly, the use of bio-stimulants for amelioration would result in higher establishment rates of plants on gold TSFs.

1.4 Dissertation structure and content

The emergence and establishment of coated grass seed compared to that of uncoated grass seed was the central theme of this study. Chapter 2 reviews literature regarding various aspects of TSF rehabilitation. Main discussion points include (1) background of mine rehabilitation in South Africa, (2) an overview of mine tailings as a growth medium concerning its physical, chemical and biological traits and (3) a discussion of phytostabilisation and the possible influence of seed coating and seeding rate on TSF rehabilitation. Chapter 3 describes the material and methods used as well as the location of the study sites. Results obtained from the vegetation trials are discussed in Chapter 4. Chapter 5 contains concluding remarks on the study and provides recommendations for future trials. Chapter 6 contains a list of all the references used in this dissertation. Finally, additional documents containing information regarding the trials are attached in the Annexures.

Chapter 2: Literature review

In this section the practice of mine TSF rehabilitation in South Africa is discussed and literature regarding the physical, chemical and biological properties of gold and platinum mine tailings contained within TSFs is reviewed. Seed coating technology, seeding rate and seed germination for the establishment of vegetation on gold and platinum TSFs is also discussed.

The literature review aims to determine the shortfalls and challenges faced when gold and platinum TSfs are used as a growth medium for the implementation of phytostabilisation practices.

2.1 Mine rehabilitation in South-Africa

Globally, the mining industry extracts metals and mineral resources vital for components in products that improve people's lives and contribute to national economies (Beylot & Villenueve, 2017:139; Cooke & Johnson, 2002:42; Kossoff *et al*., 2014:230). The mining industry ensures employment to over 40 million individuals and indirectly supports 200–250 million people worldwide (Kossoff *et al.*, 2014:230).

Previously, South Africa had the world's largest reserves of gold, manganese, chrome, vanadium and platinum. It was the world's lead producer of gold in 1886 and contributed to an estimated 40% of the global resource reserves (Orlowska *et al.*, 2010:185). Mining has consequently been a cornerstone of the South African economy for more than a century, providing social improvements such as employment, business opportunities and medical facilities (Durand, 2012:24; Kossoff *et al.*, 2014:230). According to the *Mine SA 2016* pocketbook, mining operations contributed ZAR 286 billion to the South African gross domestic product (GDP) in 2015, which equates to 7.1% of the total GDP while directly employing 457 698 individuals (Chamber of Mines of South Africa, 2017:6). Mining operations therefore have a large impact on the socio-economic aspects in South Africa.

Although mining is known to degrades the environment at an alarming rate, modern society is unable to function without this sector (Bradshaw, 1997:256, 2000:89; Dai *et al.*, 2002:223; Kossoff *et al.*, 2014:230; Mhlongo & Amphonsa-Dacosta, 2016:281; Schoenberger, 2016:119; Van der Walt *et al.*, 2012:103). Bradshaw (1997:89) and Cooke and Johnson (2002:43) further explain that the mining industry has a negative effect on natural vegetation and soils due to the large operations and construction of TSFs. Alberts *et al.* (2016:268) reported that there are approximately 6 000 ownerless and derelict mines in South Africa and that the total financial cost of the rehabilitation thereof is projected at ZAR 30 billion. Cooke and Johnson (2002:42) note that

the total area of environmental degradation due to mining operations in South Africa has been estimated at around 200 000 ha.

In 2006, annually around 315 million tonnes of tailings are produced by the South African mining sector (Weiersbye *et al.*, 2006:101). Weiersbye *et al.* (2006:101) also reported that 105 million tonnes of the aforementioned tailings use to be attributed to gold mining in the Witwatersrand Basin, which is produced at a rate of 200 000 tonnes of tailings per tonne of gold extracted (Weiersbye *et al.*, 2006:101). According to Mhlongo and Amphonsa-Dacosta (2016:281) and Weiersbye *et al.* (2006:101), this vast amount of tailings has led to approximately 40 000 ha of the Witwatersrand Basin goldfields being covered by TSFs or their residual footprints, in which 6 billion tonnes of gold and uranium tailings are retained.

According to Weiersbye *et al.* (2006:101), legislation to reduce or control the impacts of mining on the surrounding environment was lacking prior to 1991, but presently South Africa has some of the most stringent environmental legislation frameworks in the world, directed at the mining industry. Alberts *et al.* (2017:267) supports Weiersbye *et al.* (2006:101) by describing the environmental legislation of South Africa as comprehensive. These authors also argue that it is severely complicated and confusing due to the many individual regulations, laws and guidelines, as they function in conjunction with the various ministries that are involved in this industry, such as Agriculture, Environment and Water affairs.

Marais (2014:1) states that companies are forced to ensure good environmental practices in all of their operations by addressing and reducing environmental impacts. This author further explains that the ‘duty of care’ principle contained within Section 28 of the National Environmental Management Act No. 107 of 1998 (NEMA), places the responsibility on each person who has caused or may cause severe pollution or environmental degradation to take satisfactory measures to prevent, mitigate or rectify the effect of the relevant activity on the surrounding environment (. This corresponds to Section 34 of NEMA and Section 38 of the Mineral and Petroleum Resources Development Act No. 28 of 2002. These regulations make it possible for the directors of a mining company and members of a corporation associated with the mining industry to be held responsible for environmental impacts; they are therefore open to prosecution for any unacceptable negative environmental impacts. This includes environmental damages, degradation and any aspects causing pollution advertently or inadvertently (Marais, 2014:1).

It is therefore evident that the scope for environmental rehabilitation in South Africa is substantial, considering the extent of mining activities and the requirements by legislative Frameworks.

2.2 Reclamation, rehabilitation and restoration

The Society for Ecological Restoration (SER) (2004:4) defines ecological restoration as the process of helping the recovery of an ecosystem that has been damaged, degraded or destroyed. The SER further explain that rehabilitation and ecological restoration share a fundamental focus on the pre-existing ecosystem as a model of reference, but that the two activities differ from one another in their goals and strategies (SER, 2004:13). Rehabilitation focuses on the reinstitution of an ecosystem's processes, productivity and services. The goals of restoration, on the other hand, additionally focus on the re-establishment of a disturbed ecosystem's biotic integrity regarding the historical plant composition and community structure (SER, 2004:13). The SER (2004:13) continues to explain that the term 'reclamation', which is commonly used within the context of mined land and the rehabilitation of TSFs, has an even broader application than rehabilitation. The main objectives of reclamation are to stabilise the terrain surface, ensure public safety, improve site aesthetics in order to return the land to what is considered within the regional context as 'useful'. Various authors such as Alberts *et al*. (2017:269), Cooke and Johnson (2002:45) and Shrestha *et al.* (2005:1043) share similar views to those of SER (2004:13) and agree that reclamation should include a process by which the degraded land surface is stabilised and transformed to some form of beneficial use. When the reclamation process is guided by ecological principles, 'restoration' is the more fitting term.

Singh *et al.* (2002:1437) provide a slightly different view concerning the outcomes of rehabilitation, reclamation and restoration, stating that these practices can be seen as a 'continuum of outcomes'. Whereby the end result is a measure of the land's similarity to the pre-mining ecosystem and where rehabilitation yields the least similar and restoration the most similar ecosystem to the pre-mining ecosystem. Bradshaw (2002:4) agrees with the views of Singh *et al.* (2002:1437), in that the goals of rehabilitation and restoration distinguish the practices from one another, but argues that the above may be termed 'reclamation', because these practices ensure the re-use of the damaged land. Cooke and Johnson (2002:45), however, state that the terms 'reclamation', 'restoration' and 'rehabilitation' are commonly used interchangeably in practice and likened to the re-setting of an ecological clock.

Van Deventer and Hattingh (2009:151) argue that within the context of the mining industry, TSFs are permanent structures and unless it proves beneficial to return the mine tailings within these structures underground, they will inevitably remain on the surface and occupy vast areas of land. These authors continue to explain that while rehabilitation is able to change TSF aesthetics and have a substantially beneficial impact on the surrounding environment, it is impossible to

rehabilitate the land surface to its original state and land use. Santini and Banning (2016:44) support Van Deventer and Hattingh (2009:151), explaining that it is theoretically impossible to rehabilitate mine TSFs to the original pre-mining land use because their placement within natural landscapes irreversibly modifies the preceding conditions. This has prompted alternative considerations concerning TSF rehabilitation.

Orlowska *et al*. (2010:186) explain that in-situ waste management is the only viable rehabilitation technique applicable to TSFs because of their large size and the extensive surface areas they cover. On-site rehabilitation, in the case of TSFs, usually involves using the mine tailings as a growth medium and ameliorating it with soil amendments to introduce suitable plants and soil micro-fauna with the purpose of stabilising the TSF surface. Orlowska *et al.* (2010:186) continue to explain that this is a more realistic and affordable technique compared to other site stabilisation techniques.

Deciding against restoration and aiming for alternative rehabilitation pathways greatly expands the endpoint ecosystem options applicable to TSFs (Santini & Banning, 2016:44). With careful planning, these highly disturbed land surfaces may be rehabilitated to a state where they can be used for cash crop production or alternative land uses (Santini & Banning, 2016:44). During the rehabilitation of TSFs, the main objective, however, is to prevent wind and water erosion by increasing the surface stability of these structures (Van Deventer & Hattingh, 2009:181).

Although Godfrey *et al.* (2007:2) explains that in the case of gold TSFs, what is currently considered a waste may in future be regarded as a viable mineral resource suitable to be re-mined. These authors mention the Ergo project initiated in 1977, which entailed the reprocessing of 50 old gold TSFs to recover residual gold. The modern approach to mineral waste management in South Africa is to stockpile mineral waste in TSFs until re-mining becomes financially viable (Godfrey *et al.,* 2007:2). Therefore, the current principal aim of TSF rehabilitation is to stabilise the TSF surface and reduce environmental impacts associated with the stockpile until re-mining is financially viable. Integrally, rehabilitation is able to address environmental issues associated with TSFs, e.g. surface water quality, erosion control and surrounding air quality. This, however, does not reduce the problem of the long-term pollution potential associated with mineral waste such as mine tailings (Godfrey *et al.,* 2007:2).

2.3 An overview of mine tailings

2.3.1 Tailings material

Tailings is defined as a combination of fluids and pulverised rock that remain after the ore has been extracted from the mining resource through various processes, involving mills, washeries or concentrators (Beylot & Villenueve, 2017:139; Kossoff *et al.*, 2014:230; Mendez & Maier, 2008a:278; Mulugisi *et al.*, 2009:512; Schoenberger, 2016:119; Young *et al.*, 2013:498). The term 'tailings' is general and describes the by-product of various extractive industries that include coal, uranium, oil sands, aluminium and precious and base metals such as gold and platinum. Commonly, the ratio of mine tailings to the concentrate is within the range of 200:1. This ratio tends to increase as the grades of ores decrease with the advance of the mining process (Cooke & Jonson, 2002:42; Kossoff *et al.*, 2014:230; Santini & Banning, 2016:38; Schoenberger, 2016:119).

Currently in South Africa, the most common storage practice for tailings is to pump it in the form of a low-density aqueous slurry into large impoundment facilities where it is left to dry and harden (Fourie, 2009:1067; Kossoff *et al.*, 2014:242; Weiersbye *et al.,* 2006:103). This is an indirect disposal method and tailings slurries consist of 25–30% solids (Adiansyah *et al.*, 2015:1053). This procedure is commonly known as dry stacking (Santini & Banning, 2016:41). The resulting tailings impoundment facilities are referred to as tailings dams, tailings dumps, tailings ponds, spoils, slime dams or TSFs and are usually constructed from the mine tailings itself, other available waste rock or soil (Beylot & Villenueve, 2017:139; Cooke & Johnson, 2002:42; Kossoff *et al.*, 2014:230; Mulugisi *et al.*, 2009:512; Schoenberger, 2016:119; Touceda-Gonzãles *et al.*, 2017:301; Weiersbye *et al.*, 2006:101; Young *et al.*, 2013:408).

It is not uncommon for one of these structures to occupy extensive areas of land that range in size horizontally from hundreds of square metres to multiple square kilometres and tens of metres vertically (Kossoff *et al.*, 2014:232; Schoenberger, 2016:119).

Kossoff *et al.* (2014:233), Schoenberger (2016:119) and Fourie (2009:1067) explain that the construction of TSFs commence in stages. After the initial retaining wall has been constructed and as more mine tailings areproduced, the structure perimeter is raised. This increases the retaining capacity of the TSF and enables additional mine tailings to be pumped into the TSF. This process is repeated as needed to accommodate additional tailings material. Young *et al.* (2015:250) added that while the mine is operational, the associated TSFs are expected to remain under ponded water due to the active pumping of low-density tailings slurry. When the mine is

abandoned and operations cease, the tailings surface will likely dry, rendering the tailings vulnerable to erosion and oxidation. Meza-Figuera *et al.* (2009:141) stress that in semi-arid areas, when mine tailings dry, it becomes more vulnerable to wind erosion.

Straker *et al.* (2007:219) explain that TSFs pose unique challenges to phytostabilisation practices, because TSFs are intensively disturbed by erosive forces while simultaneously being polluted and biologically impoverished environments.

2.3.2 Mine tailings erosion

Mendez and Maier (2008a:278) explain that one of the prominent problems with TSFs is their instability and vulnerability to wind and water erosion (Fourie, 2009:1068; Young *et al.*, 2015:250). Weiersbye *et al.* (2006:101) further elaborate that the steep slope angles and elevation of TSFs above the natural ground contour render them vulnerable to erosion. Weiersbye *et al.* (2006:101) and Orlowska *et al.* (2010:185) report that there are instances where water erosion losses from the slopes of TSFs are more than 500 t/ha/y, this surpasses that of agricultural areas, which commonly have an erosion rate of 10–15 t/ha/y.

According to Van Deventer *et al.* (2008:231), erosion is caused when the energy of moving water or air is strong enough to overwhelm the interrelated forces binding soil particles together. The process of water erosion as described by Sharma (1996:125) and Singer *et al.* (1992:382) as a series of events during which soil particles are dislodged through a disturbance (water drop impacts or overland flow). The particles are then transported from their source within a flow of water (overland flow, also known as surface runoff) before being deposited at a different location (Sharma, 1996:125; Singer *et al.,* 1992:382). Surface runoff occurs when the rate of water applied to the soil surface exceeds the infiltration rate of the soil, leading to surface ponding (Van Deventer *et al.*, 2008:240). The excess water flows over the soil surface and provides sufficient energy to dislodge soil particles. The dislodged soil particles are then carried as suspended or bed loads downslope until the energy of the runoff water is diminished, at which point the soil particles are deposited (Sharma, 1996:125; Singer *et al.,* 1992:382). Sharma (1996:134) states that during the water erosion process, clay- and silt-sized particles are carried the farthest as suspended loads. The surface runoff initially flows in sheets for a short distance, known as sheet flow, after which it concentrates in small and larger channels called rills and gullies. Like water, wind is a fluid medium and the process of wind erosion is similar to that of water (Singer *et al.*, 1992:387). When the force of the wind, which is a function of its velocity, exceeds the forces holding a soil particle to the ground, it liberates and moves that soil particle (Singer *et al.*, 1992:387).

Through erosion, structural failure, acid mine drainage, pollution and environmental impacts tend to far exceed TSF borders (Bleeker *et al.*, 2002:2; Cooke & Johnson, 2002:43; Gil-Loaiza *et al.,* 2016:452; Mendez & Maier, 2008a:278; Orlowska *et al*., 2010:186; Petrisor *et al.*, 2004:2; Ye *et al.*, 2002:1103). Contamination with mine tailings and disturbances caused by mining operations have serious impacts on the nutrient cycling processes in soils of the surrounding environment. These disrupted processes include the growth and development of natural vegetation communities and the bio-geochemical cycling of potentially toxic elements (Weiersbye *et al.,* 2006:101; Young *et al.,* 2015:250). Correspondingly, Gil-Loaiza *et al.* (2016:452) and Mendez and Maier (2008a:278) stress that potentially toxic elements hazardous to human health are often associated with mine tailings. Human health risks arise from the dispersion of mine tailings particles via wind and water erosion. According to Andaros *et al.* (2016:S229), TSFs are known as dust-generating sources in South Africa, especially in the North-West, Gauteng and Freestate Provinces. TSFs contribute to unacceptable health risks to surrounding communities due to exposure to crystalline silica emanating from the TSFs (Andaros *et al.*, 2016:S229).

Van Deventer *et al.* (2008:231) explain that the process of erosion at a specific site is determined and influenced by a number of factors. These factors include climate, hydrological regime, topographical characteristics, substrate attributes and vegetation or cover conditions already in place. Vegetation is described as the factor that influences the process of erosion the most. The Universal Soil Loss Equation (USLE) Equation 1 uses various factors controlling the erosion process to calculate the average annual water erosion rate at a site (Bell, 2002:39):

Equation 1: $A = R \times K \times L \times S \times C \times P$

Where A is the calculated spatial and temporal average soil loss per area unit, commonly expressed as t/ha/y R is the rainfall-runoff erosivity factor, K is the soil erodibility factor, L is the slope length factor, S is the slope steepness factor, C is the cover-management factor and P is the support practice or cultivation factor (Bell, 2002:39). Not all the factors can be altered to influence the resulting soil loss (A), such as the climate (R), of a site. Other factors, such as slope length (L) and steepness (S), may be altered by shortening and flattening slopes and the cover (C); management (P) factors can be altered via vegetation and agricultural practices (Bell, 2002:40).

Burton *et al.* (2006:380) emphasise that within the USLE there exists a negative exponential relationship between the amount of erosion (A) and the cover (C) of living plants or plant residue covering the surface. These authors explain that in the case where bare soil surfaces have slopes greater than 9%, soil loss can be drastically decreased by 60–70% with additional plant cover,

after which increasingly larger amounts of plant or litter cover are required to achieve smaller decreases in erosion (Burton *et al.,* 2006:380).

Van Deventer *et al.* (2008:234) list six principles of erosion control aimed at preventing it entirely or subsequently reducing its intensity to protect the substrate. These principles include:

1. Protecting the substrate surface against the effect of kinetic energy of raindrops, surface water flow or wind.
2. Increasing the infiltration capacity of the substrate.
3. Improving the stability of substrate aggregates.
4. Increasing the water accumulation and retention capacity of the surface.
5. Creating a course or rough surface.
6. Improving the interception and safe conduction of erosion-triggering precipitation water.

Van Deventer *et al.* (2008:234) state that vegetation cover meets all six of the above-mentioned principles and the establishment of a dense vegetation community is therefore the best means of long-term erosion control. Orlowska *et al.* (2010:186) support Van Deventer *et al.* (2008:234) by stating that to overcome erosion on TSFs, the establishment of vegetation on the mine tailings surface is crucial. Tordoff *et al.* (2000:220) add that vegetation cover effectively provides the required surface stability, preventing the spread of windblown contaminant particles and reducing water erosion by intercepting incidental precipitation.

The establishment of vegetation or phytostabilisation on TSF surfaces can therefore be regarded as an effective method to prevent wind and water erosion. Many authors, however, explain that TSFs are inhospitable environments. The harsh physical and chemical properties frequently associated with mine tailings contained within TSFs make the establishment of a proper vegetation cover difficult (Burges *et al.*, 2016:481; De-Bashan *et al.*, 2010:343; Epelde *et al.*, 2014:1; Gil-Loaiza *et al.*, 2016:451; Grandlic *et al.*, 2009:1734; Mendez & Maier, 2008a:278; Meza-Figuera *et al.*, 2009:140; Nel *et al.*, 2014:97; Orlowska *et al.*, 2010:186; Petrisor *et al.*, 2004:1; Tordoff *et al.*, 2000:220; Touceda-González *et al.*, 2017:301; Weiersbye *et al.*, 2006:102; Ye *et al.*, 2002:1104; Yang *et al.*, 2016:427; Young *et al.*, 2013:498, Young *et al.*, 2015:250).

2.3.3 Physical characteristics

Tailings are regularly gravel-free and their particle size distribution ranges between sand and clay (2 mm–3.9 µm), with the sand fraction (2 mm–625 µm) being more abundant than that of silt (625–3.9 µm) (Kossoff *et al.*, 2014; Tordoff *et al.*, 2000:220). Santini and Banning (2016:42) describe tailings as having a massive apedal structure that retards chemical weathering processes

dependent on gas exchange. Wijesekara *et al.* (2016:127) explain that the physical properties of soils, including bulk density, porosity, infiltration, water-holding capacity and hydraulic conductivity, are interrelated and because mine tailings tend to be poorly aggregated with an ill-defined soil texture, mine tailingscommonly exhibit less favourable physical conditions for plant establishment.

Tordoff *et al.* (2000:221) state that impermeable surface barriers are associated with mine tailings due to compaction and cementation processes. Singer *et al.* (1992:391) explain that soil crusting or sealing and compaction are physical degradation processes in soils and occur where finer soil particles are sorted downwards, which plug pore spaces between soil particles, creating an impeding layer on the soil surface. Seals pertain to wet soil conditions and crusts occur on dry soils, which drastically reduce water infiltration rates and block seedling emergence.

Singer *et al.* (1992:391) and Schroeder (1984:51) define bulk density as a measure of dry soil mass per unit volume. Bulk densities of mine tailings differ according to the properties of the parent rock mined but are generally in the range of 1,8–1,9 g/cm^3. The density increases with depth in mine tailingsat a general rate of 0,09–0,17 g/cm^3 per 30 cm due to the effect of compaction, de-watering and diagenesis (Kossoff *et al.*, 2014:231; Young *et al.*, 2015:250). Santini and Banning (2016:38) mention that the average bulk density of tailings is approximately 1,5 g/cm^3. Van Deventer and Hattingh (2009:174), however, report the bulk density of dried deposited mine tailings to be between 1,4 and 3 g/cm^3.

Cooke and Johnson (2002:49) explain that the fine texture and the absence of organic matter in mine tailings result in a weakly structured medium with high bulk densities that lead to severe compaction, low water infiltration rates and poor water retention capacities. Ye *et al.* (2002:110) also conclude that root growth tends to be restricted in tailings with a silty and sandy texture and the absence of organic matter. Weiersbye *et al*. (2006:103) report that the deposition of fine particles, such as tailings, in a low-density aqueous slurry results in compaction and weak aeration within the rhizosphere. Shresta *et al.* (2005:1043) support Cooke and Johnson (2002:49), stating that soil compaction is the result of an increase in bulk density, which is a decrease in the void ratio also referred to as porosity.

Shresta *et al*. (2005:1043) report various causes for compaction in mine tailings. These include vehicular and raindrop action and the drying of settled tailings slurries, with the repeated process of wet slurry being pumped, after which the drying, compact tailings layers form on and beneath the surface through sealing and crusting (Singer *et al.,* 1992:391; Tordoff *et al.*, 2000:221).

According to Shresta *et al.* (2005:1051), the restriction of root development begins at soil bulk densities of 1,4–1,75 g/cm^3.

Bell (2002:46) explains that the temperature associated with the surfaces of mine tailings has a direct influence on the establishment of vegetation, especially in the case of direct seeding techniques. This is especially true where the mine tailings, are used as a growth medium. If the surface temperatures reach extreme values of 60°C, it has detrimental effects on seedling establishment. Bell (2002:46) adds that the main sources of radiation to the surface of mine tailings is the sun and constitutes the main driver of temperature increase. The latter is mainly controlled by the amount of radiation absorbed or reflected by the material, the moisture and air contents of soil and any factor that influences the evaporation rate of the material (Bell, 2002:46). The slope of the TSF has an influence on the amount of radiation received. For example, in the southern hemisphere, a north-facing aspect will be more exposed to the sun and therefore warmer. Furthermore, dry material is more reflective than moist material; the amount of radiation reflected from moist soil material varies between 10 and 15%. The colour of the soil material also influences the amount of radiation reflected: Darker-coloured material, such as grey platinum and black coal tailings, tend to reflect less and absorb more light radiation than lighter-coloured material such as yellow gold mine tailings (Bell, 2002:47). Tordoff *et al.* (2000:221) argue that even though the fine-textured and unstructured physical nature of mine tailings may severely inhibit vegetation establishment, it is the chemical characteristics of mine tailings contained within TSFs that principally inhibit plant growth.

2.3.4 Chemical characteristics of tailings

Chemically, TSFs are characterised by extreme acidity (pH[H_2O] two – five) or alkalinity (pH[H_2O] eight – nine), plant nutrients deficiencies, absence of organic matter, abundance in potentially toxic elements and elevated salinity levels (Gil-Loaiza *et al.*, 2016:452; Li *et al.*, 2006:43; Orlowska *et al*., 2010:186; Petrisor *et al.*, 2004:2; Santini & Banning, 2016:44; Tordoff *et al.*, 2000:221; Touceda-González *et al.*, 2017:301; Yang *et al.*, 2016:428; Ye *et al.*, 2002:1104).

- **pH of tailings material**

Soil pH is a chemical parameter that can be expressed as pH = -Log (H^+) (Sparks, 2003:267; Viljoen, 2014:2). According to Hodson and Donner (2013:218) and Sparks (2003:267), it is widely considered as the ‘master variable’ that controls both soil chemistry and plant nutrition, as it regulates key aspects of soil chemistry relevant to plant growth. These aspects include the availability of nutrients to plant roots, the growth of plant roots, the availability and toxicity of metals

and microbial activity within soil. Soil pH is used to classify soils into functional groups of acidity or alkalinity (Table 2.1).

Table 2.1: Soil pH(H2O) ranges and relevant descriptions [adapted from Hodson & Donner (2013:218) and Sparks (2003:267)].

pH range	***Description***
<4,5	Extremely acidic
4,5–5,5	Moderately acidic
5,5–6,5	Slightly acidic
6,5–7	Neutral
7,5–8,5	Slightly alkaline
8,5–9,5	Moderately alkaline
>9,5	Extremely alkaline

Certain TSFs, including those of gold, copper and coal, contain small amounts of sulphide minerals such as pyrite (FeS_2), sphalerite (ZnS), galena (PbS), arsenopyrite (FeAsS) and chalcopyrite ($CuFeS_2$). The presence of sulphide minerals such as pyrite within tailings coherently oxidise in the presence of water and air. This leads to the production of sulphuric acid, which in turn lowers the pH of the tailings material and results in moderate or extreme levels of acidity (Kossoff *et al.*, 2014:231; Li *et al.*, 2016:153; McGregor & Blowes, 2002:195; Mhlongo & Amphosa-Dacosta, 2016:4; Shu *et al.*, 2001:390; Tordoff *et al*., 2000:221; Weiersbye *et al.*, 2006:104; Yang *et al.*, 2010:852, Yang *et al.*, 2016:428). Akcil and Koldas (2006:1139) add that mining processes promote acid generation because, through the excavation and processing of minerals, it increases the surface area of sulphide minerals exposed to oxidising environments.

Akcil and Koldas (2006:1141) summarise the primary factors that control the rate of acid generation as the following: Initial pH, Oxygen content of the gas phase, Oxygen concentration in soil water, Degree of water saturation, Chemical activity of ferric iron, Surface area exposed of the metal sulphide, Temperature and bacterial activity.

The pH of gold TSFs is very low (acidic). This is because sulphide mineral oxidation and acid generation mainlyoccur through three reactions. The first reaction (Equation 2) entails the oxidation of pyrite in the presence of oxygen and water to produce sulphate (SO_4^{2-}), dissolved ferrous iron (Fe^{2+}) and free hydrogen cations (H^+) (Li *et al.*, 2016:153; Akcil & Koldas, 2006:1141); the resulting free H^+ ions produced indicate an increase in the total dissolved solids and acidity:

Equation 2: $FeS_2 + 7/2\ O_2 + H_2O \rightarrow Fe^{2+} + 2SO_4^{2-} + 2H^+$

The Fe^{2+} produced by the first oxidation reaction (Equation 2) is then oxidised to produce ferric iron (Fe^{3+}) as illustrated in Equation 3 below:

Equation 3: $Fe^{2+} + 1/4O_2 + H^+ \rightarrow Fe^{3+} + 1/2H_2O$

Through the hydrolysis and precipitation of Fe^{3+} complexes and minerals, additional sulphuric acid, Fe^{2+} and free hydrogen cations are produced (Equation 4):

Equation 4: $FeS_2 + 14Fe^{3+} + 8H_2O \rightarrow 15Fe^{2+} + 2SO_4^{2-} + 16H^+$

Li *et al*. (2016:153) continue to explain that the free Fe^{2+} obtained from sulphide oxidation may be further oxidised, hydrolysed and precipitated as ferric oxyhydroxide ($Fe(OH)_3$) (Equation 5). Akcil and Koldas (2006:1139) add that the following reaction occurs in a pH range of 2,3–3,5:

Equation 5: $Fe^{2+} + 1/4O_2 + 5/2H_2O \rightarrow Fe(OH)_3 + 2H^+$

However, it is well-known that, under acidic conditions, acidophilic bacteria catalyse the oxidation of sulphides and increase the number of sulphate and protons produced (Akcil & Koldas, 2006:1141; Elberling *et al.*, 2000:226; Li *et al.*, 2016:154; Tordoff *et al.*, 2000:221). Elberling *et al.* (2000:226) explain that, under severely acidic conditions, chemolithotrophic bacteria, such as *Thiobacillus ferrooxidans* and *Leptospirillum ferrooxidans*, catalyse the oxidation of Fe^{2+} (Equation 3). This provides more available Fe^{3+} for sulphide oxidation (Equation 4) and further lowers the pH of the tailings. Akcil and Koldas (2006:1141) state that *Acidithiobacillus ferrooxidans* is frequently used when researching the oxidation of pyrite. It has been found to be most active at a pH of below 3,2. However, if conditions are not suitable for acid-promoting bacteria, their contribution towards iron oxidation is minimal.

Acidity directly affects plants through elevated concentrations of hydrogen ions that consequently deactivate most enzyme systems, restrict respiration and decrease the uptake of minerals and salts through plant roots (Shu *et al.*, 2001:389; Yang *et al.*, 2010:852). Viljoen (2014:3) explains that high hydrogen ion concentrations, which typically occur in acidic soil conditions, damage the

root cell membranes of plants. The extent of cell membrane damage is specific to the plant species and its relevant tolerance levels.

To decrease soil acidity, the soil may be amended with alkaline materials such as lime. Lime provides conjugate bases of weak acids that consume hydrogen ions and raises the pH level. Examples of such conjugate bases are hydroxide (OH^-), carbonate (CO_3^{2-}) and silicate (SiO_3^{2-}). These bases are most commonly supplied in their calcium (Ca) or magnesium (Mg) forms such as calcitic lime ($CaCO_3$) and dolomitic lime ($CaMg(CO_3)_2$) (Brady & Weil, 2008:387). Brady & Weil (2008:387) explain that when the liming material is applied to an acidic substrate, it reacts with carbon dioxide and water to yield bicarbonate. This is illustrated in Equation 6, which shows the reaction of dolomitic lime, one of the most common limestones used for acid neutralisation. After this increase in alkalinity, the establishment of vegetation during the rehabilitation process is enhanced.

Equation 6: $CaMg(CO_3)_2 + 2H_2O + 2CO_2 \rightarrow Ca + 2HCO_3^- + Mg + 2HCO_3^-$

Ca and Mg bicarbonates are much more soluble than the carbonate, enabling it to react with the exchangeable and residual acidity. Ca and Mg ions replace the hydrogen and aluminium ions in the colloidal complex, causing the formation of solid aluminium hydroxide ($Al(OH)_3$). This decreases the percentage acid saturation in the colloidal complex, correspondingly increasing the soil solution pH (Brady & Weil, 2008:387).

However, not all tailings are acidic and even gold mine tailings may be alkaline as reported by Orlowska *et al*. (2010:186), who worked on gold mine tailings in Barberton, Mpumalanga Province, South Africa, where the pH levels were above 8,5. Platinum mine tailings derived from milling and processing of the rocks in the Bushveld Igneous Complex in South Africa tend to have alkaline pH values and do not readily acidify. The rocks that contain the platinum group of minerals are pyroxenite, norite and anorthosite (Van Deventer *et al.*, 2008:213). They are principally composed of minerals such as pyroxene, plagioclase, amphibole and olivine. When milled and processed, these minerals have a high pH of 10–12 due to hydrolysis and/or oxidation of the minerals (Van Deventer *et al.*, 2008:213). Equations 7 and 8 illustrate the hydrolysis of plagioclase and the oxidation of amphibole (Van Deventer *et al.*, 2008:213), which explains the high pH values of platinum mine tailings.

Equation 7: $Ca_2Al_2Si_2O_8 + 9H_2O \rightarrow Ca(OH)_2 + Al_2Si_2O_5(OH)_4$

In Equation 7, plagioclase ($Ca_2Al_2Si_2O_8$) is hydrolysed to produce a base ($Ca(OH)_2$) and a clay mineral ($Al_2Si_2O_5(OH)_4$) (Van Deventer *et al.*, 2008:213).

Equation 8: $4CaFeSi_2O_6 + O_2 + 4H_2CO_3 + 6H_2O \rightarrow 4FeOOH + 8H_2SiO_3 + 4CaCO_3$

Equation 8 illustrates how the mineral amphibole ($CaFeSi_2O_6$) is oxidised to produce lime ($CaCO_3$) (Van Deventer *et al.*, 2008:213).

- **Metal mobility and toxicity**

According to Hodson and Donner (2013:195), potentially toxic elements occur naturally in all soils and are toxic if exposed to plants in high enough concentrations. The mobility of a metal is dependent on its concentration in the soil solution and the movement of that solution through the soil. The availability of metal solutions to plants depends on the solubility of the metal, which, in turn, depends on the pH value controlling metal mobility and availability (Hodson & Donner, 2013:196). Hodson and Donner (2013:196) report that the phytotoxicity of metals associated with mine tailings tend to increase in acidic conditions, mainly due to an increase in their solubility (Epelde *et al.*, 2014:1; Mendez & Maier, 2008b:47; Petrisor *et al.*, 2004:1; Touceda-Gonzáles *et al*., 2017:301; Wa Ilunga *et al.*, 2015:214; Wijesekara *et al.*, 2016:102; Yang *et al.*, 2016:428). Yang *et al.* (2016:428) explain that metal ions, including Al, Cd, Hg, Cr, Cu, Fe, Mn, Pb and Zn become mobile and available to the plant in phytotoxic concentrations under strong acidic conditions. Li (2006:43) lists the metals according to their degree of toxicity to plants as Hg>Pb>Cu>Cd>Cr>Zn and mentions that almost all mine tailings contain co-existing toxic elements. Wijesekara *et al.* (2016:100) explain that a common feature of mine tailings is the presence of metal trace elements in elevated concentrations, which severely affect microbial activity and decomposition of organic matter to form humus.

George *et al.* (2012:417) state that the main constraints to plant growth within acid mineral soils are the toxicity of hydrogen ions (H^+), Al and Mn which manifests itself in plants mainly by the inhibition of plant root elongation and root death. The pH at which H^+ become toxic varies between species and the molecular mechanisms through which toxicity occurs are still not fully understood. However, three principal mechanisms are known, including (1) disruption of cell wall integrity, (2) interference with the maintenance of plant cytosolic pH and (3) inhibition of proton uptake (George *et al.*, 2012:418).

- **Nutrient status**

The lack of plant nutrients impedes plant establishment and growth, especially during the rehabilitation of TSFs (Pardo *et al.*, 2017:556; Wong, 2003:776). Jones (2012:18) provides the following three criteria for plant nutrient essentiality:

- Omission of the element must result in abnormal growth, failure of the plant to complete its life cycle or premature plant death.
- Element must be specific and not replaceable by another element.
- Element must directly impact the growth and metabolism of the plant and not have an indirect effect such as the antagonization of another element present at a toxic level.

Dickinson (2002:51) and Jones (2012:21) explain that there are 13 chemical elements that are considered as essential for the growth of higher plants. These elements are divided into two categories, namely macro- and micronutrients, based on their concentrations required by plants. These plant nutrients with their roles in plant growth are summarised in Table 2.2.

Table 2.2: Essential macro- and micronutrients for plants and their functions in plant growth (Jones, 2012:23).

Macronutrients	**Function**	**Micronutrients**	**Function**
Nitrogen	Combines with carbon, hydrogen, oxygen, and sometimes sulphur (S), to form amino acids, enzymes, nucleic acids, chlorophyll and alkaloids. Principally occurs as high-molecular-weight proteins in plants.	Boron	Involved in cellular activities, division, differentiation and maturation, amongst others. Important for synthesis of RNA. Associated with pollen germination, growth and improvement of pollen tube stability.
Phosphorus	Component of adenosine triphosphate (ATP), which is involved in various energy transactions, ribonucleic acids (RNA) and deoxyribonucleic acids, which are components of genetic information.	Chlorine	Contributes to the oxygen evolution in photosystem II during photosynthesis. Raises cell osmotic pressure. Influences stomatal regulation. Increases hydration of plant tissue.
Potassium	Maintains water status and cell turgor pressure. Regulates opening and closing of stomata. Required for the accumulation and translocation of newly formed carbohydrates.	Iron	Important component of many plant enzyme systems such as electron transport and the terminal respiration step. Component of protein ferredoxin and required for NO_3 and SO_4 reduction, N assimilation and energy production. Functions as a catalyst for chlorophyll formation.

Macronutrients	Function	Micronutrients	Function
			Thought to be involved in protein synthesis and root-tip meristem growth.
Calcium	Involved in maintenance of cell integrity and membrane permeability.	Manganese	Involved in oxidation-reduction processes in photosynthetic electron transport system. Essential in photosystem II for photolysis and acts as a bridge for ATP and enzyme complexes.
Magnesium	Component of the chlorophyll molecule. Cofactor in most enzymes activating phosphorylation processes bridging structures of ATP or adenosine diphosphate and enzyme molecules	Molybdenum	Component of two major enzyme systems, namely nitrogenase and nitrate reductase.
Sulphur	Involved in protein synthesis. Forms part of amino acids cysteine and thiamine. Reduces the disease incidence.	Zinc	Has similar enzymatic functions to manganese and Mg, with only carbonic anhydrase being activated by zinc.
		Copper	Constituent of the chloroplast protein plastocyanin. Participates in protein and carbohydrate metabolism. Involved in the desaturation and hydroxylation of fatty acids.

Plants obtain plant nutrients to grow and complete their life cycle from the soluble soil solution. A relatively small number of nutrients are available in soluble form to plants when compared to the number that are associated with organic and inorganic soil solids. Consequently, as plant roots extract and deplete nutrients from the soil solution, it must be constantly replenished (Brady & Weil, 2008:25). The main mechanism through which this is accomplished is through ion exchange (Brady & Weil, 2008:25; Kabata-Pendias, 2011:57; Sparks, 2003:187).

Tiny colloidal-sized particles, such as clay and humus, have charged surfaces, where oppositely charged ions can be adsorbed and held as exchangeable ions. Through exchange with other ions

from the soil solution, the adsorbed elements can escape this state of electrostatic adsorption to the particle surface into the soil solution, where it is available to plant roots for uptake (Brady Weil, 2008:25; Kabata-Pendias, 2011:57). The capacity of soil to adsorb and exchange cations is known as the cation exchange capacity (CEC) and is measured in centimoles of charge per kg ($cmol_c/kg$) (Brady & Weil, 2008:337; Sparks, 2003:187).

The availability of plant nutrients varies according to the soil solution pH. A nutrient availability chart obtained from the Fertilizer Society of Southern Africa handbook (FSSA, 2007) that depicts the behaviour of macro- and micronutrients in terms of plant availability is given in Figure 2.1.

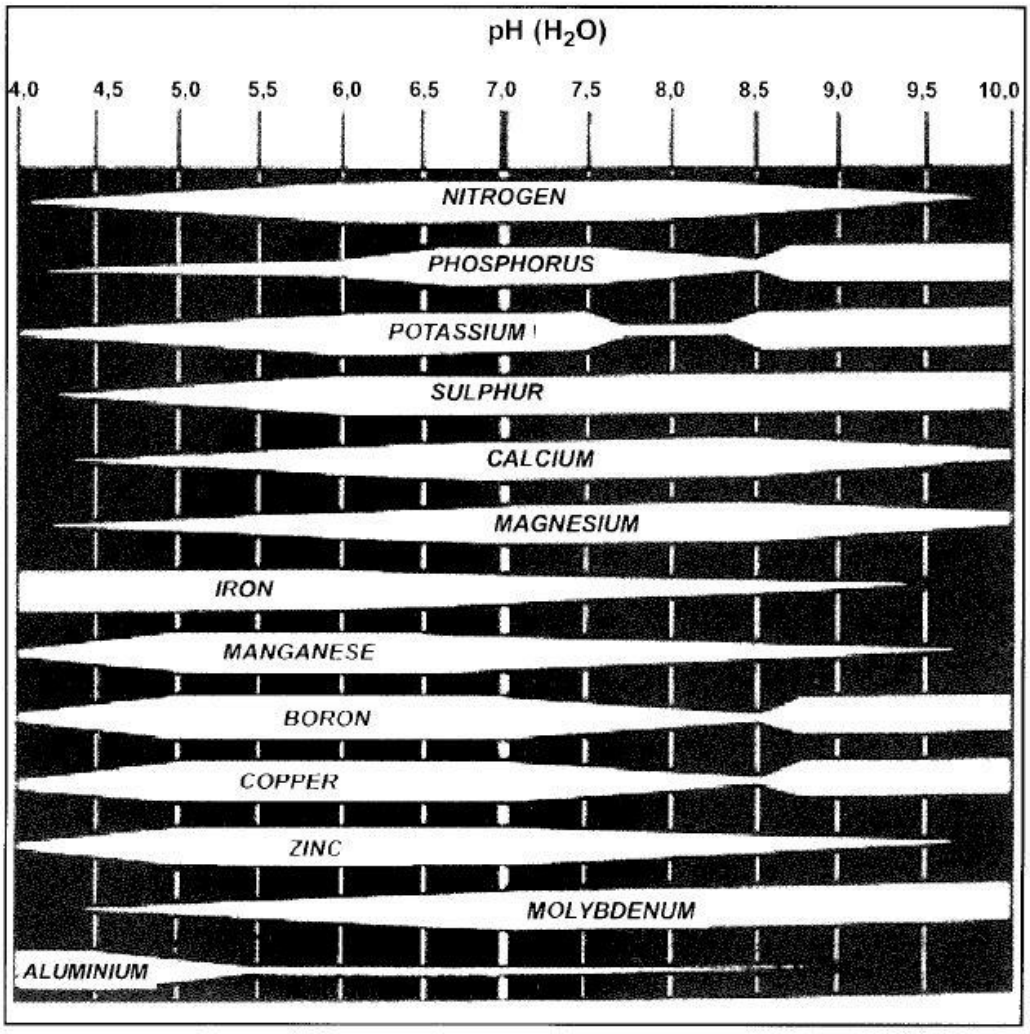

Figure 2.1: Nutrient availability chart (FSSA, 2007:94).

It is evident that the availability of each nutrient is unique at different pH levels. The sharp decrease in the availability of the macro-nutrients nitrogen (N), phosphorus (P), potassium (K), S,

Ca and Mg is notable below a pH of 5,5 and the optimal pH for the plant availability of both macro- and micronutrients is between a pH of 6,5 and 7 (FSSA, 2007:93).

Dickinson (2002:52) explains that N is a plant nutrient required in large amounts. N is absorbed as ammonium (NH_4^+) and nitrate (NO_3^-). The main sources of N in soils are decomposed organic materials and N - fixation by legumes (Dickinson, 2002:56). However, organic matter is deficient in most TSFs (Dickinson, 2002:56). Orlowska *et al.* (2010:186) state that mine tailings are almost devoid of plant nutrients, principally macro-nutrients such as N, P and K. Straker *et al.* (2007:219) support Orlowska *et al.* (2010:186) in that gold mine tailings are highly polluted environments where especially P and N are not sufficiently present.

- **Salinity**

The degree of salinity, which is characterised by the concentration of soluble salts, is expressed in terms of the electrical conductivity (EC) of a saturated extract (George *et al.*, 2012:456; Viljoen, 2014:4). The soil solution extracted from a soil at its saturated water content is the saturation extract (George *et al.*, 2012:456). The EC of the soil solution, measured in the SI unit siemens (S), is most commonly used as the working unit mS/cm or dS/m (Viljoen, 2014:4). A soil is classified as saline when the saturation extract has an EC in excess of four dS/m (George *et al.*, 2012:456; Hodson & Donner, 2013:206).

Ions that usually cause saline conditions are nitrates (NO_3^-), sulphates (SO_4^{2-}) and chlorides (Cl^-) of Mg, K, Ca and sodium (N)a (Hodson & Donner, 2013:206). George *et al.* (2012:456) state that Na^+, Ca^+, Mg^{2+} and, to a lesser extent, Fe^{2+} and K^+ are common cations and Cl^-, NO_3^-, SO_4^{2-}, HCO_3^- and CO_3^{2-} are the most abundant anions occurring in soils. Straker *et al.* (2007:204) explain that the elevated EC of gold mine tailings is principally due to large concentrations of SO_4^{2-} and not Cl^-. Viljoen (2014:4) provides a general summary of the influence of different soil salt concentrations on plant growth (Table 2.3).

Table 2.3: Effect of soil salt content on plant growth (Viljoen, 2014:4).

Effect	**EC (mS/m)**
Effect of salt quality is negligible	<200
Plants sensitive to salinity are affected	200–400
Effect of salinity is more pronounced and plant growth is affected	400–800
Only species tolerant to salinity are able to survive	800–1600
Extreme salinity – no plant species are able to survive	>1600

The greatest negative effect of high soil salinity is the reduction of soil water available to plants. This is also known as the osmotic effect. The growth rate and size of plants are reduced in conditions of salinity stress that occur at EC values greater than four dS/m (George *et al.*, 2012: 456; Viljoen, 2014:4). Substrate salinity mainly constricts plant growth in three ways:

- It lowers water potential, which leads to a deficient amount of water available to plants in the rooting medium.
- It leads to the toxicity of ions resulting from the excessive uptake of mainly sodium and chloride ions.
- It results in nutrient imbalances within the plant cells, restricting nutrient uptake from the soil solution, impairing transport of nutrients through root and shoot tissues and disrupting internal distribution of nutrients throughout plant organs (George *et al*., 2012:458; Hodson & Donner, 2013:206).

2.3.5 Organic matter and microbiological factors

Powlson *et al.* (2013:86) regard soil organic matter as the difference between soil simply being a collection of inert mineral particles and it being a functioning growth medium with a recognisable physical structure. Wijesekara *et al.* (2016:100) explain that the near absence of organic matter in TSFs is a common problem and considered as one of the main reasons for tailings being poor host mediums for plant and microbial life.

- **Organic matter**

According to the FSSA (2007:27), organic matter may be seen as the total fraction of soil comprised of organic substances. This includes living organisms of various sizes, organic residues in various stages of decomposition and dark-coloured humus consisting of non-humic and humic substances. Jones (2012:65) defines soil organic matter as the sum of all the natural and thermally altered, biologically derived material, living or dead, found in the soil or on the soil surface, irrespective of its source or stage of decomposition, and excluding aboveground portions of living plants.

Due to the interrelated nature of soil physical properties, the addition of organic matter, which commonly has a much lower bulk density than most mine tailings, simultaneously enhances most physical characteristics of tailings (Wijesekara *et al.*, 2016:127). The FSSA (2011:33) lists three ways in which organic matter primarily enhances soil physical functions:

- It serves as a binding agent between mineral particles, promoting the formation of stable aggregates and resulting in lower soil densities and higher porosities.
- It improves soil structure ultimately improving water infiltration, gas exchange and root penetration.
- It improves soil stability against wind and water erosion.

The soil water-holding capacity is directly improved by the addition of organic matter due to the material's ability to absorb large quantities of water and indirectly by improving soil structure. Soil organic matter also provides soil with a darker colour, resulting in more radiation from the sun being absorbed, leading to warmer soil temperatures (FSSA, 2011:33). Jones (2012:67) supports the above-mentioned factors listed by the FSSA (2007:33) and adds that soil organic matter reduces surface crusting.

Furthermore, organic matter enhances many detrimental chemical conditions associated with mine tailings by buffering rapid pH changes, increasing CEC (as organic matter has a much higher CEC than inorganic colloids [clays]), improving nutrient content and reducing the available metal concentrations (Brady & Weil, 2008:517; Jones, 2012:67; Wijesekara *et al.*, 2016:127). Powlson *et al.* (2013:86) provide similar views to those of Wijesekara *et al.* (2016:135), stating that organic matter not only provides food and anchorage points to soil bacteria and fungi, but it is also a reservoir of many plant nutrients, such as N and P. It further provides a substantial part of a soil's CEC, which is responsible for the retention and exchange of many plant nutrients. The

interactions between organic matter, microorganisms and mineral particles form the basis of nutrient transformation on which all plant growth depends (Powlson *et al.*, 2013:86). Lastly, soil organic carbon within soil organic matter is the main energy source for soil microorganisms (Brady & Weil, 2008:517; Jones, 201267; Wijesekara *et al.*, 2016:135).

- **Soil microorganisms**

Soil microorganisms are an integral part of the plant rhizosphere, which includes free-living plant-root-associated symbiotic rhizobacteria and fungi (Khan, 2005:357; López-Bucio *et al.*, 2015:110). These soil bacteria and fungi are able to colonise plant roots and benefit the plant, for example, by improving its tolerance to disease and environmental stress as well as improving plant growth (Harmosa *et al.*, 2012:17). Plant–microbe associations involve recognition between the plant host and the microorganism. This occurs via a signalling network that is regulated by plant hormones, such as jasmonic acid, ethylene and salicylic acid (Harmosa *et al.*, 2012:17).

Plant growth-promoting rhizobacteria (PGPR) enhance plant growth in unfavourable conditions in a multitude of ways that include assisting phytohormone production, increasing specific enzymatic activity and atmospheric N fixation, providing bio-available P for plant uptake and producing antibiotic pathogen-depressing substances, such as chelating agents and siderophores, which help protect the plant against disease (Khan, 2005:357). Grandlic *et al.* (2009:1740) mention that PGPR introduced to metal-contaminated soils, such as mine tailings material, can potentially change the structure of the microbial community, enhancing plant growth. Burges *et al.* (2016:481) state that PGPR isolated from species growing in metal-contaminated soils have successfully demonstrated their potential for phytostabilisation, mainly through their ability to stimulate plant growth and protect against metal toxicity.

Khan (2005:358) further explains that N-fixing rhizobacteria are chemotactically attracted to legume roots by root exudates. The rhizobacteria attach to and colonise the root surface to activate rhizobial nodulation genes. Saba *et al.* (2012:525) explain that the fungal genus *Trichoderma* is highly interactive in plant root, soil and foliar environments. This genus consists of more than 200 molecularly defined species (López-Bucio *et al.*, 2015:110). *Trichoderma* is widely used as a bio-fertiliser or -stimulant due to its ability to establish mycorrhizal associations with plants and act as a bio-control agent against phytopathogenic fungi (Kaveh *et al.,* 2011:169). The various species of *Trichoderma* control soil-borne plant pathogens more effectively than industrial chemicals do and promote plant growth (Saba *et al*., 2012:525; Vinale *et al.*, 2014:9761). Hermosa *et al.* (2012:17) further state that certain *Trichoderma* rhizosphere-

competent strains stimulate direct effects on plants by means of root interactions. This direct root interaction increases the percentage and rate of seed germination, plant growth potential, nutrient uptake efficiency, increases fertiliser use efficiency and it stimulates plant defences against abiotic and biotic factors (Hermosa *et al*., 2012:17). López-Bucio *et al.* (2015:110) elaborate that by reprogramming the gene expression within plant roots and shoots, *Trichoderma* species are able to improve the resistance of plants to environmental stresses, such as drought and salinity. The fungal mycelium also secretes compounds that increase the branching capacity of the root system, increasing the capacity of the plant to absorb nutrients and water.

Von Mersi & Schinner (1990: 216) explain that a general criteria used to measure the biomass of microbial populations present in soil, is the dehydrogenase activity (DHA). Dehydrogenase is an intracellular enzyme catalysing the oxidation of organic compounds through the separation of hydrogen atoms to take part in biosynthesis processes. This occurs within the enzyme system of all microorganisms. The DHA is therefore a measure of the oxidative activities of microorganisms present in soils.

In a study conducted by Petrisor *et al.* (2004:9) to evaluate the artificial inoculation of sulphidic mine tailings with microbial communities, DHA was used to measure the overall microbial population size.

Organic matter and soil microorganisms are able to improve the establishment of vegetation on TSFs to increase the success of phytostabilisation.

2.4 Phytostabilisation as part of phytoremediation

Phytoremediation is an in-situ remediation technology where plants are used to clean up soils, sediments and water that have been contaminated with organic pollutants or potentially toxic elements (Khan, 2005:356). Its aim is to use metal-tolerant plants to remove, transfer or stabilise contaminants from and in soil, sediments and water within the rhizosphere (Khan, 2005:356). Wong (2003:777) describes phytoremediation as ‘the use of plants and their associated microbial, soil amendments and agronomic techniques to render contaminants harmless to the environment’. Khan (2005:357) explains that phytoremediation has five sub-categories (Table 2.4).

Table 2.4: Five sub-categories of phytoremediation (Khan, 2005:357; Wong, 2003:777-779).

Category	**Description**
Phytoextraction	Potentially toxic elements are concentrated and removed into harvestable plant parts.
Phytodegradation	Potentially toxic elements are degraded by the plants and their associated microbes.
Rhizofiltration	Potentially toxic elements are absorbed from contaminated water by plant roots.
Phytostabilisation	Potentially toxic elements are immobilised and their bioavailability decreased in the plant rhizosphere.
Phytovolatilisation	Potentially toxic elements are volatilised through plants from the soil and into the atmosphere.

Wong (2003:777) supports Khan (2005:357) by stating that phytoremediation techniques aimed at immobilising potentially toxic elements in contaminated soils mainly include phytostabilisation and phytoextraction.

2.5 Background of phytostabilisation

The basic principle of phytostabilisation is to prevent erosion and runoff or leaching of chemicals or to convert pollutants within the substrate to less bio-available forms (Orlowska *et al.*, 2010:186). When dealing with mining environments where vegetation has been removed or is entirely absent, as in the case of TSFs, natural colonisation and succession processes are too slow. Cooke and Johnson (2002:49) explain that the presence of populations or species at a specific site depends on the ability of the surrounding species' propagules that have been transported to the site to germinate, survive and reproduce. The time lag for colonisation depends on the severity of conditions associated with the site and substrate. This may leave rehabilitated mining sites devoid of vegetation for extensive periods of time, which can be resolved by phytostabilisation (Bradshaw, 2000:9; Mendez & Maier, 2008a:278; Tordoff *et al.*, 2000:220; Young *et al.*, 2013:498; Zhang *et al.*, 2001:378).

Tordoff *et al.* (2000:220) state that it is possible to stabilise mine tailings in Europe and achieve complete long-term rehabilitation by means of vegetation. Yang *et al.* (2010:852) support Tordoff *et al.* (2000:220) by stating that once a re-vegetation programme has been successfully

implemented, it will not only sufficiently achieve the objectives of stabilisation, pollution control, visual improvement and removing environmental threats to humans, but will also improve specific properties such as biodiversity, vegetation structure and ecological processes that are lost from the ecosystem through disturbance.

Mendez and Maier (2008a:279) describe phytostabilisation as 'the use of stress tolerant plants to immobilise contaminants in the plant rhizosphere'. Burges *et al.* (2016:481) support Mendez and Maier (2008a:279) by stating that phytostabilisation is regarded as an effective phytomanagement option for mine TSFs and entails the establishment of metal-tolerant plants to decrease the bioavailability and mobility of metals within the soil. Shutcha *et al.* (2015:82) describe phytostabilisation as a restoration approach where plant cover consisting of metal-tolerant species (metalophytes) is used in combination with soil amendments, which reduces metal dispersion through water and wind. The latter is also reported by Gil-Loaiza *et al.* (2016:452), Touceda-González *et al.* (2016:301) and Young *et al.* (2015:250).

Mendez and Maier (2008a:279) mentions that the established vegetation canopy reduces wind dispersion of tailings particles, while plant roots prevent water erosion. Simultaneously, metals are immobilised in the rhizosphere where they accumulate and are stabilised through adsorption and precipitation. Petrisor *et al.* (2004:2) further explain that the establishment of vegetation on TSFs not only reduces wind and water erosion, but is also aesthetically pleasing. Saadani *et al.* (2016:264) explains that species with high biomass production and greater ground cover are important for successful phytostabilisation.

The main objectives of phytostabilisation practices on TSFs are the germination and establishment of a vegetative cap that, through plant succession, leads to a stable vegetation community over time (Gil-Loaiza *et al.*, 2016:452). This encourages soil development processes, increases microbial diversity and eventually leads to the rehabilitation of soil ecosystem functionality to a self-sustainable state (Mendez & Maier, 2008a:279). Various studies have demonstrated the positive use of phytostabilisation as a phytomanagement tool in the rehabilitation of TSFs.

Field trials conducted by Touceda-González *et al.* (2016) on bare copper mine tailings in Spain amended with compost prove that the physio-chemical properties of the tailings, as well as the establishment of biogeochemical cycles that were previously absent, were improved by phytostabilisation (Touceda-González *et al.*, 2016:312). Likewise, Yang *et al.* (2016) successfully established five metal-tolerant plants in acidic pyritic metal-contaminated mine soil amended with

lime and chicken manure as part of a case study. Yang *et al.* (2016:433) further report that the establishment of good vegetation cover within six months, which may lead to improved organic matter content and nutrient status, is coherent with vegetation development over a two-year study period. The first step towards successful phytostabilisation of TSFs is to ensure the germination of sown seed.

2.6 Seed germination in TSF environments

Desai (2004:51) describes seed germination as the emergence of an embryo from the seed by the beginning of various catabolic and anabolic activities after water absorption, which include respiration, protein synthesis and food reserve mobilisation. Temperature, oxygen, light, carbon dioxide and factors that influence the availability of water are some of the main environmental factors that control seed germination. These factors also lead to secondary dormancy of seed, which can be described as dormancy caused by unfavourable environmental conditions after seed dispersal (Desai, 2004:74)

In arid and semiarid regions vegetation establishment on TSFs is further challenged by environmental factors that include low precipitation, high temperatures at the tailings surface, and high winds (Mendez & Maier, 2008a:278). Water stress has a significant influence on seed development and maturation. With an increase in water stress, germination, which is often characterised by radicle emergence, may be delayed, thereby impeding the germination process. Extreme temperatures may also exacerbate adverse osmotic effects, which differ for various species and cultivars (Desai, 2004:74). High evaporation rates and low water infiltration promote the development of saline conditions (Mendez & Maier, 2008a:278). Stress caused by high salinity, as found in TSFs, often prevents the uptake of water due to the unfavourable water potential gradient between the seed and the surrounding soil. It also increases the absorption of toxic ions into the developing embryo or seedling (Desai, 2004:77). Seed germination and seedling development are much more susceptible to salinity stress than subsequent growth phases are (Desai, 2004:77; Viljoen, 2014:4). To alleviate difficulties associated with seed germination on TSFs seed can be enhanced by coating it with various beneficial materials.

2.7 Seed coatings to increase vegetation establishment on TSFs

Scott (1998:198) defines seed coating as the application of finely ground and dissolved solids or liquids that form a continuous layer covering the natural seed coat (testa). Scott (1998) further states that, depending on the coating material, the coating may provide the seed with nutrients for better establishment following germination. Leinauer *et al.* (2010:179) explain that seed

coating was originally introduced by the vegetable industry between the late 1930s and early 1940s for the purpose of increasing seed size and achieving seed germination uniformity. This practice has expanded over time and seed coatings are now used to supply various beneficial compounds that may include macro- and micronutrients, insecticides, fungicides, growth regulators, pesticides and a variety of chemicals to improve the micro-environment surrounding the seed during germination, establishment and growth. Pederini *et al.* (2017:106) agree with Scott (1998:198) and Leinauer *et al.* (2010:179) and explain seed coating is the practice of covering seed with external materials to improve various physical aspects of the seed, which include the handling, protection and delivery of active ingredients, such as fertiliser and pesticides.

In trials conducted by Vartha and Clifford (1973:42), the coating of grass seed resulted in improved establishment. Vartha and Clifford (1973:42) conclude that the beneficial effects of grass seed coating are species-specific, especially regarding moisture uptake for germination and initial nutrition for the seedlings. Similar results were obtained by Dowling (1978:163) in a study carried out in New South Wales, New Zealand. The effect of seed coating on grass seeds was found to be dependent on both the grass species and time of sowing. Dowling (1978:166) further reports that the highest establishment of seed was during months with temperatures exceeding 18°C. The results obtained by Leinauer *et al.* (2010:180), who used a starch coating incorporating fertiliser, revealed a significant two-way interaction between the plant species and type of coating used. Vartha and Clifford (1973:42) conclude that improved grass establishment is not only species-specific, but also coating-specific. However, according to Dowling (1978:166) and Vartha and Clifford (1973:42), the effect of coatings on the survival of established grass seedlings is minimal. From the studies mentioned above, seed coating can enhance seed germination and seedling establishment and growth, but has a less positive effect on the long-term survival of the plants used in rehabilitation of TSFs. In summary, Dowling (1978), Vartha and Clifford (1973) and Leinauer *et al.* (2010) are positive about this intervention, but agree that there are many unanswered questions concerning the mode of action when using seed coatings to improve grass establishment.

Pederini *et al.* (2017:110) explain that the application of micronutrients such as P and K through seed coatings tends to improve the yield and growth of crops, but increases the risk of deleterious effects on seed germination through nutrient-induced osmotic stress. Pederini *et al.* (2017:106) elaborate that the market for the coating materials (polymers, colourants and bulking agents) in developing countries is expected to reach USD 1.63 billion annually by 2020. These authors add that major opportunities exist for the application of coating technologies in the field of restoration

and rehabilitation ecology, which can reach up to an estimated USD 18 billion per year, if the market by private companies is fully explored.

Considering the success rate for seedling establishment in rehabilitation and restoration programmes and that the United Nations has a global target to rehabilitate up to 150 million ha of degraded land by 2020, it is important to do more research on seed-based rehabilitation and restoration projects. Better seed-coating technologies can be a solution for improving seed-based restoration projects (Pederini *et al.*, 2017:107).

AgriCOTE is a modern seed coating application technology used to apply various beneficial agricultural products to the exterior wall of the seed. Materials such as growth stimulants, nutrients, pesticides, fungicides and protective polymers are applied in a gradual layering by a computer-assisted automated batching process using rotary equipment. This ensures consistent and evenly sized seed (AGT Foods Africa, 2010). Figure 2.2 is an illustration of the various layers surrounding the seed once coated using the AgriCOTE technology.

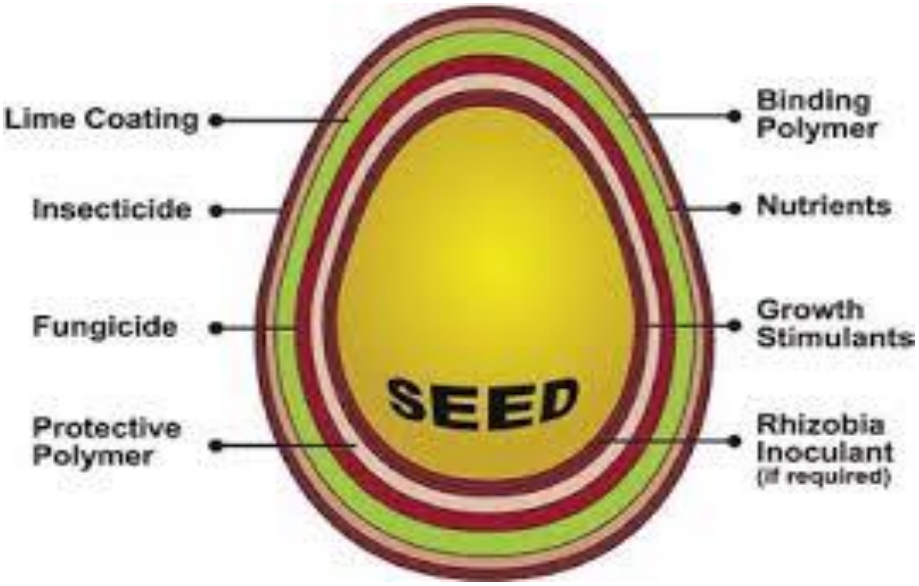

Figure 2.2: AgriCOTE coated seed displaying various coating layers (Nel, 2014).

According to Nel (2014), the use of AgriCOTE coated seed has multiple benefits:

- Coated seed are heavier, more uniform in size and less 'fluffy'[1]. This improves physical handling and assists uniform distribution of seed during sowing.
- The seed coating provides a nutrient reservoir for emerging seedlings.

[1] Seeds with awns or that are covered in hair (NRCS, 2010).

- Seed coatings improve seed-soil contact, allowing better access to water and facilitating nutrient uptake.
- Fungicides and pesticides in the coating protect seed against fungal attack and predation by insects.

In a study conducted by Westcott (2011:93) at the North-West University however, she concluded that the seed coating process stimulates germination metabolism within seeds which only lasts for a finite period of time before stored energy within the seeds becomes depleted and oxidising enzymes result in loss of seed viablility. Westcott (2011:93) recommends that coated seed be sold and used as soon as possible after the coating process to ensure high and rapid germination rates.

2.8 Seeding rate

Burton *et al.* (2006:379) is of the opinion that the goals for establishing and managing grass and legume mixtures for the rehabilitation of TSFs are mainly to maximise foliage production, which is influenced by the seeding rate, which is the amount of seed applied to specific area. Results obtained by Burton (2003:1) suggest that seeding rate is an important consideration when attempting to re-vegetate rehabilitated land. If the seeding rate is too low, plant species will not fully occupy the growing space available and if it is too high, intense competition of seedlings may inhibit the growth and development of individual seedlings, mainly due to moisture stress (Harris, 1996:74). Under extremely high densities, plants undergo density-dependant mortality, indicating a waste of the original seed input, which may lead to severe economic losses (Burton *et al.*, 2006:379).

According to Burton *et al.* (2006:379), there is currently no standard methodology or guidelines in either the agronomy or mining sectors for the rehabilitation or restoration of degraded land or TSFs regarding the seeding rate that should be used in re-vegetation projects. The seeding rate varies according to TSF location, the aim of the re-vegetation project and the inherent physical and chemical properties of the tailings material contained in the TSF. Currently, there are no standards regarding seeding rates for the rehabilitation of TSFs or the use of coated seed mixtures for such projects and much research on these topics is still required. This gap is what led to the formulation of the aims and objectives of this study.

Chapter 3: Materials and methods

This Chapter describes the location of the study sites and materials used for the vegetation trials, as well as the methods used during the monitoring process to achieve the study objectives.

3.1 Study sites

The study was carried out at two gold mine TSFs at the Crown (Mooifontein) and Rooikraal Mines, respectively, and at the North-West University (NWU) Potchefstroom Campus's nursery for soil and plant research for mine rehabilitation.

3.1.1 Location of study sites in South Africa

Both the gold mine TSF sites were located in the Gauteng Province (Figure 3.1 to Figure 3.3), while the nursery site, where pot and platinum trials were conducted, was located in Potchefstroom, North-West Province (Figureb 3.1).

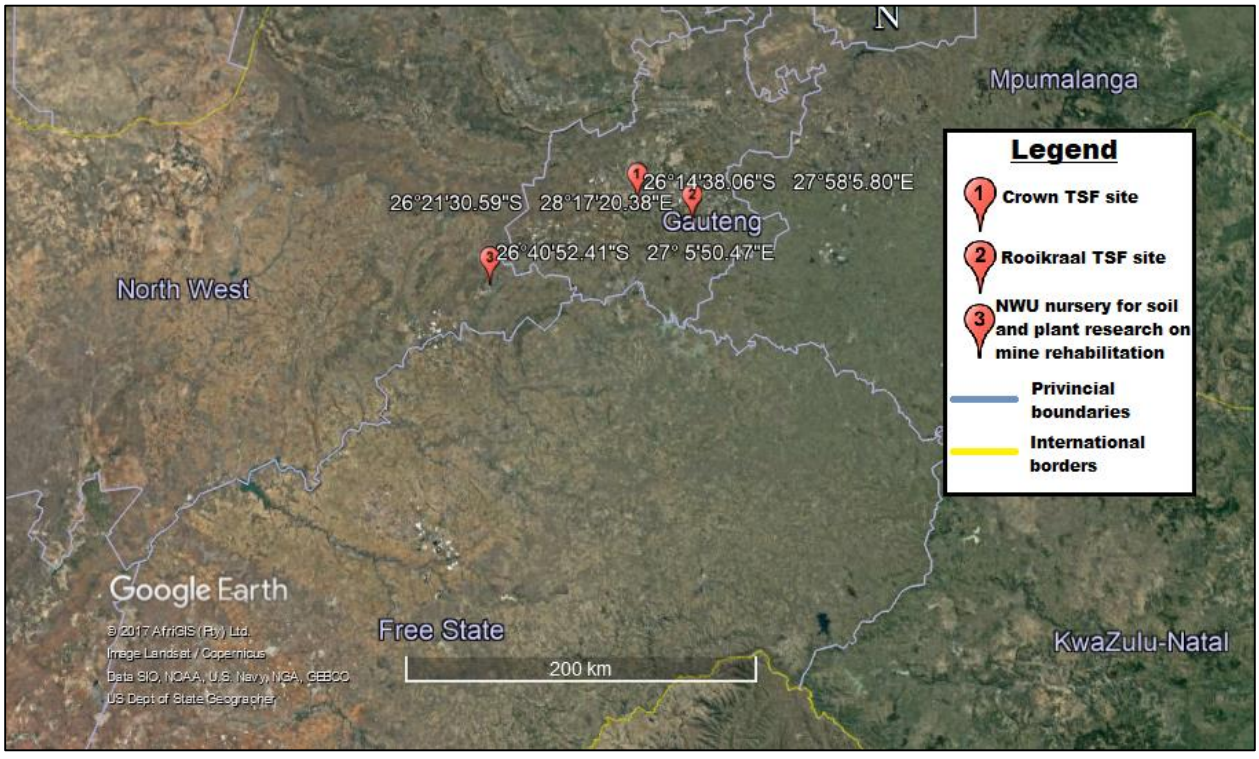

Figure 3.1: Location of study sites in South Africa (Google Earth, 2016a).

In July 1909, Crown Mines Ltd became the largest gold mining company in the world as the result of amalgamating various smaller mining companies, which included the Robertson Mine and the

original Crown Mine. The mining company ceased operation in 1977, at which time it had excavated 1 600 km of tunnels, some of which were up to three km underground.

The area used for the trial plots in this study was located on the southern part of the top-flat area of the TSF (highlighted in green in Figure 3.2). The area bordered with orange is the top-flat area of the TSF (Figure 3.2). The TSF's footprint covers an area of 171 ha (highlighted in red in Figure 3.2).

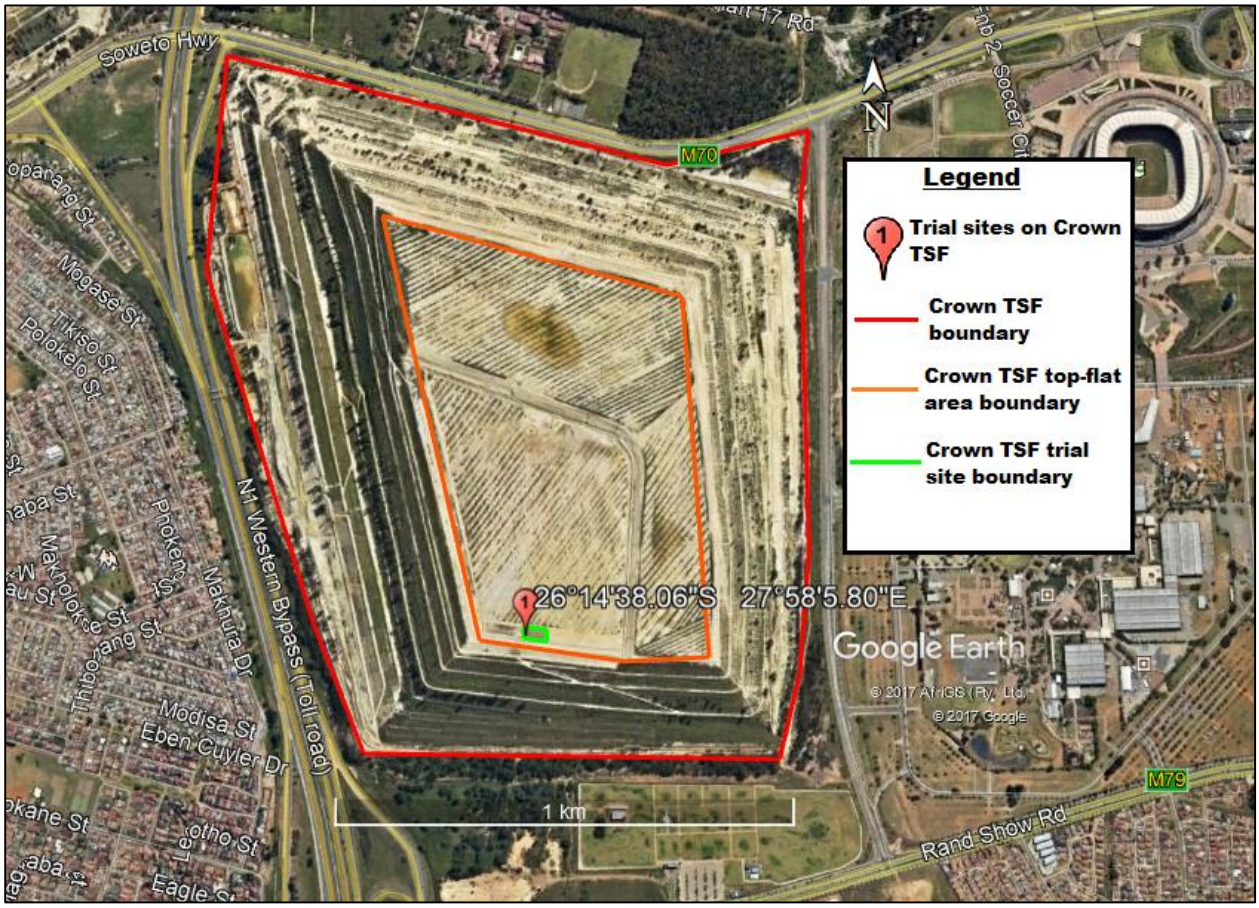

Figure 3.2: An aerial image of the Crown gold mine Mooifontein TSF used for the vegetation trials. The image shows the coordinates of the trial area (1) (Google Earth 2015a). The image is viewed at an altitude of 3,17 km.

Currently, rehabilitation of the TSF is carried out by Agreenco rehabilitation company[2], which is re-vegetating the slope, berm and top-flat areas of the TSF with a grass–legume mixture after

[2] Holdings 467, Vyfhoek, Potchefstroom, 2531.
PO Box 19896, Noordbrug, 2522.
www.agreencogroup.com

ameliorating and setting up irrigation lines. Previously established rows of *Eragrostis curvula* occur across the top-flat area north of the trial plot sites that were used for this study.

According to Botha and Van der Walt (2016:15), the Crown and Rooikraal gold TSF sites are located in the Mesic Highveld Grassland Bioregion of the Grassland Biome and the NWU nursery trial site in the Dry Highveld Grassland Bioregion of the Grassland Biome. According to Mucina and Rutherford (2006:48), the mean annual precipitation for the Mesic Highveld Grassland Bioregion is 726 mm/a, with a variation coefficient of 25% and the majority of rainfall events occurring between October and March. Thunderstorms generally occur in the area, which greatly increase erosion, especially in the slope areas of the TSF. The mean annual potential evaporation is recorded as 1958 mm/a, which is more than double the soil moisture supply by precipitation (e.g. rainfall) occurring only on 74% of days in the year. The mean annual temperature is 14,7°C, with 36 recorded frost days per annum (Mucina & Rutherford, 2006:48).

Tailings in the Rooikraal TSF were mined from the East Rand Goldfield, which extends from Benoni and Brakpan in a south-easterly direction towards Nigel and Heidelberg, Gauteng Province (McCarthy, 2011:1; Meyer & Stewart, 2012:3). The trial plots were located on the north-western berm area of the TSF. The TSF covers an area of 149,82 ha (Figure 3.3).

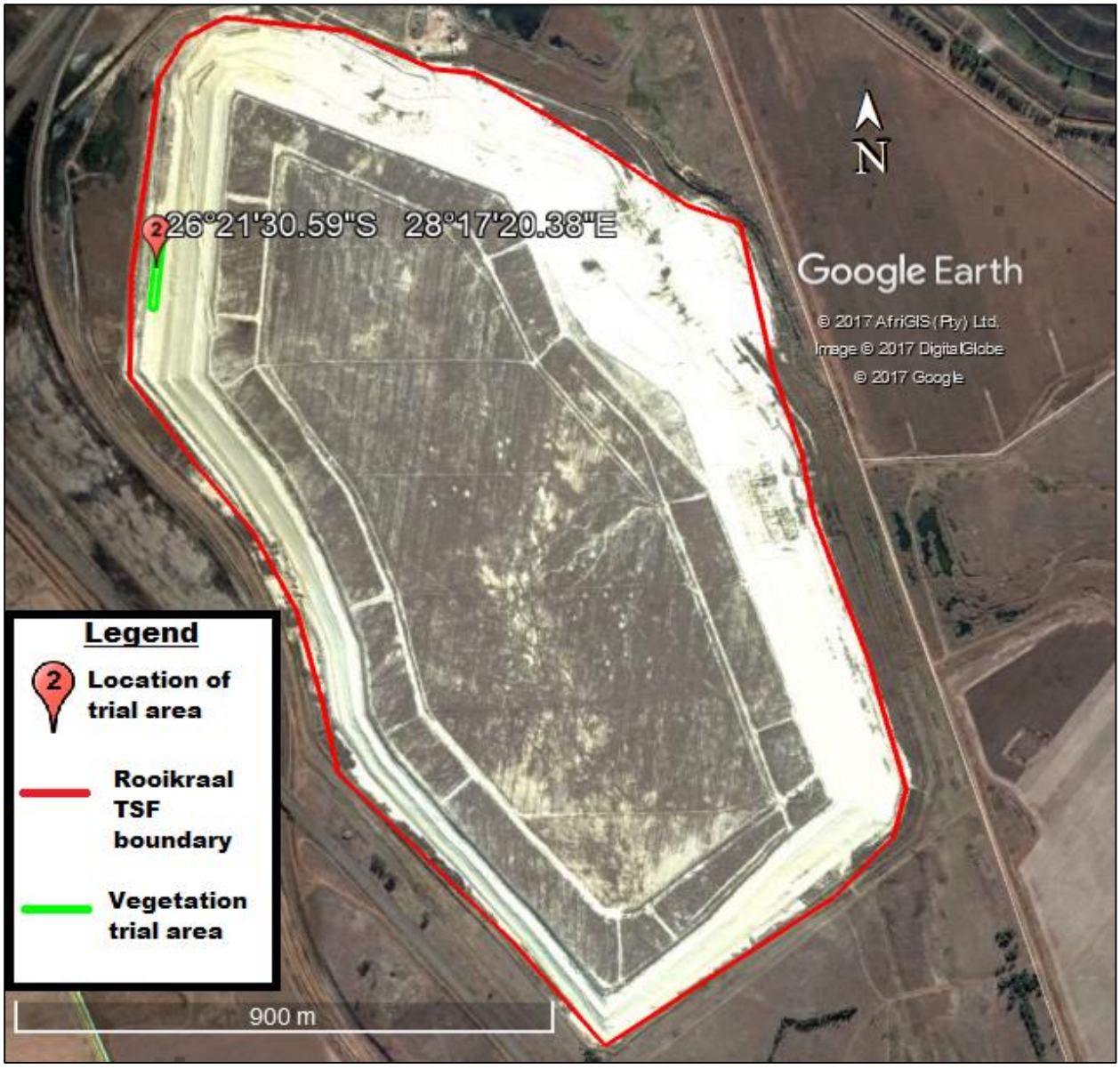

Figure 3.3: Aerial image of the Rooikraal Gold Mine tailings storage facility (TSF) site displaying the study area and GPS coordinates of the study site plots. (Google Earth 2015b) This image is viewed at an altitude of 2,83 km.

3.1.2 Location of study sites at gold mine TSFs

The berm area of the Rooikraal gold TSF, where the trial plots were situated, had an average slope of 10,8° at the top, decreasing to an average slope of 1,5° towards the north-west.

Slopes adjacent to the trial plot areas were devoid of vegetation and characterised by rill and gully erosion, the slopes are indicated in Figure 3.4 by a black arrow. The top-flat area of the TSF was well-vegetated, with *Eragrostis curvula* being the dominant species. It must be emphasised that the gold TSF sites at Crown Mooifontein and Rooikraal do not aim for closure. Rehabilitation of

the TSF sites is being carried out for the stabilisation of the TSF surfaces for re-mining in the future (Van Deventer, 2005)

Figure 3.4: Ameliorated trial plots, indicated by red arrows, during mulching after seed treatments were sown at the Rooikraal gold TSF next to TSF slopes characterised by gully and rill erosion on the right. TSF slopes are indicated by a black arrow. Photo taken by C.A. Kruger, 9/3/2017.

The Dry Highveld Grassland Bioregion has a mean annual precipitation of 498 mm/a with a variance coefficient of 31%. The mean potential annual evaporation is listed as 2327 mm with 80% of days in the year measuring evaporation rates double that of the soil moisture supply. The average annual temperature is recorded at 15,7°C with 42 annual frost days, where temperatures fall below 0°C (Mucina & Rutherford, 2006:48).

3.1.3 Geology of gold and platinum tailings

The Crown Mooifontein and Rooikraal TSFs are situated on the Witwatersrand Supergroup and is composed of tailings that were mined from gold-bearing conglomerate rock (McCarthy & Rubidge, 2005:106; McCarthy, 2011:2; Naicker *et al.*, 2003:29). The Witwatersrand Supergroup has two main subdivisions, namely the Upper and Lower West Rand Group. The West Rand Group is further divided into three Subgroups, namely the Hospital Hill, Government Reef and Jeppestown Subgroups. Subgroups of the Central Rand Group are the Johannesburg and

Turffontein Subgroups. The richest gold deposits occurred in the Johannesburg Subgroup and have been extensively mined throughout the Witwatersrand Basin (Durand, 2012:25; McCarthy & Rubidge, 2005:101). The Witwatersrand conglomerates are associated with river transport and deposition. They occur in layers that vary in thickness from a few centimetres to tens of metres. The layers consist of quartz pebbles typically one– three cm in diameter that are set in a matrix of quartz sand. This matrix commonly contains up to 3% pyrite and smaller amounts of various other sulphide minerals that cause acidity through oxidation in gold mine tailings (Naicker *et al.*, 2003:29; McCarthy & Rubidge, 2005:106).

According to Meyer and Stewart (2012:4), the gold mines at Rooikraal form part of the East Rand Goldfield. The mines in this area have produced over 9 million tonnes of gold after gold production commenced in 1888, first at the Nigel gold mine, followed by the Van Ryn Estates in 1892, which lasted until 1983. The majority of the gold was mined from the conglomerate layer known as the Nigel Reef displayed in Figure 3.5 (Meyer & Stewart, 2012:3).

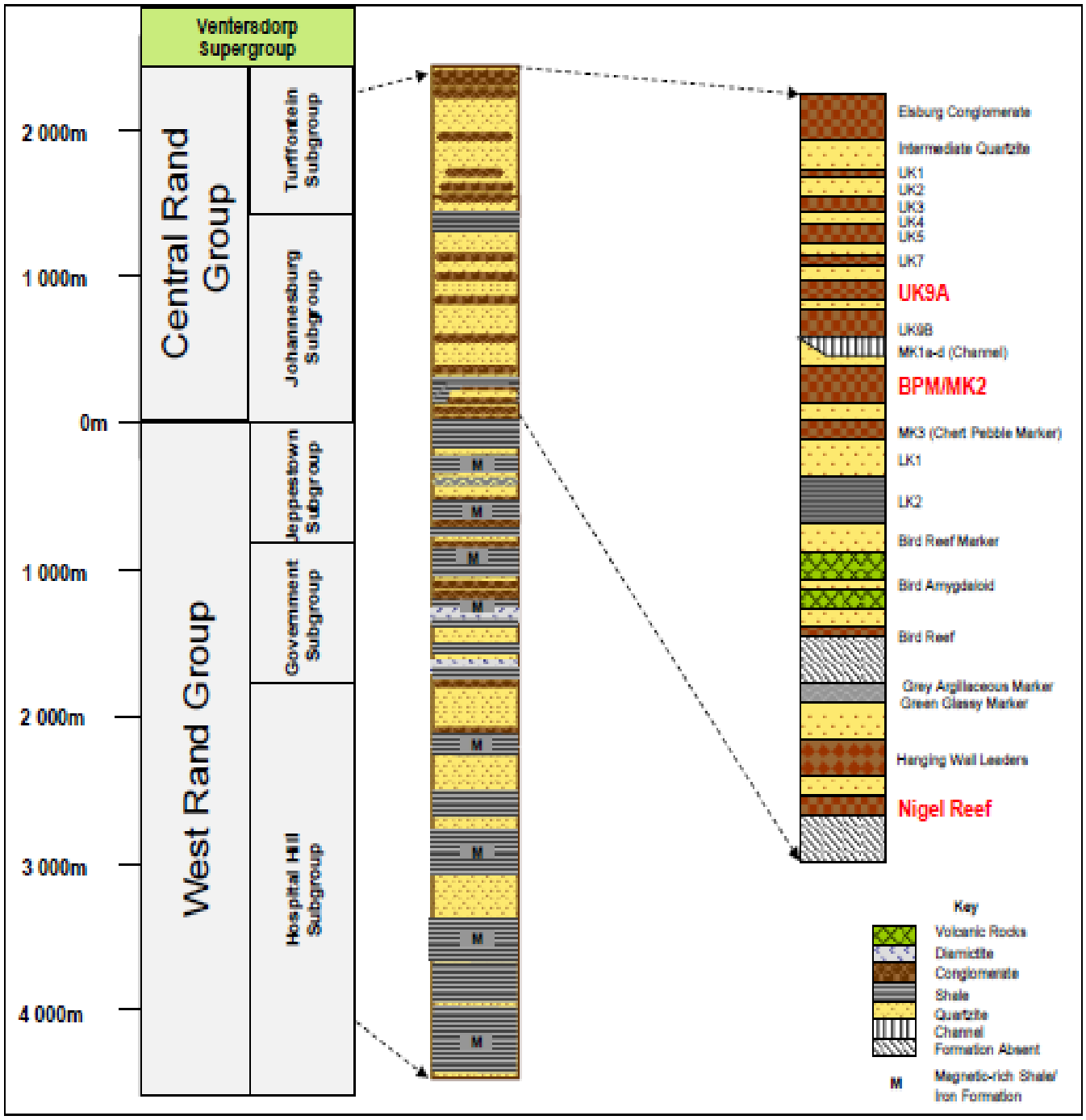

Figure 3.5: Stratigraphic column of the East Rand Goldfield (Meyer & Stewart, 2012:4).

3.1.4 NWU nursery for soil and plant research for mine rehabilitation

The NWU's nursery for soil and plant research for mine rehabilitation was the location of Phase 1 for the platinum trials and Phase 2 for the pot trials (Figure 3.6).

Figure 3.6: The location of the North-West University (NWU) nursery for soil and plant research for mine rehabilitation in Potchefstroom where the platinum trials and pot trials were carried out (Google Earth, 2016b).

- **Trials in Paardekraal platinum tailings**

The tailings material used for Phase 1 platinum trials was obtained from the Paardekraal Platinum Mine TSF outside Rustenburg, North-West Province. Paardekraal is one of the production areas of Anglo Platinum's Rustenburg section located on the western limb of the Bushveld Igneous Complex (Mabedla & Trofimczyk, 2009:1). The ore containing platinum-group metals is extracted from a pyroxenite-rich rock layer known as the Merensky Reef and a specific chromitite rock layer known as the Upper No. 2 (UG2) Reef (McCarthy & Rubidge, 2005:124). Both these layers are located in the critical zone of the Rustenburg Layered Suite, one of three components of the Bushveld Igneous Complex (McCarthy & Rubidge, 2005:121; Pretorius, 2014:21). The trials at the Paardekraal Platinum Mine were discontinued due to security reasons and forbidresearchers from entering the mining area.

Platinum mine tailings obtained from Paardekraal were available for trials at the NWU nursery trial site. The tailings were placed in bulk bags with a volume of one m x one m x 0,25m and used

as trial plots for seed trials platinum mine tailings from the TSF at the Aquarius platinum mine outside of Rustenburg were used in Phase 2 pot trials. The Aquarius platinum tailings are similar to those from Paardekraal as the operations are located on the western lobe of the Bushveld Igneous Complex and are both mined from the Merensky and the UG2 Reefs (Aquarius platinum limited, 2011:29).

- **Growth mediums for supporting pot trials**

Four growth mediums were selected and used in the Phase 2 supporting pot trials. These included tailings material from the Crown Mooifontein TSF top-flat area, the Rooikraal gold TSF berm area, platinum tailings from the Aquarius platinum mine and a red apedal B-horizon from a Hutton soil, which served as a control growth medium. The pot trials were conducted to compare the emergence and development of specific grass species used in the field trials on gold and platinum tailings when using coated and uncoated seed.

3.2 Total rainfall and average daily temperature

Weather data for the trial areas were requested from the South-African Weather Service. The total monthly rainfall (mm) and the average maximum and minimum daily temperatures (ºC) from January 2016 to June 2017 are displayed in Figure 3.7 to Figure 3.9.

The lowest temperatures recorded for the Crown gold TSF site was from May to August 2016 and May to July 2017 with temperatures below 10°C. The lowest recorded temperature was 4,5ºC during June 2016 as shown in Figure 3.7. The warmest recorded months were January 2016 to March 2016 and again October 2016 to March 2017 with temperatures greater than 24°C. Rainfall coincided with the warmer summer months during January 2016 to March 2016 and November 2016 to February 2017 (Figure 3.7).

At the Rooikraal gold TSF site very cold minimum temperatures below 5°C were recorded from June to August 2016 and again in April to June 2017 (Figure 3.8). The coldest minimum temperature was 0,4°C measured in July 2016. The warmest months were January to April 2016 and September 2016 to March 2017 (> 25 °C). Rainfall mostly coincided with the warmer months January to March 2016 and November 2016 to February 2017. Rain also occurred during the winter months of May 2016, June 2016 and July 2016.

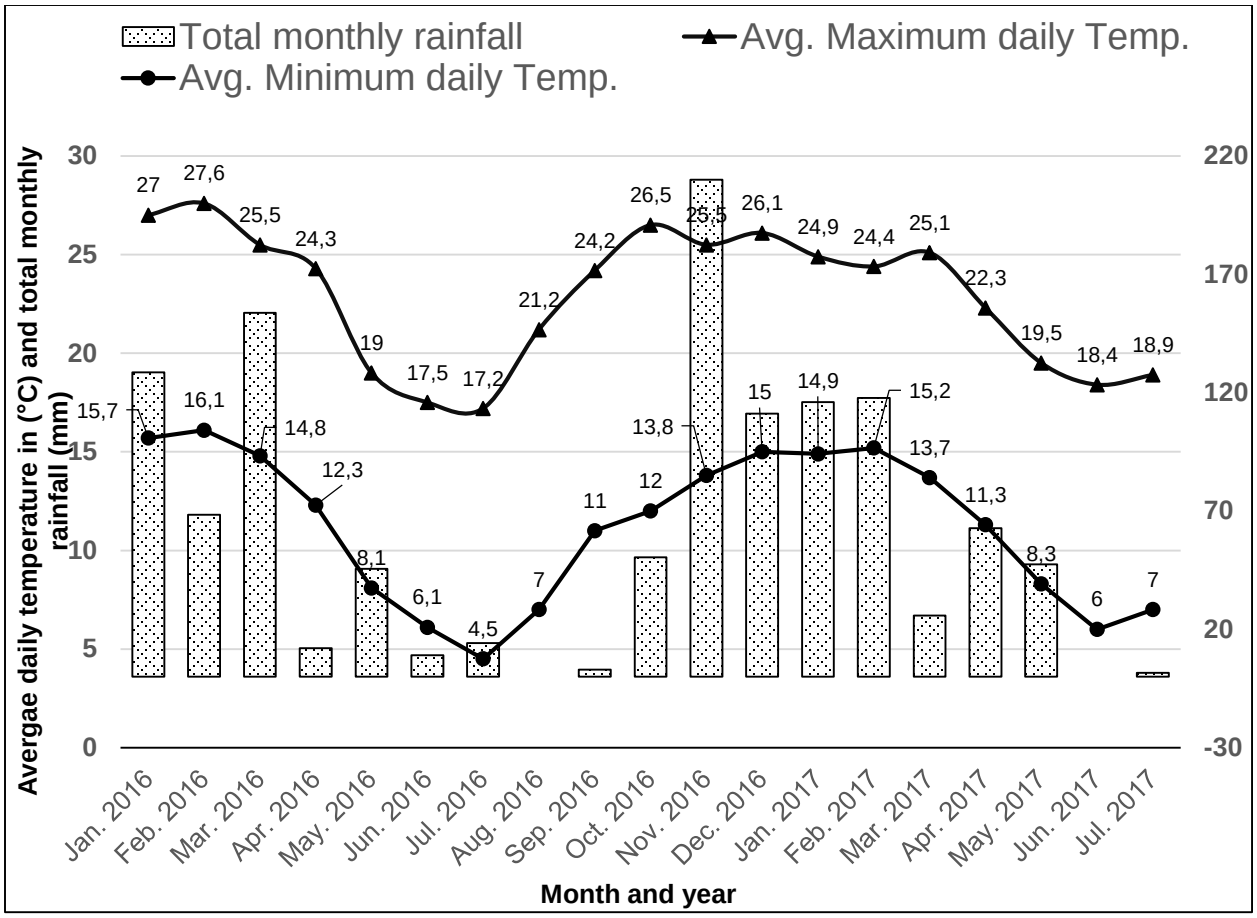

Figure 3.7: Monthly rainfall and average maximum and minimum daily temperatures for Crown gold TSF site from January 2016 to July 2017 (SAWS, 2017a).

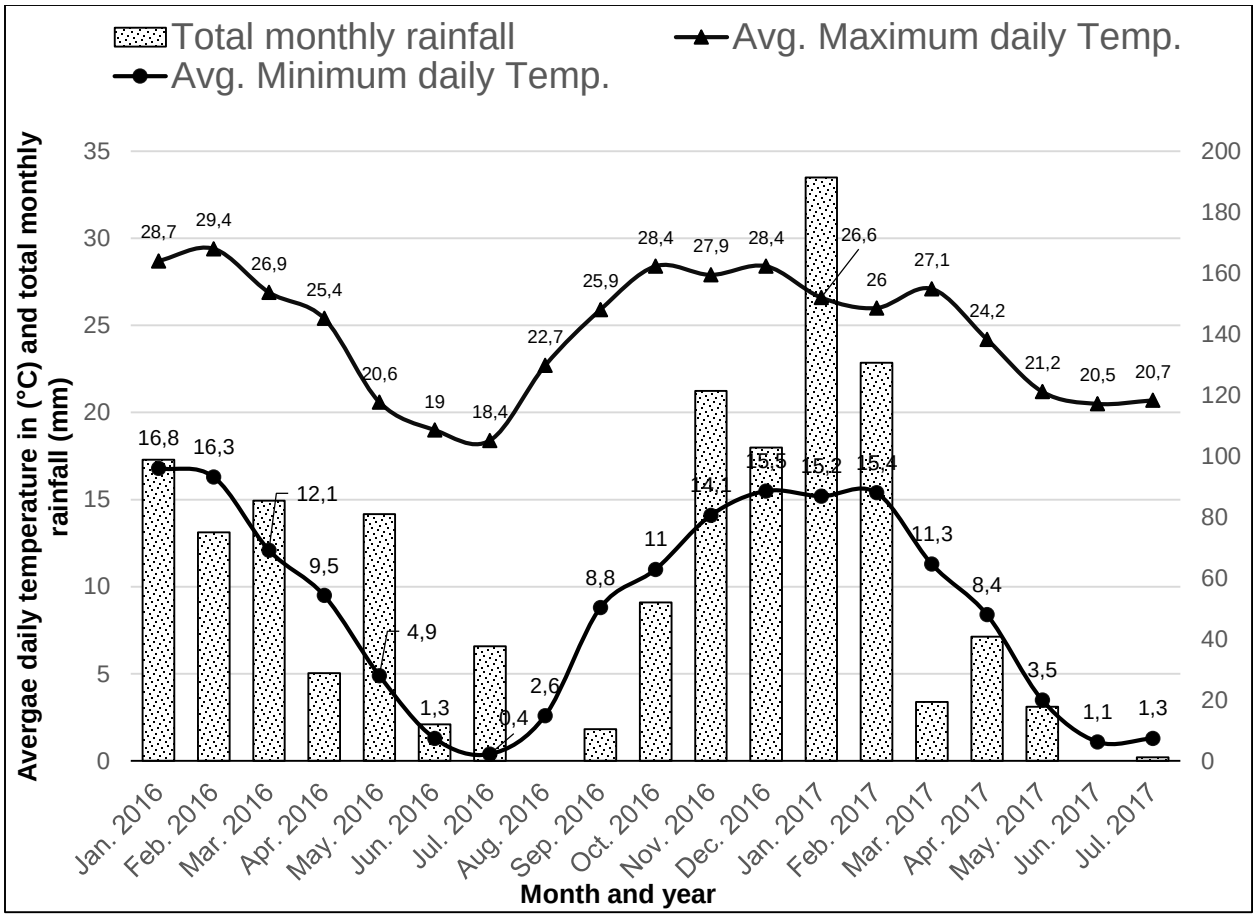

Figure 3.8: Monthly rainfall and average maximum and minimum daily temperatures for Rooikraal gold TSF site from January 2016 to July 2017 (SAWS, 2017b).

The coldest months for Potchefstroom with minimum temperatures below 5°C were in June to August 2016 and April to July 2017 (Figure 3.9). The coldest month in 2016 was June with an average minimum daily temperature of 0°C. The warmest months with an average daily maximum temperature above 25°C were January to April 2016 and September 2016 to March 2017. Rainfall coincided with the warmer summer months. The highest maximum temperature (32,4°C) was in February 2016.

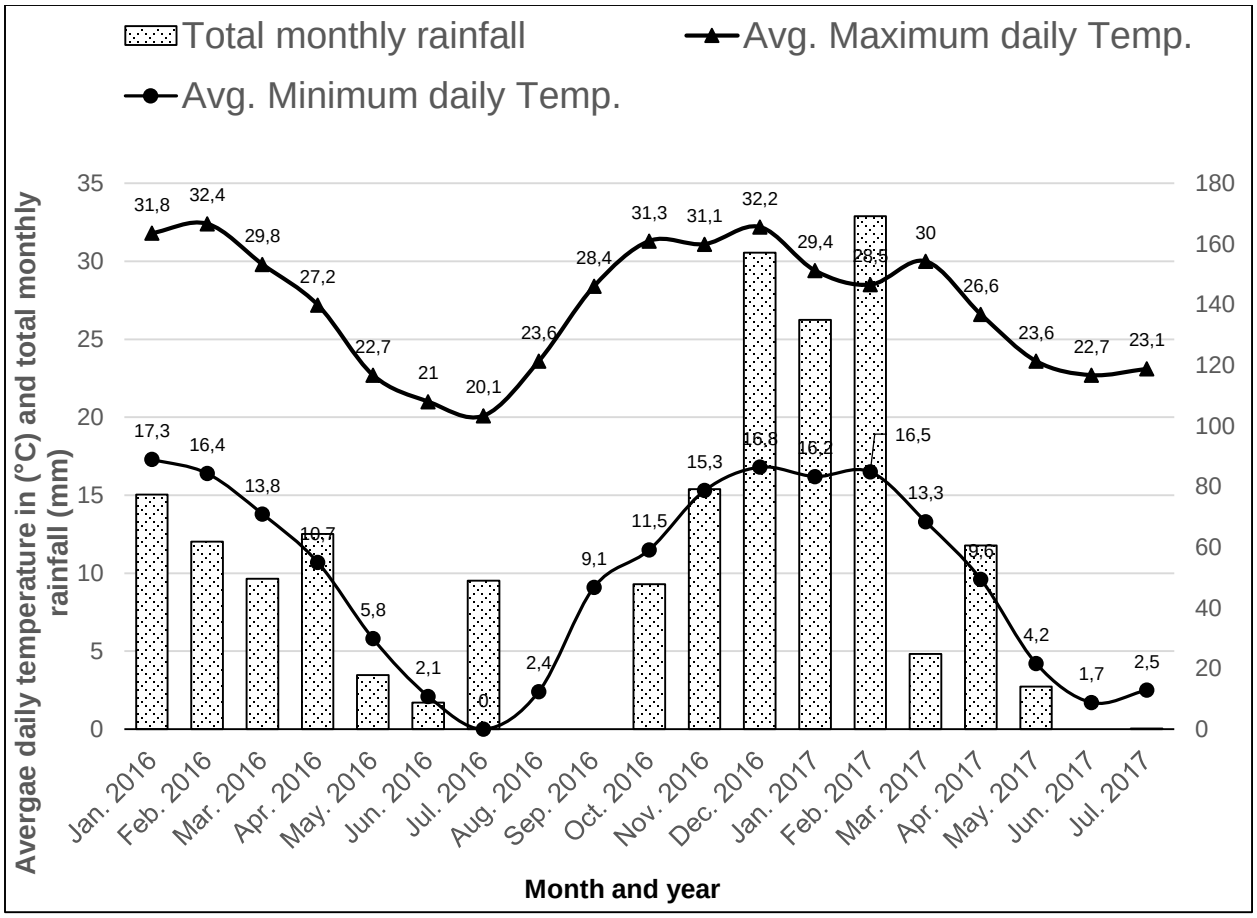

Figure3.9: Monthly rainfall and average maximum and minimum daily temperatures for Potchefstroom from January 2016 to July 2017 (SAWS, 2017c)

The weather data for the three sites show typical rainfall and temperature patterns of Mesic-and Dry Highveld bioregions (Mucina & Rutherford, 2006:48). The cold temperatures during the winter months may lead to dormancy of some of the plant species.

3.3 Plant species used in the trials

The plant species used in the seed mixtures for the project were chosen by AGT Foods Africa Pty Ltd[3] (hereafter AGT Foods), because these species have been used previously in rehabilitation projects.

[3] 8 Jacobs Street, Chamdor, Krugersdorp, 1739
P.O. Box 414, Krugersdorp, 1740,
Telephone no.: +27 011 762 5261/2
Fax no.: +27 011 762 411

- ***Cynodon dactylon***

C. dactylon, also known as Bermuda or couch grass, is a perennial grass (Horowitz, 1996:306). This species is prostrate and fine-leaved, has a creeping growth form spreading via stolons on the soil surface and can have scaly rhizomes underneath the soil surface (Horowitz, 1996:306). The stolons and rhizomes root at the nodes to form dense mats, making this species an excellent soil stabiliser (Jones, 1985:14; Van Oudtshoorn, 2004:229). The culms are erect and range in height from 50–500 mm. Each culm carries a digitate inflorescence with three to seven branches that can be 30–100 mm in length (Jones, 1985:14).

Van Oudtshoorn (2004:229) describes *C. dactylon* as a pioneer grass with an increaser 2 grazing status (increase due to the effects of overgrazing), which occurr in all soil types in degraded and disturbed environments. According to Van Oudtshoorn (2004:26), pioneer grasses are species that are hardened and able to grow in unfavourable conditions. After any disturbance, pioneers are usually the first species to occupy an area, improving the growth and habitat conditions of the area for sub-climax and perennial climax species to establish (Van Oudtshoorn, 2004:26). Furthermore, *C. dactylon* is a species generally used in cultivated pastures. It can endure heavy grazing and remains green until late in winter. The rhizomes and stolons enable *C. dactylon* to occupy less favoured niches and provide effective erosion control in the areas where other species do not establish well (Coaltech research association and Chamber of mines, 2007:117).

- ***Digitaria eriantha***

Other names for *D. eriantha* are Smutsvinger and common finger grass (Van Oudtshoorn, 2004:229). It is a strong perennial, densely tufted climax grass, commonly growing in sandy and gravelly soils throughout semi-arid parts of South Africa (Van Oudtshoorn, 2004:229). Although it occurs in a wide range of habitats, it is mainly present in undisturbed veld. It is classified as a decreaser grazing status species, as it decreases under over-grazing conditions (Van Oudtshoorn, 2004:229). Because of its characteristics, *D. eriantha* is regarded as a good pasture species. It is widely cultivated for grazing in southern Africa and known to react well to fertilisation (Van Oudtshoorn, 2004:229). The inflorescences of this grass are digitate to semi-digitate with long thin racemes that are carried upon unbranched culms 400–1800 mm high. The leaf blade is folded or open with a fringed ligule hosting hairs (Van Oudtshoorn, 2004:229).

- ***Eragrostis curvula***

Eragrostis curvula is also known as weeping love grass. It is a densely tufted perennial grass known to grow in disturbed soils, especially on roadsides and is often associated with overgrazed and trampled veld (Van Oudtshoorn, 2004:177). This species is known to tolerate drought, frost, salinity, close grazing and high temperatures (Jones, 1985:15). Furthermore, it is considered one of the most important cultivated pasture species in the Highveld regions of South Africa, as it is palatable, is easy to establish, emerges in early spring and reacts well to fertilisers. It is able to provide good hay if not cut too late before the winter season and is classified as a subclimax or climax species with an increaser two grazing status (Van Oudtshoorn, 2004:177). This species' leaves are concentrated at the base of the plant and are loose and hanging. The inflorescences are open panicles carried upon culms ranging between 300 and 1200 mm in height. The panicle can be loose and spread open or narrow and dense. Leaf blades are rolled or open up to 300 mm in length and 1–5 mm in width; the ligule consists of a ring of hairs (Jones, 1985:15; Van Oudtshoorn, 2004:177).

- ***Eragrostis tef***

E. tef is also referred to simply as 'tef'. It is a small, fine-stemmed, annual tufted grass growing 20–90 mm tall (Jones, 1985:9). It is often cultivated in South Africa and India as an annual pasture grass and used as hay for additional fodder. This species grows well in disturbed areas and in most soil types (Jones, 1985:9; Van Oudtshoorn, 2004:139). *Eragrostis tef* is often used in grass seed mixtures with perennial, climax species, such as *D. eriantha*, as it establishes well in the short term, covers the soil surface and increases the functionality (nutrient content) of the disturbed soil for the perennial and climax species to establish over the long term (Van Oudtshoorn, 2004:139). It is an exotic pioneer grass species originating in the north-eastern parts of Africa, where it was domesticated (Jones, 1985:9; Van Oudtshoorn, 2004:139). The entire plant is light green when young with leaf blades being open and hairless, except for the ligule where a ring of hairs occurs. It flowers from November to May (Van Oudtshoorn, 2004:139). The inflorescence is an open panicle and the seed are extremely small, weighing 0.25–0.3 mg each (Jones, 1985:9).

- ***Sorghum bicolor***

S. bicolor, also known commonly as grain sorghum, great millet and guinea corn, is an annual grass species from the Panicoideae subfamily originating in Ethiopia. This species has been

cultivated as a cereal for over 6000 years (Jones, 1985:6). Within the *Sorghum* genus, approximately 25 species are cultivated throughout the world due to their high yield (Głąb *et al.*, 2017:47; Jones, 1985:6). Sorghum, depending on the cultivar, may grow to heights of 500–6000 mm carrying compact panicles, which produce seed that are four–eight mm in diameter and weighing 15–40 mg each (Jones, 1985:6). Głąb *et al.* (2017:47) describes sorghum as having a wide spectrum of applications that range from human consumption to animal fodder and even construction. It therefore has a high resilience and can easily adapt to changing habitat conditions, particularly drought and saline soils (Bafeel, 2014:300; Jones, 1985:7; Peerzada *et al.*, 2017:75;). Habyarimana *et al.* (2017:1) explain that *S. bicolor* can be perennial as it has the capacity to survive the winter season through rhizomes.

S. bicolor is incorporated into the seed treatment to encourage the establishment of perennial species by providing additional root mass that in turn increases soil organic matter and encourages the build-up of beneficial soil microbial populations (Björkman & Shail, 2010:1; Gahur & Adholeya, 2004:532).

- ***Medicago sativa***

M. sativa, also known as legume, alfalfa or lucerne, is capable of establishing a symbiotic relationship with soil N-fixing rhizobacteria that occur in soil-root ecosystems. These rhizobacteria can increase plant growth by increasing the available N in the soil, which reduces the need for fertiliser application (Saadani *et al.*, 2016:264). The capacity of legumes to fix N through symbiotic relationships renders them ideal pioneers for the restoration of N-deficient environments such as metal-contaminated soils (Saadani *et al.*, 2016:264). Naidu and Harwood (1997:368) explain that the inclusion of legumes in the revegetation of degraded land should be a priority due to their ability to fix atmospheric N. Once established, legumes contribute significantly to the growth of grasses and prevent erosion when compared to vegetated areas that did not include legumes. *M. sativa* is an important perennial herbaceous forage legume that has been cultivated for the past 150 years mainly for soil improvement through N fixation (Liu *et al.*, 2017:205). It is however, not tolerant to drought conditions (Coaltech research association & Chamber of mines, 2007:118).

- **Coated and uncoated seed**

Coated and uncoated seed of the species mentioned above were used in this study. The species and whether the seed were coated and uncoated is summarised in Table 3.1.

Table 3.1: Coated and uncoated seed used for selected grass and crop species.

Species type	Coated	Uncoated
D. eriantha	**x**	**x**
E. curvula	**x**	**x**
C. dactylon	**x**	**x**
E. tef		**x**
S. bicolor	**x**	**x**
M. sativa	**x**	

In previous studies carried out by the NWU for AGT Foods, the use of coated and uncoated seed for the rehabilitation for gold and platinum mine tailings were mostly carried out in pot trials, with species separated from one another, either in a nursery or greenhouse (Muller, 2014:39; Pretorius, 2014:37, Westcott, 2011:30). In this study, coated and uncoated seed and mixtures for gold and platinum TSF rehabilitation were also used to carry out on-site trials under natural conditions.

One major difference between coated and uncoated seed is the weight of the seed and subsequently the number of seed per unit of weight. The material applied to the exterior of the coated seed adds to the weight of each seed and causes coated seed to be heavier than uncoated seed. Therefore, the number of coated seed in the same weight measurement is less than that of uncoated seed. This weight difference also varies for individual species due the differences in seed size and morphology. Some seeds are 'fluffy' and weigh much less, whereas the seed coat of other seeds is smooth and weighs much more. Some seeds are small, e.g. that of *E. tef*, whereas other seeds are larger such as that of *S. bicolor* and *D. eriantha*. It is therefore necessary to evaluate the germination and emergence of coated and uncoated seed mixtures that contain seeds of various weights and morphology, not only in pot trials, but also in the field on the TSF, where rehabilitation is carried out.

3.4 Experimental design

This section of the chapter provides an overview of the various phases of the study and the steps taken within each phase to evaluate the use of coated and uncoated seed in the revegetation of gold and platinum TSFs. The experimental trials were divided into three phases: (1) field trials, (2) corresponding pot trials with the same tailings material and (3) additional field trials on the

Rooikraal gold TSF to evaluate lower seeding rates and the use of bio-stimulants for soil amelioration. The supporting pot trials and additional field trials were carried out to support and refine the data retrieved from Phase 1 field trials.

3.4.1 Phase 1: Field trials

The first phase of the study entailed the evaluation of coated and uncoated seed ratios of the selected species on TSFs after amelioration and preparation according to current rehabilitation practices. Seed emergence and growth as well as changes in plant composition over time were monitored. The first step was setting up the seed treatments to be sown. This entailed the weighing and counting of coated and uncoated seed of the various species and calculating the seeding ratio. An amount of 0,5 g of each seed type was weighed, using an Adam Highland HCB602H 600 g x 0,01 g precision balance scale. The number of seed was counted to determine how many seed of each species were represented in 0,5 g. The count of coated and uncoated seed for the same species was used to calculate the coating ratios for the specific species and the number of seed per unit weight. The coating ratio was calculated according to Equation 9:

Equation 9: Coating ratio = *Nuc*/*Nc*

Where *Nuc* is the number of uncoated seed and *Nc* is the number of coated seed of a species at a specific weight.

- **Selected seed treatments**

To evaluate the use of coated and uncoated seed, five different seed mixtures were used. The weight of seed per species used for the different seed treatments (kg/ha) and the ratios of coated to uncoated seed for species are summarised in Table 3.2.

The amount of coated and uncoated seed for Treatments 1 (T1C) and 2 (T2UC), respectively, were the same (19 kg/ha). The only difference was that seed mixture Treatment 1 contained coated seed of *D. eriantha*, *E. curvula*, *C. dactylon* and *S. bicolor*, whereas Treatment 2 contained uncoated seed of *D. eriantha*, *E. curvula*, *C. dactylon* and *S. bicolor*. Both these treatments were set up as baseline treatments to compare the use of coated and uncoated seed. For Treatment 3 (T3C) the coated seed of *D. eriantha*, *E. curvula*, *C. dactylon* and *S. bicolor* were increased according to the coating ratio to provide the same number of seed as was used in the uncoated Treatment 2. The total seeding rate per weight for Treatment 3 was therefore 34,4 kg/ha (Table 3.2). For Treatment 4 (T4C), the total seed weight was kept at 19 kg/ha, the same as for Treatment 1, but the species in the mixture were set up according to their coated ratios. Treatment 5 (T5UC)

was a replicate of Treatment 4 using uncoated seed, also with a total of 19 kg/ha of uncoated seed. Uncoated *E. tef* seed and coated *M. sativa* seed were used in each of the five seed treatments (Table 3.2).

Table 3.2: Selected seed mixtures per coated and uncoated seed weight used for the five treatments. Amounts are given in kg/ha.

Species	**Ratio**	**T1C**	**T2UC**	**T3C**	**T4C**	**T5UC**
D. eriantha	**2.9**	4	4	11.6	5.7	5.7
E. curvula	**2.1**	4	4	8.4	4	4
C. dactylon	**1.7**	4	4	6.8	3.3	3.3
E. tef	**1 (Uncoated seed only)**	1	1	1	1.9	1.9
S. bicolor	**1.2**	3	3	3.6	2.3	2.3
M. sativa	**1 (Coated seed only)**	3	3	3	1.9	1.9
	Total in kg/ha	19	19	34.4	19	19

The calculation for each of the species in the mixture is shown in Equations 10 and 11. In Equation 10, the sum of the coating ratios of the species was determined. The coating ratios of *E. tef* and *M. sativa* were taken as 1, as only uncoated seed of *E. tef* and only coated seed of *M. sativa* was used.

Equation 10: $T_{CR} = D.\ eriantha_{CR} + E.\ curvula_{CR} + C.\ dactylon_{CR} + E.\ tef_{CR} + S.\ bicolor_{CR} + M.\ sativa_{CR}$

Where T_{CR} is the sum of the coating ratios of all the species and $Species_{CR}$ is the coating ratio of individual species. T_{CR} was then used in Equation 11 to calculate the weight of seed for the specific species ($Species_{wt}$) in Treatment 4:

Equation 11: $(Species_{CR}/T_{CR}) \times 19\ kg/ha = Species_{Wt}$

As an example, *D. eriantha* is used to demonstrate Equation 11:

D. eriantha: (2,9/9,9) × 19 kg/ha = 5,6 kg/ha

Figure 3.10 is an illustration of the weight difference in kg/ha for seed sown in the different seed treatments.

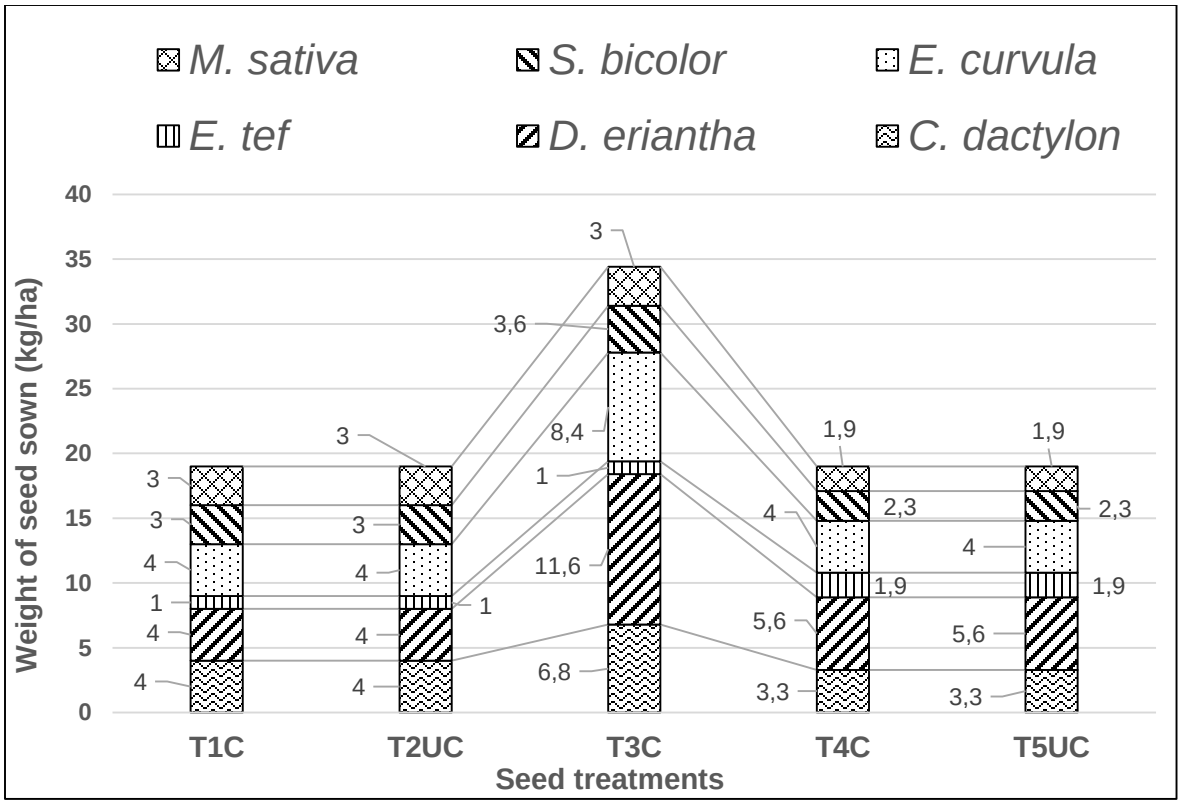

Figure 3.10: Weight of seed per seed type (coated [C] and uncoated [UC]) sown in the five treatments (T1–T5) (Table 3.2) during Phase 1 of the study.

- **Seed purity and viability**

The seed batches of each species used for the trials were analysed for their purity and viability at the AGT Foods seed testing laboratories. Seed purity is the percentage of seed excluding the accompanying inert material such as seed husks seeds of other species and additional plant material, whereas seed viability is known as the germination potential of the seed. The percentages of normal seed (high purity and viability), abnormal seed, dead seed and hard seed for the coated and uncoated seed batches of the various grass species were determined and are illustrated in figure 3.11. The seed testing laboratory is registered with the Department of Agriculture's ISTA-accredited seed testing station in terms of the Plant Improvement Act No. 53 of 1976. The seed analysis results received from AGT Foods seed testing laboratory are provided as annexures (A – K) in section five of this document.

The seed analysis reveal that the coated and uncoated *C. dactylon*, *E. curvula*, *S. bicolor*, uncoated *E. tef* seed and coated *M. sativa* seed batches mostly consisted of normal seed (>72%), meaning that these seed types are able to germinate and potentially establish. The coated and

uncoated seed of *D. eriantha* were mostly dead (77%), as only 22% of the seed were analysed as normal, meaning that the germination potential of this seed type is much lower.

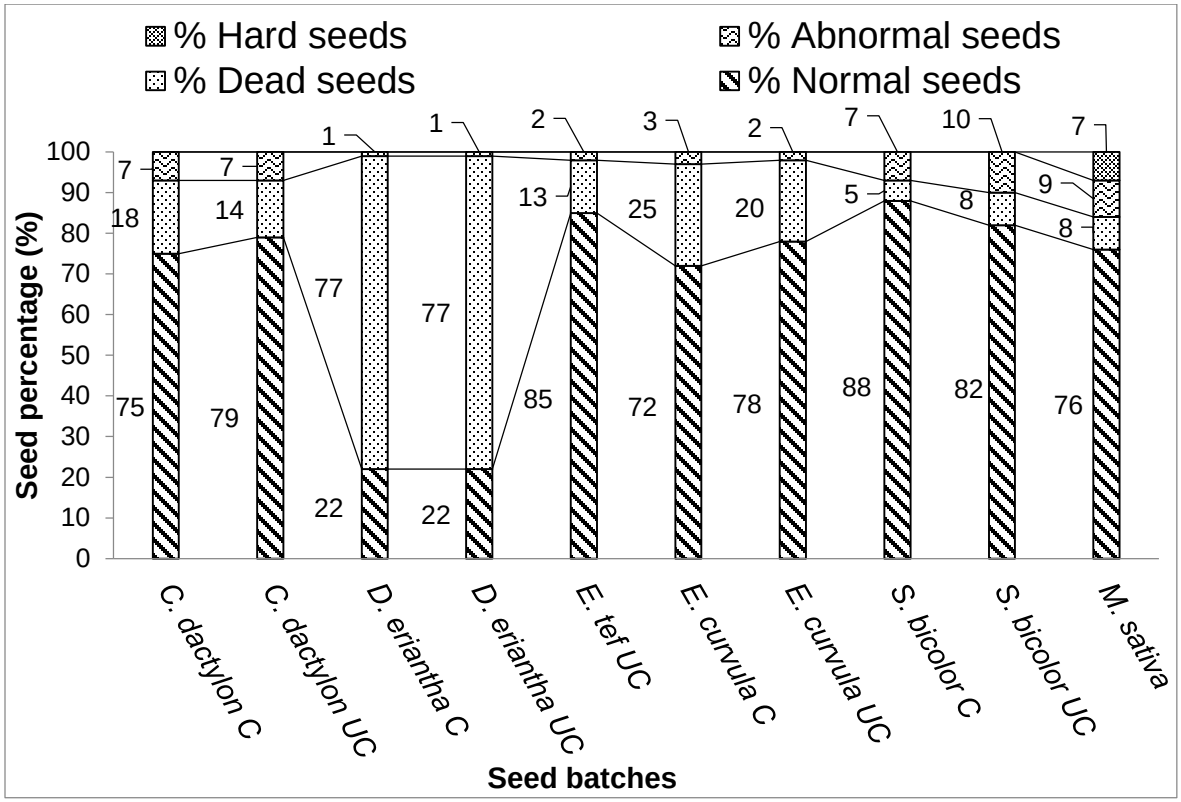

Figure 3.11: Percentage of hard, abnormal, dead and normal seed for each uncoated (UC) and coated (C) seed type per seed batch as determined by the AGT Foods seed testing laboratories

Figure 3.12 is an illustration of the number of viable seed sown for each species in the seed treatments in Phase 1 trials. It illustrates the influence of the weight difference between coated and uncoated seed for the number of seed sown.

If T1C, which incorporated coated seed of *C. dactylon*, *E. curvula*, *D. eriantha* and *S. bicolor*, is compared to the equivalent T2UC, it is evident that a much larger number of viable seed was sown in T2UC. The T3C contained the same number of coated seed as T1C did, but the weight of coated seed for specific species (*C. dactylon*, *E. curvula*, *D. eriantha* and *S. bicolor*) was increased according to the coating ratios calculated in Table 3.2 to obtain the same number of coated seed as uncoated seed in the T2UC treatment. This experiment was carried out to evaluate whether there was a difference in establishment rates if the number of coated seed were increased to the same number as that of uncoated seed. Due to the difference in the percentage of normal seed between the coated and uncoated seed batches (Figure 3.11), the number of viable seed in T3C differed from that in T2UC (Figure 3.12).

The T4C contained the same total weight of seed per ha as T1C did, namely 19 kg/ha seed, but the weight of seed per species within the mixture was adjusted according to the coating ratio of the species (Figure 3.12). This provides T4C with different seeding rates than for species in T1C, to see whether this adjustment of seeding rates would lead to a difference in the establishment rates of the species in the trials. The T5UC used the same amount and seed ratio as the T4C did.

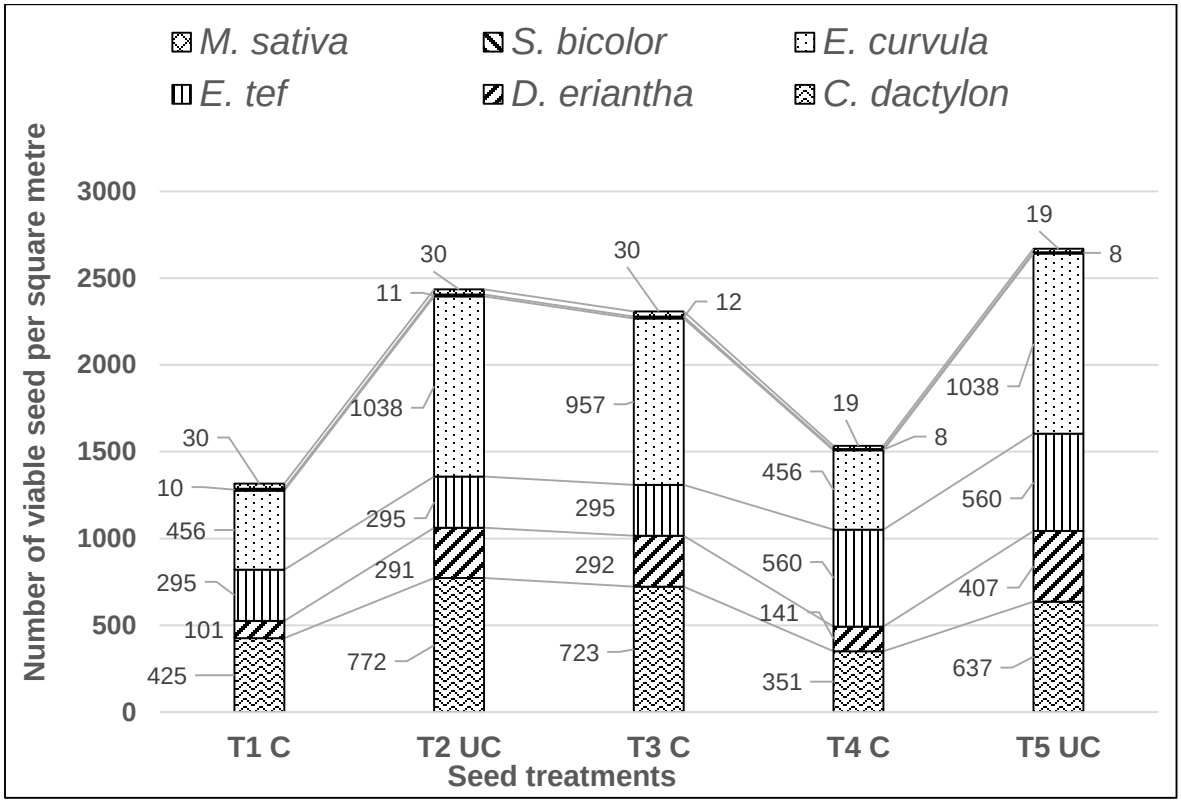

Figure 3.12: Average number of viable seed sown per square metre in the five treatments (T1–T5) during Phase 1 of the study. C: coated seed; UC: uncoated seed.

- **Trial plot preparation**

Before sowing the field trials with the relevant seed treatments, the selected areaswere ameliorated with lime, compost and fertiliser according to the recommendations provided by the Agreenco rehabilitation company working at the Crown gold TSF site (Table 3.3).

The acidity of the tailings material of the gold TSFs of the Crown and Rooikraal sites was neutralised with dolomitic lime (Table 3.3). Dolomitic lime and compost were applied evenly across the tailings surface over the designated areas (Figure 3.13), after which it was cultivated using a ripper implement in the tailings subsurface to a depth of 30 cm. At the Crown top-flat site,

a tractor with a ripper implement was available, but at the Rooikraal berm site, the tailings surface was ‘ripped’ using garden forks.

Table 3.3: Type and amount of ameliorants used to prepare trial plots in field trials.

Ameliorant	**Amount**
Dolomitic lime	85 t/ha
Well-decomposed mushroom compost	40 t/ha
4:3:4: (34) fertiliser	400 kg/ha
Superphosphate fertiliser	600 kg/ha
Mulch	60% coverage
1:0:1 (32) fertiliser	200 kg/ha

Figure 3.13: Dolomitic lime applied to the bare top-flat trial site on Crown gold TSF site to neutralise the tailings acidity. Photo taken by C.A. Kuger, 20/2/2016.

Figure 3.14 illustrates the trial plots at the Crown site after lime was applied and ripped into the tailings surface by a tractor. The cultivation and amelioration was carried out in February 2016.

The sites were left for a period of four weeks for the lime to react and neutralise the tailings material before the 4:3:4 (34) and superphosphate fertilisers were applied across the tailings surface of the trial areas. The platinum tailings that were placed in the bulk bags did not require any lime due to the high pH of the material. Only compost was applied to these tailings material before adding the 4:3:4 (34) and superphosphate fertilisers.

After the fertiliser was applied to the sites on the TSF surfaces and in the bulk bags, the seed treatments were sown by hand. Seed was mixed in a bag and broadcast evenly across the surface of the trial plot area. After sowing, mulch was applied to cover approximately 60% of the surface of the trial plots. All these applications were completed in March 2016. A follow-up fertiliser dressing using 1:0:1 (32) fertiliser was applied in September 2016 to replenish depleted nutrient reserves of tailings.

Figure 3.14: Top-flat trial plots on the Crown gold TSF after lime and compost were applied and ripped into the tailings surface. Photo taken by C.A. Kruger, 4/3/2016.

- **Crown gold TSF site**

Trial plots at the Crown gold TSF site were eight × eight m (64 m^2) in size with four replicate plots for each seed treatment. No irrigation was used to water the trial area as illustrated in Figure 3.15.

Figure 3.15: Trial plots at the Crown gold TSF site with mulch applied after amelioration and sowing. Mulch overlaying plots are indicated with red arrows. Photo taken by C.A. Kruger, 9/3/2016.

- **Rooikraal gold TSF site**

The size of the Rooikraal gold TSF site trial plots was reduced to five × five m (25 m^2) because the ameliorants (dolomitic lime, compost and fertiliser) had to be transported from the Crown gold TSF site and worked into the material by hand, which was labour-intensive. Four replicate plots for each seed treatment were prepared without any irrigation (Figure 3.16).

Figure 3.16: Lime and compost being worked into the tailings surface manually after application at the Rooikraal gold TSF site. Photo taken by C.A. Kruger, 24/2/2016.

- **Platinum trials**

As previously mentioned, the Paardekraal platinum mine TSF was intended to be the location for platinum field trials, but due to security and access issues, platinum tailings from the Aquarius platinum mine was transported to the NWU nursery in Potchefstroom and placed in bulk bags instead (Figure 3.17). Woven polyethylene bulk bags were filled with platinum tailings ameliorated with compost and fertiliser in amounts according to Table 3.3. The tailings material was ameliorated and the seed treatments sown in April 2016. One bulk bag was prepared for each seed treatment. The trials were irrigated with an average 45 mm of water per month (Figure 3.17).

Figure 3.17: Platinum tailing material trials in bulk bags after amelioration, mulch application and sowing. Photo taken by C.A. Kruger, 4/6/2016.

In November 2016 it was visually observed that the emerged *M. sativa* plants were growing exceptionally well and hindering the emergence and growth of grass seedlings in the platinum trials (Figure 3.18). It was decided to cut the *M. sativa* plants at the base of the stems on 8 November 2016 to create opportune space for the emergence and growth of grass seedlings (Figure 3.19).

Figure 3.18: *M. sativa* growth overshadowing grasses in platinum tailing trials before cutting in November 2016. Photo taken by C.A. Kruger, 8/11/2017.

Figure 3.19: Platinum tailing trial seed treatment 2 after *M. sativa* was cut on 8 November 2016. Photo taken by C.A. Kruger, 8/11/2017.

3.4.2 Phase 2: Supporting pot trials

Pot trials were established to support the results obtained from Phase 1 field trials. In Phase 2, the establishment and growth of coated and uncoated seed of the selected grass species at the Crown and Rooikraal gold TSF field trial sites as well as the platinum tailings and control (red Hutton soil) was evaluated. Amelioration of the pot trials with lime was carried out according to the lime requirement proposed by Geolab soil and environmental laboratory[4] after the soil was analysed (Table 3.4). Lime amelioration was initiated in October 2016, one month in advance of seeding the trial sites. The platinum growth medium did not receive any lime due to its high pH of 9.15.

[4] P.O Box 5546
Kockspark 2523
Tel: +27 83379 6540

Table 3.4: Net Acid Potential (NAP) analysis results. Residual titratable acidity (Titr. Acid): amount of lime required to neutralise the active acidity (pH KCl) of tailings. Lime requirement 1 (Lime req.1): amount of lime required to neutralise future acid generation. Nett lime req.: total amount of lime to neutralise active and future acidity.

Sample no.	**pH(KCl)**	**Titr. Acid**	**Lime req. 1**	**Nett lime req.**
		t/ha lime	**t/ha**	**t/ha**
Crown gold	2,7	9	26	35
Rooikraal Gold	3,6	3	3	6
Control soil	4,1	6	0	6

After amelioration with the required amount of lime, well-decomposed mushroom compost (40 t/ha), 4:3:4 (34) fertiliser (400 kg/ha) and superphosphate fertiliser (600 kg/ha) were added to each of the pots containing the different growth mediums. Figure 3.20 shows the fertiliser and the compost being mixed into the gold mine tailings during preparation for the pot trials. The fertiliser can be seen as a white powder and the compost can be seen as a dark grey powder. Both the fertiliser and compost are ontop of the gold-brown gold mine tailings.

A total of 224 pots were prepared for the trial. Each of the four growth mediums was placed in 56 pots, each numbered using a permanent marker. The volume of each pot was five l, with a diameter of 23cm at the top, 18cm at the bottom and a height of 17,5cm. The top surface area of the plant pots used for the study calculated by the formula $A = \pi \times r^2$, was 0,04 m^2. Ten uncoated and coated seed of *C. dactylon*, *D. eriantha* and *E. curvula* and 10 uncoated seed of *E. tef* were separately sown in eight replicate pots containing the different growth mediums. Soil chemical analyses were carried out for the growth mediums in the pots before amelioration. The pH and EC were measured on the sowing date (1 November 2016) and at the end of the trial period in March 2017 using a pH and EC multimeter in 1:2.5 mass soil to de-ionised water. After the seed were sown, the pots were irrigated and covered with compost, which served as mulch. The trial pots were randomly arranged using Microsoft Excel's RAND function to generate a random number between zero and one next to the pot number in the adjacent column. The random numbers were then sorted from lowest to highest, which arranged the pots in a random order.

Figure 3.20: Fertiliser and compost being mixed into gold mine tailings growth medium using a cement mixer. Photo taken by C. A. Kruger, 27/9/2016.

- **Vegetation monitoring of Phase 2 pot trials**

After sowing, seedling emergence was monitored weekly for four weeks by counting the emerged seedlings in each pot (Figure 3.21). Two months after emergence (January 2017) the culm height of *D. eriantha*, *E. curvula* and *E. tef* and the stolon length of *C. dactylon* were measured (cm) using a measuring tape (Figure 3.22). The tuft diameters of the established individual species were measured (cm) using a digital calliper (Figure 3.23).

Figure 3.21: Emerging seedlings in pot trials during emergence counts. Photo taken by C.A. Kruger, 8/11/2016.

Plant height and diameter (cm) were measured monthly for a period of three months (January–March 2017). The dry matter (DM) produced by the grass species in each pot was then determined by harvesting the above ground plant material of the grasses by cutting it 4 cm above the soil surface and drying it at 65°C for 24h in an air-drying oven. The dried plant material was weighed on an Adam Highland HCB602H 600 g × 0.01 g precision balance scale to determine the DM produced. The average weekly emergence for each species in the pot trials was calculated for the different growth mediums to evaluate the emergence rate of seedlings from uncoated and coated seed of the different species. The average height measurements were multiplied with the average width measurements of each species from coated and uncoated seed to provide a height and width index for each seed type in the different growth mediums. The average DM weight produced by plants was calculated per pot and the results of coated seed were compared to those of uncoated seed in the different growth mediums. The results were used to evaluate and compare the performance of seedlings from coated and uncoated seed of individual species in the various growth mediums.

Figure 3.22: Tape measure used to measure the culm height of *D. eriantha* in pot trials. Photo taken by C.A. Kruger, 9/1/2017.

Figure 3.23: Basal diameter of a grass tuft measured with a digital calliper. Photo taken by C.A. Kruger, 11/1/2017.

3.4.3 Phase 3: Additional field trials on Rooikraal gold TSF site

The additional vegetation trials conducted for Phase 3 were carried out to evaluate the effect of decreasing seed densities in coated and uncoated in seed mixtures and the effect of bio-stimulants as amelioration on the seed emergence of each species. These trials were conducted at the Rooikraal gold TSF berm site, as no space was available on the Crown gold TSF site due to the ongoing rehabilitation of the TSF. Forty trial plots of four × four m (16 m^2) each were established on the berm area of the Rooikraal gold TSF site and ameliorated according to the same procedure as that implemented at the Rooikraal gold TSF site trials in Phase 1. The quantity and type of ameliorants that were used are summarised in Table 3.3. Trial plots were ameliorated with lime and composted in November 2016. The material was left for six weeks to neutralise before being ameliorated with fertiliser.

The weight of the coated and uncoated seed used for the various additional trials on the Rooikraal gold TSF berm site is presented in Table 3.5.

Table 3.5: Weight of seed for each species used in the various seed treatments in Phase 3, namely the additional field trials at the Rooikraal gold TSF berm site. LD19C, LD12C and LD5C: lower-density coated seed treatments of 19, 12 and 5 kg/ha, respectively. LD19UC, LD12UC and LD5UC: lower-density uncoated seed treatments of 19, 12 and five kg/ha, respectively. MA 1–4: microbial ameliorant trial seed treatments 1–4.

Seed in kg/ha							
Species	**LD19C**	**LD19UC**	**LD12 C**	**LD12UC**	**LD5C**	**LD5UC**	**MA1–MA4**
C. dactylon	4	4	2,53	2,53	1,05	1,05	1,05
D. eriantha	4	4	2,53	2,53	1,05	1,05	1,05
E. tef	1	1	0,63	0,63	0,26	0,26	0,26
E. curvula	4	4	2,53	2,53	1,05	1,05	1,05
S. bicolor	3	3	1,89	1,89	0,79	0,79	0,79
M. sativa	3	3	1,89	1,89	0,79	0,79	0,79
Total (kg/ha)	**19**	**19**	**12**	**12**	**5**	**5**	**5**

In the lower-density (LD) seeding trials, six seed treatments were set up to evaluate the emergence percentage of coated and uncoated seed treatments when the seeding rate was decreased. The weight of the seed treatments is illustrated in Figure 3.24 and the average number of viable seed sown per square meter is illustrated in Figure 3.25.

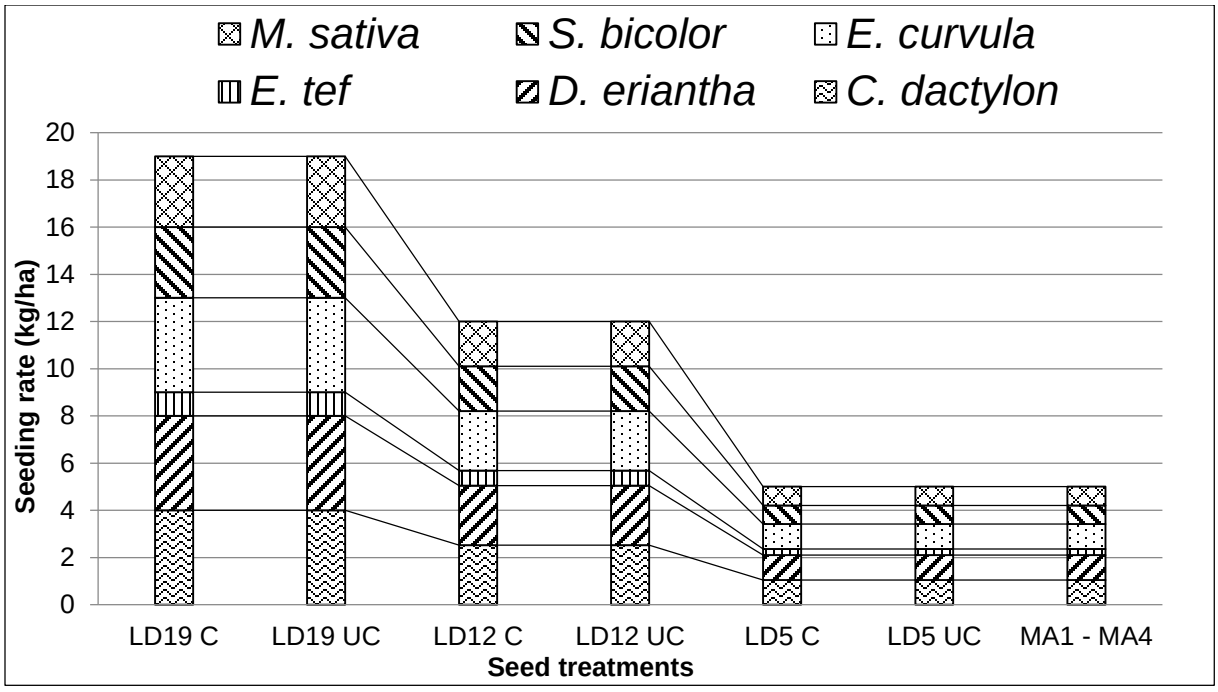

Figure 3.24: Seeding rate for coated (C) and uncoated (UC) lower density seed treatments (LD 19, 12 and 5 kg/ha) and microbial ameliorant trials trial treatments (MA1–MA4) applied to Rooikraal TSF in January 2017.

The LD treatment containing 19 kg/ha coated seed (LD19C) and the LD treatment containing 19 kg/ha uncoated seed (LD19UC) had the highest numbers of seed (Figure 3.25) for lower density seeding rate trials. The 12 kg/ha LD coated seed treatment (LD12C) and the 12 kg/ha LD uncoated seed treatment (LD12UC) contained 37% less seed than their 19 kg/ha. The five kg/ha LD coated seed treatment (LD5C) contained 74% less seed than LD19C did and the five kg/ha LD uncoated seed treatment (LD5UC) contained 74% less seed than LD19UC did.

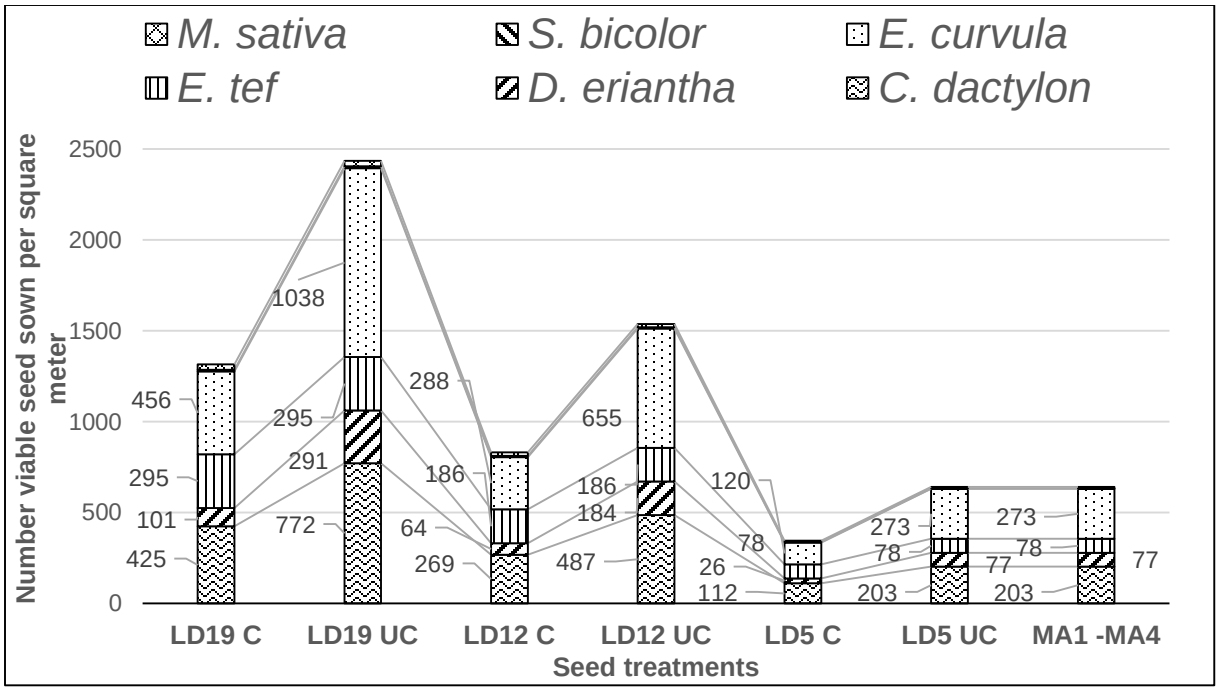

Figure 3.25: Average number of viable seed coated and uncoated lower density seed treatments (LD 19, LD12, LD5) and microbial ameliorant trials trial treatments (MA1-MA4) sown on Rooikraal gold TSF site in January 2017.

Only uncoated seed was used at a seeding rate of five kg/ha in the bio-stimulant trials, the bio-stimulant trial treatments were named and refered to as microbial ameliorants treatments one to four (MA1–MA4) in Table 3.5. In treatment MA1, a fungal growth stimulant in powder form was applied directly to the seeds by mixing them in a bag and shaking the bag for 30 seconds. The fungal *Trichoderma* growth stimulant was added at a rate of two g powder per 500 g seeds. In treatment MA2, the growth stimulant was used as in MA1; it was directly applied to the seed and the trial plot area was ameliorated with a liquid food source for the fungi via a garden spray (Figure 3.26) at a rate of 250 ml per 400 m^2. In treatment MA3, a bacterial growth stimulant with its corresponding food source was applied to the specific plot areas using a garden spray (Figure 3.26) and the area was then sown with the uncoated seed mixture. The application rate of the bacterial ameliorant was one l/ha with a food source also applied at one l/ha. In treatment MA4 both the fungal and microbial growth stimulants were applied together with their food sources at the same application rates as mentioned above. After the seed were sown, mulch was used to cover the trial plot areas to approximately 60%. The trial plots were randomly arranged using Microsoft Excel's RAND function as explained in Section 3.4.2. The results were used to evaluate the effect of decreased sowing densities on the seed emergence of coated and uncoated seed

mixtures and whether the application of bio-stimulants increases the emergence and establishment of seed mixtures sown on gold TSFs.

Figure 3.26: Liquid food source for the fungal bio-stimulant being applied to the microbial ameliorant trial plots on the Rooikraal gold TSF site during Phase 3 seed trials. Photo taken by C.A. Kruger, 19/1/2017.

The results were used to evaluate the effect of decreased sowing densities on coated and uncoated seed mixtures and if the application of bio-stimulants increase the emergence and establishment of seed mixtures sown on gold TSFs.

3.4.4 Vegetation monitoring of field trials

- **Monitoring of Phase 1 field trials**

During Phase 1 field trials, species density was monitored using 0,25 m^2 steel quadrants; only plants rooted within the quadrat area were counted. The quadrats were laid along three transects on the Crown and Rooikraal gold TSF site trial plots (Figures 3.27). In the platinum trials, the bulk bag area (one m^2) was divided into four 0,25 m^2 areas and treated as a quadrat. The length of the transects and the intervals differed between the Crown and Rooikraal plots due the difference in plot sizes at the three sites. Nine quadrats per trial plot were delineated at the Crown and Rooikraal plots and four quadrants per trial plot were delineated on the platinum trials. The transect length was 8 m for the Crown gold TSF site plots and quadrats were placed at two m, four m and six m. On the Rooikraal TSF trial plots, quadrats were placed at 1,5 m, 2,5 m and 3,5 m.

Figure 3.27: The 0,25 m^2 steel quadrant used on transect line to determine the density of the species at the Crown and Rooikraal gold TSF trial plots. Photo taken by C.A. Kruger, 15/11/2016.

The canopy and basal cover of the vegetation were determined using the line intercept method (Godínez-Alvarez *et al.*, 2009:1003). A metal pin was lowered at 20 points along a transect line.

Basal cover was determined by the hits of the living basal tuft of each grass and the canopy cover was recorded when the living above-ground material of the plant touched the metal shaft of the pin. If no plant material touched the metal pin, bare ground was recorded. The percentage canopy and basal cover per species in each trial plot was calculated. Six line transects were laid out per plot, with three lines perpendicular to the other three on the Crown gold TSF site, Rooikraal gold TSF site and platinum trials. The length of transects used for species cover estimation on the Crown gold TSF site plots was four m divided into 20 cm intervals, where the pin was lowered. At the Rooikraal TSF site, the line transect length was two m divided into 10 cm intervals. In the platinum bulk bags, each line was 50 cm long and divided into 2,5 cm sections. To avoid the edge effect, vegetation monitoring was not carried out within 1,5 m from the nearest trial plot edge on the Crown gold TSF site and within 1 m at the Rooikraal TSF site. Figure 3.28 illustrates the grid layout used to monitor the vegetation at the Crown and Rooikraal plots.

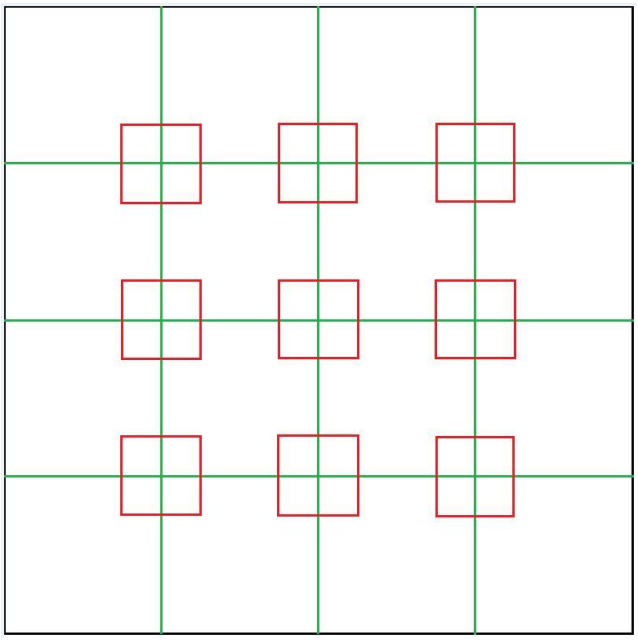

Figure 3.28: Illustration of the layout of quadrats and transects used for vegetation sampling in field trials on Crown and Rooikraal gold TSFs. The green lines represent the transect lines used to place the 0,25 m^2 quadrants and determine the basal and canopy cover with the intercept method. The red squares represent the 0,25 m^2 quadrants used for vegetation counts.

Vegetation monitoring for Phase 1 at the Crown and Rooikraal TSF sites was carried out in March 2016 and seven weeks later in April 2016. The Crown gold TSF site plots were monitored for a longer time period in June 2016, September 2016, November 2016, March 2017 and ending in June 2017. The Rooikraal gold TSF site plots were monitored in June 2016, September 2016, November 2016 and January 2017. Between September and November 2016, a large rainfall

event caused tailings from the slope adjacent to the trial plots at the Rooikraal TSF to bury the trial plots under 10–20 cm of none-ameliorated gold mine tailings. Since the majority of the trials was buried under tailings material, the trials were only monitored until January 2017. Figure 3.29 shows the buried trial site next to the eroded TSF slope on the right and Figure 3.30 shows the trial plot vegetation buried under 20 cm of raw gold mine tailings.

Figure 3.29: Buried trial plots on berm area of the Rooikraal TSF site next to eroded TSF slope (right) with *Eragrostis curvula* grass canopies projecting above the deposited mine tailings. *E. curvula* grass canopies are indicated by a red arrow and the eroded TSF slope is indicated by a black arrow. Photo taken by C.A. Kruger, 15/11/2016.

Figure 3.30: *Eragrostis curvula* and *Cynodon dactylon* grass of uncoated seed Treatment 4 on the Rooikraal gold TSF site buried beneath 20 cm of none-ameliorated gold mine tailings. Buried grass are indicated by red arrows. Photo taken by C.A. Kruger, 17/11/2016.

The platinum bulk bag trial seed treatments were sown in April 2016 and monitoring commenced seven weeks later in June 2016. Monitoring continued in August 2016, September 2016, November 2016, January 2017 and ended in May 2017.

The vegetation counts conducted in the quadrats were used to calculate the seedling emergence percentage in April 2016 and survival in September 2016 on the Crown and Rooikraal gold TSF sites. The emergence percentage in June 2016 and survival percentage in November 2016 were also calculated for the platinum trials. The change in average density of each species and change

in total cover contribution (aerial and basal cover) over time for each species present was calculated in percentage.

- **Monitoring of Phase 3 additional field trials on Rooikraal gold TSF site**

The emerging vegetation was monitored in March 2017 and six weeks after treatments were sown in January 2017 in the additional field trials of the Rooikraal gold TSF site during Phase 3 of the study. Similar to the monitoring described above, the species density was determined using four quadrats of 0.5 × 0.5 m (0.25 m^2) per plot. To avoid the edge effect, areas of one m were avoided between the plots. The average number of emerged seedlings was calculated for each trial and used to evaluate the influence of lower seeding densities and the application of bio-stimulants to the seed and soil.

3.4.5 Soil sampling and analysis

Composite soil samples of 1 kg each were collected at a depth of 30 cm using a soil auger (1) before amelioration, (2) after amelioration on the sowing day and (3) at the end of the trials on last vegetation sampling date. Each soil sample was analysed for chemical and physical parameters at EcoAnalytica[5] soil analysis laboratories at the Centre for Water Science and Management, Potchefstroom. Samples were air-dried at 65°C for 24 h and sieved through a 2 mm sieve according to the soil preparation procedure described by the The Non-Affiliated Soil Analysis Work Committee (1990) before the analysis. The soil was analysed for the following parameters:

- **pH(H_2O)**

Soil pH(H_2O) determines the pH of a soil sample in a 1:2.5 soil/water ratio suspension on a mass basis (The Non-Affiliated Soil Analysis Work Committee, 1990:3/1).

- **pH(KCl)**

Soil pH(KCl) determines the pH of a soil in suspension in 1 mol/dm^3 KCl. Potassium chloride (KCl) masks the variation in salt concentration caused by fertiliser residues, irrigation water and microbial decomposition (The Non-Affiliated Soil Analysis Work Committee, 1990:2/1).

[5] Vyfhoek plot 491
Thabo Mbeki Drive (R501)
Potchefstroom

- **EC**

Soil EC was measured using a pH/EC multimeter. Before measuring supernatant EC, the multimeter was calibrated with a 0.01 mol/dm^3 KCl solution, which has an EC of 141.18 mS/m^1 at 25°C. The measured conductivity was multiplied by a correction factor of 2,3 before being recorded in mS/m (The Non-Affiliated Soil Analysis Work Committee, 1990:4/3).

- **Extractable P: Bray-1 solution**

A known mass of prepared soil (dried and sieved <2 mm) was shaken manually with a Bray-1 solution. The contact time between soil and the extractant did not exceed 60 s. This ensured that more soluble P was extracted. The total inorganic phosphates in the extracts were determined by automated colorimetric analysis by first converting the condensed phosphates present to orthophosphates through hydrolysis with sulphuric acid at 90°C. Subsequently, the total phosphate concentration was determined by reduction of phosphomolybdic acid using the reducing agent 1-amino-2-naphthol-4-sulfonic acid. This yields an intense blue colour that is suitable for phytometric determination at 660 nm (The Non-Affiliated Soil Analysis Work Committee, 1990:20/1).

- **Cation exchange capacity (CEC) and water-soluble cations: Ammonium acetate (1 m/dm^3, pH 7)**

To determine the CEC and water-soluble cations, an ammonium acetate solution (one mol/dm^3) was used as an extractant for exchangeable and water-soluble cations. Water-soluble cations were determined separately in soils containing high quantities of soluble salts (resistance < 460 ohms). These salts were subtracted from the amount of extractable cations to obtain the amount of exchangeable cations. After the exchange complex was saturated with the index cation, the adsorbed cation and the small amount of solution entrained by the soil after centrifuging can be directly displaced by another salt solution, such as KCl. Ammonia was separated through steam distillation and taken equal to the CEC of the soil (The Non-Affiliated Soil Analysis Work Committee, 1990:12/1).

- **Particle size distribution**

The particle size distribution of the soil expresses the fractions of particles of different sizes it contains. The method of fractionation and particle size analysis is limited to sieving and sedimentation procedures (The Non-Affiliated Soil Analysis Work Committee, 1990:35/1).

- **Net acid potential and lime requirement**

Lime requirements of the gold mine tailings were determined by a method developed by Bloem (2007). This method consists of two components: First the active acidity (titratable acidity) is determined, which provides the lime requirement to raise the sample pH(KCl) to neutral (Bloem, 2017). Second, the latent acidity that is caused by the future oxidation of pyrite is determined by following the modified acid-base accounting procedure described by Usher *et al.* (2003:72); the neutralising potential is then determined. This provides the amount of lime required to neutralise acid generation in the future.

- **Dehydrogenase activity (DHA)**

Three soil samples were collected from each plot of the uncoated and coated LD5 treatments and uncoated seed treatments ameliorated with bio-stimulants (MA1–MA2). A sample of the bare non-ameliorated tailings and a natural soil sample approximately 300 m away from the Rooikraal TSF was also collected to compare the dehydrogenase activity of natural soil to that of the Rooikraal TSF material. The top 15 cm of the soil surface was collected randomly at three places within each trial plot and sampling site using a soil auger. The samples were mixed and combined to create a composite sample for each treatment from the four replicate plots. The soil samples were taken in April 2017, three months after treatments were sown. The soil samples were kept cool in a cooler with ice and taken to the EcoAnalytica laboratory later that same day to be analysed for dehydrogenase activity. The dehydrogenase activity of the samples was determined using iodonitrotetrazolium violet (Von Mersi & Schinner, 1991:217).

3.4.6 Data analysis

The vegetation data for all three vegetation trial phases were captured using Microsoft Excel 2013 and used to construct graphs and tables that illustrate the results of the three trial phases. To identify significant differences in the emergence and survival between coated and uncoated seed treatments sown on the field trial sites on Crown and Rooikraal TSF sites as well as the platinum bulk bags. A one-way analysis of variance (ANOVA) at a confidence level of 95% was applied using Stat Soft, Inc (2014) STATISTICA version 12 data analysis software. The post-hoc Tukey HSD test was applied where significant differences were found (p <0,05). Bar graphs were drawn to illustrate and describe the seedling emergence percentage, seedling survival percentage, change in species density and change in total cover contribution of species throughout the trial period (March 2016–June 2017).

To identify if the growth medium or the seed type (coated and uncoated) have and influence on the weekly emergence percentage and the change in the height and width indices of *C. dactylon, D. eriantha* and *E. curvula* a repeated measures ANOVA was applied to the data at a confidence level of 95%. The repeated measures ANOVA was also used to identify if the growth medium had an influence on the weekly emergence and the change in the height and width index of *E. tef.*

The DM yield from coated and uncoated seed for the species used during the pot trials were analysed using a two – way ANOVA at a confidence level of 95%. Line graphs were drawn to illustrate the weekly emergence of species and the change in height and width indices over time and bar graphs were drawn to illustrate the average DM produced per trial pot.

The emergence percentage of the lower seeding density trials and the bio-stimulant trials used in Phase 3 were compared in a bar graph and one-way ANOVA was used (confidence level 95%) to determine significant differences between the seedling emergence of seed treatments, followed by the post-hoc Tukey HSD test if these differences were significant.

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Chapter 4: Results and Discussion

4.1 Soil analyses

The degree of acidity, salinity, nutrient status, available P and exchangeable cation ratios of the soil analytical data for the:

- Phase 1 field trial plots before amelioration, on the sowing date and at the end of the trial period.
- Phase 2 supporting pot trial growth mediums on the sowing date after amelioration.
- Phase 3 additional trial plots after amelioration on the sowing date.

Are displayed in Tables 4.1–4.6 in Sections 4.1.1–4.1.3 and were interpreted using guidelines set out in the Fertilizer handbook (FSSA, 2007).

4.1.1 Phase 1 field trials (Soil analysis)

The none-ameliorated Crown gold TSF site tailings have a loamy sand texture (79,6% sand, 14,4% silt and 6% clay; Table 4.4) (The Non-Affiliated Soil Analysis Work Committee, 1990:35.9) and are very acidic, with a pH(H_2O) of 3,5 and pH(KCl)of 3,4) and a nett acid-producing potential of 35 tons/ha (Table 4.1 and Table 3.4). The severe acidity of the tailings and the high amount of sulphate (11425 mg/l) can be ascribed to the presence and oxidation of pyrite within the tailings (Table 4.1 and 4.2). Dolomitic lime amelioration raised the pH to an acceptable 6,4 at the time of seeding. The pH continued to rise and measured 7,1 at the end of the trials in June 2017 (Table 4.1).

Concerning the exchangeable cation ratios of the none-ameliorated Crown gold TSF site tailings, the calcium:magnesium (Ca:Mg) ratio is on the lower acceptable threshold of 1,5. The magnesium:potassium (Mg:K) ratio is much higher than the acceptable ratio of 4 and the K CEC percentage is below the normal threshold of 3–7. After amelioration these ratios improved to acceptable levels, Ca:Mg (3,4), Mg:K (5,1), K as percentage of CEC (3,1) (Table 4.1).

The EC of the Crown gold TSF tailings was 434 mS/m, which is above the allowable limits of 200–400 mS/m for sensitive plants (FSSA, 2007:55). After amelioration it decreased but still remained at a level where sensitive plants could be affected. The nutrient status results do not reveal any major deficiencies or excessive amounts of Ca, Mg, and K but P (7 mg/kg) was below the lower threshold of 8 mg/kg. Fertilisation did however improve the available P to very acceptable levels above 35 mg/kg (Table 4.2).

In summary, the Crown gold TSF site site was acidic, saline and mainly deprived of P. The amount of lime (85 tons/ha), compost and fertiliser (Table 3.3) addressed the acidic pH and improved the tailings nutrient status to acceptable norms for plant growth. The EC decreased due to the breaking of the surface crust when the tailings were ripped that allowed precipitation of rainwater to leach the salts from the surface and near subsurface.

The Rooikraal gold TSF site trials have a sandy texture (97,5% sand): (The Non-Affiliated Soil Analysis Work Committee, 1990:35.9) and were very acidic with pH levels well below 5 (Table 4.1 and Table 4.4). The nett acid potential of the tailings are 6 tons lime per ha, much less than the 35 tons per ha of the Crown gold TSF (Table 3.4). The exchangeable Mg:K (0,6) cation ratio is less than recommended (3–4) before as well as after amelioration with dolomitic lime and fertiliser, but was higher (10,7) at the end of the trials in January 2017. The percentage K occupying the CEC (K% of CEC) (1,8%) was lower than recommended before amelioration, but with amelioration it increased (10,6%) to surpass the recommended percentage of 3–7%; it then decreased to a mere 0,2% when monitoring stopped in January 2017 (Table 4.1). The decrease is due to K-fixation through jarosite formation during the oxidation of pyrite.

Table 4.1: CEC, pH, and exchangeable cation ratios of Phase 1 field trial sites, Crown gold TSF site, Rooikraal gold TSF site and Paardekraal platinum tailings before amelioration with lime compost and fertiliser (Section 3.4.1), after amelioration on sowing date and at the end of the trials.

Trial site	**pH and Exchangeable cation ratios**										
Crown Gold TSF	**pH(H2O)**	**pH(KCl)**	**CEC**	**Ca:Mg**	**Mg:K**	**Ca+Mg:K**	**K% of**	**Ca%**	**Mg%**	**K%**	**Na%**
			cmol(+)/kg				**CEC**	**as % of S value**			
Before amelioration 01/2016	3,4	3,5	5,7	1,5	15,1	38,4	2,6	59,0	38,4	2,5	0,0
After amelioration on sowing date 03/2016	6,4	5,7	11,2	3,4	5,1	22,5	3,1	73,0	21,2	4,2	1,6
At end of trials 06/2017	7,1	6,6	5,9	6,2	4,7	34,0	1,9	83,0	13,4	2,8	0,7
Rooikraal gold TSF	**pH(H_2O)**	**pH(KCl)**	**CEC**	**Ca:Mg**	**Mg:K**	**Ca+Mg:K**	**K% of**	**Ca%**	**Mg%**	**K%**	**Na%**
			cmol(+)/kg				**CEC**	**as % of S value**			
Before amelioration 01/2016	4,01	3,80	7,66	2,7	0,6	2,2	1,8	50,2	18,4	30,9	0,5
After amelioration on sowing date 03/2016	7,47	7,37	6,83	1,8	1,3	3,6	10,6	46,6	25,7	19,9	7,8
At end of trials 01/2017	6,12	4,42	7,66	9,1	10,7	108,2	0,2	88,8	9,7	0,9	0,5
Paardekraal platinum tailings	**pH(H_2O)**	**pH(KCl)**	**CEC**	**Ca:Mg**	**Mg:K**	**Ca+Mg:K**	**K% of**	**Ca%**	**Mg%**	**K%**	**Na%**
			cmol(+)/kg				**CEC**	**as % of S value**			
Before amelioration 01/2016	6,94	6,81	11,47	2,5	3,1	10,6	2,4	62,9	25,6	8,4	3,1
After amelioration on sowing date 04/2016	7,48	7,29	6,83	2,1	0,8	2,5	15,6	43,3	20,1	25,5	11,1
At end of trials 05/2016	7,49	7,33	6,60	2,9	5,9	22,8	2,1	70,6	24,7	4,2	0,5

Tailings from the Rooikraal TSF trial site did not pose any salinity risks with the EC well below 400 mS/m. The cation concentrations were deficient for Ca (45 mg/kg) and Mg (10 mg/kg) and P was virtually absent (0,00 mg/kg), K was just above the lower threshold set at 40 mg/kg. After the trial site was ameliorated the cation status was adequately raised to sufficient concentrations for Ca (341 mg/kg), Mg (114 mg/kg), K (283 mg/kg) and P (118 mg/kg). At the end of the trials the

concentrations of Mg (15 mg/kg) and K (4,5 mg/kg) fell below the sufficient thresholds of 50 mg/kg for Mg and 40 mg/kg for K. The decrease in Mg could be due to leaching of the sandy material from the tailings and the decrease in K due to K-fixation when jarosite is formed as a by-product of pyrite oxidation (Keene *et al.*, 2004:1).

The trial area measured a moderate acidic pH(KCl); this and the sharp decrease in the concentrations of Mg and K were most likely caused by the excess tailings material deposited over the trials from the adjacent TSF slope during an intense rainfall event.

The Paardekraal platinum tailings measured a pH(H_2O) and –(KCl) of 6,94 and 6,81 respectivly, slightly below neutral, which is quite optimum for plant growth. After amelioration with compost and fertiliser it increased to a slightly alkaline level. The Ca:Mg ratio was sufficient at 2,5–2,9 before amelioration until the trials ended. The Mg:K ratio was efficient at 3,1 before amelioration but the K as a percentage of CEC was insufficient at 2,4%. After the fertiliser application the K as a percentage of CEC was raised to a very sufficient 15% and the Mg:K dropped to 0.8. At the end of the trials the Mg:K ratio had increased to 5,9 and the exchangeable K fell to 2%, indicating that the change in the Mg:K ratio was caused by the addition of the N:P:K fertiliser (Table 4.1).

The Paardekraal platinum tailings were classified into the textural class sand (90% sand, 6,7% silt, 3,4% clay; Table 4.4) (The Non-Affiliated Soil Analysis Work Committee, 1990:35.9) (Table 4.4). It did not pose any salinity risks before amelioration, but after it was ameliorated with compost and fertiliser it had a slightly saline EC (340 mS/m) that, according to the FSSA (2007:55), would affect the growth of saline sensitive crops. The nutrient concentrations Ca, Mg, K and P were sufficient before amelioration and increased after amelioration. K however had decreased at the end of the trials (Table 4.2).

Table 4.2: EC, Salt concentrations and nutrient status of Phase 1 field trial sites Crown, Rooikraal and Paardekraal platinum tailings before amelioration with lime compost and fertiliser (Section 3.4.1), after amelioration on sowing date and at the end of the trials.

Trial site	**EC and Anions**				**Nutrient status**				
Crown Gold TSF	**EC**	**Cl**	**NO_3**	**SO_4**	**Ca**	**Mg**	**K**	**Na**	**P-BRAY 1**
	mS/m	**mg/l**			**mg/kg**				
Before amelioration 01/2016	434	35,1	37,1	11425,2	686,5	271,0	57,5	0,5	7,0
After amelioration on sowing date 03/2016	331	73,0	9,2	2481,1	1224,5	216,0	137,0	30,0	56,6
At end of trials 06/2017	121	5,7	7,3	628,3	647,5	63,5	43,0	6,5	68,6
Rooikraal gold TSF	**EC**	**Cl**	**NO_3**	**SO_4**	**Ca**	**Mg**	**K**	**Na**	**P-BRAY 1**
	mS/m	**mg/l**			**mg/kg**				
Before amelioration 01/2016	27,00	1,58	1,27	112,52	45,0	10,0	54,0	0,5	0,0
After amelioration on sowing date 03/2016	122	123,92	9,46	176,29	341,0	114,0	283,5	65,5	118,5
At end of trials 01/2017	123	1,88	3,09	785,66	225,5	15,0	4,5	1,5	15,7
Paardekraal platinum tailings	**EC**	**Cl**	**NO_3**	**SO_4**	**Ca**	**Mg**	**K**	**Na**	**P-BRAY 1**
	mS/m	**mg/l**			**mg/kg**				
Before amelioration 01/2016	90,00	21,78	87,58	87,58	411,5	101,5	106,5	23,5	8,5
After amelioration on sowing date 04/2016	340	594,94	18,73	173,74	361,5	102,0	414,5	106,0	224,0
At end of trials 05/2016	49	32,35	0,33	98,22	478,0	101,5	55,0	4,0	131,3

To summarise: both the gold mine TSF sites were acidic, but after amelioration with 85 tons/ha of dolomitic lime (Table 3.3) the acidity was neutralised. The source of the acidity may be ascribed to the oxidation of iron-sulphide minerals.

Only the Crown gold mine tailings posed a moderate salinity risk (434 mS/m), while the ameliorated Paardekraal platinum tailings were only slightly saline (340 mS/m) (Table 4.2).

Initially K and P are the most deficient nutrients. Exchangeable K appears as the nutrient most in demand for each growth medium. It is initially not sufficiently present on the exchange complex (< 3%) after amelioration it is sufficiently present (> 3%), but is depleted (< 3%) at the end of the trials. Keene *et al.* (2004:1) explain that in sulphate acid soils, the secondary mineral jarosite forms through the oxidation of sulphide minerals. This acts as an infinite sink for K in the upper oxidative zone of the soil and reduces the amount of plant available K which would explain the reduction of K post fertilisation on the Crown and Rooikraal trial sites.

P is not sufficiently present in raw gold mine tailings (< 8 mg/kg), but with a high initial input (600 kg/ha) of superphosphate it is adequately present (Table 4.2), only requiring small top-up fertiliser dressings.

4.1.2 Phase 2 supporting pot trials (Soil analysis)

Table 4.3 and 4.4 show the results of the soil analyses for the growth mediums used in the Phase 2 pot trials before any amelioration.

The Aquarius platinum mine tailings are very alkaline with a pH(H_2O) and pH(KCl) of 8,8 and 9,15 as shown in Table 4.3. In Table 4.3 it is clear that the exchangeable Ca:Mg ratio (6,4) is adequate and the Mg:K (12.4), Ca+Mg:K (91,5) exceed the normal values of 3–4 and 10–20. The exchangeable K percentage of CEC is below the normal value of 3–7. The platinum tailings do not have a salinity risk but are deficient in K (7 mg/kg) and P (3,2 mg/kg). The large concentration of nitrate (NO_3) (56,9 mg/l) is deemed the residue from explosives used during mining (Table 4.3 and 4.4).

The control soil was very acidic (pH(H_2O) 4,1), the exchangeable Ca:Mg ratio (1,1) was insufficient. It did not have any salinity risk (EC 13 mS/m), but was deficient in Ca (88 mg/kg). The elevated nitrate concentration can be attributed to previous fertilisation applications when the soil was used for farming purposes (Table 4.3 and Table 4.4).

Table 4.3: Exchangeable cations and pH of growth mediums in Phase 2 supporting pot trials, Crown gold mine tailings, Rooikraal gold mine tailings, Aquarius platinum tailings and control soil before amelioration with ameliorants (Section 3.4.2).

Pot trial growth mediums	pH, EC and Exchangeable cation ratios										
	pH(H2O)	pH(KCl)	CEC	Ca:Mg	Mg:K	Ca+Mg:K	K% of	Ca%	Mg%	K%	Na%
			(cmol(+)/kg)				CEC	as % of S value			
Crown gold tailings	3,4	3,5	5,7	1,5	15,1	38,4	2,6	59,0	38,4	2,5	0,0
Rooikraal gold tailings	4,0	3,8	7,7	2,7	0,6	2,2	1,8	50,2	18,4	30,9	0,5
Aquarius platinum tailings	8,8	9,15	6,9	6,4	12,4	91,5	0,3	83,1	13,0	1,1	2,8
Control soil	4,1	4,1	1,4	1,1	2,5	5,3	11,4	43,6	40,2	15,8	0,4

Table 4.4: The EC, anion (Cl, NO_3, SO_4), nutrient status and particle size distribution of the growth mediums (Crown tailings, Rooikraal tailings, Platinum tailings and Control soil) used in the Phase 2 supporting pot trials.

Pot trial growth medium	EC and Anions				Nutrient status					Particle size distribution		
	EC	Cl	NO_3	SO_4	Ca	Mg	K	Na	P-Bray 1	Sand	Silt	Clay
	mS/m	mg/l			mg/kg					%		
Crown gold tailings	434,0	35,1	37,1	11425,2	686,5	271,0	57,5	0,5	7,0	79,6	14,4	6,0
Rooikraal gold tailings	27,0	1,6	1,3	112,5	4,5	10,0	54,0	0,5	0,0	97,5	1,6	0,8
Aquarius platinum tailings	39,0	28,4	56,9	41,4	284,5	27,0	7,0	11,0	3,2	90,0	6,7	3,4
Control soil	13,0	3,4	48,8	31,1	88,0	49,0	62,0	1,0	4,0	92,8	2,9	4,3

Table 4.5 illustrates the pH and EC of the growth mediums when the coated and uncoated seeds were sown in November 2017 after the gold mine tailings and the control soil were ameliorated with lime and each of the growth mediums were ameliorated with compost and fertiliser. It also illustrates the pH and EC of the growth mediums at the end of the trials in March 2017.

Crown gold mine tailings, Rooikraal gold mine tailings and the control soil were neutralised on the pot trial sowing date 1 November. The platinum tailings had a highly alkaline pH (9). Only the Crown gold mine tailings had an EC measurement that could be regarded as a salt hazard to sensitive plants (490 mS/m) the other growth mediums had EC measurements well below 200 mS/m when seeds were sown in November 2016. At the end of the trials five months later, the pH of the gold mine tailings continued to increase and become alkaline, while the platinum tailings and the control soil decreased. The Crown gold, Rooikraal gold, and Control soils EC decreased to below 60 mS/m and the platinum tailings had a slight increase in salinity. The decrease of the gold mine tailings EC reading can be explained by the leaching of salts from the growth mediums in the pots by irrigation water and rainfall events.

Table 4.5: Electrical conductivity (EC) and pH of growth mediums used in pot trials on the day seeds were sown in November 2016 and at the end of trials in March 2017.

Growth mediums	On plant day 1 November 2016		End of trial March 2017	
	pH	EC in mS/m	pH	EC in mS/m
Crown gold	6,7	490	7,92	59,8
Rooikraal gold	7,8	178	8,27	55,2
Aquarius platinum	9	55,6	8,18	94,3
Control soil	7,7	111	6,49	43,7

4.1.3 Phase 3 Rooikraal soil analysis on sowing date

The trial plots used for the Phase 3 trials were ameliorated according to the previous trials on the Rooikraal gold TSF site (Section 3.4.1, Table 3.3).

Table 4.6 displays the soil analysis results of the trial plots on the day the Phase 3 seed treatments were sown in January 2017.

According to Table 4.6 that shows the soil analysis results for the Rooikraal gold TSF site during the Phase 3 field trials after it was ameliorated. The lime application of 85 tons/ha was sufficient to neutralise the tailings acidity. The exchangeable cation ratio Ca:Mg was above the normal values prescribed (1,5-4,5). The Mg:K ratio was just below the normal ratio of three to four. The K percentage of the CEC was sufficient at 4,3%. The EC (183 mS/m) of the ameliorated tailings did not pose a salt hazard to the plants. Ca, Mg and P were sufficiently present after amelioration.

Table 4.6: Soil analysis results for Phase 3 trial sites on the Rooikraal gold TSF site after amelioration displaying the pH(H_2O and KCl), CEC, exchangeable cation ratios, EC, Anions and nutrient status for the tailings after amelioration (Section 3.4.1).

Rooikraal Phase 3 trial site										
pH and Exchangeable cation ratios										
pH(H_2O)	**pH(KCl)**	**CEC**	**Ca:Mg**	**Mg:K**	**Ca+Mg:K**	**K% of**	**Ca%**	**Mg%**	**K%**	**Na%**
		cmol(+)/kg				**CEC**	**as % of S value**			
7,30	7,03	5,50	5,1	2,7	16,9	4,3	77,8	15,1	5,5	1,6
EC and Anions				**Nutrient status**						
EC	**Cl**	**NO3**	**SO4**	**Ca**	**Mg**	**K**	**Na**	**P-BRAY 1**		
mS/m	**mg/l**			**mg/kg**						
183	44,2	130,2	511,0	671,0	79,0	92,5	16,0	84,9		

4.2 Phase 1 Field trials vegetation emergence and survival results

Results of the seedling emergence, change in plant density and cover contribution of species for the coated and uncoated seed treatments sown at the various trial areas during the Phase 1 trials are illustrated and discussed in the following part of this Section.

4.2.1 Seedling emergence at the gold TSFs

The seedling emergence data compare the emerged percentage of seedlings from coated and uncoated seed treatments and the individual plants surviving per square meter (plants/m^2) out of the total amount of seed sown plants/m^2 within the seed treatments.

The seedling emergence percentage for the coated (T1C, T3C, T4C) and uncoated (T2UC, T5UC) seed treatments sown in March 2016 at the Crown and Rooikraal gold TSF sites are illustrated in Figure 4.1. The emerged seedling density in April 2016 is shown in Figure 4.2. Significant differences between seed treatments in for the Crown gold TSF site are illustrated by bold italic symbols e.g. "***a***", the significant differences between seed treatments for the Rooikraal gold TSF site are illustrated with normal font e.g. "a".

On both the Crown and Rooikraal gold TSF sites the T1C, T3C and T4C had a higher emergence percentage than the uncoated seeds at the same sites, i.e. T2UC and T5UC. Even though the seeding rate of the coated seed were increased to sow the same amount of coated seed T3C as uncoated seed treatment T2UC, the coated seed treatment T3C still had a better emergence percentage than the uncoated seed treatment T2UC at both Crown and Rooikraal sites (Figure 4.1). This increase was however not significant.

The One-way ANOVA indicated a significant difference in the seedling emergence for treatments sown on the Crown gold TSF site (Table 4.7). According to the Tukey HSD analysis, treatment

T1C (10.9%) had a higher emergence percentage than T2UC (4,8%), T3C (6,1%), T4C (7,1%) and T5UC (5,3%) (Figure 4.1 and Table 4.7).

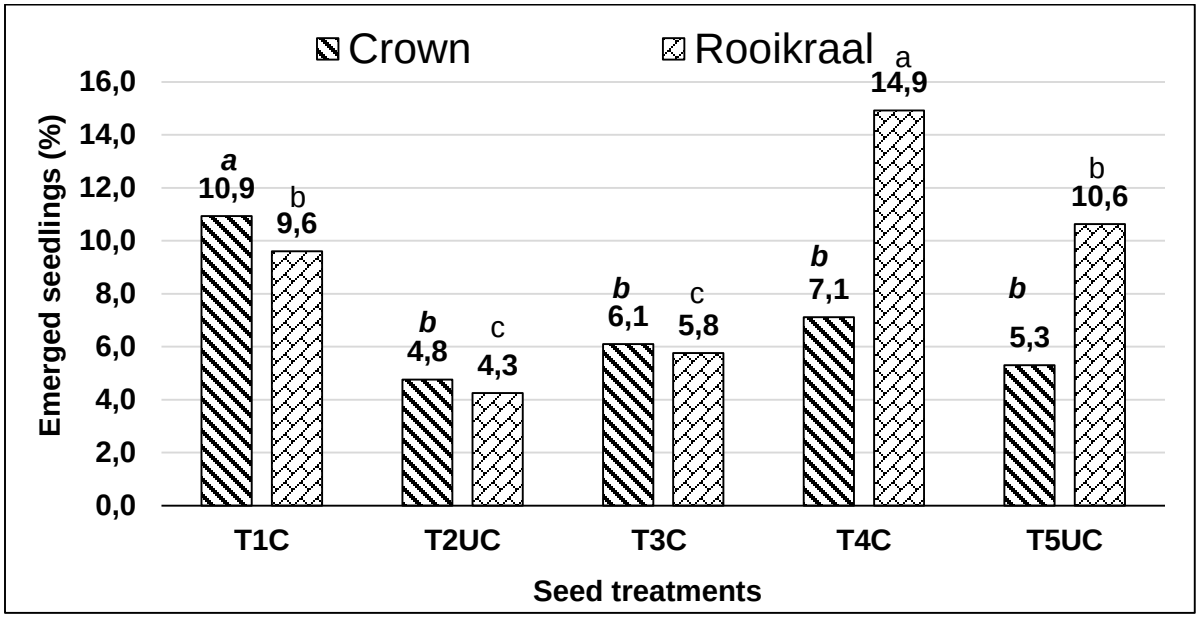

Figure 4.1: Seedling emergence percentage (%) of coated and uncoated seed treatments (T1C, T2UC, T3C, T4C, T5UC – see Section 3.4.1) at the Crown and Rooikraal gold TSF sites in April 2016. Statistical significance ($p < 0,05$) between the emergence percentage of seed treatments at Crown is illustrated in bold italics i.e. "***a***". Statistical significance between emergence percentage at Rooikraal is illustrated in normal font.

According to the One-way ANOVA post hoc Tukey HSD analysis there was no significant difference between the mean emerged seedling density for seed treatments sown on Crown (Figure 4.2 and Table 4.7). Because there is no significant difference between the mean seedling emergence density (Figure 4.2) in any of the seed treatments sown at Crown and the coated seed treatment, T1C (10,9%) had a significantly higher seedling emergence percentage (Figure 4.1) than coated seed treatment 3 (T3C) which was sown at an increased seeding rate (34,4 kg/ha) according to the coated and uncoated seed ratios (Figure 3.10, 3.12 and Table 3.2). An increased seeding rate according to the seedling emergence percentage at the Crown site did not increase seedling establishment density. At the Rooikraal site the mean emerged seedling density of T5C (283,7 plants/m^2) and T4C (228,9 plants/m^2) was significantly higher than T3C (132,9 plants/m^2), T1C (126,4 plants/m^2) and T2UC (103,7 plants/m^2) (Figure 4.2 and Table 4.7). This is most likely caused by the higher rate of *E. tef* seed sown (1,9 kg/ha) in these treatments compared to the seeding rate of *E. tef* sown (1 kg/ha) in the other three seed treatments (T1C, T2UC and T3C) (Figure 3.10 and Table 3.2).

At the Rooikraal TSF site, coated seed treatment 1 (T1C) (9,6%) had a significant higher seedling emergence (Figure 4.1 and Table 4.7) than T2UC (4,7%) and T3C (5,8%), but there was no significant difference between the average emerged seedling densities in March 2016 for T1C (126,4 plants/m^2), T2UC (103,7 plants/m^2) and T3C (132,9 plants/m^2) (Figure 4.2 and Table 4.7). This indicates that the increase in the seeding rate of coated seed to equal the number of seed contained within an uncoated seed treatment did not result in improved emergence results for the coated seed treatment at the Rooikraal gold site (Figure 4.1, 4.2 and Table 4.7).

Coated seed treatment 4 (T4C) (14,9%) had the best seedling emergence percentage and was significantly higher than all the seed treatments at Rooikraal, T1C (9,6%), T2UC (4,3%), T3C (5,8%) and T5UC (10,6%) (Figure 4.1). However, when the density of seedlings present in the treatment trials are considered (Figure 4.2), there was no significant difference between the mean seedling emergence density of T5UC (283,7 plants/m^2) and T4C (228,9 plants/m^2) (Figure 4.2). Both these seed treatments (T5UC and T4C) had a significantly higher emerged seedling density in March 2016 than T1C (126,4 plants/m^2), T2C (103,7 plants/m^2) and T3C (132,9 plants/m^2) (Figure 4.2 and Table 4.7). This can be attributed to the higher *E. tef* seeding rate (1,9 kg/ha) within T4C and T5UC (Figure 3.11 and Table 3.2).

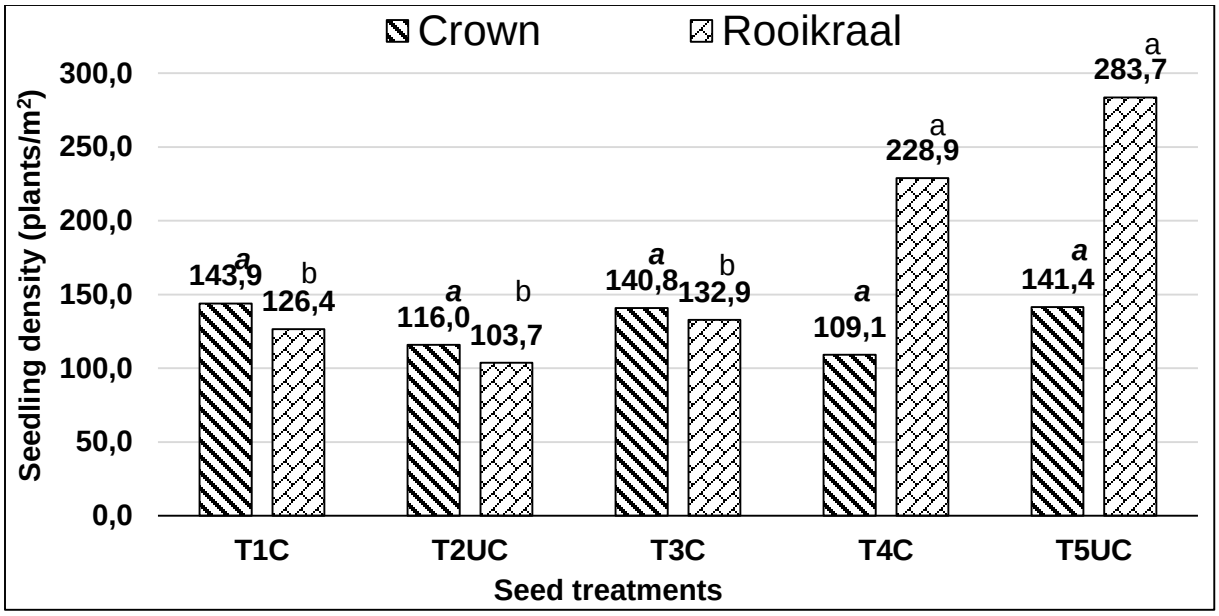

Figure 4.2: Average seedling emergence density in plants per square meter (plants/m2) in April 2016 for coated and uncoated seed treatments (T1C, T2UC, T3C, T4C, T5UC see Section 3.4.1. sown at the Crown and Rooikraal TSF sites. Statistical significance ($p < 0,05$) between the emergence density of seed treatments at Crown are illustrated in bold italics i.e. “a”. Statistical significance between emergence density of seed treatments at Rooikraal are illustrated in normal font.

Table 4.7: One-way ANOVA and post Hoc Tukey HSD results illustrating the statistical significance ($p < 0,05$) of variance between the mean emergence percentage (%) and seedling density (plants/m^2) of seed reatments (T1C, T2UC, T3C, T4C, T5UC see Section 3.4.1) in April 2016 at the Crown and Rooikraal TSF sites.

Crown			
Treatments	Emergence (%)	Treatments	Density (plants/m^2)
T1C	10,9 ***a***	T1C	143,9 ***a***
T4C	7,1 ***b***	T5UC	141,4 ***a***
T3C	6,1 ***b***	T3C	140,8 ***a***
T5UC	5,3 ***b***	T2UC	116,0 ***a***
T2UC	4,8 ***b***	T4C	109,1 ***a***
$p < 0,0001$		$p = 0,253$	
Significant	Yes	Significant	No
Rooikraal			
Treatments	Emergence (%)	Treatments	Density (plants/m^2)
T4C	14,9 a	T5UC	283,7 a
T5UC	10,6 b	T4C	228,9 a
T1C	9,6 b	T3C	132,9 b
T3C	5,8 c	T1C	126,4 b
T2UC	4,3 c	T2UC	103,7 b
$p < 0,0001$		$p < 0,0001$	
Significant	Yes	Significant	Yes

4.2.2 Seedling survival at the gold TSFs

The seedling survival percentage in September 2016 at the Crown and Rooikraal gold TSF sites for the various seed treatments sown in March 2016 are presented in Figure 4.3.

In September 2016, the T1C (6,8%-Crown and 6,2%-Rooikraal) and T4C (5,8%-Crown and 7%-Rooikraal) had a slightly higher plant survival compared to the treatments, T2UC (4,2% – Crown – and 3.1% – Rooikraal) and T5UC (4,8% – Crown and 4,2% – Rooikraal), which were sowedat the Crown and Rooikraal gold TSF sites (Figure 4.3).

At the Crown gold TSF site the surviving percentage of seedlings at treatment T1C (6,8%) was significantly higher than for treatments T2UC (4,2%), T3C (3,7%) and T5UC (4,8%), and the survival percentage of T4C (5,8%) was significantly higher than T3C (3,6%). However, there was only a significant difference in the mean density of surviving seedlings for T5UC (126,9 plants/m^2) and T3C (85,9 plants/m^2) (Figure 4.4 and Table 4.8). The seedling survival of T1C (6,8%) sown at a seeding rate of 19 kg/ha did result in significantly higher seedling survival compared to the seedling survival of the equivalent T2UC (4,2%) (Table 4.8). There is not a significant difference between the mean surviving seedling densities of T1C (90,1 plants/m^2), T2UC (102,2 plants/m^2), T3C (85,9 plants/m^2) and T4C (89,3 plants/m^2) (Figure 4.4 and Table 4.8), but the surviving seedling percentage of T1C (6,8%) is significantly higher than T3C (3,7%) (Figure 4.3 and Table 4.8). An increase in the seeding rate of T3C did not result in higher seedling survival densities between March and September 2016 at the Crown site (Figure 4.3, 4.4 and Table 4.8).

At the Rooikraal gold TSF site, the mean seedling survival of T1C (6,2%) and T4C (7%) was significantly higher than the seedling survival of T2UC (3,1%) and T5C (4,2%) and T3C (3,6%) that was sown at a higher seeding rate (34,4 kg/ha) (Figure 4.3 and Table 4.8). According to the post hoc Tukey analysis, only the mean survival density of seedlings in the T5UC (112,1 plants/m^2) and T2UC (74,3 plants/m^2) was significantly different, with no differences between the mean seedling survival density of seeds sown in the afore-mentioned seed treatments T5UC and T2UC and T1C (82,2 plants/m^2), T3C (82,3 plants/m^2) and T4 (108,1 plants/m^2) (Figure 4.4 and Table 4.8). The significantly higher seedling survival density of T5UC (112,1 plants/m^2) compared to treatment T2UC (74,3 plants/m^2) is likely due to the higher seeding rate of *E. tef* (1,9 kg/ha) in T5UC compared to T2UC (1 kg/ha), as this pioneer grass (*E.tef)* tends to emerge very fast (3 to 7 days) after sowing (Evert, *et al*., 2009:236; Kassier, 2002:32).

The mean survival of seedlings for T1C (6,8%) is significantly higher than the mean seedling survival of the equivalent weight T2UC (4,2%). Similarly, the mean seedling survival for the coated seed treatment T4C (5,8%) is significantly higher than the equivalent weight of uncoated seed sown in T5UC (4,2%) (Figure 4.3). It therefore seems that in treatments where coated seed was used, the seedling survival was higher than in uncoated seed treatments, even if the same weight of seed was used (i.e. 19 kg/ha uncoated seed) at especially the Rooikraal gold TSF site (Figure 4.3 and Table 4.8).

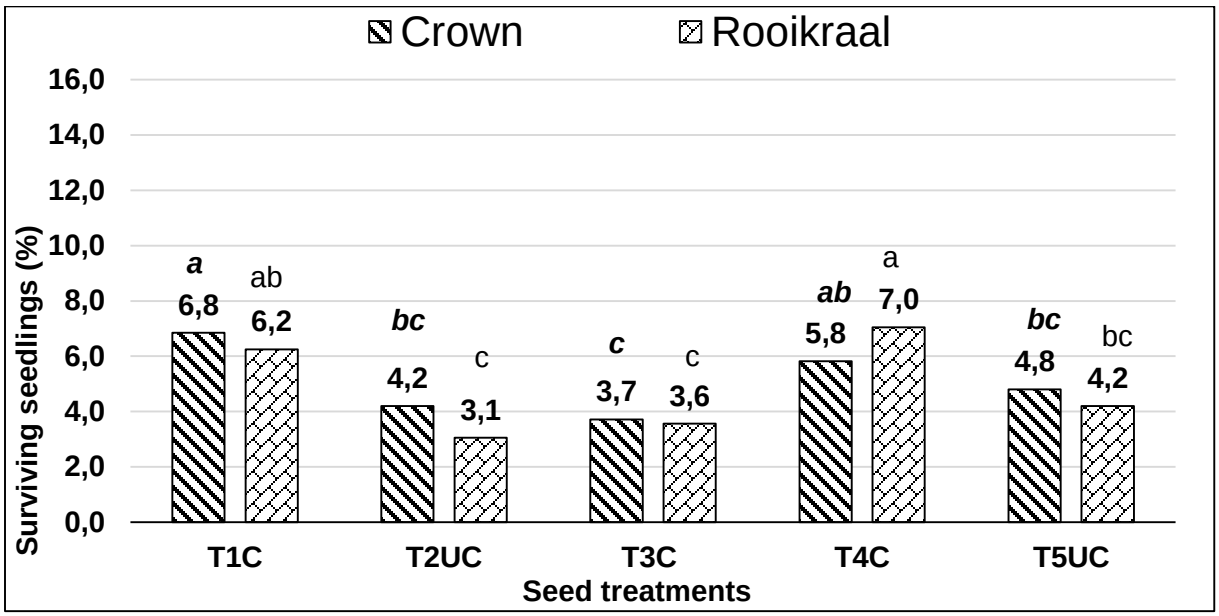

Figure 4.3: Average seedling survival percentage in September 2016 for coated and uncoated seed treatments (T1C, T2UC, T3C, T4C, T5UC Section 3.4.1) sown at the Crown and Rooikraal gold TSF sites. Statistical significance ($p < 0,05$) between the survival percentage of seed treatments at Crown is illustrated in bold italics i.e. "***a***". Statistical significance between survival percentage of seed treatments at Rooikraal is illustrated in normal font.

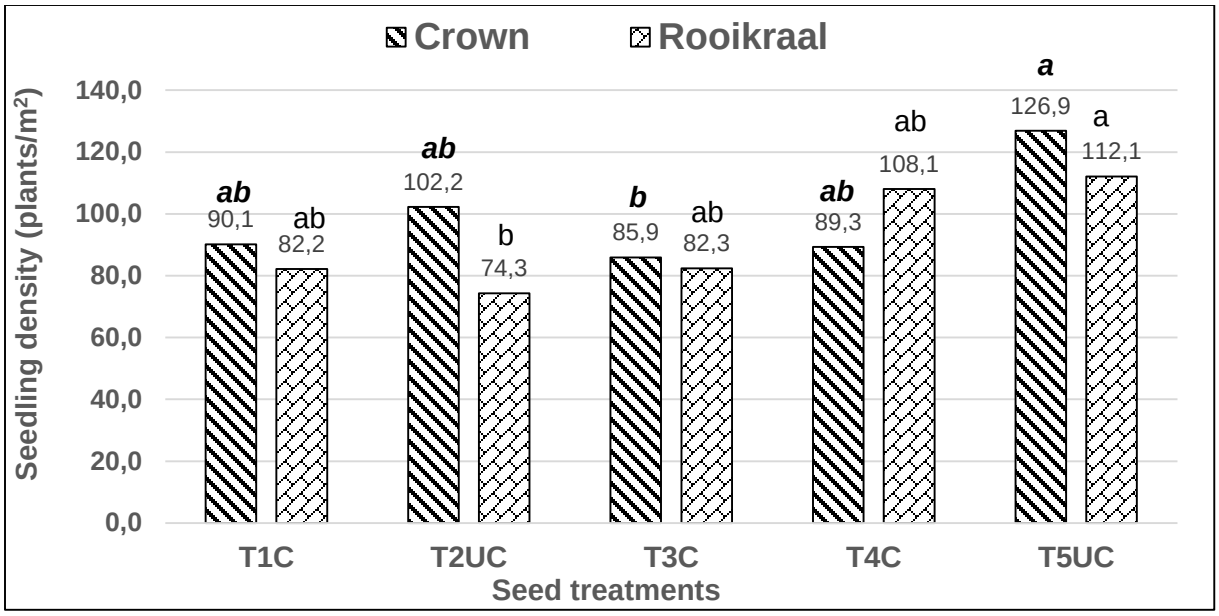

Figure 4.4: Average seedling survival density of seedlings in plants per square meter (plants/m^2) in September 2016 for coated and uncoated seed treatments (T1C, T2UC, T3C, T4C and T5UC Section 3.4.1) sown at the Crown and Rooikraal gold TSF sites. Statistical significance (p < 0,05) between the survival densities of seed treatments at Crown are illustrated in bold italics i.e. "***a***". Statistical significance between survival densities of seed treatments at Rooikraal is illustrated in normal font.

Table 4.8: One-way ANOVA and post Hoc Tukey HSD results illustrating the statistical significance (p < 0,05) of variance between the mean emergence percentage (%) and seedling density (plants/m^2) of seed treatments (T1C, T2UC, T3C, T4C and T5UC Section 3.4.1) in September 2016 at the Crown and Rooikraal TSF sites.

Crown			
Treatments	Survival (%)	Treatments	Density (plants/m^2)
T1C	6,8 ***a***	T5UC	126,9 ***a***
T4C	5,8 ***ab***	T2UC	102,2 ***ab***
T5UC	4,8 ***bc***	T1C	90,1 ***ab***
T2UC	4,2 ***bc***	T4C	89,3 ***ab***
T3C	3,7 ***c***	T3C	85,8 ***b***
p = 0,000		p = 0,026	
Significant	Yes	Significant	Yes
Rooikraal			
Treatments	Survival (%)	Treatments	Density (plants/m^2)
T4C	7,0 a	T5UC	112,1 a
T1C	6,2 ab	T4C	108,1 ab
T5UC	4,2 bc	T3C	82,3 ab
T3C	3,6 c	T1C	82,2 ab
T2UC	3,1 c	T2UC	74,3 b
p = 0,000		p = 0,017	
Significant	Yes	Significant	Yes

At the Crown and Rooikraal gold TSF sites, the species survival of coated seed in treatment T1C was significantly higher than the species survival of the uncoated seed treatment T2UC even if the same sowing rate of uncoated seed as coated seed in T1C (19 kg/ha) was used. However, an increase in the coated seed seeding rate according to the coated and uncoated seed ratio of the species selected for the field trials (Figures 3.10, 3.12 and Table 3.2) to a total seeding rate of 34,4 kg/ha in T3C, did not result in a significantly higher seedling survival percentage or a significantly higher surviving seedling density compared to the coated seed sown in T1C (19 kg/ha) and the uncoated seed sown in T2UC (19 kg/ha). This suggests that the seed survival percentage in September 2016 at the Crown and Rooikraal TSF sites for treatment T1C and T4C compared to T2UC and T5UC was significantly higher after the emergence in April 2016, due to the larger amount and weight of the coated seed compared to uncoated seed used. The amount of coated seed per unit weight is normally much lower than the amount of uncoated seed per unit weight. This results in a smaller number of coated seed per unit weight being sown compared to the number of uncoated seed in the same weight uncoated seed that is sown, but still provides the same survival results.

4.2.3 Seedling emergence and survival in the platinum trials

The seedling emergence in June 2016 for T1C (5,6%) was significantly higher than for the T2UC (2,2%), T3C (1,7%) and T5UC (2,7%) treatments in the platinum bulk bags trials as indicated in Figure 4.5. However, there was not a significant difference between the mean densities of emerged seedlings in any of the seed treatments. This indicates that a higher seeding rate of coated seed does not result in a significant increase in seedling emergence on platinum tailings (Figure 4.5).

The emergence percentages of seedlings for each of the five seed treatments in the platinum trials presented in Figure 4.5 are notably lower than that for the gold TSF trials at Crown and Rooikraal as illustrated in Figure 4.1. This is most probably due to the delayed sowing time carried out for the platinum trials, as the time for these trials was much shorter and more towards the winter time (May and June 2016) when the temperatures were already much cooler (Figure 3.9). In trials conducted by Evert *et al.* (2009:235) to evaluate the influence of temperature and planting depth on *E. tef*, Evert *et al.* (2009:235) concluded that when cool conditions (15 to 19°C) are experienced the emergence of *E. tef* was notably slower.

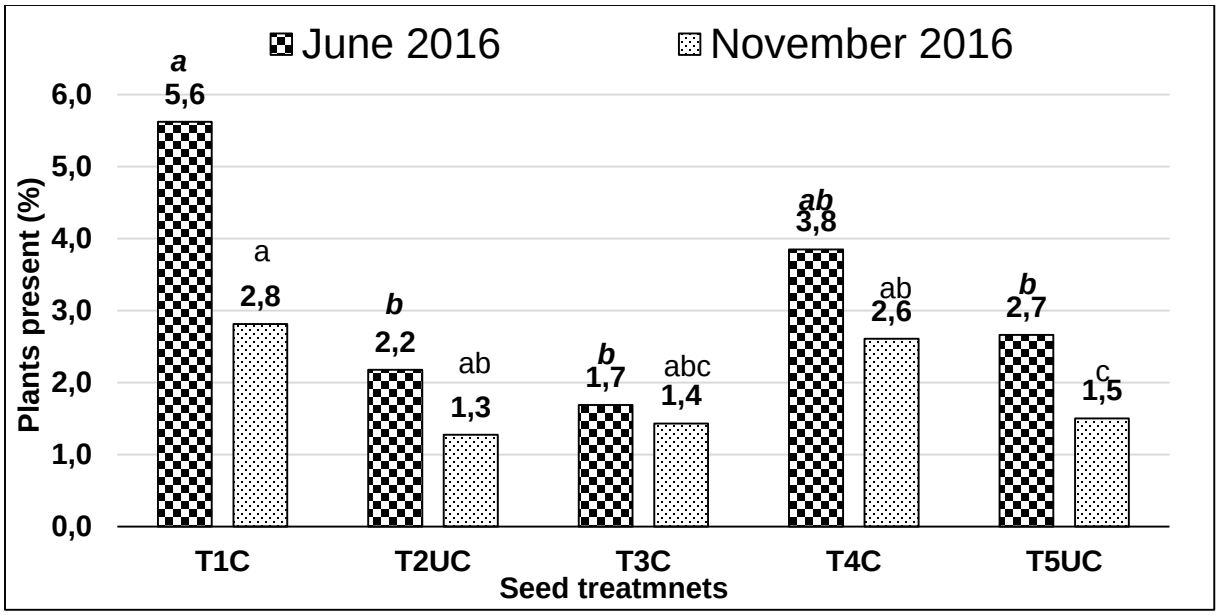

Figure 4.5: Average seedling emergence percentage in June 2016 and survival percentage in November 2016 for coated and uncoated seed treatments (T1C, T2UC, T3C, T4C and T5 UC Section 3.4.1) sown in platinum trials. Statistical significance ($p < 0,05$) between the emergence percentage of seed treatments in June 2016 are illustrated in bold italics i.e. "***a***". Statistical significance between survival percentage of seed treatments in November are illustrated in normal font.

In November 2016 the seedling survival % of T1C was 2,8% with 37 plants/m^2 and T4C was 2,6% with 40 plants/m^2 which was significantly higher than the seedling survival of T5UC that had a survival % of 1,5% and 10 plants/m^2 (Figure 4.6 and Table 4.9). No significant differences occurred in the mean survival of seedlings for treatments T1C (2,8% and 37 plants/m^2), T2C (1,3% and 31 plants/m^2), T3C (1,4% and 33 plants/m^2) and T4C (2,6% and 40 plants/m^2) (Figure 4.6 and Table 4.9).

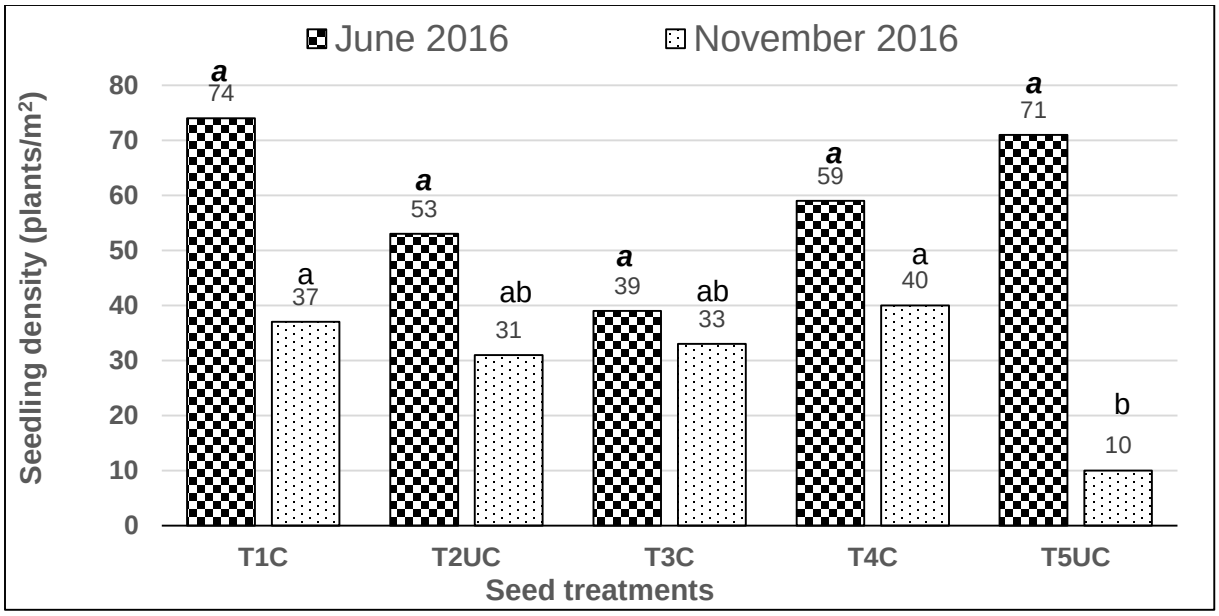

Figure 4.6: Average seedling emergence density in plants per square meter (plants/m²) in June 2016 and survival density in November 2016 for coated and uncoated seed treatments (T1C, T2UC, T3C, T4C and T5UC Section 3.4.1) sown in platinum trials. Statistical significance ($p < 0,05$) between the emergence density of seed treatments in June 2016 is illustrated in bold italics i.e. "***a***". Statistical significance between survival densities of seed treatments in November is illustrated in normal font.

Table 4.9: One-way ANOVA and post Hoc Tukey HSD results illustrating the statistical significance ($p < 0,05$) of variance between the mean emergence and survival percentage (%) and seedling density plants/m²) of seed treatments (T1C, T2UC, T3C, T4C and T5UC Section 3.4.1) in patinum trials in June 2016 and November 2016.

Platinum emergence			
Treatments	Emergence (%)	Treatments	Density (plants/m²)
T1C	5,6 a	T1C	74,0 a
T4C	3,8 ab	T5UC	71,0 a
T5UC	2,7 b	T4C	59,0 a
T2UC	2,2 b	T2UC	53,0 a
T3C	1,7 b	T3C	39,0 a
$p = 0,003$		$p = 0,21$	
Significant	Yes	Significant	No
Platinum survival			
Treatments	Survival (%)	Treatments	Density (plants/m²)
T1C	2,8 a	T4C	40,0 a
T4C	2,6 ab	T1C	37,0 a
T3C	1,4 abc	T3C	33,0 ab
T2UC	1,3 bc	T2UC	31,0 ab
T5UC	0,4 c	T5UC	10,0 b
$p = 0,000$		$p = 0,014$	
Significant	Yes	Significant	Yes

In summary: T3C did have higher seedling emergence in April 2016 compared to T2UC, even though it had the same amount of seeds. The higher seedling emergence for treatments T1C and T4C compared to treatments T2UC and T5UC is most likely caused by the lower amount of seed sown in the coated seed treatments, as the weight of coated seed is much higher. There is however not a significant difference between the seedling emergence density of coated and uncoated seed if the treatments T1C, T2UC and T3C on the Crown and Rooikraal gold TSF sites and in any treatments on the platinum bulk bags are considered.

As there is no significant difference between the mean average seedling densities during emergence when using coated or uncoated seeds, it is suggested that the same seedling emergence and survival results can be obtained if the seeding rate of seed is reduced. This matter is further discussed in Section 4.5

4.3 Change in species density, surviving plant composition and cover contribution of species for the Phase 1 field trials

The change in the species density and seedling composition for the Phase 1 field trials are discussed for each of the five seed treatments at the Crown gold TSF, the Rooikraal gold TSF sites and the platinum tailing bulk bag trials. A figure for the change in species density and the change in plant composition percentage is provided to illustrate the results for each seed treatment at the three different sites. A table summarising the plant density and plant composition averages is also provided for each treatment.

4.3.1 Crown gold TSF site species density and plant composition

Tufts of rye grass (*L. perenne*) were present in the treatments at a very low density of 1,8 plants/m^2. This was a surprise, since rye grass did not form part of the seed mixture that was originally sown. A possible source of the rye grass was that the dormant seeds were present in the compost used for the amelioration during the Phase 3 trials, as rye grass was only observed in these sites, or the seed mixture sown was not totally clean seed, but contaminated with seed from rye grass.

The species density for T1C is displayed in Figure 4.7 and Figure 4.8 illustrates the plant composition percentage of T1C. Table 4.10 ilustrates both the species density and plant composition of T1C illustrated in Figures 4.7 and 4.8.

Initially for the T1C treatment, all the species sown, *E. curvula, C. dactylon, E. tef*, *D. eriantha, S. bicolor* and *M. sativa* were present in June 2016, four months after sowing. The highest overall plant density occurred in June 2016, with a total of 110 plants/m^2 (Figure 4.7 and Table 4.10). The vegetation density gradually decreased from June 2016 to as low as 47 plants/m^2 in March 2017 *E. curvula* had the highest plant density per square meter of all the species at each sampling time throughout the trial period with a high plant density of 52,9 plants/m^2 as illustrated in Figure 4.7. *E. curvula* represented the largest fraction of the plant composition (Figure 4.8 and Table 4.10. The second highest recorded density was initially recorded for *E. tef at 28,7 plants/m^2* in June 2016 and 19,8 plants/m^2 in September 2016, but it died and disappeared from the plant composition in November 2016. *C. dactylon* had the highest density in June 2016 noted at 15,9 plants/m^2, but then dropped to as low as 8,1 plants/m^2 before increasing again to a density of 12,9 plants/m^2 in March 2017, after which it slightly decreased again to a density of 8 plants/m^2 (Figure 4.7).The lowest density noted for *D. eriantha* was 7,9 plants/m^2 in June 2016 after which the density of this grass rapidly decreased to 1,0 plants/m^2 in September 2016 In June 2017, it was totally absent (Figure 4.7).

Initially *S. bicolor* with a density of 2,9 plants/$m^2$and *M. sativa* with a density 0,8 plants/m^2 had the lowest densities of all the species sown (Figure 4.7 and Table 4.10. The density of *S. bicolor* slightly decreased to 0,8 plants/m^2 in September 2016, after which it increased to 1,2 plants/m^2 in November 2016. The lowest density for sorghum was recorded in March 2016 at 0,2 plants/m2 before it increased to 0,8 plants/m2 towards the end of the trail. *M. sativa* continued to decrease until it was totally absent in June 2017 (Figure 4.7).The density of the rye grass increased to a maximum of 3,1 plants/m^2 in November 2016 before decreasing to 0,2 plants/m^2 in March 2017 becoming totally absent in June 2017 (Figure 4.7).

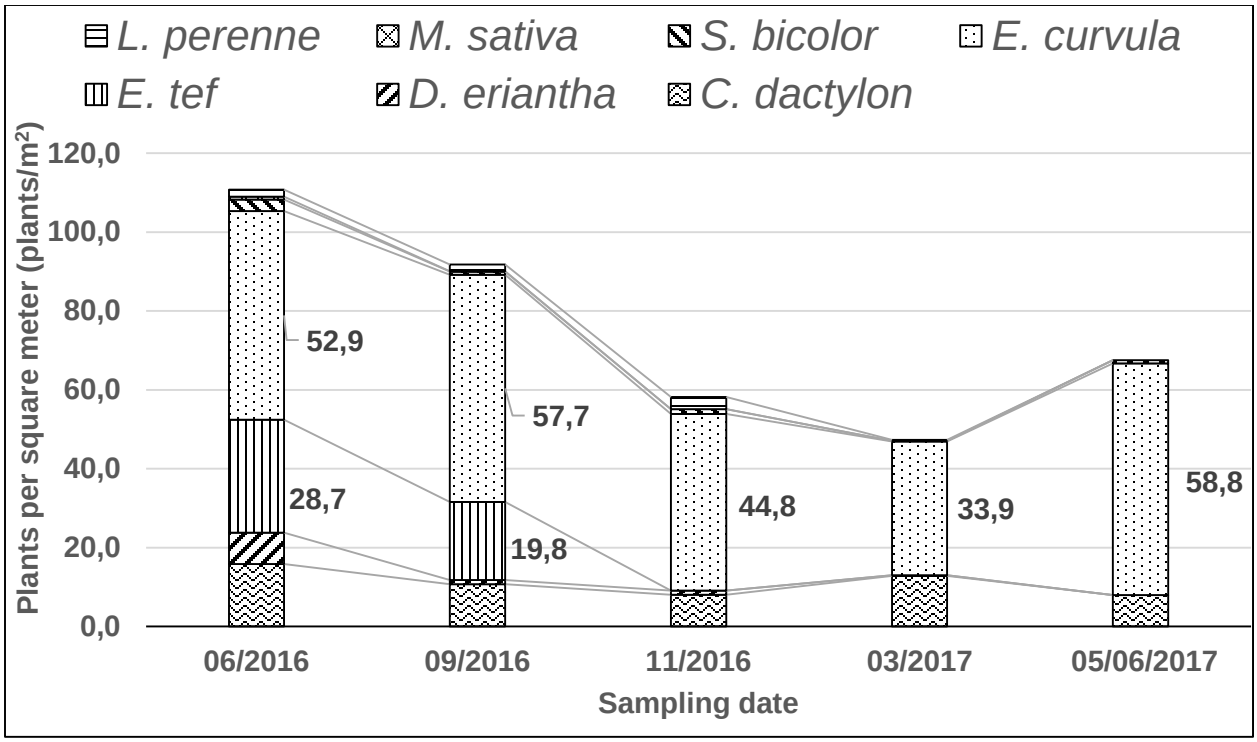

Figure 4.7: Change in density (plants/m^2) of species used (*C. dactylon, D. eriantha, E. curvula, E. tef, S. bicolor, M. sativa*) in T1C and *L.* perenne, at the Crown gold TSF site from June 2016 to June 2017.

In June 2016, the plant composition was dominated by *E. curvula* (47,7%), *E. tef* (25,9%) and *C. dactylon* (14,3%) (Figure 4.8). *E. Tef* first decreased to 21,5% before becoming totally absent in November 2016 (Figure 4.8). The plant composition was then dominated by *C. dactylon* and *E. curvula* until the end of the trial in June 2017 (Figure 4.8 and Table 4.10).

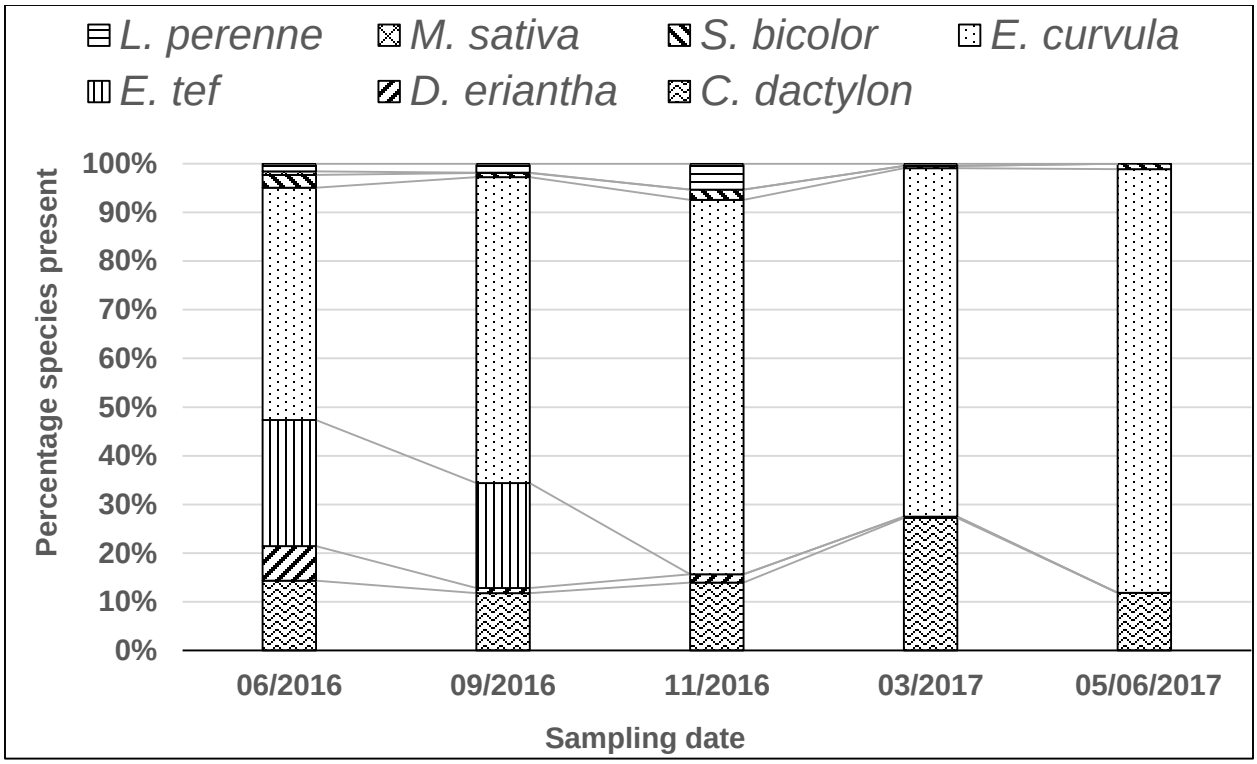

Figure 4.8: Change in plant composition percentage of species used (*C. dactylon, D. eriantha, E. curvula, E. tef, S. bicolor, M. sativa* and *L. perenne*) inT1C *at* the Crown gold TSF site from June 2016 to June 2017

Table 4.10: Summary of plant density (plants/m^2) and plant composition (%) results for *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor, M. sativa* and *L. perenne* in T1C at the Crown gold TSF site from June 2016 to June 2017.

T1C	06/2016		09/2016		11/2016		03/2017		06/2017	
Species	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**
C. dactylon	15,9	14,3	10,8	11,7	8,1	13,9	12,9	27,2	8,0	11,9
D. eriantha	7,9	7,1	1,0	1,1	1,0	1,7	0,1	0,2	0,0	0,0
E. tef	28,7	25,9	19,8	21,5	0,0	0,0	0,0	0,0	0,0	0,0
E. curvula	52,9	47,7	57,7	62,8	44,8	76,9	33,9	71,6	58,8	87,0
S. bicolor	2,9	2,6	0,8	0,8	1,2	2,1	0,2	0,5	0,8	1,1
M. sativa	0,8	0,7	0,1	0,1	0,0	0,0	0,0	0,0	0,0	0,0
L. perenne	1,8	1,6	1,7	1,8	3,1	5,3	0,2	0,5	0,0	0,0
Total	**110,8**	**100,0**	**91,8**	**100,0**	**58,2**	**100,0**	**47,3**	**100,0**	**67,5**	**100,0**

The species density and the plant composition percentage for T2UC is illustrated in Figure 4.9 and Figure 4.10, the results are also summarised and displayed in Table 4.11.

In the T2UC, the species with the highest recorded initial densities in June 2016 were *E. curvula* (37,7 plants/m^2), *E. tef* (28,6 plants/m^2) and *C. dactylon* (17,4 plants/m^2) (Figure 4.9 and Table 4.11). The density of *E. tef* decreased from its initial density of 28,6 plants/m^2 in June 2016 until died and was no longer present in November 2016 (Figure 4.9 and Table 4.11). The density for

E. curvula decreased to 37,9 plants/m^2 in November 2016 before increasing again to 398 plants/m^2 in June 2017. *C. dactylon* decreased to a low of 13.1 plants/m^2 in September 2016 before increasing to 17,3 plants/m^2 in November 2016 and then decreasing again to 13.8 plants/m^2 in June 2017 (Figure 4.9 and Table 4.11).

D. eriantha initially had the lowest density of the grasses in treatment T2UC (2,3 plants/m^2), but then slightly increased to 2,4 plants/m^2 in September 2016 before decreasing again to 0,2 plants/m^2 in March 2017. In June 2017 this species was totally absent (Figure 4.9 and Table 4.11).

M. sativa had a higher initial density of 3 plants/m^2 in treatment T2UC in June 2016, compared to T1C treatment of 0,8 plants/m^2 (Table 4.10 and 4.11). However, it decreased sharply before to be totally absent in March 2017 (Figure 4.9). *S. bicolor* had a lower initial density of 1,3 plants/m^2 compared to the density in June 2016 for treatment T1C 2,9 plants/m^2 (Table 4.10 and 4.11). It decreased to 0,2 plant/m^2 in March 2017 before disappearing in June 2017 (Figure 4.9 and Table 4.11). *L. perenne* was initially present at a density of 1 plant/m^2 in June 2016 and then increased to 1,7 plants/m^2 in September 2016, whereafter it decreased and was totally absent in March 2017 (Figure 4.9 and Table 4.11).

Initially the plant composition mainly comprised of *E. curvula* (41,2%), *E. tef* (31,3%) and *C. dactylon* (19,1%) (Figure 4.10 and Table 4.11. The abundance of *E. tef* decreased over time until it was totally absent (0%) from the established vegetation community in November 2016. *E. curvula* and *C. dactylon* were the only surviving species at the end of the trial (Figure 4.10 and Table 4.11).

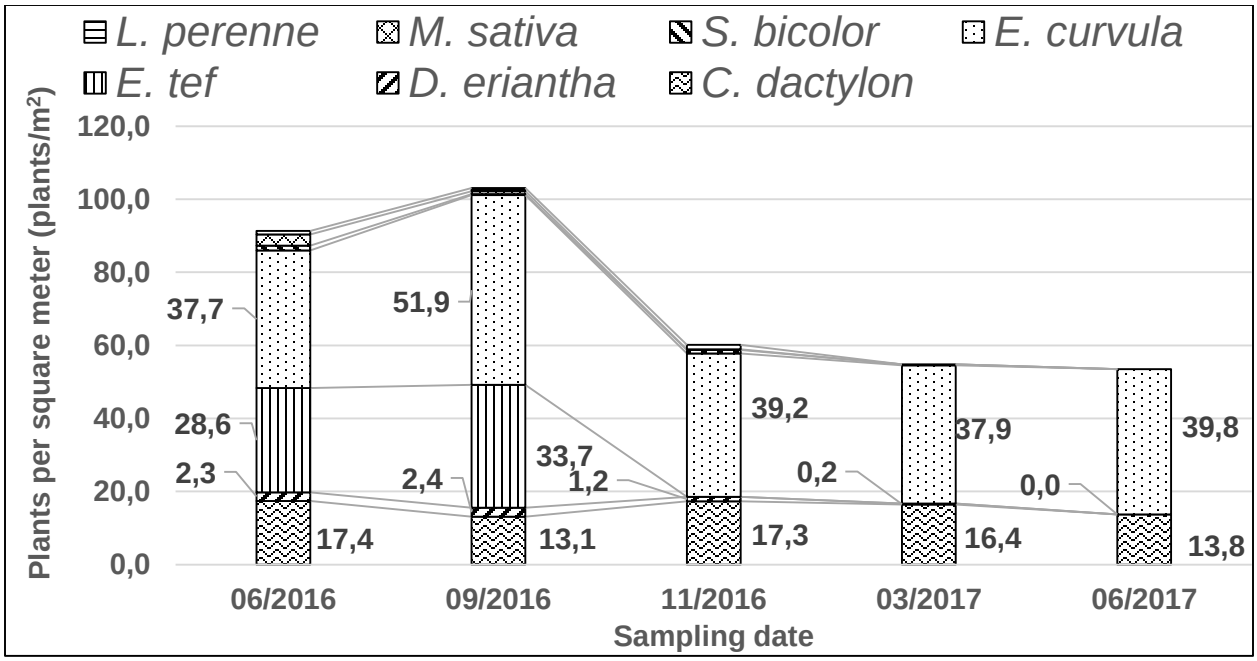

Figure 4.9: Change in plant (plants/m^2) of species used (*C. dactylon, D. eriantha, E. curvula, E. tef, S. bicolor, M. sativa* and *L. perenne*) in T2UC at the Crown gold TSF site from June 2016 to June 2017.

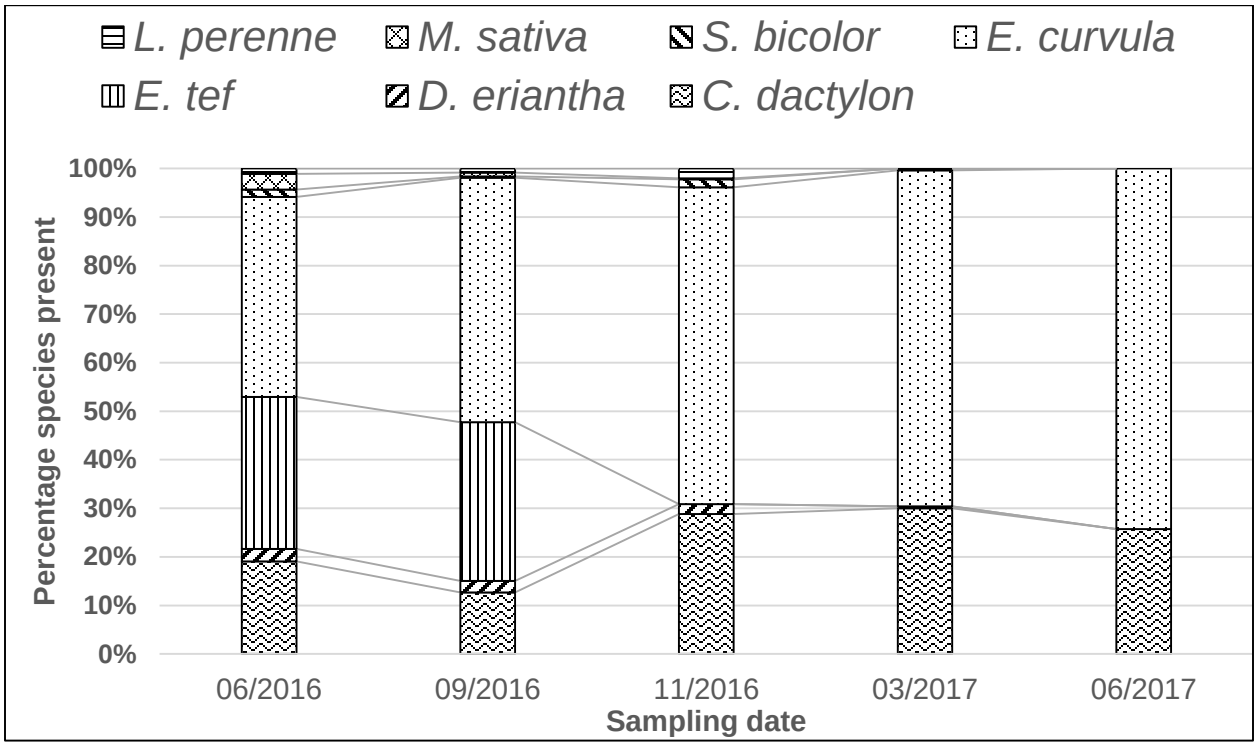

Figure 4.10: Change in plantplant composition percentage of species used (*C. dactylon, D. eriantha, E. curvula, S. bicolor, M. sativa* and *L. perenne*) in T2UC on the Crown gold TSF site from June 2016–June 2017.

Table 4.11: Summary of plant density (plants/m^2) and plant composition (%) results for *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor, M. sativa* and *L. perenne* in T2UC at the Crown gold TSF site from June 2016 to June 2017.

T2UC	06/2016		09/2016		11/2016		03/2017		06/2017	
Species	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**
C. dactylon	17,4	19,1	13,1	12,7	17,3	28,8	16,4	30,0	13,8	25,7
D. eriantha	2,3	2,6	2,4	2,4	1,2	2,0	0,2	0,4	0,0	0,0
E. tef	28,6	31,3	33,7	32,7	0,0	0,0	0,0	0,0	0,0	0,0
E. curvula	37,7	41,2	51,9	50,3	39,2	65,2	37,9	69,2	39,8	74,3
S. bicolor	1,3	1,5	0,3	0,3	1,0	1,7	0,2	0,4	0,0	0,0
M. sativa	3,0	3,3	0,8	0,8	0,1	0,2	0,0	0,0	0,0	0,0
L. perenne	1,0	1,1	0,9	0,9	1,2	2,0	0,0	0,0	0,0	0,0
Total	**91,3**	**100,0**	**103,1**	**100,0**	**60,1**	**100,0**	**54,8**	**100,0**	**53,5**	**100,0**

The plant density and the plant composition for T3C where the weight of the coated seed was increased according to the coating ratios which can be seen in Table 3.2 and Figure 3.11 of Section 3.4.1, are illustrated in Figure 4.11 and Figure 4.12 and Table 4.12.

Initially the density as for treatments T1C and T2UC, for *E. curvula* was 52,7 plants/m^2, *E. tef* 27,8 plants/m^2, and *C. dactylon* 23,3 plants/m^2 were the three dominant species and *D. eriantha* with the lowest density of 5,6 plants/m^2 for the four grasses. *E. tef* decreased until it was no longer present in November 2016 however, dormant *E. tef* seeds germinated afterwards and according to Table 4.12 *E. tef* was present in March 2017 a density of 0,3 plants/m^2. The density of *D. eriantha* was very low at 0,3 plants/m^2 and *E. tef* was no longer present at the end of the trail period in June 2017. *E. curvula* and *C. dactylon* were the dominant surviving grass species at the end of the trials (Figure 4.11 and Table 4.12).

S. bicolor and *M. sativa* were initially present at densities of 1,3 plants/m^2 and 1,4 plants/m^2 respectively, in June 2016, whereafter *M. sativa* decreased until totally absent in June 2017 and *S. bicolor* still had a slight density of 0,3 plants/m^2 in June 2017 (Figure 4.11 and Table 4.12). *L perenne* was initially present at a density of 0,6 plants/m^2 in June 2017, whereafter it increased to 1,7 plants/m^2 before decreasing until it was totally absent in March 2017 (Figure 4.11 and Table 4.12).

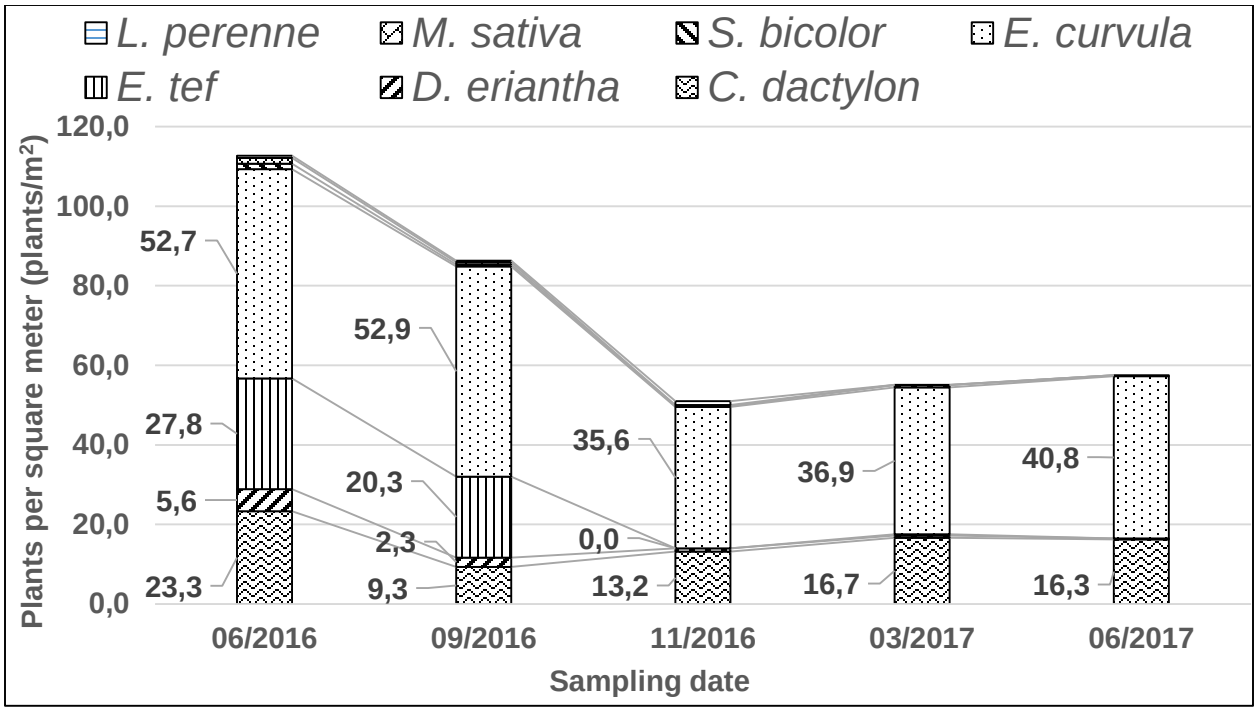

Figure 4.11: Change in density (plants/m^2) of species used (*C. dactylon, D. eriantha, E. curvula, E. tef, S. bicolor, M. sativa* and *L. perenne*) in T3C at the Crown gold TSF site from June 2016 to June 2017.

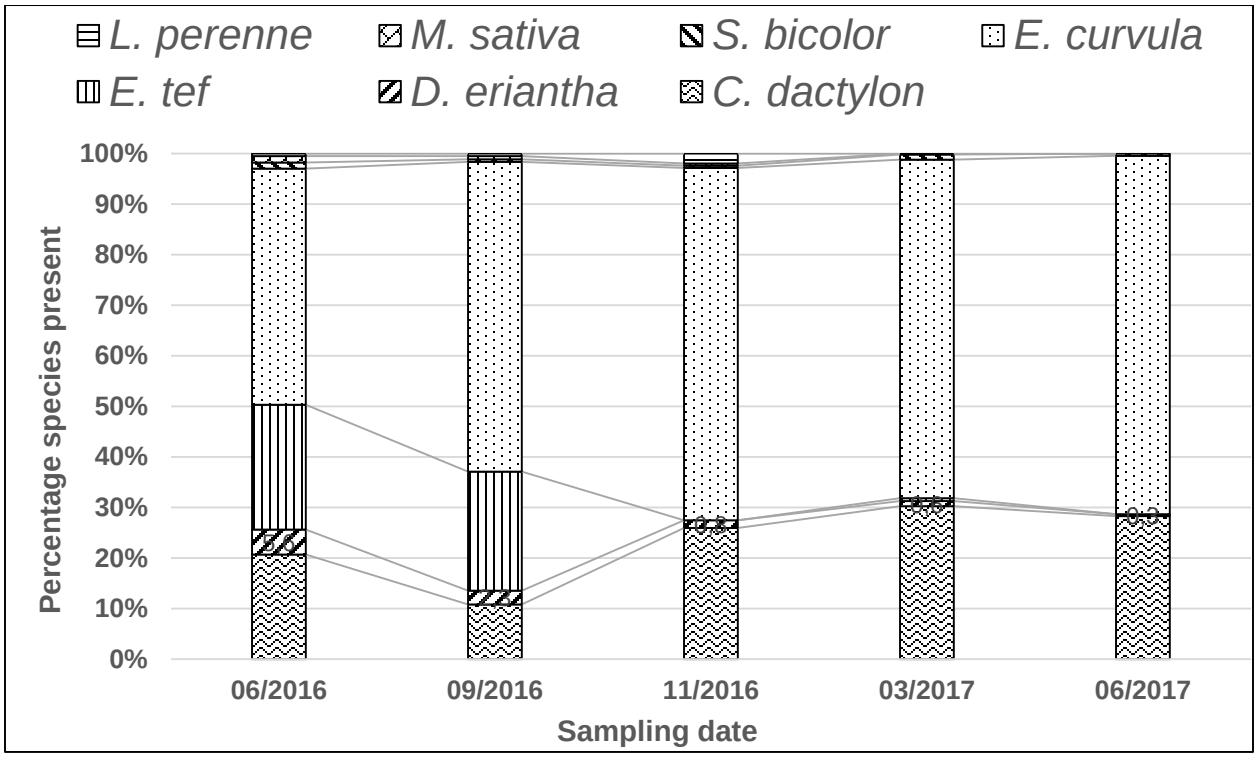

Figure 4.12: Change in plant composition percentage of species used (*C. dactylon, D. eriantha, E. curvula, E. tef, S. bicolor, M. sativa* and *L. perenne*) in T3C at the Crown gold TSF site from June 2016–June 2017.

The plant composition for T3C followed the same pattern as for treatments T1C and T2UC. In Figure 4.12 and Table 4.12, *E.s curvula* (46,7,%), *E. tef* (24,7%) and *C. dactylon* (20,7%) were the dominant species in June 2016. *E. tef* then died after March 2017 and leaving *E. curvula* (70,9%) and *C. dactylon* (28,3%) as the dominant species at the end of the trials in June 2017(Figure 4.12 and Table 4.12).

Table 4.12: Summary of plant density (plants/m^2) and plant composition (%) results for *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor, M. sativa* and *L. perenne* in T3C at the Crown gold TSF site from June 2016 to June 2017.

T3C	06/2016		09/2016		11/2016		03/2017		06/2017	
Species	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**
C. dactylon	23,3	20,7	9,3	10,8	13,2	25,9	16,7	30,2	16,3	28,3
D. eriantha	5,6	4,9	2,3	2,7	0,8	1,5	0,6	1,0	0,3	0,4
E. tef	27,8	24,7	20,3	23,6	0,0	0,0	0,3	0,6	0,0	0,0
E. curvula	52,7	46,7	52,9	61,3	35,6	69,7	36,9	66,9	40,8	70,9
S. bicolor	1,3	1,2	0,4	0,5	0,2	0,4	0,6	1,0	0,3	0,4
M. sativa	1,4	1,3	0,6	0,6	0,2	0,4	0,1	0,2	0,0	0,0
L. perenne	0,6	0,5	0,4	0,5	1,0	2,0	0,0	0,0	0,0	0,0
Total	**112,7**	**100,0**	**86,3**	**100,0**	**51,0**	**100,0**	**55,1**	**100,0**	**57,5**	**100,0**

The change in plant density (plants/m^2) and plant composition (%) for T4C, which contained the same total weight of coated seed (19 kg/ha), but with the seeding rate of *D. eriantha*, *C. dactylon, E. curvula* and *S. bicolor* adjusted to the coated and uncoated seeding ratios of the species (Table 3.2 and Figure 3.11) are given below in Figures 4.13 and 4.14 and Table 4.13.

Initially in June 2017 *E. curvula* was present at a density of 34 plants/m^2, *C. dactylon* was present at 15,4 plants/m^2 and *E. tef* was present at 46,2 plants/m^2 (Figure 4.13). Although *E. tef* was present at a notably higher density of 46,2 plants/m^2 than in treatments T1C (28,7 plants/m^2), T2UC (28,6 plants/m^2) and T3C (27,8 plants/m^2), it was absent from the community in November 2016 except during March 2017 at T3C. The initial increase in *E. tef* compared to the three other seed treatments was most likely caused by the increased seeding rate of 1,9 kg/ha for *E. tef* in treatment T4C compared to a smaller seeding rate of 1 kg/ha *E. tef* seed in treatments T1C, T2UC and T3C (Table 3.2 and Figure. 3.10).

E. curvula had a notably lower density of 34 plants/m^2 compared to treatments T1C 52,9 plants/m^2, T3C 52,7 plants/m^2 and T5UC 51 plants/m^2 initialy in June 2016. *C. dactylon* was present at its highest density of 18 plants/m^2 in November 2016 before it decreased to 16,9 plants/m^2 at the end of the trials in June 2017 (Figure 4.13 and Table 4.13).

D. eriantha had an initial density of 3,1 plants/m^2 in June 2016, after which it decreased to 0,3 plants/m^2 in June 2017 (Figure 4.13 and Table 4.13).

M. sativa and *S. bicolor* had the highest density of 2,2 plants/m^2 and 2,1 plants/m^2 in June 2016 respectively. The density of *M. sativa* decreased until it was absent in June 2017, but the *S. bicolor* density decreased to 0,9 plants/m^2 in March 2017 before increasing to 1,5 plants/m^2 in June 2017 again (Figure 4.13 and Table 4.13.

L. perenne was also present in June 2016 (1,3 plants/m^2), after which it increased until November 2016, before decreasing until totally absent in June 2017 (Figure 4.13 and Table 4.13).

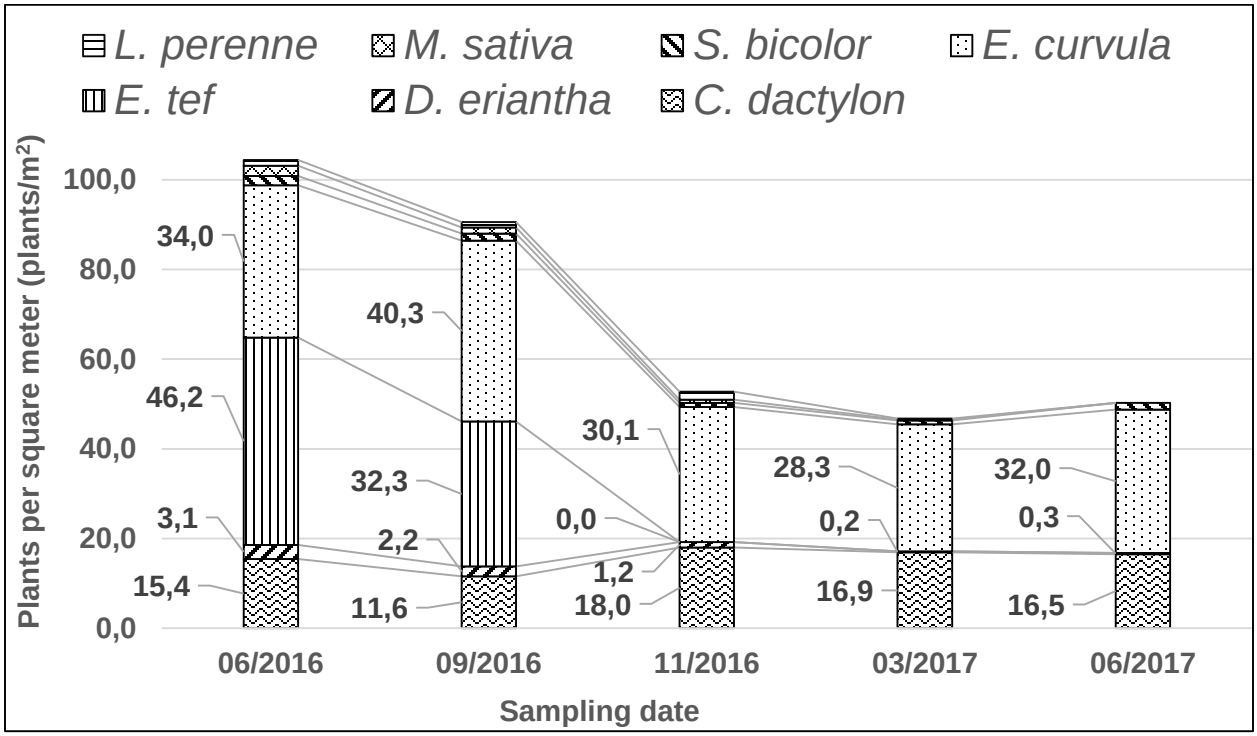

Figure 4.13: Change in density (plants/m^2) of species used (*C. dactylon, D. eriantha, E. curvula, E. tef, S. bicolor, M. sativa* and *L. perenne*) in T4C at the Crown gold TSF site from June 2016 to June 2017.

The plant composition in T4C is slightly different to that of T1C, T2UC and T3C, due to the larger percentage *E. tef* of 44,3% in the community (Figure 4.14). The abundance of *E. tef* disappeared in November 2016 with *E. curvula* (57,1%) and *C. dactylon* (34,1%) as the dominant species for the rest of the trial period, while *M. sativa* (0%), *S. bicolor* (3%), and *D. eriantha* (0,5%) were virtually absent from the plant composition (Figure 4.14 and Table 4.13).

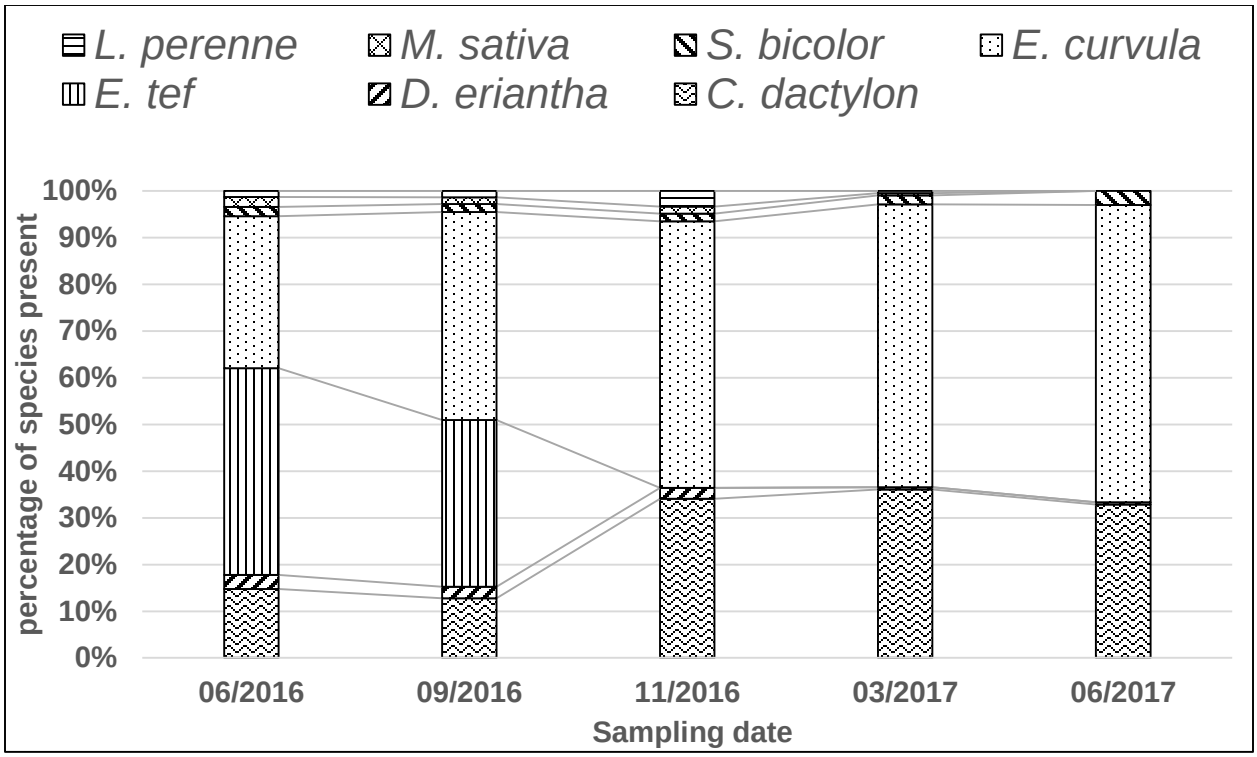

Figure 4.14: Change in plant composition percentage of species used (*C. dactylon, D. eriantha, E. curvula, E. tef, S. bicolor, M. sativa* and *L. perenne*) in T4C at the Crown gold TSF site from June 2016 to June 2017.

Table 4.13: Summary of plant density (plants/m^2) and plant composition (%) results for *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor, M. sativa* and *L. perenne* in T4C at the Crown gold TSF site from June 2016 to June 2017.

T4C	06/2016		09/2016		11/2016		03/2017		06/2017	
Species	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**
C. dactylon	15,4	14,8	11,6	12,8	18,0	34,1	16,9	36,1	16,5	32,8
D. eriantha	3,1	3,0	2,2	2,5	1,2	2,3	0,2	0,5	0,3	0,5
E. tef	46,2	44,3	32,3	35,7	0,0	0,0	0,0	0,0	0,0	0,0
E. curvula	34,0	32,6	40,3	44,5	30,1	57,1	28,3	60,6	32,0	63,7
S. bicolor	2,1	2,0	1,6	1,7	0,9	1,7	0,9	1,9	1,5	3,0
M. sativa	2,2	2,1	1,3	1,5	0,8	1,5	0,2	0,5	0,0	0,0
L. perenne	1,3	1,3	1,2	1,3	1,8	3,4	0,2	0,5	0,0	0,0
Total	**104,4**	**100,0**	**90,6**	**100,0**	**52,8**	**100,0**	**46,8**	**100,0**	**50,3**	**100,0**

T5UC used the same rate uncoated seed as coated seed used T4C and the emergence results are illustrated in Figure 4.15, 4.16 and Table 4.14.

Initialy the species with the highest densities were *E. curvula* with 51 plants/m^2, *E. tef* 44 plants/m^2, *C. dactylon* 21,1 plants/m^2 and *D. eriantha* with 16,3 plants/m^2. Like for treatment T4C, *E. tef* had a higher initial density in June 2016 and September 2016 when compared to treatments T1C, T2UC and T3C (Figure 4.15 and Table 4.14). This was most likely caused by the higher seeding

rate of *E. tef* (1.9 kg/ha) in T5UC compared to seed treatments T1C, T2UC and T3C in which *E. tef* was sown at a rate of 1 kg/ha.

The densities of all the other species sown decreased towards the end of the trial, except for *E. curvula,* which increased from 38,1 plants/m^2 to 39,3 plants/m^2 between March and June 2017 (Figure 4.15 and Table 4.14).

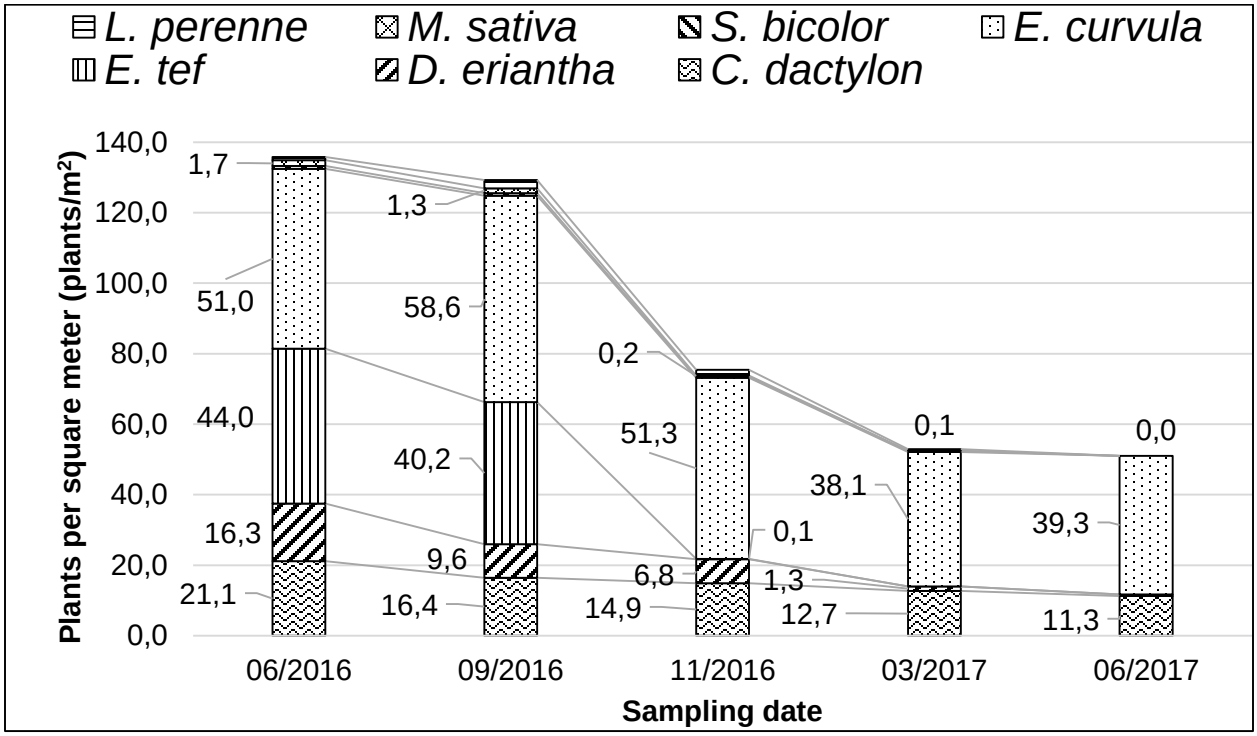

Figure 4.15: Change in density (plants/m^2) of species used (*C. dactylon, D. eriantha, E. curvula, E. tef, S. bicolor, M. sativa* and *L. perenne*) in T5UC at the Crown gold TSF site from June 2016 to June 2017.

Regarding the plant composition, the percentages of species remained constant, except for *D. eriantha* occupying a notably larger percentage (12%) of the plant composition compared to the other treatments. The succession of species still followed the same pattern as previous treatments discussed, with *E. tef* disappearing in November 2016, and *E. curvula* and *C. dactylon* being the dominant surviving species (Figure 4.16 and Table 4.14).

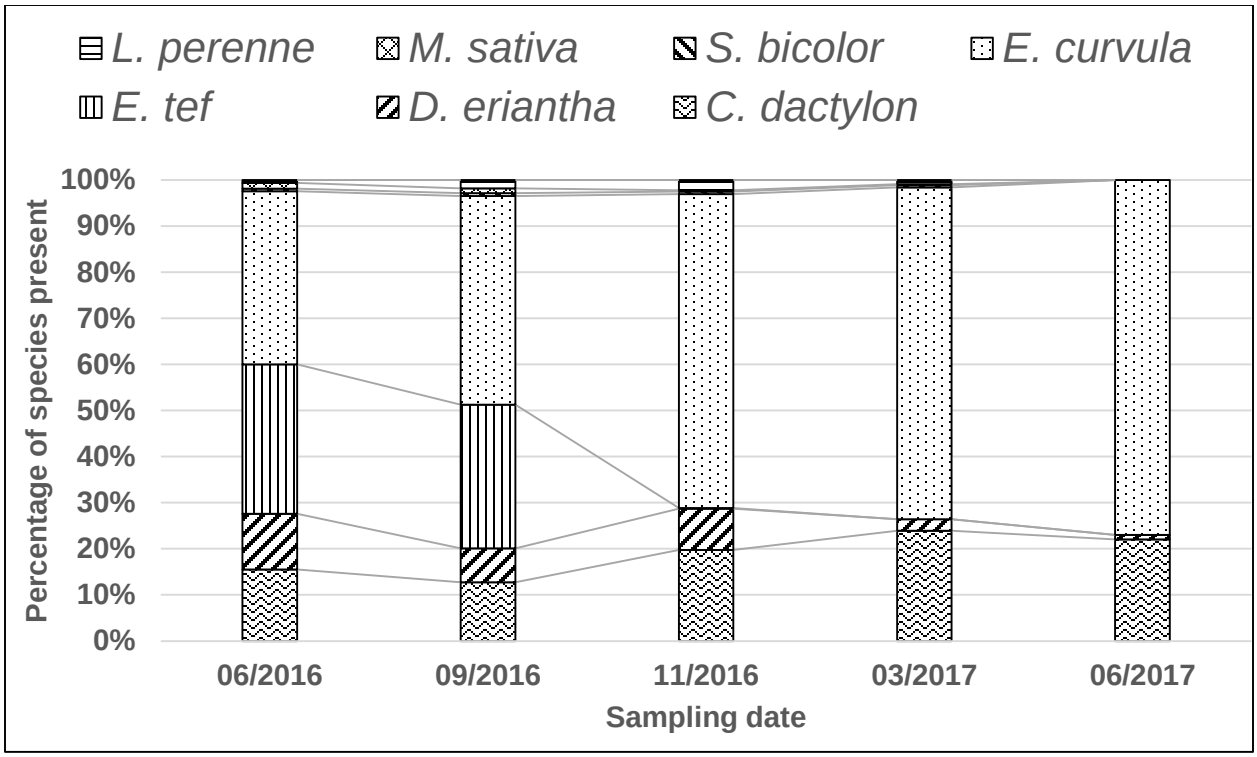

Figure 4.16: Change in plant composition percentage of species used (*C. dactylon, D. eriantha, E. curvula, E, tef, S. bicolor, M. sativa* and *L. perenne*) in T5UC at the Crown gold TSF site from June 2016 to June 2017.

Table 4.14: Summary of plant density (plants/m^2) and plant composition (%) results for *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor, M. sativa* and *L. perenne* in T5UC at the Crown gold TSF site from June 2016 to June 2017.

T5UC	**06/2016**		**09/2016**		**11/2016**		**03/2017**		**06/2017**	
Species	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**
C. dactylon	21,1	15,5	16,4	12,7	14,9	19,7	12,7	23,9	11,3	22,1
D. eriantha	16,3	12,0	9,6	7,4	6,8	9,0	1,3	2,5	0,5	1,0
E. tef	44,0	32,4	40,2	31,1	0,1	0,1	0,0	0,0	0,0	0,0
E. curvula	51,0	37,6	58,6	45,3	51,3	68,0	38,1	72,0	39,3	77,0
S. bicolor	0,8	0,6	0,8	0,6	0,4	0,6	0,2	0,4	0,0	0,0
M. sativa	1,7	1,2	1,3	1,0	0,2	0,3	0,1	0,2	0,0	0,0
L. perenne	0,9	0,7	2,3	1,8	1,7	2,2	0,5	0,9	0,0	0,0
Total	**135,8**	**100,0**	**129,2**	**100,0**	**75,4**	**100,0**	**52,9**	**100,0**	**51,0**	**100,0**

In summary, the Crown gold TSF site initially had the highest total densities of species in all the treatments in June 2016, with *E. curvula*, *C. dactylon* and *E. tef* dominating the plant composition. *E. tef* is an annual pioneer species and disappears at the end of the growing season (November), which explains why this species was totally absent at the end of the trial with only the perennial species, such as *C. dactylon* and *E. curvula* remaining and dominating the plant composition at the end.

The density of *E. tef* was notably higher in treatments T4C and T5UC, which was most likely due to the increased seeding rate of 1,9 kg/ha compared to the 1 kg/ha seeding rate for the other three seed treatments.

D. eriantha had the lowest plant density of the four grass species (*E. curvula*, *C. dactylon* and *E. tef*). This grass decreased throughout the trial and failed to establish. The latter may be due to the low seed viability (22%) of the coated and uncoated seed batches (Figure 3.11). *D. eriantha* is also a climax grass species which tends to increase at a later stage of succession when growth conditions are improved by the pioneer species, such as *C. dactylon* and *E. tef* and subclimax species, such as *E. curvula* (Van Oudtshoorn, 2004:224).

M. sativa was totally absent and *S. bicolor* had very low densities of < 3 plants/m^2 after one year of the trial.

4.3.2 Change in cover and contribution of species at the Crown gold TSF site

The change in the total vegetation cover (aerial cover and basal cover) contribution for the species used in the seed treatments on the Crown gold TSF trials are displayed below in Figure 4.17 to Figure 4.21 and Table 4.15 to Table 4.20.

The cover percentage results for T1C are discussed and illustrated in Figure 4.17 and Tbale 4.21. The initial combined plant cover for T1C in June 2016 was 72,5%, with *E. tef* (29%) and *E. curvula* (21,9%) having the greatest contribution (Figure 4.17). In September 2016, the total vegetation coverage remained constant (70%), but the cover contribution of *E. tef* decreased to 14,4%, while *E. curvula* increased to 42,9%. *E. curvula* then increased to its maximum coverage of 76,1% in November 2016 before falling down to 49,6% in June 2017. The cover contribution of *C. dactylon* significantly increased between November 2016 and March 2017 to provide 42,7% total coverage before decreasing to 27,3% in June 2017 (Figure 4.17 and Table 4.15).

The cover of *L. perenne* was low during June to November 2016 (9,4% and 8%) (Figure 4.17), after which it disappeared towards March 2017 (Figure 4.17). The cover for *M. sativa*, *S. bicolor* and *D. eriantha* were very low at < 2% (Figure 4.17 and Table 4.15)

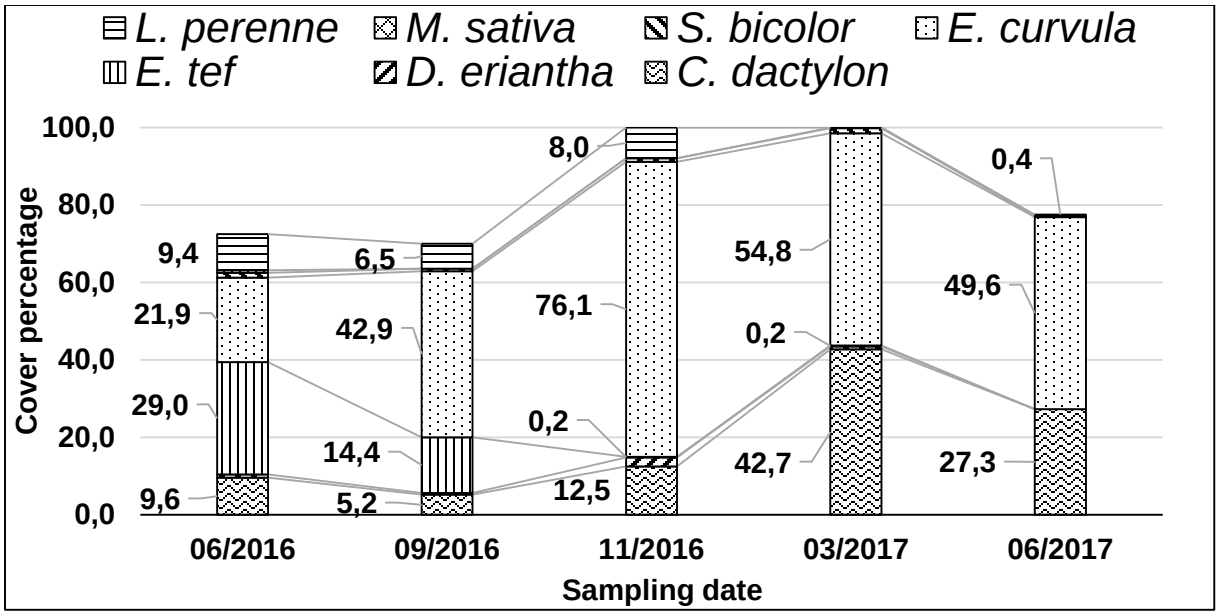

Figure 4.17: Change in total cover contribution percentage of *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor, M. sativa* and *L. perenne* in T1C at the Crown gold TSF site from June 2016 to June 2017.

Table 4.15: Change in total cover contribution % of *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor, M. sativa* and *L. perenne* in T1C at the Crown gold TSF site from June 2016 to June 2017 shown in Figure 4.17.

T1C	06/2016	09/2016	11/2016	03/2017	06/2017
Species	**%**	**%**	**%**	**%**	**%**
C. dactylon	9,6	5,2	12,5	42,7	27,3
D. eriantha	0,8	0,4	2,3	0,8	0,0
E. tef	29,0	14,4	0,2	0,2	0,0
E. curvula	21,9	42,9	76,1	54,8	49,6
S. bicolor	1,3	0,6	0,9	1,3	0,2
M. sativa	0,6	0,0	0,0	0,2	0,0
L. perenne	9,4	6,5	8,0	0,0	0,4
Total	**72,5**	**70,0**	**100,0**	**100,0**	**77,5**

T2UC, initially had a total vegetation cover of 67,3% for all the species combined. This increased to 90,8% before being completely covered (100%) in March 2017, but decreased again to 80,6% in June 2017 (Figure 4.18 and Table 4.16).

The cover contribution of *E. tef* was above 30% in September 2016, but was no longer present in November 2016. *E. curvula* and *C. dactylon* were the two primary species covering the trial plot areas from November 2016 until the end of the trials in June 2017 (Figure 4.18 and Table 4.16). *E. curvula* had the highest contribution cover in November 2016 (67,4%) and *C. dactylon* (42,4%) had the highest contribution of cover in March 2017 (Figure 4.18 and Table 4.16). The cover

contribution of *D. eriantha*, *M. sativa* and *S. bicolor* were negligible, with *D. eriantha* only attaining a cover percentage contribution of 1,9% (Figure 4.18 and Table 4.16).

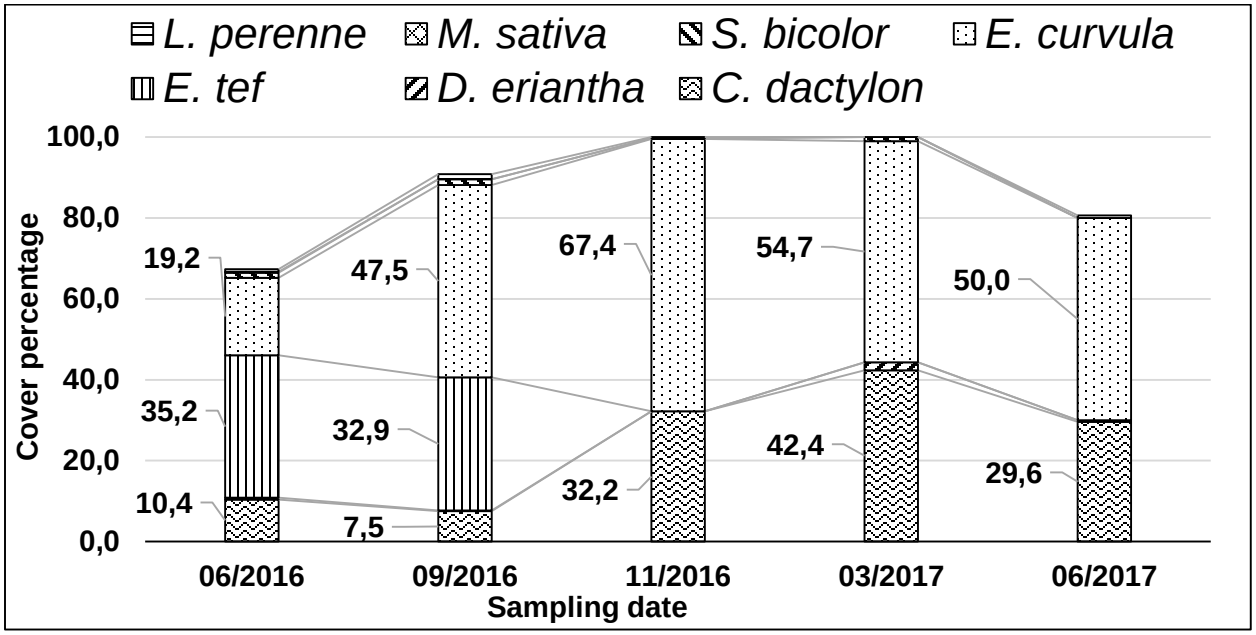

Figure 4.18: Change in total cover contribution of *C. dactylon, D. eriantha, E. tef, E. curvula, E, tef, S. bicolor, M. sativa* and *L. perenne* in T2UC at the Crown gold TSF site from June 2016 to June 2017.

Table 4.16: Change in total cover % contribution of *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor, M. sativa* and *L. perenne* in T2UC at the Crown gold TSFsite from June 2016 to June 2017 shown in Figure 4.18.

T2UC	06/2016	09/2016	11/2016	03/2017	06/2017
Species	**%**	**%**	**%**	**%**	**%**
C. dactylon	10,4	7,5	32,2	42,4	29,6
D. eriantha	0,4	0,2	0,0	1,9	0,4
E. tef	35,2	32,9	0,0	0,0	0,0
E. curvula	19,2	47,5	67,4	54,7	50,0
S. bicolor	1,3	1,5	0,2	1,0	0,0
M. sativa	0,2	0,0	0,0	0,0	0,0
L. perenne	0,6	1,3	0,3	0,0	0,6
Total	**67,3**	**90,8**	**100,0**	**100,0**	**80,6**

The T3C had a slightly lower total cover contribution than T2UC in June 2016 only contributing 53.3% of the total cover contribution (Table 4.17 and 4.19.

The combined cover contribution of the species increased to its maximum in March 2017 (100%), of which *E. curvula* attained a 63.9% coverage, and *C. dactylon* attained 33,3% coverage, before decreasing to 49,8% (*E. curvula*) and 29,2% (*C. dactylon*). *S. bicolor* and *M. sativa* had a

negligible cover contribution below 1,5% throughout the trial. *D. eriantha* increased to a maximum cover contribution of 2,2% in March 2017 (Figure 4.19 and Table 4.17).

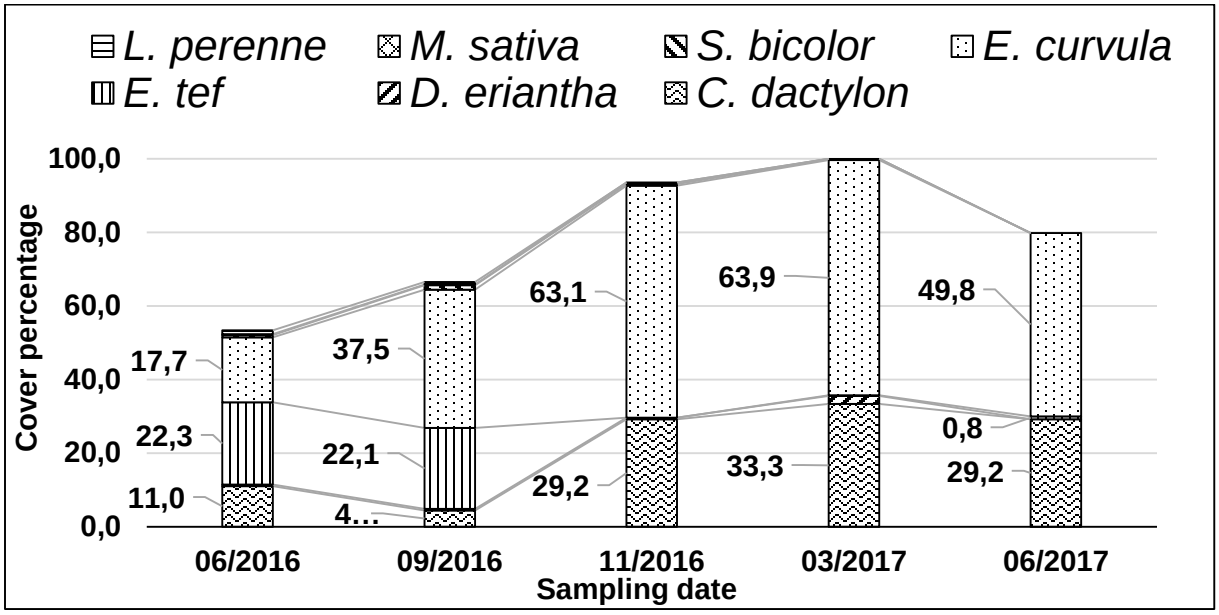

Figure 4.19: Change in total cover contribution of *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor, M. sativa* and *L. perenne* in T3C at the Crown gold TSF site from June 2016 to June 2017.

Table 4.17: Change in total cover contribution % of *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor, M. sativa* and *L. perenne* in T3C at the Crown gold TSF site from June 2016 to June 2017 shown in Figure 4.19.

T3C	06/2016	09/2016	11/2016	03/2017	06/2017
Species	**%**	**%**	**%**	**%**	**%**
C. dactylon	11,0	4,4	29,2	33,3	29,2
D. eriantha	0,4	0,4	0,4	2,2	0,0
E. tef	22,3	22,1	0,0	0,1	0,8
E. curvula	17,7	37,5	63,1	63,9	49,8
S. bicolor	0,6	1,3	0,4	0,4	0,0
M. sativa	0,2	0,2	0,4	0,0	0,0
L. perenne	1,0	0,6	0,0	0,0	0,0
Total	**53,3**	**66,5**	**93,5**	**100,0**	**79,8**

Figure 4.20 and Table 4.18, display the plant coverage percentage results for T4C. It isseen that T4C had a higher initial total vegetation coverage (81,7%) compared to T1C (72,5%), T2UC (67,3%) and T3C (53,3%) (Tables 4.15, 4.16 and 4.17). Initialy, in T4C, *E. tef* contributed 49,2% vegetation cover; this then decreased to 42,9% in September 2016 before it no longer existed in the community and provided no vegetation cover from November 2016 (Figure 4.20 and Table 4.18).

E. curvula continued to increase towards November 2016 when it attained a maximum coverage of 59,6% before decreasing to 53,8% total cover in June 2017. *C. dactylon* contributed 12,5% total coverage in June 2016 before increasing to a maximum of 42,4% in March 2017 and then decreasing to 41,9% in June 2017 (Figure 4.20 and Table 4.18). *E. tef*'s coverage contribution percentage decreased from an initial high of 49,2% in June 2016 to 42,9% in September 2016 afterwards it did not contribute any coverage throughout the rest of the trial period (Figure 4.20 and Table 4.18). *D. eriantha* provided 0,2% cover in June 2016 which then increased to 3,4% in November 2016 however this decreased to no coverage in June 2017 (Figure 4.20 and Table 4.18).

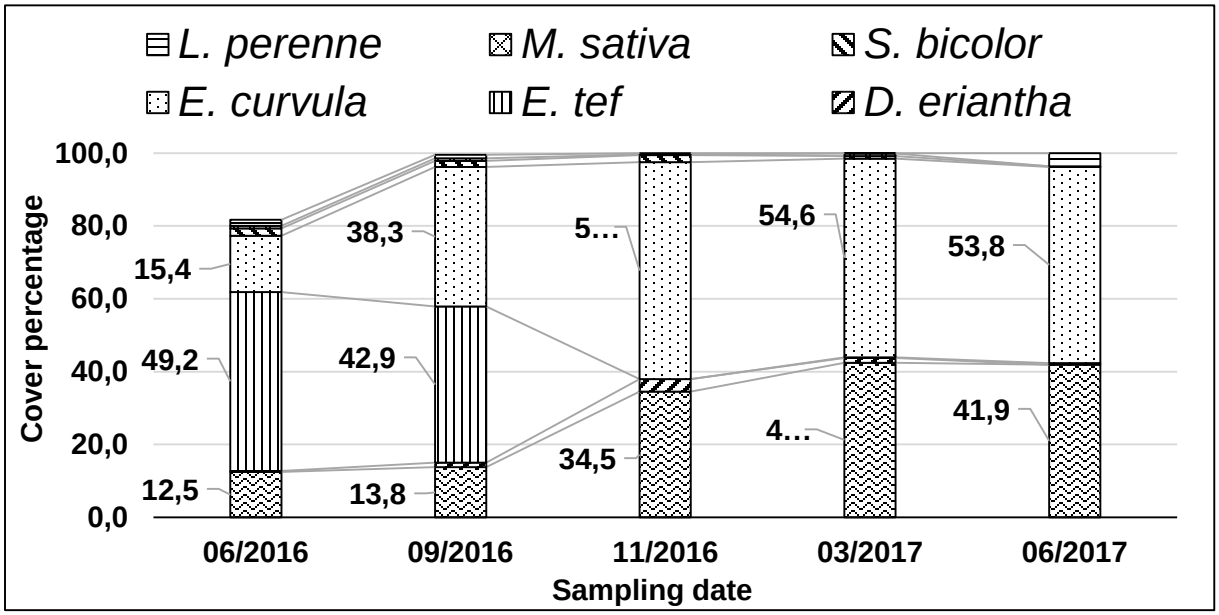

Figure 4.20: Change in total cover contribution of *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor, M. sativa* and *L. perenne* in T4C at the Crown gold TSF site from June 2016 to June 2017.

Table 4.18: Change in total cover contribution % of *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor, M. sativa* and *L. perenne* in T4C at the Crown gold TSF site from June 2016 to June 2017 shown in Figure 4.20.

T4C	06/2016	09/2016	11/2016	03/2017	06/2017
Species	**%**	**%**	**%**	**%**	**%**
C. dactylon	12,5	13,8	34,5	42,4	41,9
D. eriantha	0,2	1,3	3,4	1,3	0,0
E. tef	49,2	42,9	0,0	0,2	0,4
E. curvula	15,4	38,3	59,6	54,6	53,8
S. bicolor	2,1	1,7	1,9	0,7	0,2
M. sativa	0,6	0,6	0,2	0,7	0,0
L. perenne	1,7	1,0	0,4	0,1	3,6
Total	**81,7**	**99,6**	**100,0**	**100,0**	**100,0**

In the T5UC, the same trend is visible as with T4C and previously discussed treatments, with *E. tef* (48,5%), *E. curvula* (24,5%) and *C. dactylon* (16,7%) being the primary species covering the ground in June 2016 This then changed with *E. tef* 's coverage decreasing to 29,6% before not being present in November 2016. *E. curvula* provided the majority of the ground coverage (68,1%, 56,4%, 66,9%) throughout the trial period, followed by *C. dactylon* (21%, 37%, 29%) (Figure 4.21 and Table 4.19).

D. eriantha had the highest cover contribution (5,7%) in T5UC compared to the other seed treatments (0,8% – T1C, 1,9% – T2UC, 2,2% – T3C, 1,3% – T4C) in March 2017 (Figure 4.21 and Table 4.19).

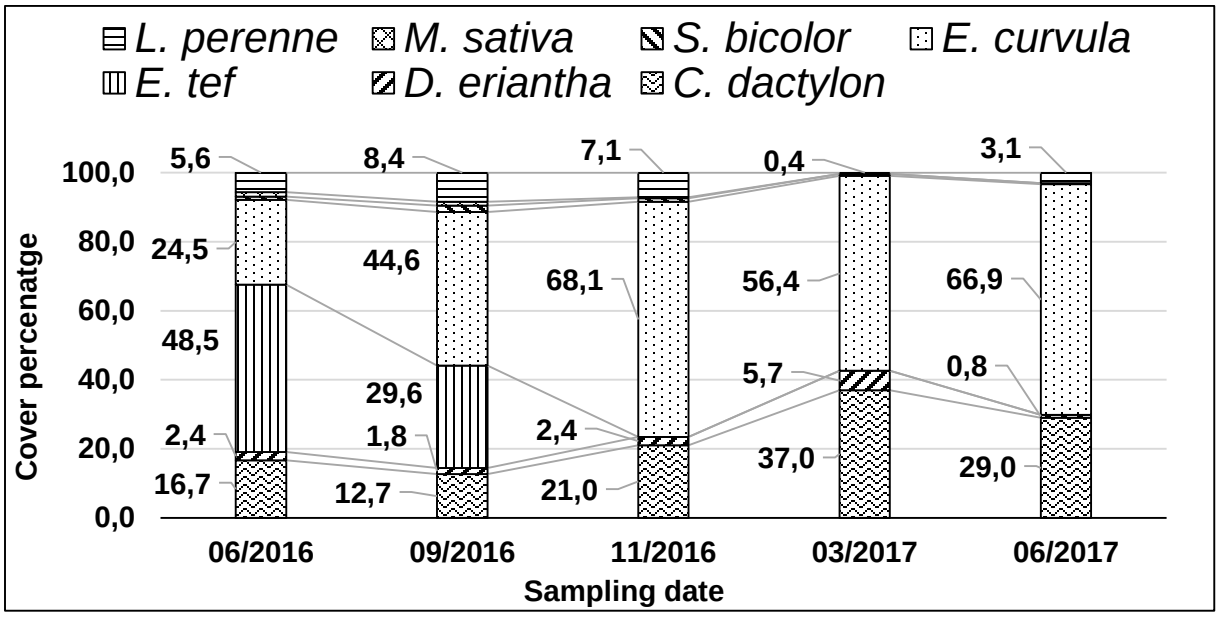

Figure 4.21: Change in total cover contribution of *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor, M. sativa* and *L. perenne* in T5UC at the Crown gold TSFsite from June 2016 to June 2017.

Table 4.19: Change in total cover contribution % of *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor, M. sativa* and *L. perenne* in T5UC at the Crown gold TSF sitefrom June 2016 to June 2017 shown in Figure 4.21

T5UC	06/2016	09/2016	11/2016	03/2017	06/2017
Species	**%**	**%**	**%**	**%**	**%**
C. dactylon	16,7	12,7	21,0	37,0	29,0
D. eriantha	2,4	1,8	2,4	5,7	0,8
E. tef	48,5	29,6	0,0	0,0	0,0
E. curvula	24,5	44,6	68,1	56,4	66,9
S. bicolor	0,9	1,8	1,0	0,6	0,3
M. sativa	1,3	1,2	0,3	0,0	0,0
L. perenne	5,6	8,4	7,1	0,4	3,1
Total	**100,0**	**100,0**	**100,0**	**100,0**	**100,0**

In summary of the change in total cover percentage of specie for the Crown gold TSFsite trials, the initial vegetation coverage in June 2016 was provided primarily by *E. tef* and *E. curvula*. The *E. tef* decreased towards September 2016 and was no longer present in November 2016. This was expected, since *E. tef* was the annual species in the mixture with the purpose of acting as a pioneer species to improve the growth conditions for the subclimax and climax species (*E. curvula*, *C. dactylon* and *D. eriantha*) to establish. With the decrease of *E. tef*, *E. curvula* then became the dominant species providing the largest cover percentage of all the species, followed by *C. dactylon* in March 2017 and June 2017. The total vegetation coverage reached a maximum in March 2017 during which the trial plots were completely covered with vegetation before the total vegetation coverage receded in June 2017. This was a natural cycle where grass growth increased during the warmer summer temperatures and rainfall before species wither and become dormant during the colder dry winter months (Figure 3.7).

The initial cover contribution of *E. tef* appeared higher in treatment T4C and T5UC (> 49%) compared to T1C (29%), T2UC (35,2%) and T3C (22,3%). This is due to *E. tef* being sown at a larger seeding rate (1,9 kg/ha) compared to the 1 kg/ha seeding rate in T1C, T2UC and T3C (Figure 3.10. This led to T4C and T5UC having nearly twice the amount of *E. tef* seeds incorporated into the seed treatment.

D. eriantha provided the smallest cover contribution of all the grass species within each seed treatment, which could be ascribed to the low seed viability of both the coated and uncoated seed batches (Figure 3.11).

4.3.3 Rooikraal gold TSF site species density and plant composition

The severe erosion of the adjacent unvegetated TSF slopes caused a drastic decrease in the plant densities between September and November 2016 (Figure 4.22). The raw unvegetated gold tailing material buried the majority of the trial plots up to 20 cm deep.

The species density and plant composition of T1C at the Rooikraal gold TSF site are illustrated in Figure 4.22, 4.23 and Table 4.20.

In June 2016, *E. curvula* had the highest plant density of 47,9 plants/m^2, which increased to 58,4 plants/m^2 in September 2016 (Figure 4.22). *E. tef* was the second most abundant species at 20,8 plants/m^2 before it decreased again to 11,7 plants/m^2 in September 2016. *C. dactylon* had an initial density of 19,6 plants/m^2 before decreasing to 8 plants/m^2, while *D. eriantha* had an initial density of 11,1 plants/m^2 before decreasing to 0,7 plants/m^2 in September 2016 (Figure 4.22 and Table 4.20).

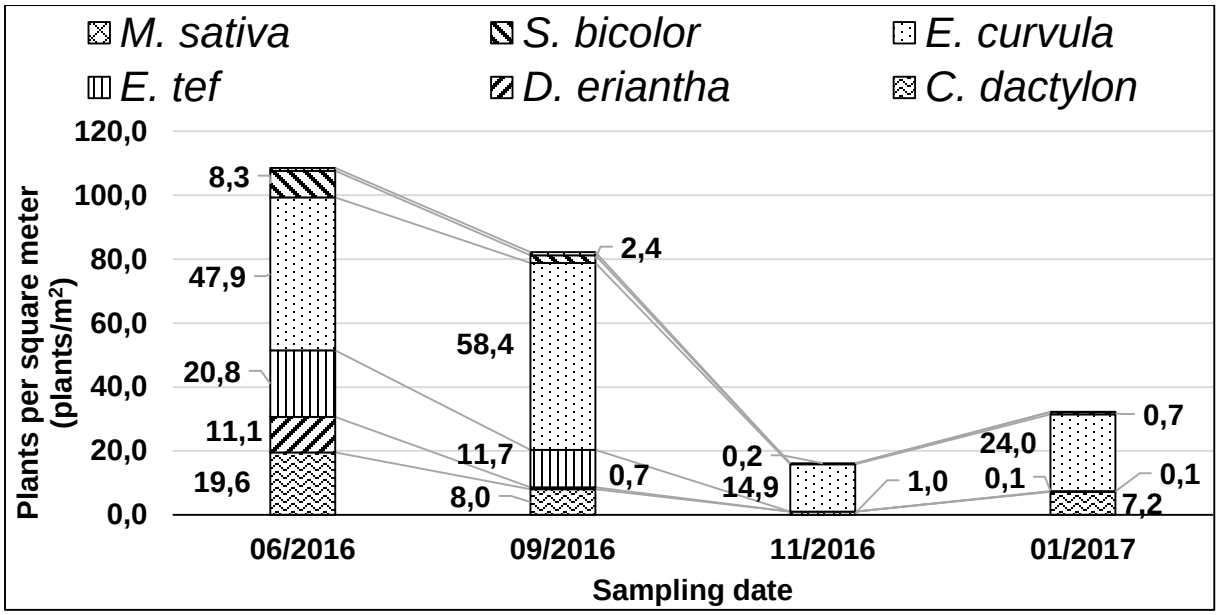

Figure 4.22: Change in plant density (plants/m^2) of species used (*C. dactylon, D. eriantha, E. curvula, E. tef, S. bicolor, M. sativa*) in T1C at the Rooikraal gold TSF site from June 2016 to January 2017.

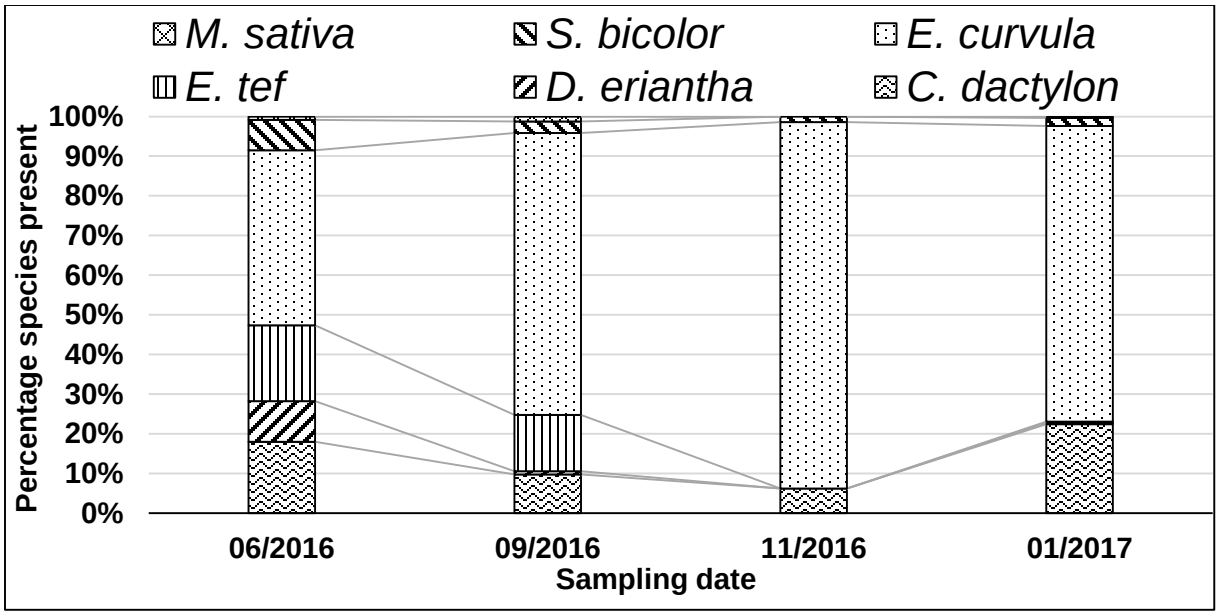

Figure 4.23: Change in plant composition of species used (*C. dactylon, D. eriantha, E. curvula, E. tef, S. bicolor, M. sativa*) in T1C at the Rooikraal gold TSF site from June 2016 to January 2017.

In Figure 4.22 it can be seen that after the erosion event, which buried the majority of the species, the total plant density dropped from 82,2 plants/m^2 in September 2016 to 16,1 plants/m^2 in November 2016. *E. curvula* and *C. dactylon* were the species that showed the greatest recovery. From visual, qualitative observations, it seemed that if the culms and leaves of *E. curvula* were still visible and protruded from the tailings material, it was likely to survive. *C. dactylon* had an advantage with much faster regrowth, since this grass has stolons that are able to grow underground which can resurface again. *S. bicolor* also survived, which is not surprising, since its thicker stem and taller growth form allowed it to resist the burial of the tailings material.

Initially *S. bicolor* was present at a density of 8,3 plants/m^2, but later decreased to 2,4 plants/m^2 in September 2016 and 0,7 plants/m^2 in January 2017 (Figure 4.22).

M. sativa was present at a density of 1 plant/m^2 in September 2016 (Figure 4.22 and Table 4.20). The plant composition changed with *E. tef* disappearing before November 2016 and *E. curvula* increasing to be the dominant species in January 2017 (Figure 4.23 and Table 4.20)

Table 4.20: Summary of plant density (plants/m^2) and plant composition (%) results for *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor,* and *M. sativa* in T1C at the Rooikraal gold TSF site from June 2016 to January 2017.

T1C	06/2016		09/2016		11/2016		01/2017	
Species	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**
C. dactylon	19,6	18,0	8,0	9,7	1,0	6,2	7,2	22,4
D. eriantha	11,1	10,2	0,7	0,8	0,0	0,0	0,1	0,3
E. tef	20,8	19,1	11,7	14,2	0,0	0,0	0,1	0,3
E. curvula	47,9	44,1	58,4	71,1	14,9	92,4	24,0	74,5
S. bicolor	8,3	7,7	2,4	3,0	0,2	1,4	0,7	2,1
M. sativa	0,9	0,8	1,0	1,2	0,0	0,0	0,1	0,3
Total	**108,6**	**100,0**	**82,2**	**100,0**	**16,1**	**100,0**	**32,2**	**100,0**

The results of T2UC are given in Figures 4.24 and 4.25 and Table 4.21. *E. curvula* was initially the dominant species in June 2016, comprising 37,4% of the community at a density of 39,6 plants/m^2 (Figure 4.24 and Table 4.21). *E. tef* had the second highest density (29,6 plants/m^2), comprising 27,9% of the community, but decreased to 15,3 plants/m^2 before disappearing in November 2016 (Figure 4.24 and Table 4.21).

C. dactylon was initially present at a density of 18,4 plants/m^2 (June 2016) before decreasing to 9.1 plants/m^2 (Figure 4.24). *D. eriantha* had a density of 3,4 plants/m^2, comprising 3,3% (Figure 4.25) of the established community before decreasing (1,2 plants/m^2) in November 2016 (Figure 4.24). This species did not establish after the erosion event. *S. bicolor* and *M. sativa* were present at the similar densities of 7,7 plants/m^2 and 7,2 plants/m^2 in June 2016 respectively (Figure 4.24 and Table 4.21.

Only *E. curvula* (12,7 plants/m^2), *C. dactylon* (0,1 plants/m^2) and *S. bicolor* (0,3 plants/m^2) survived the erosion event in November 2016, after which the density of *E. curvula* (13,8 plants/m^2) and *C. dactylon* (1,1 plants/m^2) increased and the density of *S. bicolor* (0,1 plants/m^2) decreased (Figure 4.24 and Table 4.21).

Initialy in June 2016 all six species sown in T2UC were present however, in Novemeber 2016 the plant composition was comprised mainly of *E. curvula* (96,6%), *C. dactylon* (0,8%) and *S. bicolor* (2,5%) were also present (Figure 4.25 and Table 4.21). The only species surviving at the end of the trial period were *E. curvula* constituting 91,9% of the plants present, *C. dactylon* with comprising 7,4% and *S. bicolor* with 0,7% (Figure 4.25 and Table 4.21).

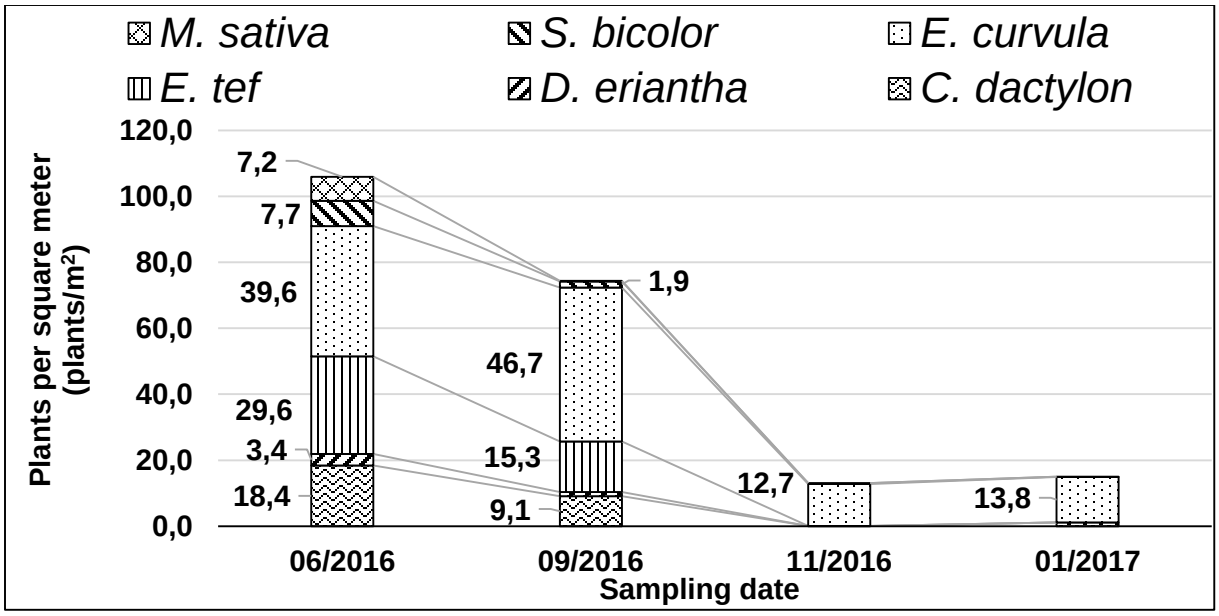

Figure 4.24: Change in plant density (plants/m^2) of species used (*C. dactylon, D. eriantha, E. curvula, E. tef, S. bicolor, M. sativa*) in T2UC at the Rooikraal gold TSF site from June 2016 to January 2017.

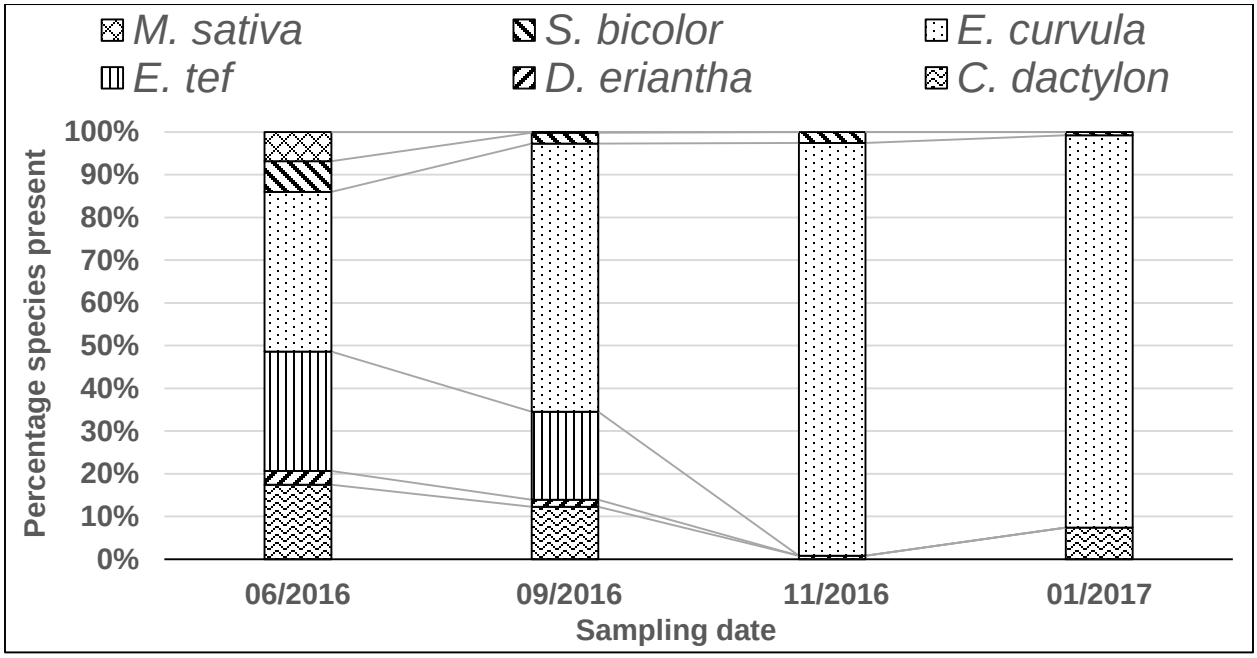

Figure 4.25: Change in plant composition of species used (*C. dactylon, D. eriantha, E. curvula, E. tef, S. bicolor, M. sativa*) in T2UC at the Rooikraal gold TSF site from June 2016 to January 2017.

Table 4.21: Summary of plant density (plants/m^2) and plant composition (%) results for *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor,* and *M. sativa* in T2UC at the Rooikraal gold TSF site from June 2016 to January 2017.

T2UC	06/2016		09/2016		11/2016		01/2017	
Species	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**
C. dactylon	18,4	17,4	9,1	12,3	0,1	0,8	1,1	7,4
D. eriantha	3,4	3,3	1,2	1,6	0,0	0,0	0,0	0,0
E. tef	29,6	27,9	15,3	20,6	0,0	0,0	0,0	0,0
E. curvula	39,6	37,4	46,7	62,8	12,7	96,6	13,8	91,9
S. bicolor	7,7	7,2	1,9	2,5	0,3	2,5	0,1	0,7
M. sativa	7,2	6,8	0,1	0,1	0,0	0,0	0,0	0,0
Total	**105,9**	**100,0**	**74,3**	**100,0**	**13,1**	**100,0**	**15,0**	**100,0**

The species density results of T3C are illustrated in Figures 4.26 and 4.27 and summarised in table 4.22. T3C had the same amount of seed as the T2UC but had a higher total plant density (148 plants/m^2) than T2UC (105,9 plants/m^2) (Figure 4.26 and Table 4.22).

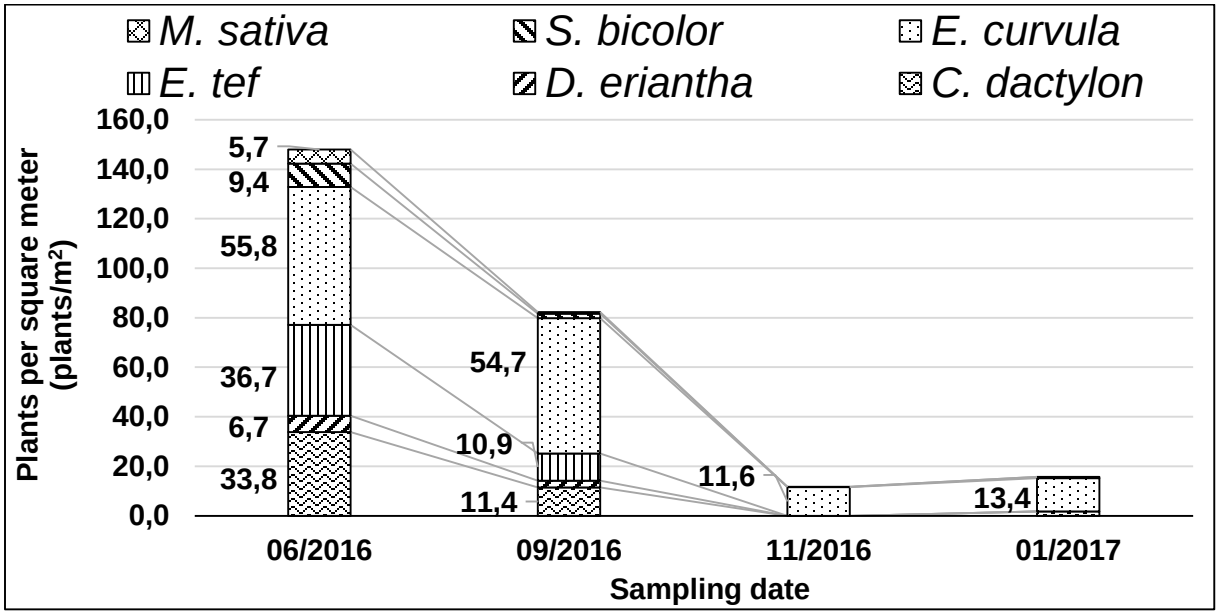

Figure 4.26: Change in plant density (plants/m^2) of species used (*C. dactylon, D. eriantha, E. curvula, E. tef, S. bicolor, M. sativa*) in T3C at the Rooikraal gold TSF site from June 2016 to January 2017.

E. curvula had a density of 55,8 plants/m^2, *E. tef*,a density of 36,7 plants/m^2 and *C. dactylon* a density of 33,8 plants/m^2. These three grasses were also the dominant plant species in June 2016, with *E. curvula* comprising 37,7%, *C. dactylon* 22,8% and *E. tef* 24,8% of the vegetation community (Figure 4.26, 4.27 and Table 4.22.

M. sativa was present in June 2016 at a density of 5,7 plants/m^2 in treatment T3C, but decreased to a density of 0,7 plants/m^2 in September 2016, after which is disappeared in November 2016.

S. bicolor was present at a density of 9,4 plants/m^2 in June 2016, but decreased to as low as 0,1 plants/m^2 in November 2016 before increasing to 0,4 plants/m^2 in January 2017 again (Figure 4.26 and Table 4.22).

D. eriantha had a density of 6,7 plants/m^2 in June 2016 before decreasing to 2,8 plants/m^2 in September 2016.

The only species surviving in November 2016 were therefore, *E. curvula, C. dactylon* and *S. bicolor*. After the erosion event when many species were buried, *E. curvula* and *C. dactylon* increased and *S. bicolor* decreased as mentioned.

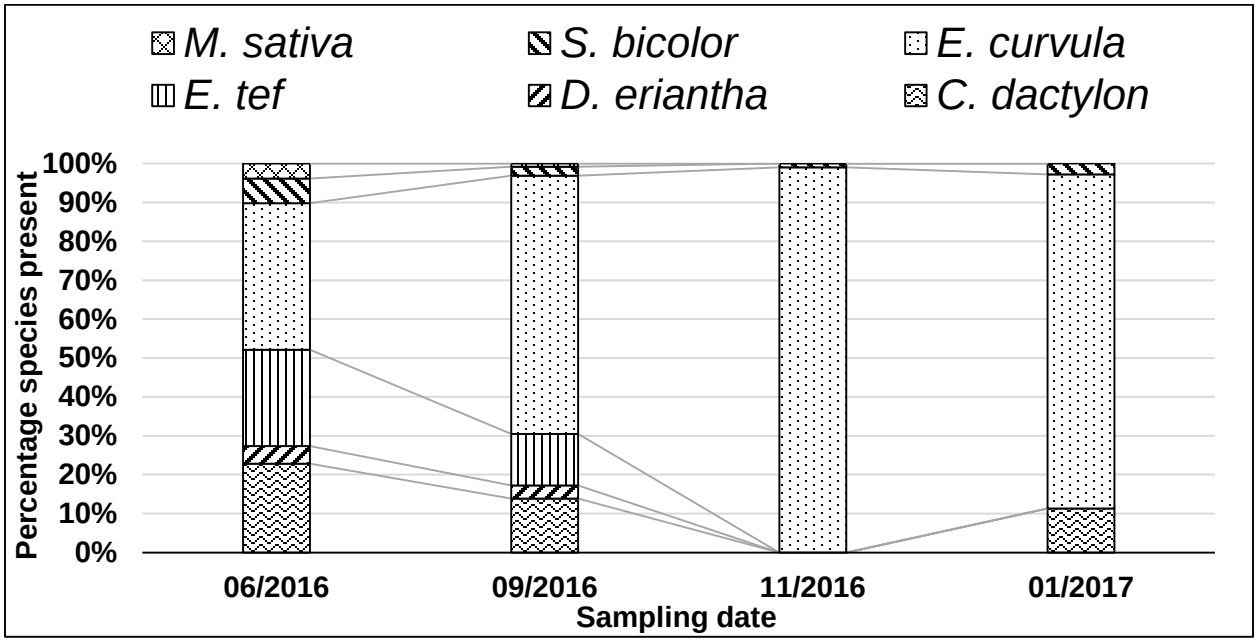

Figure 4.27: Change in plant composition of species used (*C. dactylon, D. eriantha, E. curvula, E. tef, S. bicolor, M. sativa*) in T3C at the Rooikraal gold TSF site from June 2016 to January 2017.

Table 4.22: Summary of plant density (plants/m^2) and plant composition (%) results for *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor,* and *M. sativa* in T3C at the Rooikraal gold TSF site from June 2016 to January 2017.

T3C	06/2016		09/2016		11/2016		01/2017	
Species	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**
C. dactylon	33,8	22,8	11,4	13,9	0,0	0,0	1,8	11,3
D. eriantha	6,7	4,5	2,8	3,4	0,0	0,0	0,0	0,0
E. tef	36,7	24,8	10,9	13,2	0,0	0,0	0,0	0,0
E. curvula	55,8	37,7	54,7	66,4	11,6	99,0	13,4	85,8
S. bicolor	9,4	6,4	1,9	2,3	0,1	1,0	0,4	2,8
M. sativa	5,7	3,8	0,7	0,8	0,0	0,0	0,0	0,0
Total	**148,0**	**100,0**	**82,3**	**100,0**	**11,7**	**100,0**	**15,7**	**100,0**

The change in the species density for T4C is shown in Figure 4.28, the plant composition percentages is illustrated in Figure 4.29 and Table 4.23 is summary of the results illustrated in the aforementioned figures.

The density of *E. tef* (of139,3 plants/m^2 in June 2016 was much higher than the density at the same time for treatments T1C, T2UC and T3C. In September 2016 the density of *E. tef* (decreased radically to 27,9 plants/m^2 and in November 2016 to 1,2 plants/m^2, before disappearing in January 2017 (Figure 4.28 and Table 4.23.

The density of *E. curvula* increased from 50,7 plants/m^2 in June 2016 to 67,4 plants/m^2 in September, after which it decreased to only 2,2 plants/m^2 in November 2016 (Figure 4.28 and Table 4.23.

The initial density of *C. dactylon* was 15,3 plants/m^2 in June 2016 (Figure 4.28). This density decreased to 8,2 plants/m^2 in September 2016 before disappearing in November 2016 (Figure. 4.28).

D. eriantha had a density of 10,3 plants/m^2, before decreasing to 2,2 plants/m^2 in September 2016 and then disappearing towards the end of the trial. The plots of treatment T4C were the worst hit by the erosion event since only one specie survived at the end of the trail (Figure 4.28 and Table 4.23).

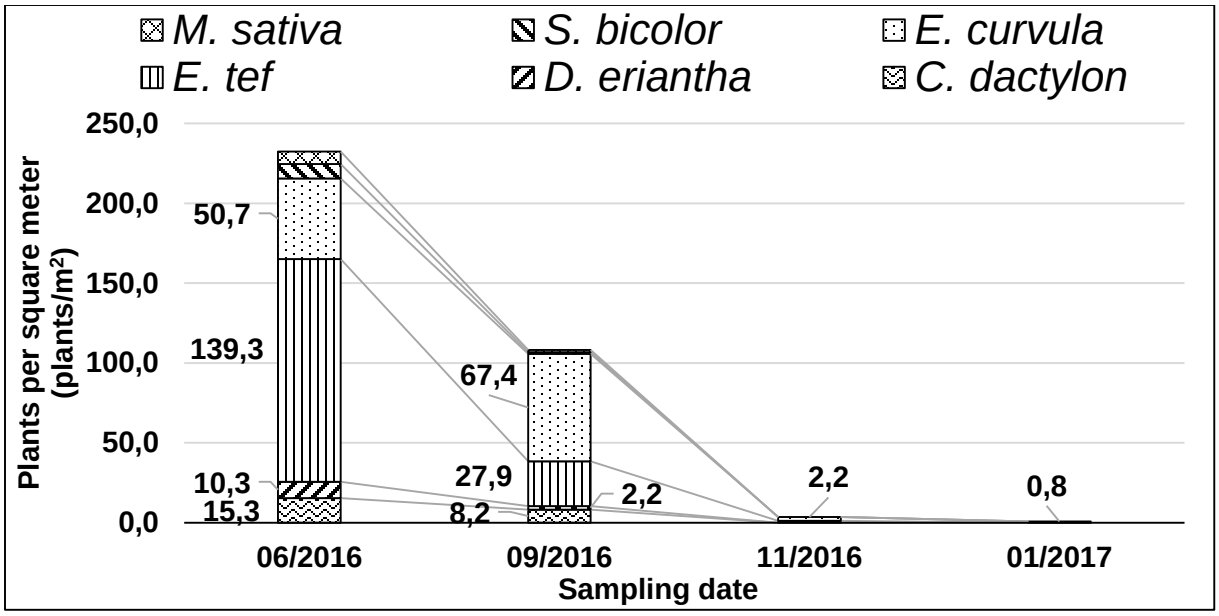

Figure 4.28: Change in plant density (plants/m^2) of species used (*C. dactylon, D. eriantha, E. curvula, E. tef, S. bicolor, M. sativa*) in T4C at the Rooikraal gold TSF site from June 2016 to January 2017.

Initially in June 2016 the plant composition of T4C was dominated by *E. tef* comprising 59,9% of the plant community followed by *E. curvula* with 21,8%, *C. dactylon* made up 6,6% of the plant community, *S. bicolor* 3,8%, *D. eriantha* 4,4% and *M. sativa* 3,4% (Figure 4.29 and Table 4.23). The presence of *E. tef* decreased to 25,8% in September 2016 before it increased to 35,5% in November 2016 before dying off ahead of January 2017. *E. curvula* increased in September 2016 to 62,4% and 64,5% in November 2016 before comprising 100% of the plant community in January 2017 (Figure 4.29 and Table 4.23). *C. dactylon*, *S. bicolor, D. eriantha* and *M. sativa* died off ahead of November 2016 (Figure 4.29 and Table 4.23).

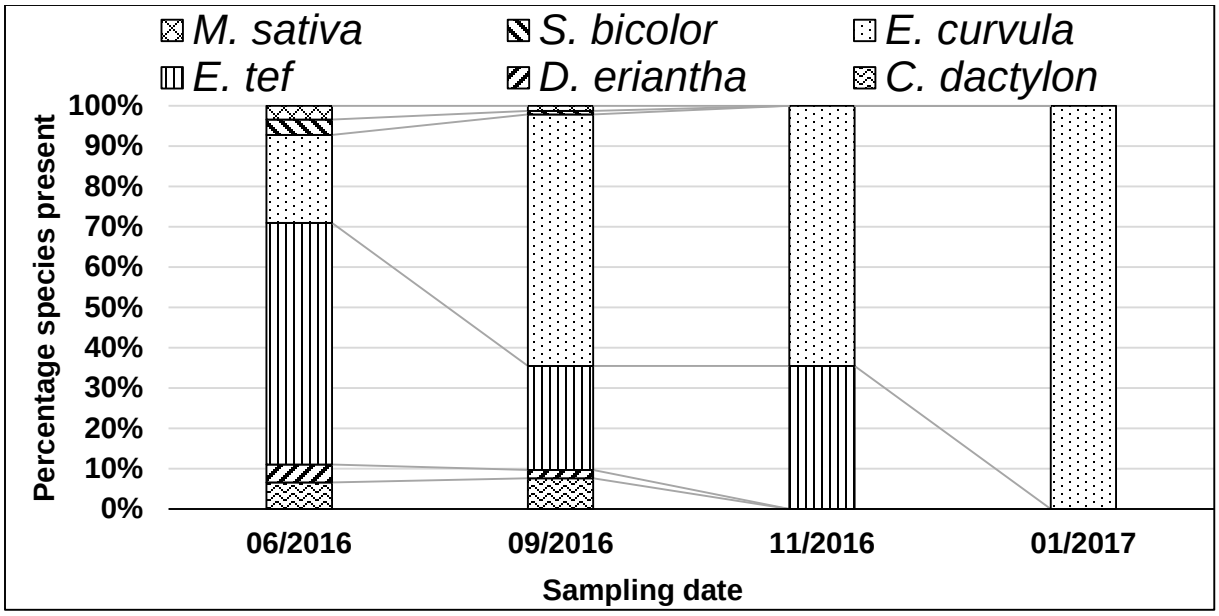

Figure 4.29: Change in plant composition of species used (*C. dactylon, D. eriantha, E. curvula, E. tef, S. bicolor, M. sativa*) in T4C at the Rooikraal gold TSF site from June 2016 to January 2017.

Table 4.23: Summary of plant density (plants/m^2) and plant composition (%) results for *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor*, and *M. sativa* in T4C at the Rooikraal gold TSF site from June 2016 to January 2017.

T4C	06/2016		09/2016		11/2016		01/2017	
Species	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**
C. dactylon	15,3	6,6	8,2	7,6	0,0	0,0	0,0	0,0
D. eriantha	10,3	4,4	2,2	2,1	0,0	0,0	0,0	0,0
E. tef	139,3	59,9	27,9	25,8	1,2	35,5	0,0	0,0
E. curvula	50,7	21,8	67,4	62,4	2,2	64,5	0,8	100,0
S. bicolor	8,9	3,8	1,0	0,9	0,0	0,0	0,0	0,0
M. sativa	7,9	3,4	1,3	1,2	0,0	0,0	0,0	0,0
Total	**232,4**	**100,0**	**108,1**	**100,0**	**3,4**	**100,0**	**0,8**	**100,0**

The results of the specie density change for T5UC at the Rooikraal gold TSF site are shown in Figures 4.30 and 4.31 and Table 4.24.

The species densities in T5UC and T4C both were larger at and 232,4 plants/m^2 and 247,9 plants/m^2 than for treatments T1C (108,6 plants/m^2), T2UC (105,9 plants/m^2) and T3C (148 plants/m^2). In June 2016, the density of *E. tef* of 113,3 plants/m^2 was the highest compared to the other species and made out 45.8% of the vegetation composition. *E. curvula* initially made out 35,5% of the plant composition with a density of 87,9 plants/m^2 (Figure 4.30 and figure 4.31).

While the density of *E. tef* decreased from 113,3 plants/m^2 to 14,1 plants/m^2, the density of *E. curvula* remained generally constant from 87,9 to 88,2 plants/m^2. This made the *E. curvula* the

dominant species in the vegetation composition (78,7%) in September 2016. *D. eriantha* was present at a density of 16,4 plants/m^2 in June 2016, before decreasing and disappearing with the majority of the species in November 2016 (Figures 4.30 and Table 4.24.

C. dactylon decreased from an initial density of 16,3 plants/m^2 in June 2016 to 4,9 plants/m^2 in September 2016 and was still present in November 2016 (0,9 plants/m^2), after which it increased to 1,4 plants/m^2 (Figure 4.30).

D. eriantha had an initial density of 16,4 plants/m^2 in June 2016 before decreasing to 4,2 plants/m^2 in September 2016 and disappearing in November 2016 (Figure 4.30).

In treatment T5UC *M. sativa* was present at a density of 4,1 plants/m^2, which decreased to 0,1 plants/m^2 in September 2016. *S. bicolor* was also present at a density of 9,4 plants/m^2 in June 2016, which then decreased to 0,6 plants/m^2 in September 2016. Neither *S. bicolor* nor *M. sativa* survived after November 2016 (Figures 4 30 and 4.31 and Table 4.24).

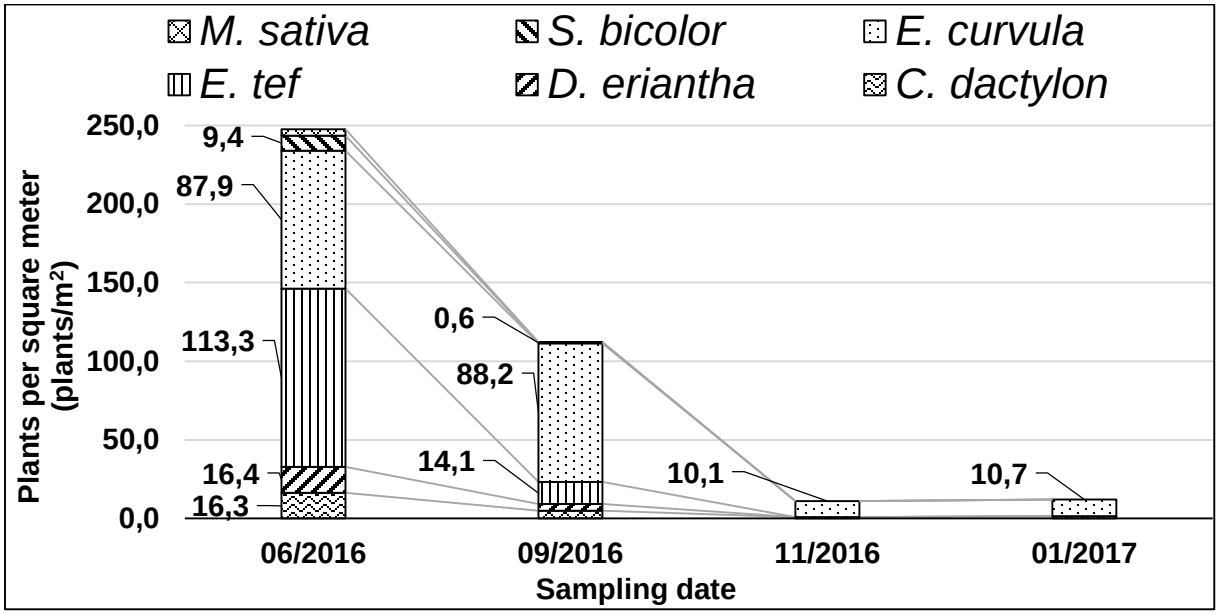

Figure 4.30: Change in plant density (plants/m^2) of species used (*C. dactylon, D. eriantha, E. curvula, E. tef, S. bicolor, M. sativa*) in T5UC at the Rooikraal gold TSF site from June 2016 to January 2017.

The plant composition was initialy dominated by *E. curvula* (35,5%) and *E. tef* (45,8%) in June 2016. *E. curvula* increased to contribute 78,7% of the plant composition in September 2016 and reached a maximum of 91,9% in November 2016 (Figure 4.31 and Table 4.24).

The presence of *E. tef* had decreased to 12,6% in Septemeber 2016 and was no longer present from November 2016 until the end of the trial in January 2017 (Figure 4.31 and Table 4.24).

C. dactylon (6,6%), *D.* eriantha (6,6%), S*. bicolor* (3,8%) and *M. sativa* (1,7%) were present intialy in June 2016. However, the presence of *C. dactylon* (4,4%), *D. eriantha* (3,8%), *S. bicolor* (0,5%) and *M. sativa* (0,1%) decreased (Figure 4.31 and Table 4.24).

D. eriantha, *S. bicolor* and *M. sativa* were no longer present in November 2016. *C. dactylon* increased from 4,4% in September 2016 to 8,1% in November 2016 and finaly comprised 11,9% of the plant composition in January 2017 (Figure 4.31 and Table 4.24).

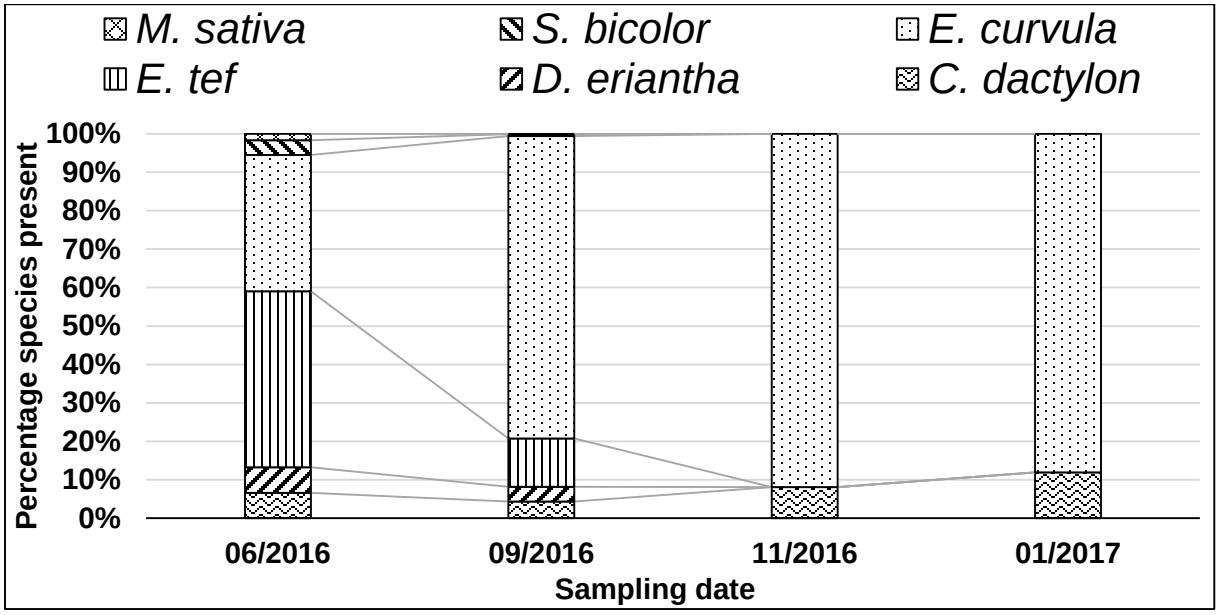

Figure 4.31: Change in plant composition of species used (*C. dactylon, D. eriantha, E. curvula, E. tef, S. bicolor, M. sativa*) in T5UC at the Rooikraal gold TSF site from June 2016 to January 2017.

Table 4.24: Summary of plant density (plants/m^2) and plant composition (%) results for *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor,* and *M. sativa* in T5UC at the Rooikraal gold TSF site from June 2016 to January 2017.

T5UC	**06/2016**		**09/2016**		**11/2016**		**01/2017**	
Species	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**
C. dactylon	16,3	6,6	4,9	4,4	0,9	8,1	1,4	11,9
D. eriantha	16,4	6,6	4,2	3,8	0,0	0,0	0,0	0,0
E. tef	113,3	45,8	14,1	12,6	0,0	0,0	0,0	0,0
E. curvula	87,9	35,5	88,2	78,7	10,1	91,9	10,7	88,1
S. bicolor	9,4	3,8	0,6	0,5	0,0	0,0	0,0	0,0
M. sativa	4,1	1,7	0,1	0,1	0,0	0,0	0,0	0,0
Total	**247,6**	**100,0**	**112,1**	**100,0**	**11,0**	**100,0**	**12,1**	**100,0**

To summarise the change in species densities and the plant composition of the seed treatments on the Rooikraal gold TSF site is as follows. Initially, in June 2016, *E. curvula* and *E. tef* were the

two dominating species. However, *E. tef* then decreased due to the annual life stage, while the *E. curvula* persevered due to its being perennial.

The T4C and T5UC nearly had double the emergence and survival of *E. tef* compared to the other three treatments. These results showed that *E. tef* had a much higher initial total plant density and had being the dominant species at the beginning of the trials.

E. curvula, *C. dactylon* and *S. bicolor* were the species most likely to survive after being buried during the erosion event, which showed that these species have a unique advantage of resprouting even if covered with material after erosion.

D. eriantha had the lowest density of the grass species in each of the treatments. *S. bicolor* and *M. sativa* emerged, but were mostly unable to survive later than November 2016.

4.3.4 Rooikraal gold TSF site total cover contribution of species

The change in total cover (basal and canopy) contribution of species for seed treatments at the Rooikraal gold TSF site are illustrated in Figures 4.32 to 4.36 and Tables 4.25 to 4.29.

Figure 4.32 and Table 4.25 illustrate the plant cover results for T1C. Initially T1C had a total vegetation coverage of 50,6%. *E. curvula* (18,8%), *E. tef* (14,2%) and *C. dactylon* (11,5%) were the species that contributed the most to the total cover. In September 2016, the total vegetation cover dropped to 39,8%. This was caused by the decrease in cover of *E. tef* and *C. dactylon*, whereas the cover of *E. curvula* increased to 32,7% (Figure 4.32 Table 4.25).

In November 2016 the total vegetation cover decreased to 31,7% with *E. tef* being totally absent. In January 2017, the cover increased again to 79,6%, mostly as a result of the increase in cover by *E. curvula* to 59,8% and *C. dactylon* to 16% (Figure 4.32 and Table 4.25).

S. bicolor provided 4,6% coverage in June 2016; this decreased to 1,5% in September 2016 and 1% in November 2016, but increased to 3,8% in January 2017 (Figure 4.32 and Table 4.25).

D. eriantha and *M. sativa* contributed the least to the total cover, 1.5% and 0.2%, respectively (Figure 4.32 Table 4.25). The cover then decreased again in September 2016, with only *E. curvula* and *C. dactylon* providing cover until January 2017 (Figure 4. 32 and Table 4.25.

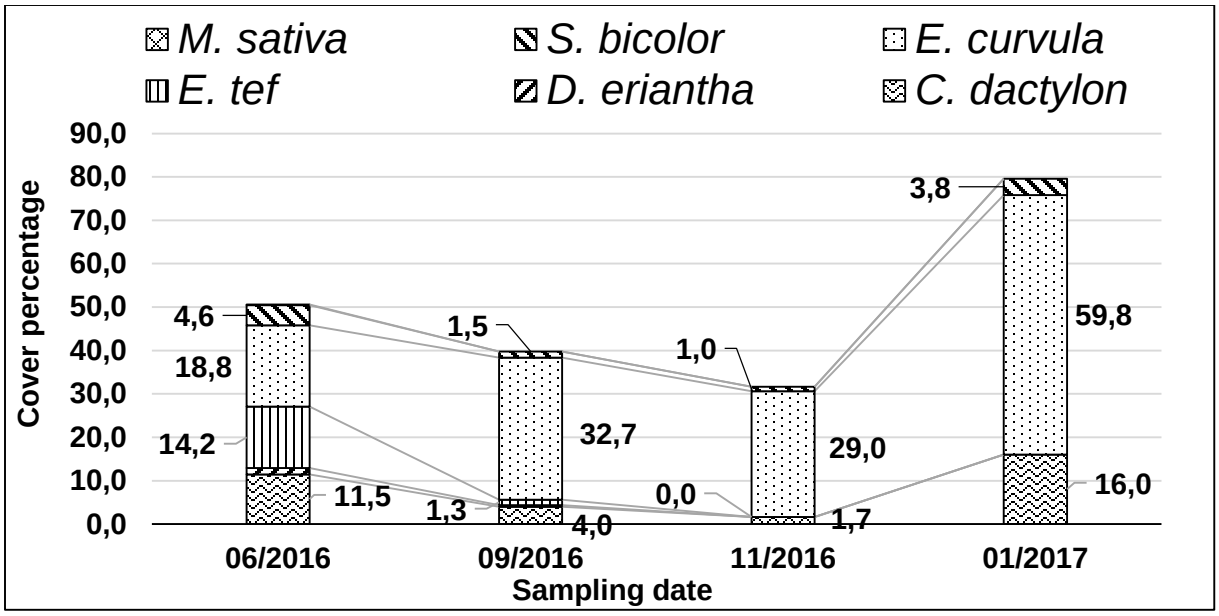

Figure 4.32: Change in total cover contribution of *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor* and *M. sativa* in T1C at the Rooikraal gold TSF site from June 2016 to January 2017.

Table 4.25: Change in total cover contribution of *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor and M. sativa* in T1C at the Rooikraal gold TSF site from June 2016 to January 2017 shown in Figure 4.32.

T1C	06/2016	09/2016	11/2016	01/2017
Species	**%**	**%**	**%**	**%**
C. dactylon	11,5	4,0	1,7	16,0
D. eriantha	1,5	0,4	0,0	0,0
E. tef	14,2	1,3	0,0	0,0
E. curvula	18,8	32,7	29,0	59,8
S. bicolor	4,6	1,5	1,0	3,8
M. sativa	0,2	0,0	0,0	0,0
Total	**50,6**	**39,8**	**31,7**	**79,6**

Figure 4.33 and Table 4.26 below illustrate the cover of species for T2UC at the Rooikraal gold TSF site.

In June 2016 the total vegetation coverage of the species was 63,1%. *E. tef* (26,9%), *E. curvula* (17,5%) and *C. dactylon* (13,5%) were the species with the highest cover contribution at that time (Figure 4.33 and Table 4.26).

In September 2016 the total vegetation cover decreased to 34,8% (Figure 4.33). The cover for grasses such as *E. tef* was 4% and *C. dactylon*, 5% respectively, while the cover for *E. curvula* increased to 24,4% (Figure 4.33 and Table 4.26).

In November 2016, the total cover decreased to 20,2%, after the erosion event that occurred during this month (Figure 4.33 and Table 4.26).

In January 2017 the vegetation cover doubled to 42,3%, with *E. curvula* (39,4%) forming the largest part of the cover, while the cover for *C. dactylon* was only 2,7% (Figure 4.33 and Table 4.26).

D. eriantha and *M. sativa* were again the two species with the least amount of cover in June 2016 (Figure 4.33 and Table 4.26).

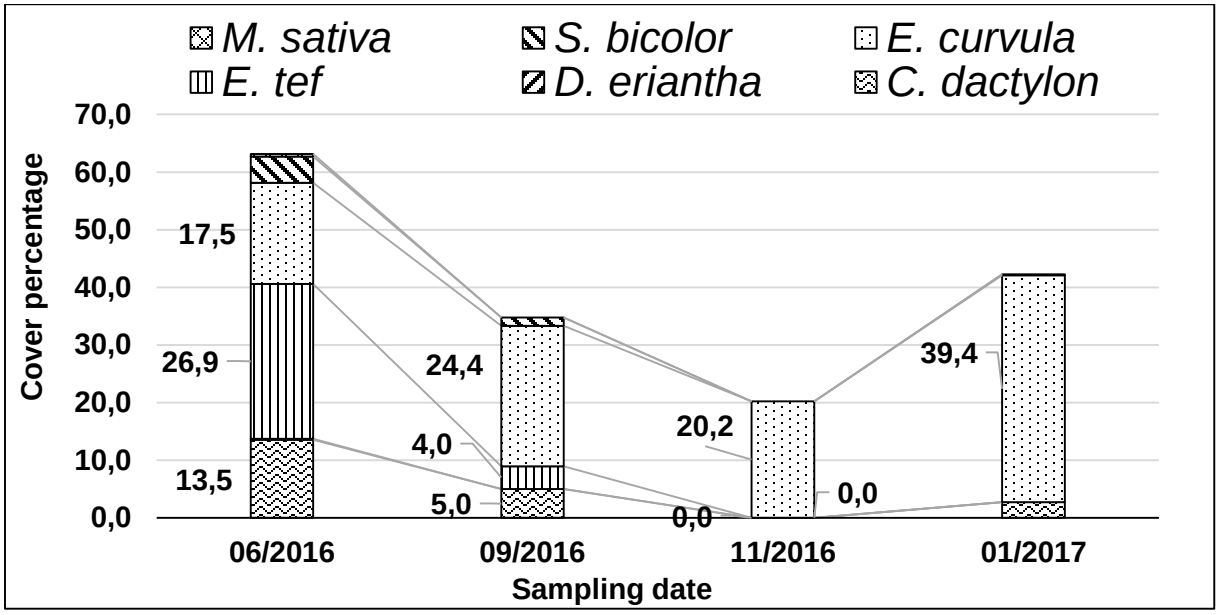

Figure 4.33: Change in total cover contribution of *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor* and *M. sativa* in T2UC at the Rooikraal gold TSF site from June 2016 to January 2017.

Table 4.26: Change in total cover contribution percentage of *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor* and *M. sativa* in T2UC at the Rooikraal gold TSF site from June 2016 to January 2017 shown in Figure 4.33.

T2UC	06/2016	09/2016	11/2016	01/2017
Species	**%**	**%**	**%**	**%**
C. dactylon	13,5	5,0	0,0	2,7
D. eriantha	0,2	0,0	0,0	0,0
E. tef	26,9	4,0	0,0	0,0
E. curvula	17,5	24,4	20,2	39,4
S. bicolor	4,6	1,5	0,0	0,2
M. sativa	0,4	0,0	0,0	0,0
Total	**63,1**	**34,8**	**20,2**	**42,3**

The vegetation cover for T3C, in which the weight of coated seed was increased to match the number of seeds sown in T2UC, is shown in Figure 4.34 and Table 4.27

The total vegetation cover was the highest in June 2016 (66,9%) with *E. tef* (24,6%), *E. curvula* (19,4%) and *C. dactylon* (20%) contributing to most of the cover (Figure 4.34). The total cover decreased to 46,9% in Septembers 2016, with *E. tef* (3,3%) and *C. dactylon* (7,9%) decreasing and the cover of *E. curvula* (35%) increasing (Figure 4.34 and Table 4.27).

In November 2016, after the erosion event, the total vegetation coverage decreased to a low of 26% with *E. curvula* accounting for nearly all the coverage (25,8%) and *S. bicolor* a minor 0,2% (Figure 4.34 and Table 4.27).

In January 2017 the vegetation coverage increased to 42,1%. *E. curvula* (37,7%) and *S. bicolor* (0,6%) increased since November 2016 and *C. dactylon* returned, providing 3,8% vegetation coverage (Figure 4.34 and Table 4.27).

D. eriantha (0,6%) and *M. sativa* (0,2%) provided the least cover and were absent from November 2016 (Figure 4.34 and Table 4.27).

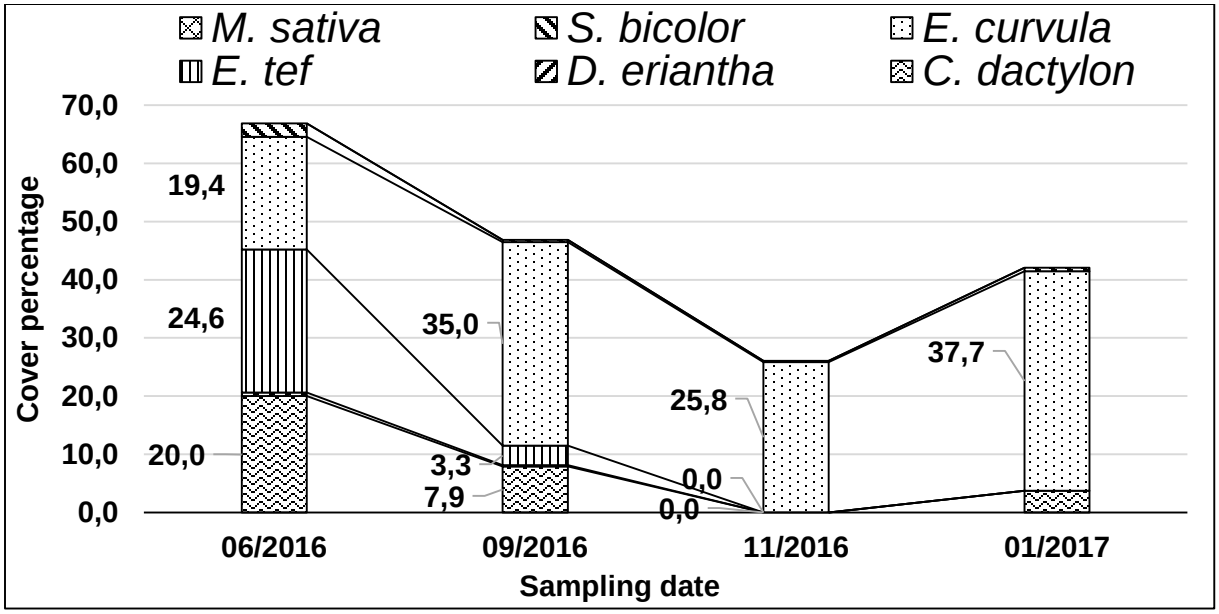

Figure 4.34: Change in total cover contribution of *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor* and *M. sativa* in T3C at the Rooikraal gold TSF site from June 2016 to January 2017.

Table 4.27: Change in total cover contribution percentage of *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor and M. sativa* inT3C at the Rooikraal gold TSF site from June 2016 to January 2017 shown in Figure 4.34

T3C	06/2016	09/2016	11/2016	01/2017
Species	**%**	**%**	**%**	**%**
C. dactylon	20,0	7,9	0,0	3,8
D. eriantha	0,6	0,2	0,0	0,0
E. tef	24,6	3,3	0,0	0,0
E. curvula	19,4	35,0	25,8	37,7
S. bicolor	2,3	0,4	0,2	0,6
M. sativa	0,0	0,0	0,0	0,0
Total	**66,9**	**46,9**	**26,0**	**42,1**

Figure 4.35 and Table 4.28 illustrate the change in vegetation cover for T4C from June 2016 to January 2017. T4C had the highest total vegetation cover (91,3%) in June 2016, with *E. tef* (64,8%) providing the largest amount of cover. *E. curvula* (16%), *C. dactylon* (4,8%) and *S. bicolor* (4,2%) also accounted for a part of the vegetation cover (Figure 4.35).

The total vegetation cover decreased in Septembers 2016 to 53,1% for T4C. The cover of *E. curvula* increased to 31,7%, whereas the cover of *E. tef* (19,4%) and *C. dactylon* (1,9%) decreased (Figure 4.35 and Table 4.28).

After the erosion event in November 2016, the whole plot only had a vegetation cover of 2,7%, mainly due to the cover of *E. curvula*. The cover further decreased to 1,3% in January 2017 (Figure 4.35 and Table 4.28).

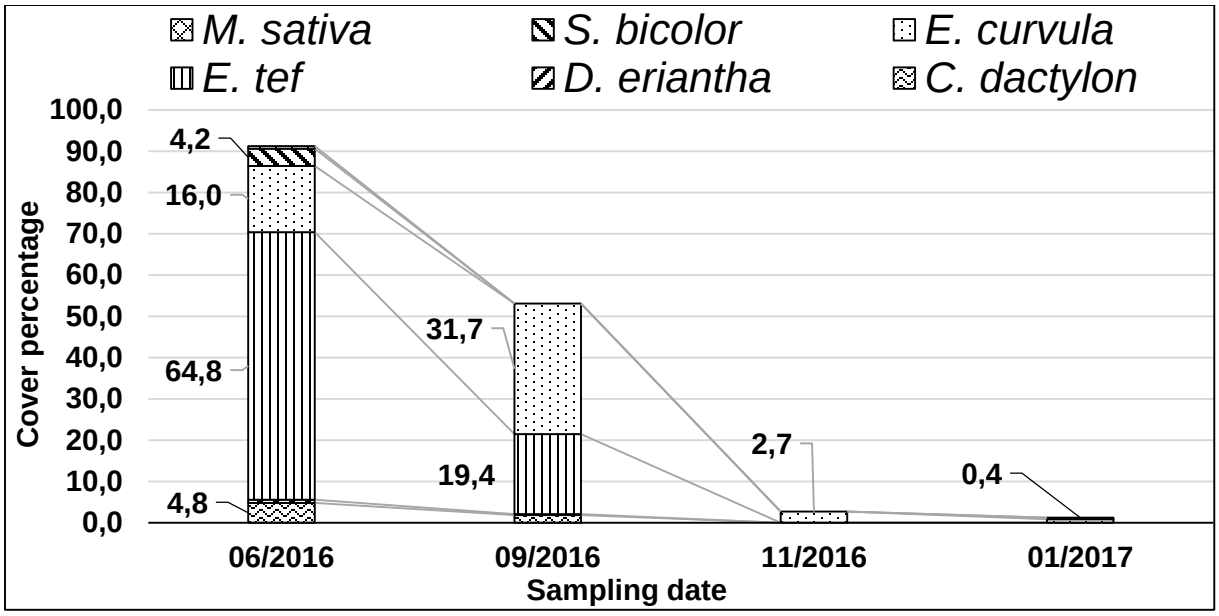

Figure 4.35: Change in total cover contribution of *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor* and *M. sativa* in T4C at the Rooikraal gold TSF site from June 2016 to January 2017.

Table 4.28: Change in total cover contribution percentage of *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor and M. sativa* in T4C at the Rooikraal gold TSF site from June 2016 to January 2017 shown in Figure 4.35.

T4C	06/2016	09/2016	11/2016	01/2017
Species	**%**	**%**	**%**	**%**
C. dactylon	4,8	1,9	0,0	0,0
D. eriantha	0,8	0,2	0,0	0,0
E. tef	64,8	19,4	0,0	0,0
E. curvula	16,0	31,7	2,7	0,8
S. bicolor	4,2	0,0	0,0	0,4
M. sativa	0,6	0,0	0,0	0,0
Total	**91,3**	**53,1**	**2,7**	**1,3**

The vegetation cover for T5UC is illustrated in Figure 4.36 and Table 4.29). The highest vegetation cover was present in June 2016 (78,1%), mostly accounted for by *E. tef* (43,8%) and *E. curvula* (26,9%), with minor contributions by *C. dactylon* (3,8%), *S. bicolor* (2,1%) and *D. eriantha* (1,5%) (Figure 4.36 and Table 4.29).

In Figure 4.36 and Table 4.29 The total vegetation cover decreased to 56% in September 2016. Now *E. curvula* provided the most vegetation cover of 49%; the cover of *E. tef* decreased to 5,6%, as well as *C. dactylon* decreased to 0,4%, *S. bicolor* decreased to 0,4% and *D. eriantha* decreased to 0,6% (Figure 4.36 and Table 4.29).

After the erosion event in November 2016, the total vegetation coverage decreased to 23,3%, with mostly *E. curvula* left with a total cover of 22,5% (Figure 4.36 and Table 4.29).

The total vegetation cover then increased to 25,8% with an increase in cover of *E. curvula* (24%) and *C. dactylon* from not being present in November 2016 to 1,9% in January 2017 (Figure 4.36 and Table 4.29).

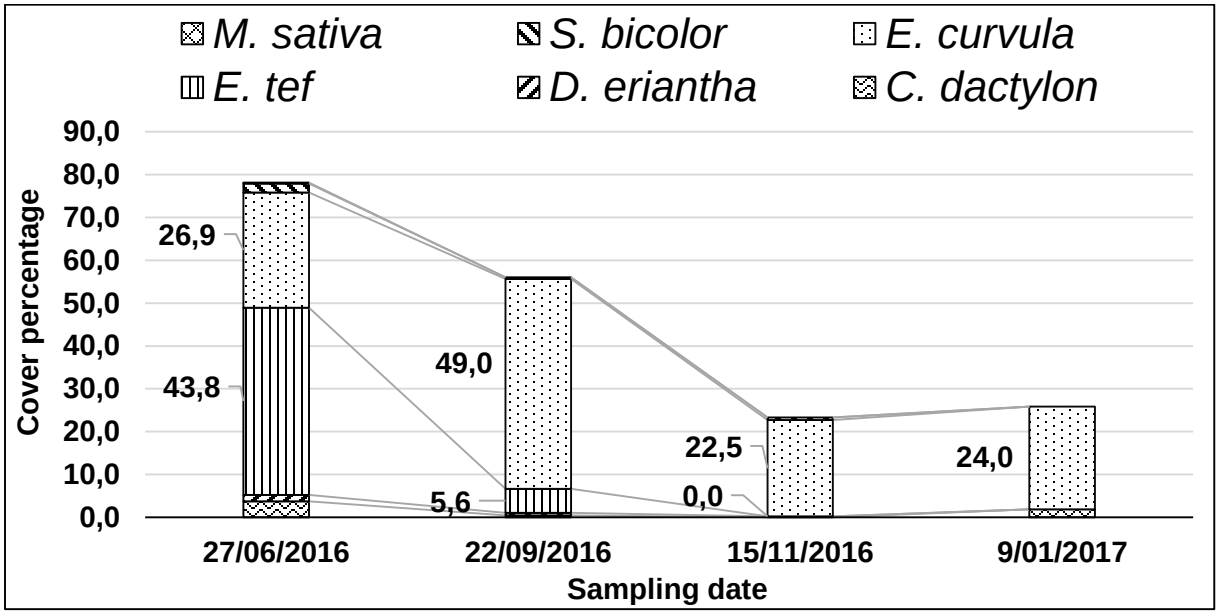

Figure 4.36: Change in total cover contribution of *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor* and *M. sativa* in T5UC at the Rooikraal gold TSF site from June 2016 to January 2017.

Table 4.29: Change in total cover contribution percentage of *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor and M. sativa* in T5UC at the Rooikraal gold TSF site from June 2016 to January 2017 shown in Figure 4.36.

T5UC	06/2016	09/2016	11/2016	01/2017
Species	**%**	**%**	**%**	**%**
C. dactylon	3,8	0,4	0,0	1,9
D. eriantha	1,5	0,6	0,2	0,0
E. tef	43,8	5,6	0,0	0,0
E. curvula	26,9	49,0	22,5	24,0
S. bicolor	2,1	0,4	0,6	0,0
M. sativa	0,2	0,0	0,0	0,0
Total	**78,1**	**56,0**	**23,3**	**25,8**

To summarise the vegetation cover results for the seed treatments sown at the Rooikraal gold TSF site, the following is given:

The change in vegetation coverage provided by the individual species followed the same pattern throughout each of the five seed treatments. *E. Tef*, *E. curvula* and occasionally *C. dactylon* had the highest cover in June 2016. Towards September 2016, the cover was mostly provided by *E. curvula,* with a decrease in the cover of *E. tef* (Figure 4.32 to 4.36 and Table 4.24 to 4.29). Thereafter (November 2016 to January 2017) *E. curvula* provided the highest amount of cover, with *C. dactylon* and *S. bicolor* also increasing.

The initial high total vegetation coverage in June 2016 decreased following the decrease of cover by *E. tef* towards September 2016. This was expected, as *E. tef* is a pioneer, annual grass of which the cover and abundance decreases towards the end of the season. The increase of cover by *E. curvula* was also expected, since this is a sub-climax, perennial grass which grows more than one year (Van Oudtshoorn, 2004:177).

Some trials were affected more than others by the erosion event in November 2016. *E. curvula* was one of the species that recovered and could protrude out of the covered deposited material, mainly due to the high culm (30–1200 mm) and leaf (up to 400 mm) lengths (Van Oudtshoorn, 2004:177). *S. bicolor* and *C. dactylon* also survived and in some cases even increased after the erosion event. *S. bicolor,* like *E. curvula,* is tall (culms 300–3000 mm) allowing it to continue growing after the burial of the material caused by the erosion event. *C. dactylon*, on the other hand, has a mat forming growth form carrying its inflorescence on much shorter flimsy culms (50–400 mm), making it unlikely to protrude from deposited gold mine tailings, except where the rhizomes were already present which helped this species to stay alive and recover (Van Oudtshoorn, 2004:229).

D. eriantha had the least cover after the erosion event. This could be attributed to low plant density caused by the very low seed viability of 22% for the seed batch used for this species (Figures 4.22, 4.24, 4.26, 4.28, 4.30 and 3.11).

M. sativa also had a low cover for each of the treatments, which can also be attributed to the low emergence and plant density of this species.

Both T4C and T5UC had a higher vegetation coverage (≥ 78%) than the other three treatments T1C, T2UC and T3C in June 2017 with *E. tef* accounting for most of the cover. This can most likely be due to a higher *E. tef* seeding rate of 1,9 kg/ha in T4C and T5UC that led to more *E. tef* seedlings being densely present in T4C and T5UC. 3.

4.3.5 Platinum trials species density and plant composition

In this Section, the plant density and plant composition of the seed treatments sown on the patinum trials are discussed.

In Figures 4.37 to 4.38 and Table 4.30 the average density of the species sown are illustrated, as well as the plant composition for T1C.

Initially in August 2016, the total plant density was 39 plants/m^2 (Figure 4.37 and Table 4.30). The total plant density then increased to 66 plants/m^2 in September 2016, after which it decreased to 28 plants/m^2 in March 2017and to 16 plants/m^2 in May 2017 (Figure 4.37).

In August 2016, *E. curvula* had the highest density (22 plants/m^2) compared to other species. *E. curvula* increased in September 2016 (35 plants/m^2) before decreasing and reaching its lowest density at the end of the trials in May 2017. *C. dactylon* had a density of 8 plants/m^2 which doubled in September 2016 to 16 plants/m^2, before decreasing to 7 plants/m^2 till the end of the trail in May 2017 (Figure 4.37).

D. eriantha was present at 1 plant/m^2 in August 2016 it then increased to 6 plants/m^2 in September 2016 before decreasing to 1 plant/m^2 in November 2016 and dying out before May 2017. *E. tef* had a density of 4 plants/m^2 occupying 10,3% of the community but was no longer present in November 2016. *M. sativa* was also present at 4 plants/m^2 in August 2016, after which it decreased to 3 plants/m^2 in May 2016 (Figure 4.37 Table 4.30).

The only three species that were present at the end of the trail in May 2017, included *E. curvula* at 3 plants/m^2, *C. dactylon* at plants/m^2 and *M. sativa* at 3 plants/m^2 (Figure 4.37 and Table 4.30).

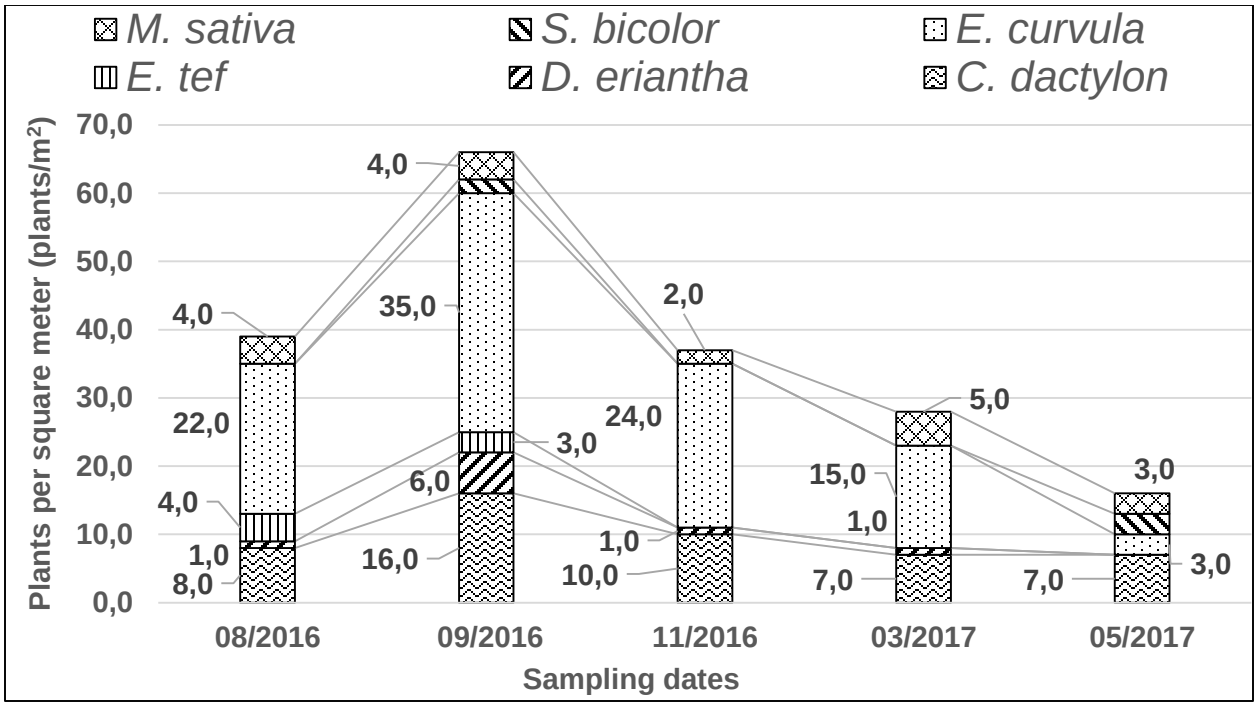

Figure 4.37: Change in plant density (plants/m^2) of species used (*C. dactylon, D. eriantha, E. curvula, E. tef, S. bicolor, M. sativa*) in T1C in the platinum trials from August 2016 to May 2017.

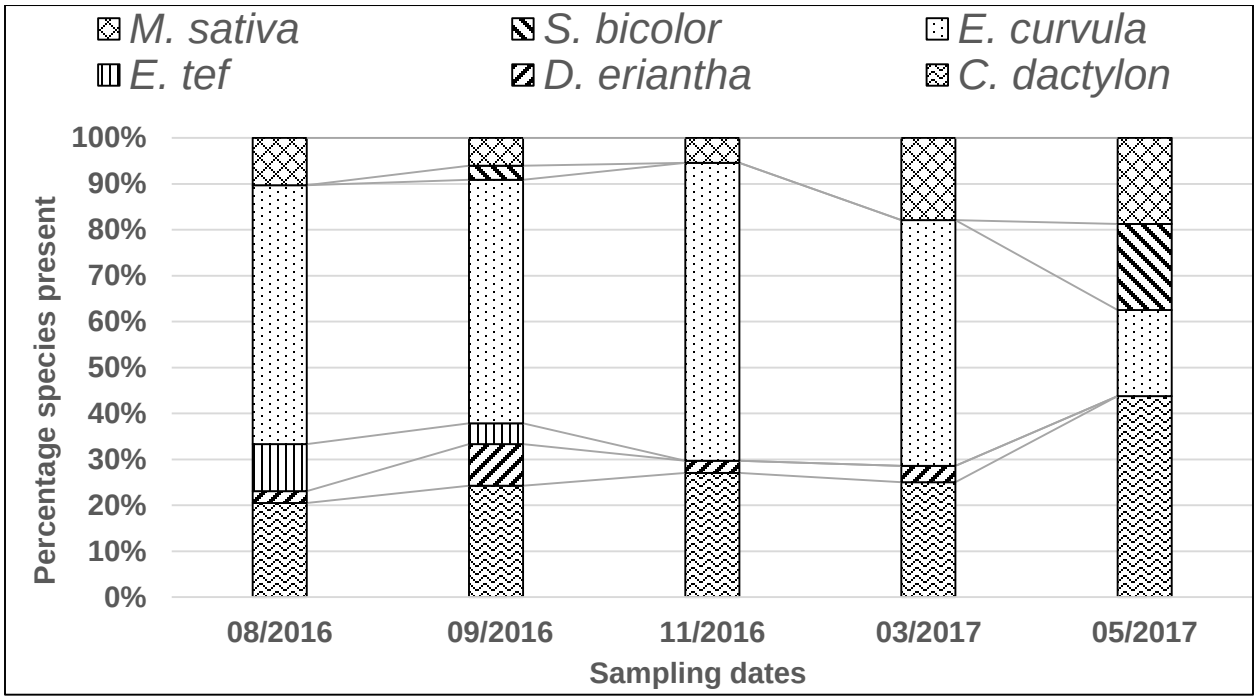

Figure 4.38: Change in plant composition of species used (*C. dactylon, D. eriantha, E. curvula, E. tef, S. bicolor, M. sativa*) in T1C in the platinum trials from August 2016 to May 2017.

E. curvula was the dominant species (64,9%–53%) from August 2016 until March 2017, *C. dactylon* (43,8%) then became the dominant species in May 2017 followed by *M. sativa*, *S. bicolor* and *E. curvula* each contributing 18,8% to the plant composition (Figure 4.38 and Table 4.30

Table 4.30: Summary of plant density (plants/m^2) and plant composition (%) results for *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor* and *M. sativa* in T1C in the platinum trials from August 2016 to May 2017.

T1 C	08/2016		09/2016		11/2016		03/2017		05/2017	
Species	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**
C. dactylon	8,0	20,5	16,0	24,2	10,0	27,0	7,0	25,0	7,0	43,8
D. eriantha	1,0	2,6	6,0	9,1	1,0	2,7	1,0	3,6	0,0	0,0
E. tef	4,0	10,3	3,0	4,5	0,0	0,0	0,0	0,0	0,0	0,0
E. curvula	22,0	56,4	35,0	53,0	24,0	64,9	15,0	53,6	3,0	18,8
S. bicolor	0,0	0,0	2,0	3,0	0,0	0,0	0,0	0,0	3,0	18,8
M. sativa	4,0	10,3	4,0	6,1	2,0	5,4	5,0	17,9	3,0	18,8
Total	**39,0**	**100,0**	**66,0**	**100,0**	**37,0**	**100,0**	**28,0**	**100,0**	**16,0**	**100,0**

The change in the density of species and the plant composition for T2UC in the platinum trials are illustrated in Figure 4.39, 4.40 and Table 4.31.

In August 2016, the total vegetation density was 37 plants/m^2 in T2UC (Figure 4.39 and Table 4.31) This density increased to 56 plants/m^2 before decreasing to 31 plants/m^2 in November, with further decrease to 25 plants/m^2 at the end of the trial in May 2017 (Figure 4.39 and Table 4.31).

The density of *E. curvula* increased from 19 plants/m^2 in August 2016 to 23 plants/m^2 in September 2016, before decreasing to a low of 11 plants/m^2 in May 2017 (Figure 4.39). The density for *C. dactylon* was initially 15 plants/m^2 (August 2016), after which the destiny increased to 28 plants/m^2 in September 2016 before decreasing to 9 plants/m^2 in May 2017 (Figure 4.39). The density of *D. eriantha* was zero in August 2016 it then increased to 2 plants/m^{2}and remained constant from September 2016 to March 2017 before increasing to 4 plants/m^2 in May 2017. The density of *M. sativa* remained constant throughout the trial at 1 plant/m^2 except for an increase to 2 plant/m^2 in November 2016 before returning to 1 plant/m^2 in March 2017 (Figure 4.39).

E. tef was not present in August 2016 however, only 2 plants/m^2 were then presentin September 2017 after which it died (Figure 4.39 and Table 4.31).

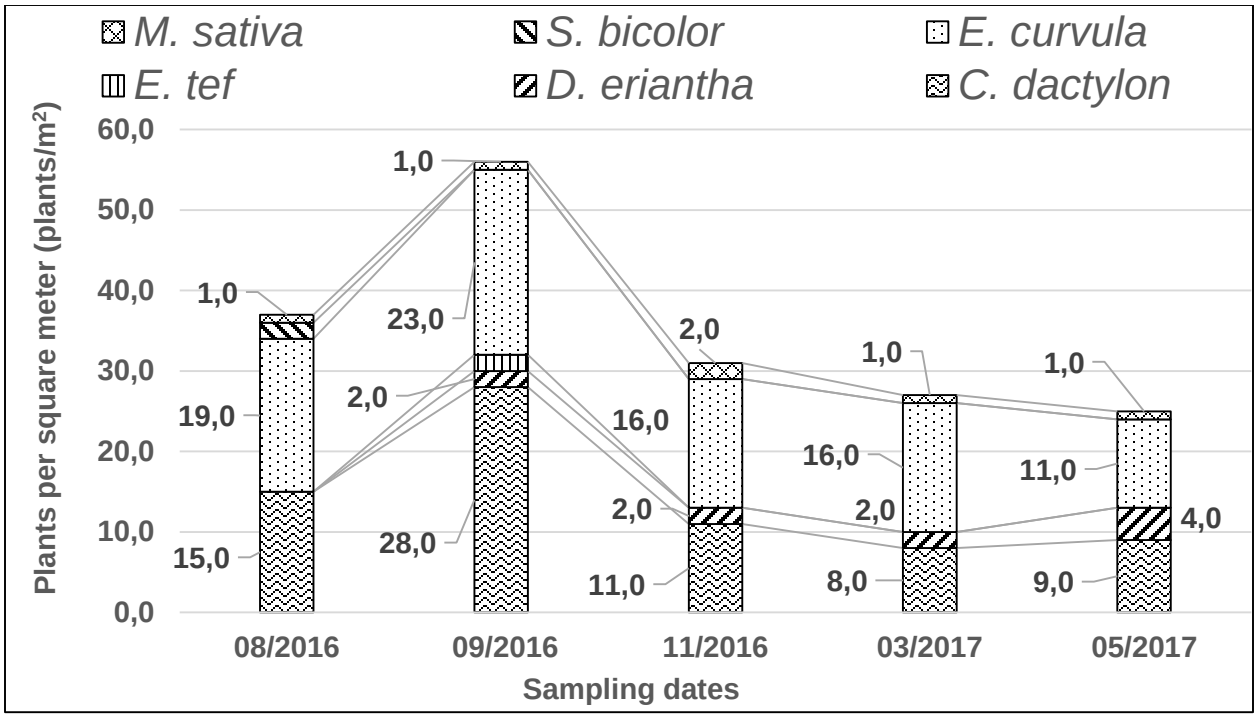

Figure 4.39: Change in plant density (plants/m^2) of species used (*C. dactylon, D. eriantha, E. curvula, E. tef, S. bicolor, M. sativa*) in T2UC in the platinum trials from August 2016 to May 2017.

The plant composition was dominated by *E. curvula* and *C. dactylon* throughout the trial. (Figure 4.40 and Table 4.28). The contribution of *D. eriantha* to the plant composition was initialy zero in August 2016 however, it comprised 3,6% of the plant composition in September 2016 and gradualy dominated a larger fraction of the plant composition as the trial progressed (Figure 4.40 and Table 4.31). The presence of *M. sativa* in the plant community remained small but was consistently present between 1,8% and 6,5% throughout the trial period (Figure 4.40 and Table 4.31). *S. bicolor* was only present in August 2016 comprising 5,4% of the plant community and *E. tef* was only present in September 2016 comprising 3,6% of the plant community (Figure 4.40 and Table 4.31).

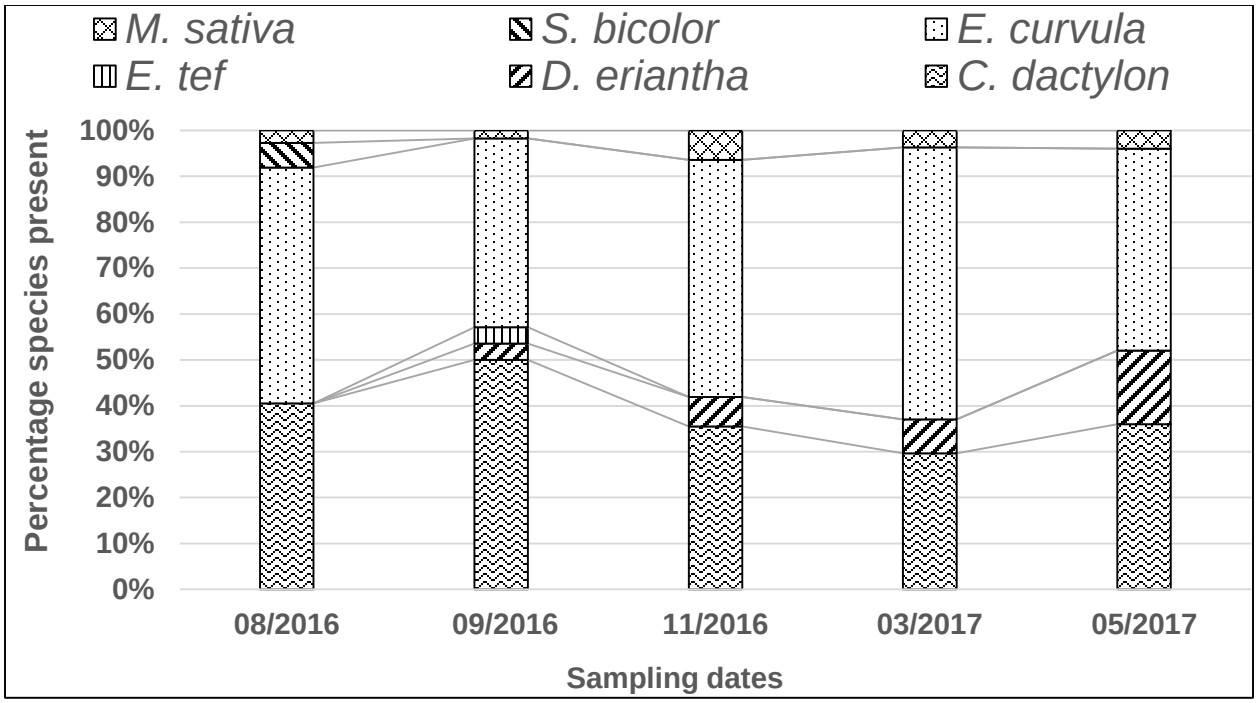

Figure 4.40: Change in plant composition percentage of species used (*C. dactylon, D. eriantha, E. curvula, E. tef, S. bicolor, M. sativa*) in T2UC in the platinum trials from August 2016 to May 2017.

Table 4.31: Summary of plant density (plants/m^2) and plant composition (%) results for *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor* and *M. sativa* in T2UC in the platinum trials from August 2016 to May 2017.

T2UC	08/2016		09/2016		11/2016		03/2017		05/2017	
Species	plants/m²	%	plants/m²	%	plants/m²	%	plants/m²	%	plants/m²	%
C. dactylon	15,0	40,5	28,0	50,0	11,0	35,5	8,0	29,6	9,0	36,0
D. eriantha	0,0	0,0	2,0	3,6	2,0	6,5	2,0	7,4	4,0	16,0
E. tef	0,0	0,0	2,0	3,6	0,0	0,0	0,0	0,0	0,0	0,0
E. curvula	19,0	51,4	23,0	41,1	16,0	51,6	16,0	59,3	11,0	44,0
S. bicolor	2,0	5,4	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0
M. sativa	1,0	2,7	1,0	1,8	2,0	6,5	1,0	3,7	1,0	4,0
Total	**37,0**	**100,0**	**56,0**	**100,0**	**31,0**	**100,0**	**27,0**	**100,0**	**25,0**	**100,0**

The species density and plant composition results of T3C for the platinum trials are illustrated in Figures 4.41, 4.42 and Table 4.32. T3C had a total vegetation density of 49 plants/m^2 in August 2016 before the density increased to a maximum of 51 plants/m^2 in September 2016 (Figure 4.41). The density then decreased to 33 plants/m^2 in November 2016, 29 plants/m^2 in March 2016 and 18 plants/m^2 in May 2017 (Figure 4.41 and Table 4.32).

The density of *E. curvula* remained constant at 22 plants/m^2 in August and September 2016 before decreasing to 7 plants/m^2 in November 2016, then increasing to 17,0 plants/m2 in March 2017 and then decreasing again to 6 plants/m^2 in May 2017 (Figure 4.41). The density of *E. tef*

was 1 plant/m^2 in August 2016 and increased to 2 plants/m^2 in September 2016, but disappeared November 2016 (Figure 4.41). The density for *C. dactylon* was initially 14 plants/m^2 in August 2016, after which the density increased to 22 plants/m^2 in November 2016 before decreasing again to 6 plants/m^2 in May 2017 (Figure 4.41 and Table 4.32).

M. sativa had an initial density of 5 plants/m^2 in August 2016, which decreased to 3 plants/m^2 in May 2016 (Figure 4.41). *D. eriantha* had an initial density of 7 plants/m^2 in August 2016, after which the density decreased in November 2016 (1 plants/m^2) before increasing towards May 2017 to 6 plants/m^2 (Figure 4.41 and Table 4.32). The density of *E. curvula* and *C. dactylon* was high throughout the trial (Figure 4.41 and Table 4.32).

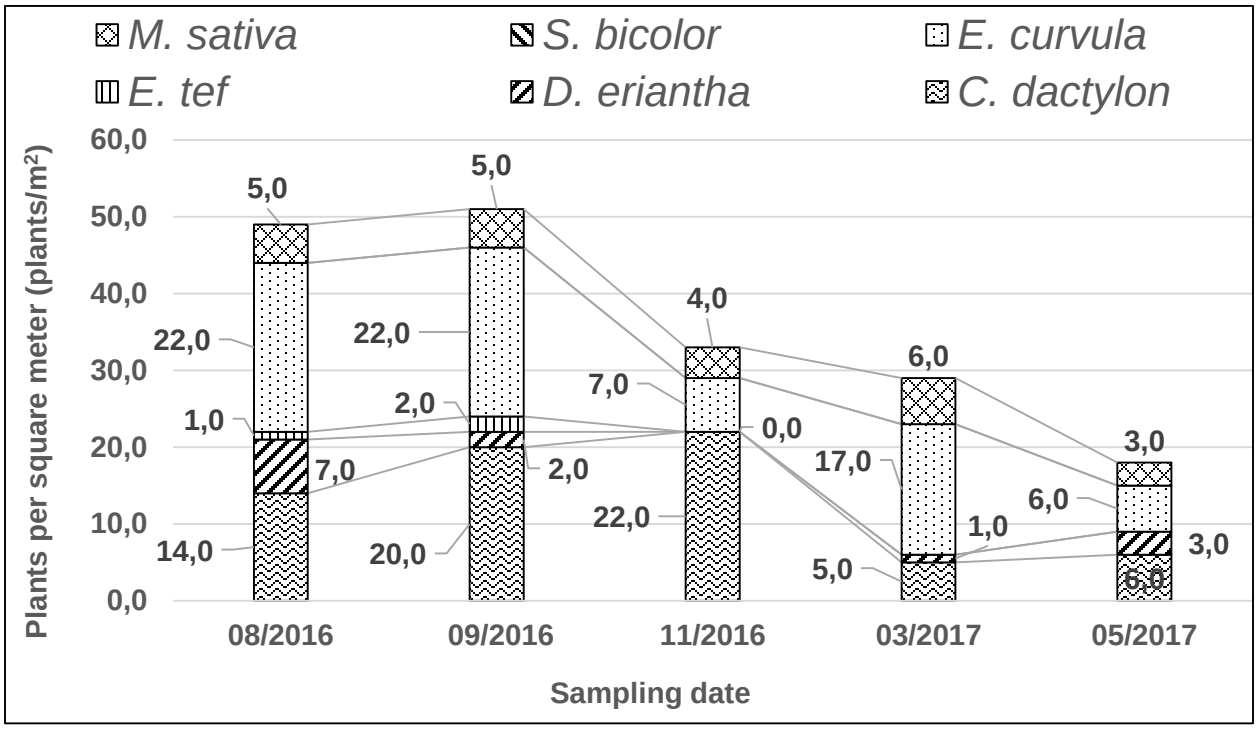

Figure 4.41: Change in plant density (plants/m^2) of species used (*C. dactylon, D. eriantha, E. curvula, E. tef, S. bicolor* and *M. sativa*) in T3C in the platinum trials from August 2016 to May 2017.

E. curvula and *C. dactylon* were the dominant species in T3C at any giver time throughout the trial period when compared to *D. eriantha*, *M. sativa* and *E. tef*. *E. curvula* comprised 44,9% of the plant community initialy in August 2016, it reached a minimum of 21,2% in Novemeber 2016 and a maximum of 58,6% in March 2017 before decreasing to 33,3% in may 2017 (Figure 4.42 and Table 4.32). *C. dactylon* initialy comprised 28,6% of the plant composition in August 2016, it increased through September to reach a maximum of 66,7% in November 2016 before

decreasing to a minimum contribution to the plant community of 17,2% in March 2017 and increasing to 33,3% of the plant community in May 2017 (Figure 4.42 and Table 4.32).

M. sativa contribution to the plant community remained constant at 10,2% and 9,8% in August and September 2016, this increased to 12,1% in November 2016 and reached a maximum of 20,7% in March 2017 before decreasing to 16,7% in May2017 (Figure 4.42 and Table 4.32).

S. bicolor was not once present in T3C during the trial and *E. tef* only comprised 2% of the plant community in August 2016 and 3,9% in September 2016 before dying off and no longer being present (Figure 4.42 and Table 4.32).

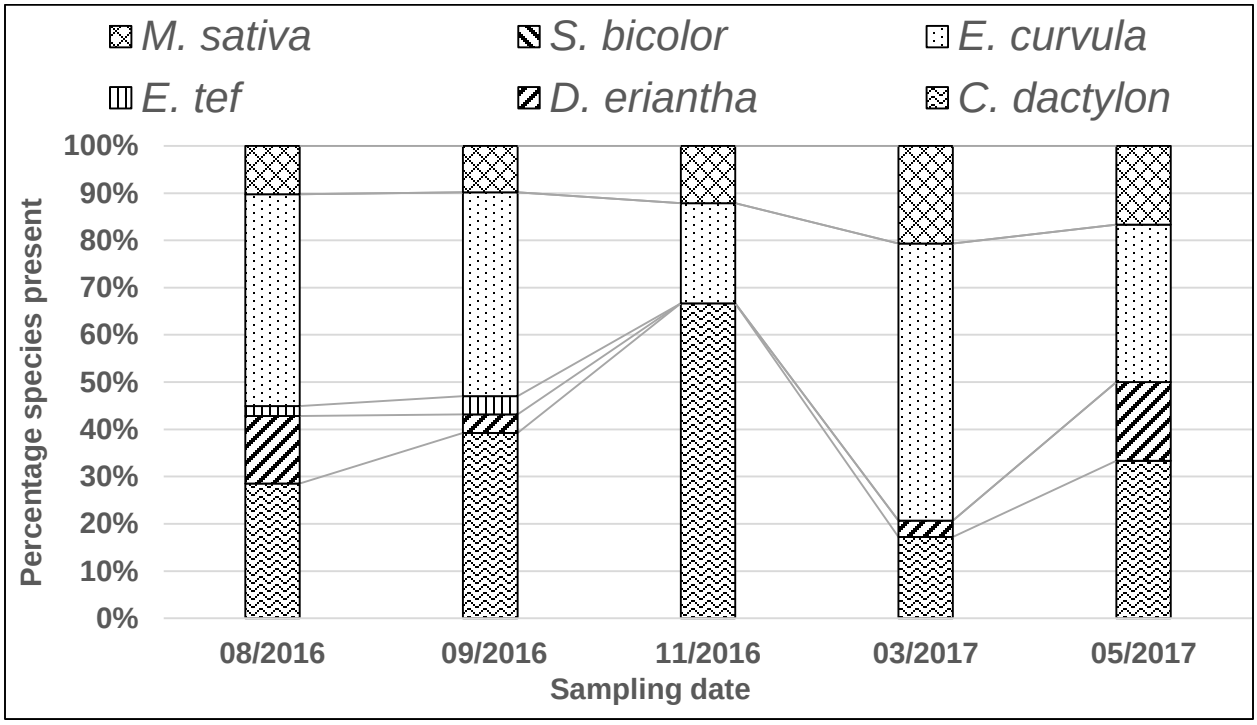

Figure 4.42: Change in plant composition of species used (*C. dactylon, D. eriantha, E. curvula, S. bicolor* and *M. sativa*) in T3C in the platinum trials from August 2016 to May 2017.

Table 4.32: Summary of plant density (plants/m^2) and plant composition (%) results for *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor* and *M. sativa* in T3C in the platinum trials from August 2016 to May 2017.

T3C	08/2016		09/2016		11/2016		03/2017		05/2017	
Species	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**
C. dactylon	14,0	28,6	20,0	39,2	22,0	66,7	5,0	17,2	6,0	33,3
D. eriantha	7,0	14,3	2,0	3,9	0,0	0,0	1,0	3,4	3,0	16,7
E. tef	1,0	2,0	2,0	3,9	0,0	0,0	0,0	0,0	0,0	0,0
E. curvula	22,0	44,9	22,0	43,1	7,0	21,2	17,0	58,6	6,0	33,3
S. bicolor	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0
M. sativa	5,0	10,2	5,0	9,8	4,0	12,1	6,0	20,7	3,0	16,7
Total	**49,0**	**100,0**	**51,0**	**100,0**	**33,0**	**100,0**	**29,0**	**100,0**	**18,0**	**100,0**

The species density and plant composition results of T4C are illustrated in Figures 4.43, 4.44 and Table 4.33. T4C had a total plant density of 34 plants/m^2 in August 2016, which increased to 55 plants/m^2 in September 2016 and then decreased again to 22 plants/m^2 in May 2017 (Figure 4.43 and Table 4.33).

The density of *E. curvula* remained constant in August 2016 (15 plants/m^2) and September 2016 (14 plants/m^2) before increasing in November 2016 to 28 plants/m^2 and afterwards decreasing towards May 2017 to 8 plants/m^2 (Figure 4.43 and Table 4.33). The density of *C. dactylon* increased from 12 plants/m^2 in August 2016 to 33 plants/m^2 in September 2016, before decreasing to 6 plants/m^2 in November 2016 and doubling to 13 plants/m^2 in March 2017. The density of *C. dactylon* however decreased to 8 plants/m^2 in May 2017 (Figure 4.43). As for the other trials, the density of *E. tef*, decreased until this grass was totally absent in March 2017 (Figure 4.43). The density of *D. eriantha* changed from 1 plant/m^2 in November 2016 to 3 plants/m^2 in March 2017 before decreasing to 2 plants/m^2 in May 2017. As for the other trials, the low abundance of *D. eriantha* can be ascribed to poor seed with low viability.

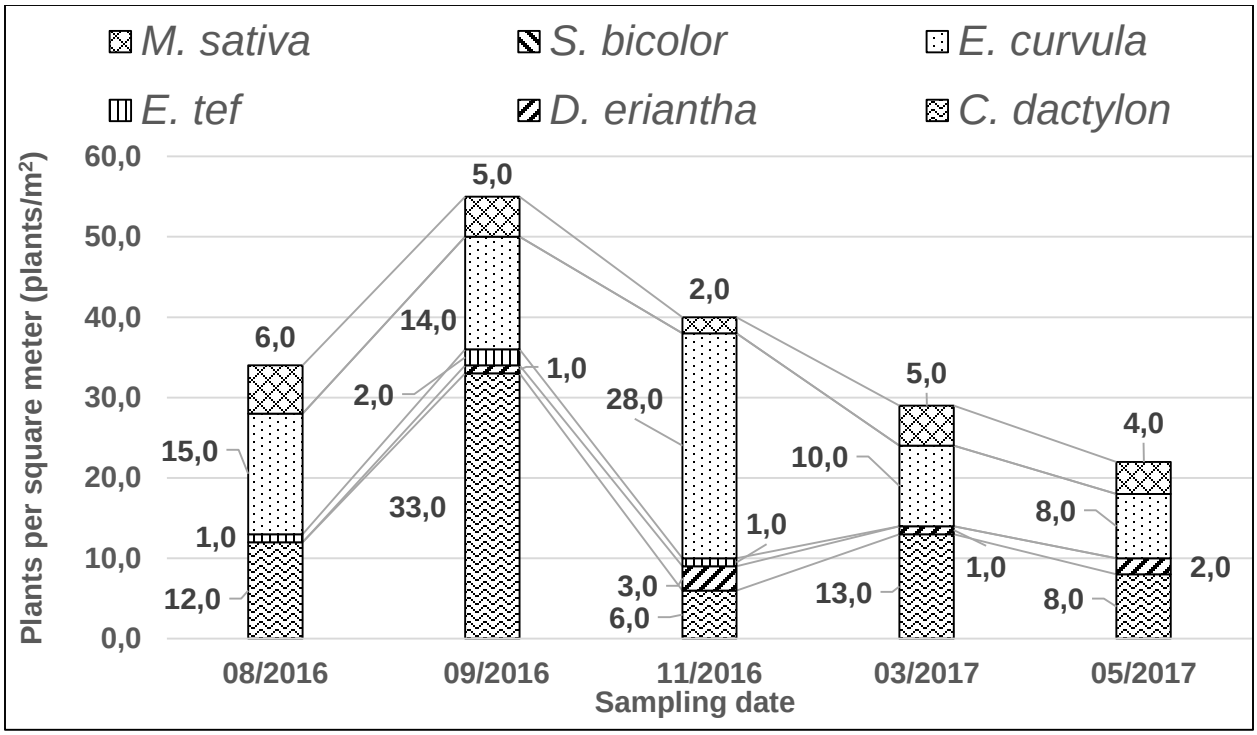

Figure 4.43: Change in plant density (plants/m^2) of species used (*C. dactylon, D. eriantha, E. curvula, E. tef, S. bicolor and M. sativa*) in T4C in the platinum trials from August 2016 to May 2017.

The plant composition was primarily dominated by *E. curvula* grass and *C. dactylon* throughout the trial period with *E. curvula* reaching a maximum of 70% of the plant composition in November 2016 and *C. dactylon* reaching a maximum of 60% in September 2016 (Figure 4.44 and Table 4.33). *D. eriantha* was not present in August 2016 and its contribution to the plant community fluctuated, increasing to 1,8% in September 2016 increasing to 7,5% in November 2016 before decreasing to 3,4% in March 2017 and increasing again to a maximum of 9,1% in May 2017 (Figure 4.44 and Table 4.33).

M. sativa initialy comprised 17,6% of the plant composition in August 2016, it reached a minimum of 5% in November 2016 and a maximum of 18,2% in May 2017 at the end of the trial period (Figure 4.44 and Table 4.33).

S. bicolor failed to emerge and *E. tef* did emerge but only comprised between 2,5% and 3,6% of the plant community from August 2016 to November 2016 before dying off (Figure 4.44 and Table 4.33).

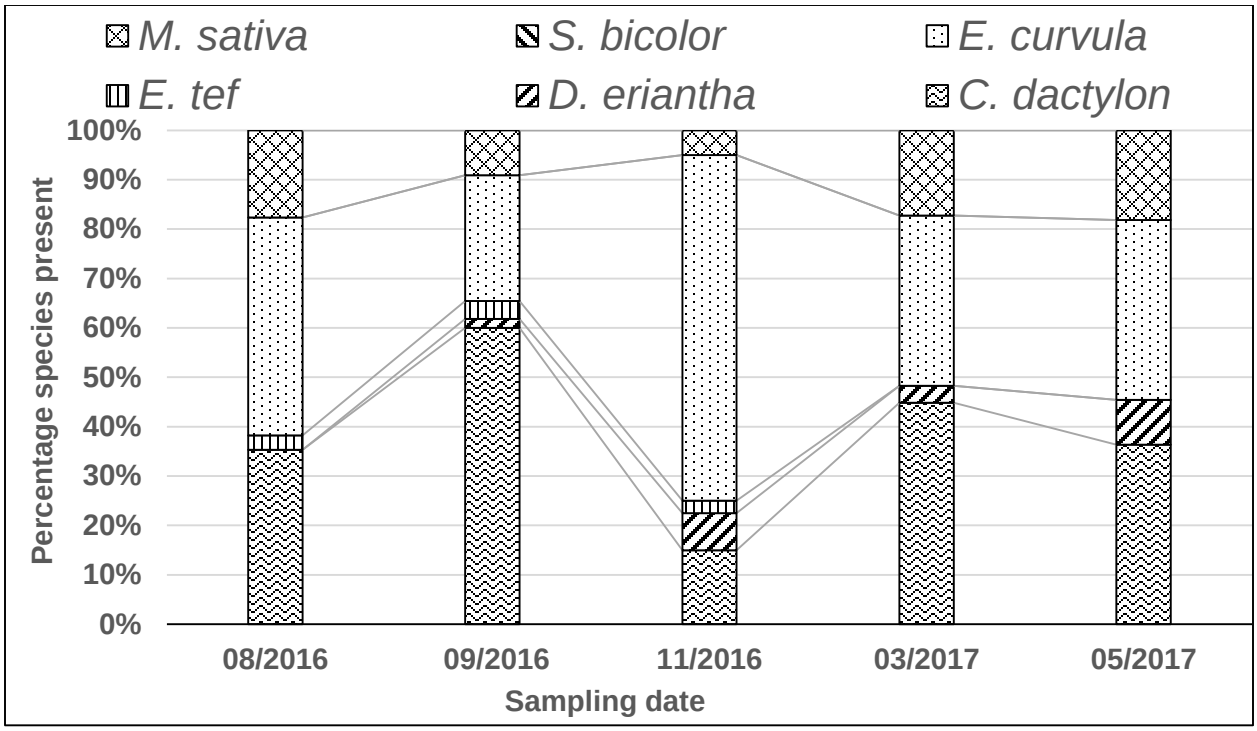

Figure 4.44: Change in plant composition of species used (*C. dactylon, D. eriantha, E. curvula, E. tef, S. bicolor* and *M. sativa*) in T4C in the platinum trials from August 2016 to May 2017.

Table 4.33: Summary of plant density (plants/m^2) and plant composition (%) results for *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor* and *M. sativa* in T4C in the platinum trials from August 2016 to May 2017.

T4C	08/2016		09/2016		11/2016		03/2017		05/2017	
Species	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**
C. dactylon	12,0	35,3	33,0	60,0	6,0	15,0	13,0	44,8	8,0	36,4
D. eriantha	0,0	0,0	1,0	1,8	3,0	7,5	1,0	3,4	2,0	9,1
E. tef	1,0	2,9	2,0	3,6	1,0	2,5	0,0	0,0	0,0	0,0
E. curvula	15,0	44,1	14,0	25,5	28,0	70,0	10,0	34,5	8,0	36,4
S. bicolor	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0
M. sativa	6,0	17,6	5,0	9,1	2,0	5,0	5,0	17,2	4,0	18,2
Total	**34,0**	**100,0**	**55,0**	**100,0**	**40,0**	**100,0**	**29,0**	**100,0**	**22,0**	**100,0**

The species density and plant composition results of T5UC are illustrated in Figure 4.45, 4.46 and Table 4.34. The density of T5UC was initially 62 plants/m^2 in August 2016, which decreased to 40 plants/m^2 in November 2016 and continued to decrease to 22 plants/m^2 in May 2017 (Figure 4.45).

The destiny of *E. curvula* was 42 plants/m^2 in August 2016, which then decreased to 39 plants/m^2 in September 2016 and continued to decrease until its lowest density of 11 plants/m^2 in May 2017 (Figure 4.45). The density of *C. dactylon* was initially 12 plants/m^2 August 2016, which then

increased to 14 plants/m^2 in September 2016, before decreasing to 8 plants/m^2 in March 2017 and May 2017 (Figure 4.45).

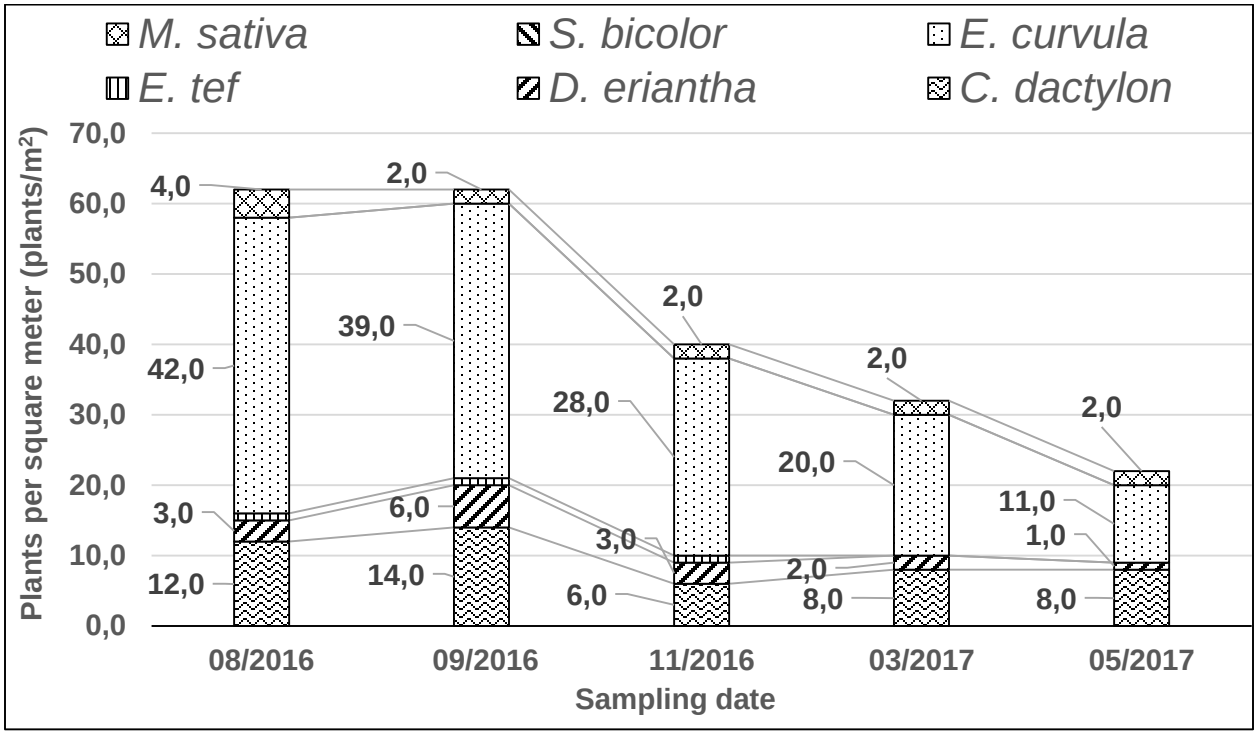

Figure 4.45: Change in plant density (plants/m^2) of species used (*C. dactylon, D. eriantha, E. curvula, E. tef, S. bicolor, M. sativa*) in T5UC in the platinum trials from August 2016 to May 2017.

The plant composition of treatment T5UC remained quite stable throughout the trial period with *E. curvula* as the dominant species (70–50%), followed by *C. dactylon* grass (19,4–36,4%) and *D. eriantha* (10–4,5%) (Figure 4.46).

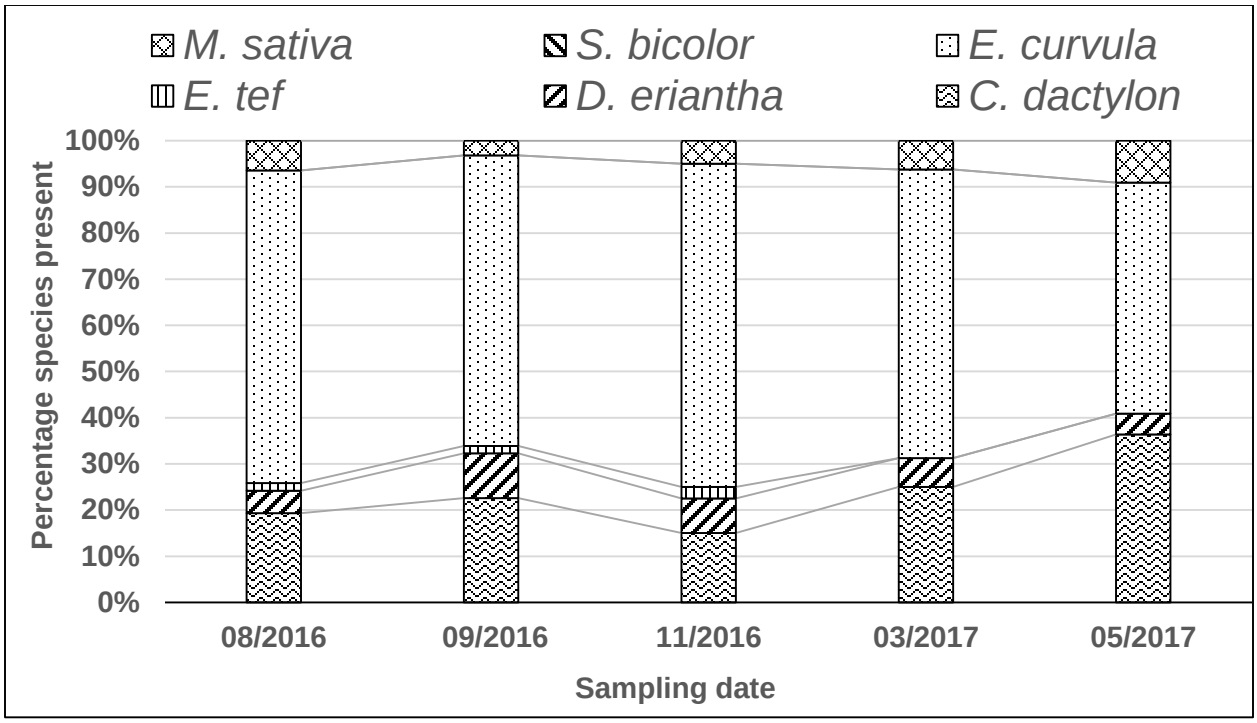

Figure 4.46: Change in plant composition of species used (*C. dactylon, D. eriantha, E. curvula, E. tef, S. bicolor, M. sativa*) in T5UC in the platinum trials from August 2016 to May 2017.

Table 4.34: Summary of plant density (plants/m^2) and plant composition (%) results for *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor* and *M. sativa* T5UC in the platinum trials from August 2016 to May 2017.

T5UC	08/2016		09/2016		11/2016		03/2017		05/2017	
Species	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**	**plants/m²**	**%**
C. dactylon	12,0	19,4	14,0	22,6	6,0	15,0	8,0	25,0	8,0	36,4
D. eriantha	3,0	4,8	6,0	9,7	3,0	7,5	2,0	6,3	1,0	4,5
E. tef	1,0	1,6	1,0	1,6	1,0	2,5	0,0	0,0	0,0	0,0
E. curvula	42,0	67,7	39,0	62,9	28,0	70,0	20,0	62,5	11,0	50,0
S. bicolor	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0
M. sativa	4,0	6,5	2,0	3,2	2,0	5,0	2,0	6,3	2,0	9,1
Total	**62,0**	**100,0**	**62,0**	**100,0**	**40,0**	**100,0**	**32,0**	**100,0**	**22,0**	**100,0**

To summarise the results for the platinum trials: The total plant densities of the treatments tended to reach a maximum in September 2016 before readily decreasing towards May 2017. The density of *E. tef* was minimal in August 2016, after which it died off in November 2016 (Figure 4.46). According to Baskin & Baskin (1998:40) and O'Connor & Everson (1998:349), the low emergence densities of *E. tef* may be attributed to the conditional dormancy brought forth by decreasing temperatures in April 2016 and June 2016 (see Section 3.2 Figure 3.9). O'Connor & Everson (1998:349) explain that in temperate regions seeds of summer annual grasses such as *E. tef* come out of dormancy during winter and enter a conditional dormancy phase during which they

only germinate at high summer temperatures. Baskin & Baskin (1998:41) continue to explain that during the conditional dormancy phase seeds of summer annuals are unlikely to germinate in late autumn months because temperatures are below those required for germination.

E. curvula and *C. dactylon* were the dominant species in each of the five seed treatments of which the density of *E. curvula* generally decreased throughout the trial period. *S. bicolor* did not establish well in any of the treatments and *M. sativa* remained present at low densities throughout the trial period. The density of *C. dactylon* tended to increase in September 2016 for all the treatments.

The main influence on the species emergence and the change in plant composition was the extensive growth of *M. sativa* in the limited growth space (1 m^2) provided by the bulk bags. De Kock (2012) explains that the growth of *M. sativa* is not limited as much by temperature as it is limited by moisture, requiring large quantities of water for optimal production, in the range of 750–800 kg per kg DM produced. He continues to add that for summer rainfall areas, the annual rainfall must be higher than 400–500 mm for the successful cultivation of *M. sativa*. In a study conducted by Taylor and Marble (1986: Abstract) on the influence of irrigation during bloom, Taylor and Marble found that irrigation frequency correlated positively with an increase in seed yield. *M. sativa* is also moderately frost tolerant, it has an erect growth habit, growing up to 50–70 cm with many stems originating from the crown and it grows rapidly after summer rainfall events exceeding 10 mm (Dolling, 2017).

The high growth rate of *M. sativa* was therefore primarily caused by the irrigation of the plots, as well as the protection from frost, as these trials were carried out at the nursery for soil and plant research for mine rehabilitation in Potchefstroom. The growth of *C. dactylon* was mostly caused by the growth of the rhizomes and stolons that grow either above or below the soil surface.

The *M. sativa* was cut on 8 November 2016 to open up the trial area for the other species to establish and grow due to the better light conditions. This resulted in the increase density of *E. curvula* for T4C, on 23 November 2016. The rapid regrowth of *M. staiva* and competition between individual plants within the limited growth space mainly caused the decrease of the plant density of the other species present in the seed mixtures.

4.3.6 Platinum trials total cover contribution of species

The vegetation cover results for seed treatments in the platinum trials are illustrated in Figure 4.47 to Figure 4.51 and Table 4.35 to Table 4.39 The vegetation cover in treatment T1C from August

2016 until May 2017 is given in Figure 4.47 and Table 4.32. Initially, the total vegetation cover was 17,3% in August 2016, which increased to 83,1% in September 2016 with *M. sativa* contributing 54,2% of the vegetation cover and *E. curvula* 21,3% (Figure 4.47). In November 2016, the vegetation cover of *M. sativa* decreased to 31,7% before increasing to 40,1% in May 2017 (Figure 4.47).

The coverage of *E. curvula* decreased to 36% in March 2017 from 52.8% in November 2016 before increasing to 47,1% in May 2017 (Figure 4.47). The contributions of *C. dactylon* was 15,5% in November 2016; this increased to 23,1% in March 2017 and decreased to 12,7% in May 2017 (Figure 4.47 and Table 4.35.

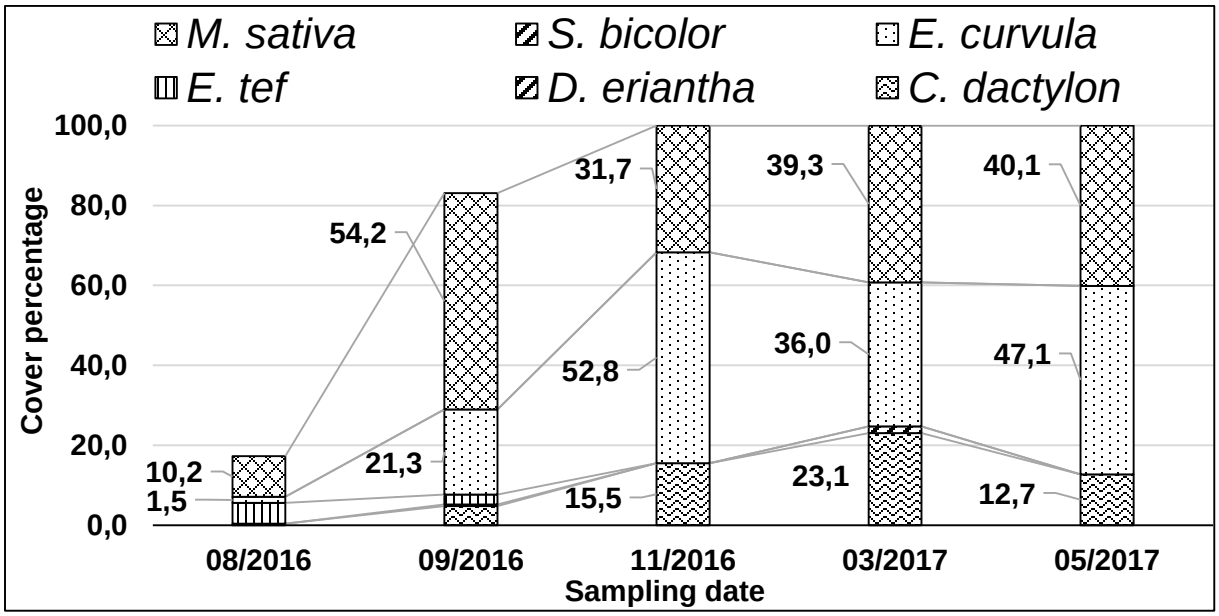

Figure 4.47: Change in total cover contribution of *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor* and *M. sativa* in T1C in the platinum trials from August 2016 to May 2017.

Table 4.35: Change in total cover contribution of *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor and M. sativa* in T1C in the platinum trials from August 2016 to May 2017 shown in Figure 4.47.

T1C	08/2016	09/2016	11/2016	03/2017	05/2017
Species	**%**	**%**	**%**	**%**	**%**
C. dactylon	0,4	4,8	15,5	23,1	12,7
D. eriantha	0,0	0,4	0,0	1,6	0,0
E. tef	5,2	2,5	0,0	0,0	0,0
E. curvula	1,5	21,3	52,8	36,0	47,1
S. bicolor	0,0	0,0	0,0	0,0	0,0
M. sativa	10,2	54,2	31,7	39,3	40,1
Total	**17,3**	**83,1**	**100,0**	**100,0**	**100,0**

The vegetation cover for T2UC is illustrated in Figure 4.48 and Table 4.36 Initially, the combined vegetation cover of the species was 15,2% in August 2016; this increased to 66,9% in September 2016, with *E. curvula* providing 27,5%, *C. dactylon* providing 21,5% and *M. sativa* providing 16,3% cover, these were the main species contributing plant cover.

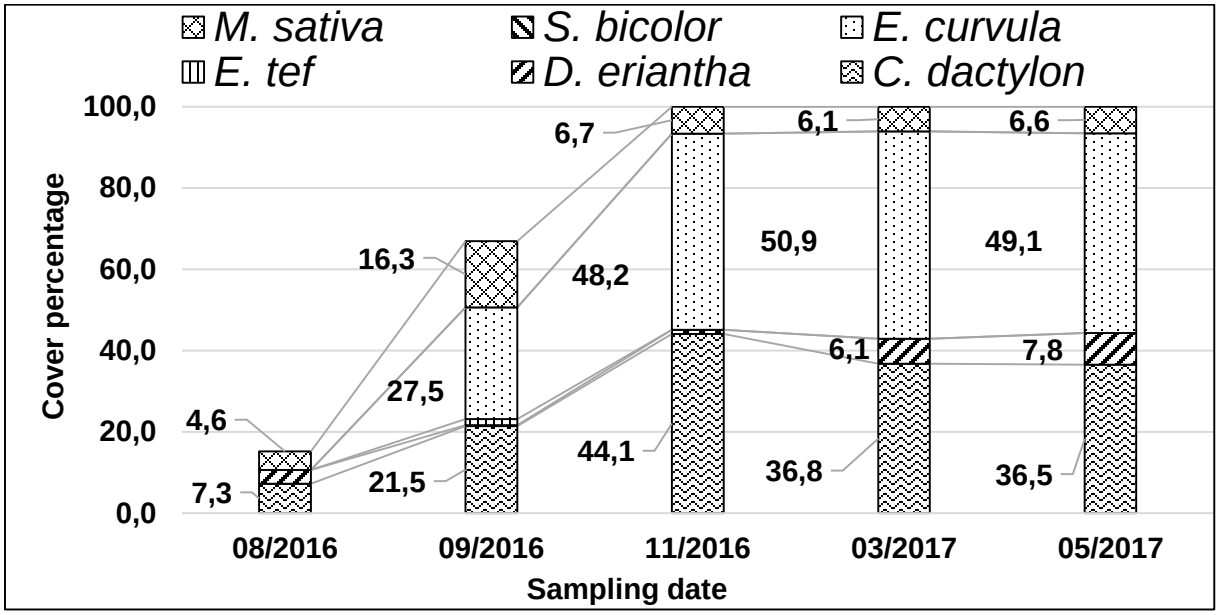

Figure 4.48: Change in total cover contribution of *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor* and *M. sativa* in T2UC in the platinum trials from August 2016 to May 2017.

In November 2016, the cover of *E. curvula* (48,2%) and *C. dactylon* (44,1%) increased, while the cover for *M. sativa* decreased (6,7%). The cover contribution of *E. curvula* increased (50,9%), leading up to March 2017 before decreasing to 49,1% in May 2017. *C. dactylon* decreased from its maximum coverage of 44,1% in November 2016 to 36,8% in March 2017 and 36.5% in May 2017. *M. sativa* remained constant between 6,7–6,6% from November 2016 to May 2017 (Figure 4.48 and Table 4.36).

In November 2016, *D. eriantha* provided some cover (1%); this increased to 6,1% in March 2017 and remained constant until May 2017.

Table 4.36: Change in total cover contribution of *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor and M. sativa* and n T2UC in the platinum trials from August 2016 to May 2017 shown in Figure 4.48.

T2UC	08/2016	09/2016	11/2016	03/2017	05/2017
Species	**%**	**%**	**%**	**%**	**%**
C. dactylon	7,3	21,5	44,1	36,8	36,5
D. eriantha	3,3	0,2	1,0	6,1	7,8
E. tef	0,0	1,5	0,0	0,0	0,0
E. curvula	0,0	27,5	48,2	50,9	49,1
S. bicolor	0,0	0,0	0,0	0,0	0,0
M. sativa	4,6	16,3	6,7	6,1	6,6
Total	**15,2**	**66,9**	**100,0**	**100,0**	**100,0**

In (T3C, *M. sativa* contributed the most cover in August 2016 (24,8%) and September 2016 (52,3%). This decreased in November 2016 (40,7%) and continued to decrease towards May 2017 (32,7%) at the end of the trial.

The coverage of *E. curvula* increased throughout the trial period to a maximum of 40% coverage in November 2017. This decreased to 30,3% in March 2017 before increasing to 39% in May 2017. *C. dactylon* increased from 4,6% coverage in September 2016, to a maximum of 22,5% in March 2017 and decreased to 12,9%% in May 2017. *D. eriantha* increased from 2,2% in November to 11,7% in March 2017 before reaching a maximum of 15,2% cover in May 2017 (Figure 4.49 and Table 4.37).

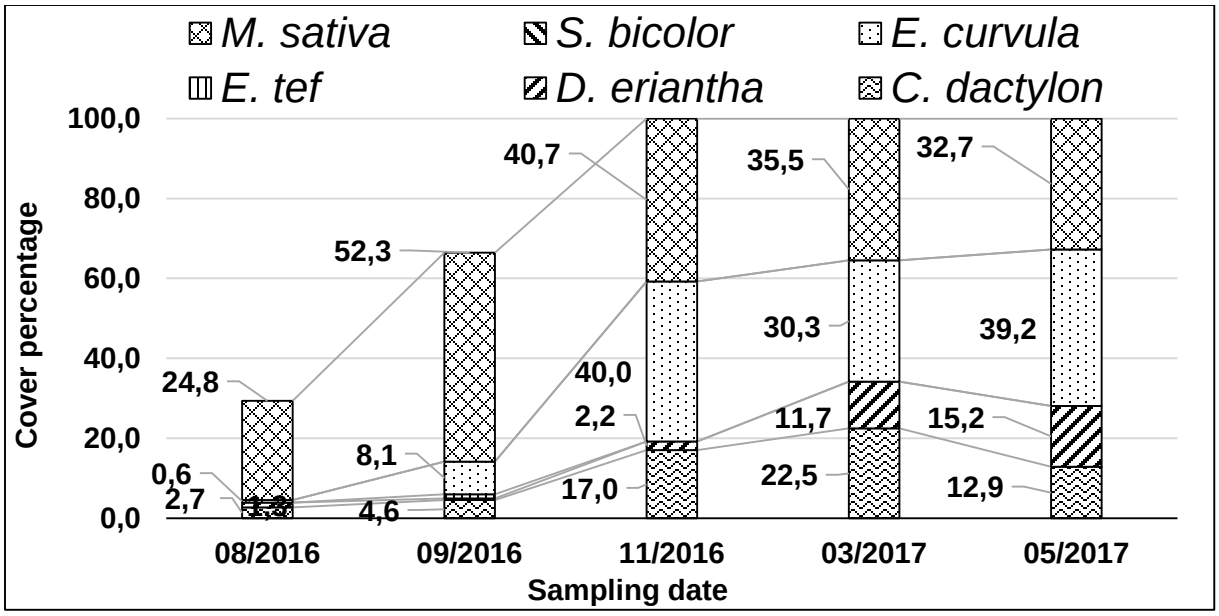

Figure 4.49: Change in total cover contribution of *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor* and *M. sativa* in T3C in the platinum trials from August 2016 to May 2017.

Table 4.37: Change in total cover contribution of *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor and M. sativa* in T3C in the platinum trials from August 2016 to May 2017 shown in Figure 4.49.

T3C	08/2016	09/2016	11/2016	03/2017	05/2017
Species	**%**	**%**	**%**	**%**	**%**
C. dactylon	2,7	4,6	17,0	22,5	12,9
D. eriantha	1,3	0,4	2,2	11,7	15,2
E. tef	0,0	1,0	0,0	0,0	0,0
E. curvula	0,6	8,1	40,0	30,3	39,2
S. bicolor	0,0	0,0	0,0	0,0	0,0
M. sativa	24,8	52,3	40,7	35,5	32,7
Total	**29,4**	**66,5**	**100,0**	**100,0**	**100,0**

In T4C *M. sativa* provided the most cover in August (26,9%) and September 2016 (73,3%); it then decreased to 37,7% in November 2016 when it was cut, before increasing to 51,3% in March 2017 and again decreasing to 35,2% at the end of the trials in May 2017 (Figure 4.50 and Table 4.38). *E. curvula* did not contribute any vegetation cover in August 2016; in September 2016 it provided 14,6% cover. This increased to 34,9% in November 2016 before decreasing to 27,8% in March, after which it increased again, reaching a maximum of 40,9% in May 2017 (Figure 4.50 and Table 4.38). *C. dactylon* contributed 0,4% cover in August 2016; this increased to a maximum of 27,4% in November 2016 before decreasing to 19,8% in May 2017 (Figure 4.50 and Table 4.38).

E. tef contributed 8,8% vegetation cover in August 2016, before decreasing to 5,1% in September 2016 and no longer provided any cover throughout the trial period. *D. eriantha* only contributed 0,5% in September 2016, which increased to 4% in May 2017 (Figure 4.50 and Table 4.38).

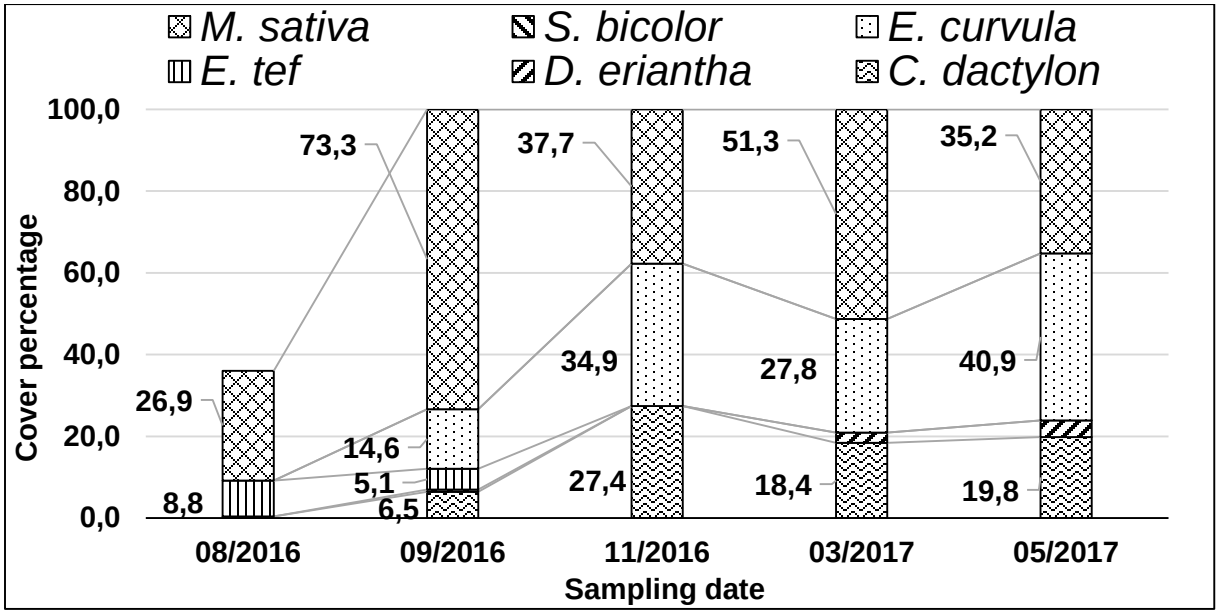

Figure 4.50: Change in total cover contribution of *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor* and *M. sativa* in T4C in the platinum trials from August 2016 to May 2017.

Table 4.38: Change in total cover contribution of *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor and M. sativa* in T4) in the platinum trials from August 2016 to May 2017 shown in Figure 4.50.

T4C	08/2016	09/2016	11/2016	03/2017	05/2017
Species	**%**	**%**	**%**	**%**	**%**
C. dactylon	0,4	6,5	27,4	18,4	19,8
D. eriantha	0,0	0,5	0,0	2,6	4,0
E. tef	8,8	5,1	0,0	0,0	0,0
E. curvula	0,0	14,6	34,9	27,8	40,9
S. bicolor	0,0	0,0	0,0	0,0	0,0
M. sativa	26,9	73,3	37,7	51,3	35,2
Total	**36,0**	**100,0**	**100,0**	**100,0**	**100,0**

Seed treatment 5TUC, had *E. curvula* providing the largest cover percentage of all the surviving species, from 54% in September 2016, 67,4% in November 2016, 54,1% in March 2017 and 69,7% in May 2017 (Figure 4.51 and Table 4.39). Initially in August 2016, the total vegetation coverage was 37,5% principally provided by *M. sativa* that contributed 17,7% and *C. dactylon* contributing 18,5%. The total vegetation covergae increased to 89,6% in September 2016 with *E. curvula* providing 54% of the cover. The combined vegetation cover of the species then remained

constant at 100% coverage from November 2016 to May 2017 (Figure 4.51 and Table 4.39). *E. curvula* increased from September 2016 (54%) to November 2016 (67,4%) before decreasing towards March 2017 (54,1%), and ending at a maximum cover contribution of 69,7% in May 2017 (Figure 4.51 and Table 4.39).

C. dactylon decreased to 6,7% cover in September 2016, after which it then increased to provide a maximum 15,5% cover in March 2017 before decreasing to 6,2% in May 2017. *D. eriantha* contributed 2,2–2,8% coverage from November 2016 to May 2017 (Figure 4.51 and Table 4.39.

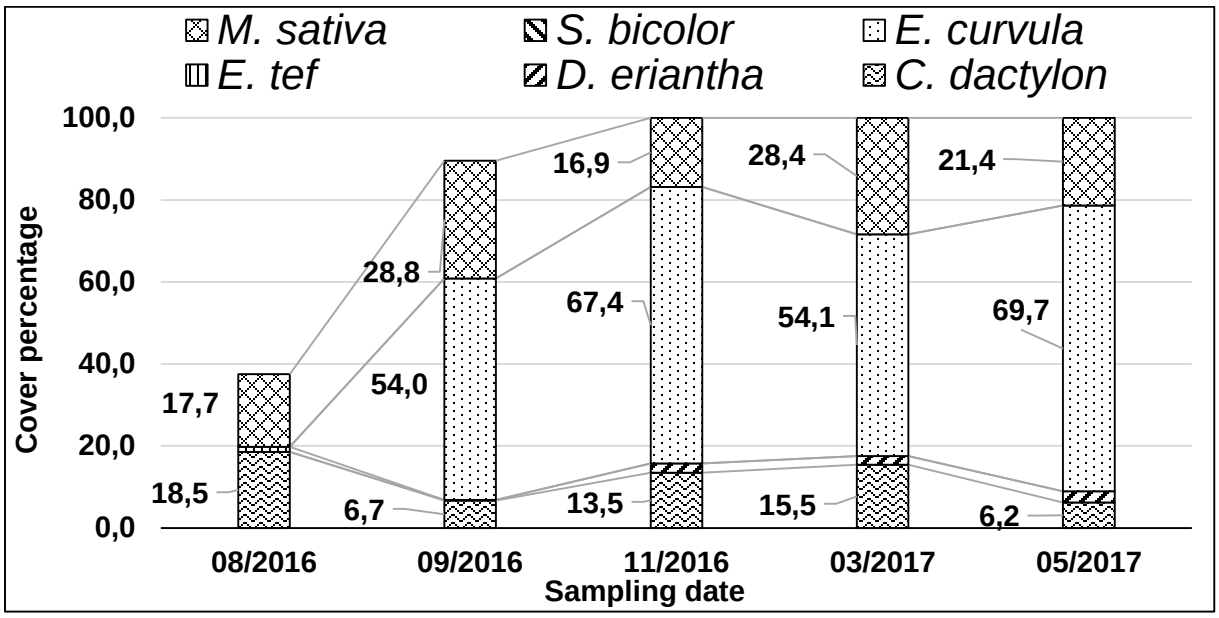

Figure 4.51: Change in total cover contribution of *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor* and *M. sativa* in T5UC in the platinum trials from August 2016 to May 2017.

Table 4.39: Change in total cover contribution of *C. dactylon, D. eriantha, E. tef, E. curvula, S. bicolor,* and *M. sativa* in T5UC in the platinum trials from August 2016 to May 2017 shown in Figure 4.51.

T5UC	08/2016	09/2016	11/2016	03/2017	05/2017
Species	**%**	**%**	**%**	**%**	**%**
C. dactylon	18,5	6,7	13,5	15,5	6,2
D. eriantha	0,0	0,2	2,2	2,1	2,8
E. tef	1,3	0,0	0,0	0,0	0,0
E. curvula	0,0	54,0	67,4	54,1	69,7
S. bicolor	0,0	0,0	0,0	0,0	0,0
M. sativa	17,7	28,8	16,9	28,4	21,4
Total	**37,5**	**89,6**	**100,0**	**100,0**	**100,0**

To summarise the cover contribution results for the treatments in the patinum trials: The main species contributing vegetation coverage throughout the trial period in the different seed

treatments were *M. sativa*, *E. curvula* and *C. dactylon*. Initially, in August 2016, the cover contribution of *E. curvula* was minimal, being dominated by *M. sativa*, and occasionally *C. dactylon*. The cover contribution of *E. curvula* then tended to increase in November 2016 (T1C, T2UC, T3C, T4C, T5UC), after *M. sativa* had been cut early in November 2016 to provide growth space for the other species (see Section 3.4.1; Figure 3.18 and 3.19). It is explained in the summary of Section 4.3.5 that the large growth form of *M. sativa* within the small space resulted in the suppression of *E. curvula* emergence. However, if the species was able to establish and grow to a size where it could not be supressed by the *M. sativa* it contributed a larger cover percentage even if the plant density was low. *D. eriantha* and *E. curvula* in Figure 4.47 to Figure 4.51 are examples of this. *C. dactylon* has the ability to creep underneath the *M. sativa* overburden plant material and establish in available growth space open to sunlight. This gave it an advantage over *E. curvula* to survive.

4.4 Phase 2 supporting pot trials vegetation results

The weekly emergence, monthly change in size (height and width index) and average DM (g) produced per pot for seedlings from coated and uncoated seed of *C. dactylon*, *D. eriantha*, *E. curvula* and uncoated *E. tef* seed within the growth mediums (Crown gold mine tailings, Rooikraal gold mine tailings, Platinum tailings, Control soil) are discussed in this Section.

4.4.1 Pot trial emergence results

The repeated measures ANOVA illustrating whether the growth medium or the seed type (coated and uncoated) demonstrated that there were statistically significant findings (p = 0,05) on the specific seedling emergence of *C. dactylon, D. eriantha*, *E. curvula* and *E. tef* is shown in Table 4.40.

It is indicated in Table 4.40 if the growth medium used during the trial or the seed type used to plant species had an effect on the emergence of the species. If the $p < 0,05$ the factor in question that is either the growth medium or the seed type had a significant influence on the seedling emergence of the species (Table 4.40).

Table 4.40: Repeated measures ANOVA results of weekly emergence for coated and uncoated seed of *C. dactylon, D. eriantha, E. curvula* and uncoated *E. tef* seed in the four growth mediums (RK – Rooikraal gold mine tailings; CRWN –Crown gold mine tailings; PT – Platinum; CTRL – Control soil). The influence of the growth medium and the seed type was significant if $p < 0,05$.

Species	Variable	*P*
C. dactylon	Growth medium	0,002
	Seed type	0,89
D. eriantha	Growth medium	0,0002
	Seedtype	0,01
E. curvula	Growth medium	0,235
	Seedtype	0,027
E. tef	Growth medium	0,902

The seedling emergence percentage of *C. dactylon* (Figure 4.52), *D. eriantha* (Figure 4.53), *E. curvula* (Figure 4.54) and *E. tef* (Figure 4.55) is shown below.

- ***Cynodon dactylon***

According to the repeated measure ANOVA (Table 4.40), the growth medium ($p = 0,048$; $F = 2,7$) did have an influence on the emergence of *C. dactylon* seedlings, but the seed type (coated and uncoated) did not ($p = 0,89$) (Figure 4.52).

In the first week following seeding (8 November 2016), the emergence of seedlings from both the coated and uncoated *C. dactylon* seed in each of the four growth mediums was low (< 10%) (Figure 4.52). In the platinum and the Rooikraal gold mine tailings, the seedling emergence of coated seed was higher than uncoated seed, whenever the seedlings were counted (11 November, 21 November and 28 November 2016).

The seedlings from uncoated *C. dactylon* seed in the Crown gold mine tailings had a higher seedling emergence percentage during the second and third weeks of the trial than seedlings from coated seed, but in the fourth week the seedlings from coated seed had a slightly higher emergence percentage than seedlings from uncoated seed (Figure 4.52). This could possibly indicate a delay in the seedling emergence of *C. dactylon* when using coated seed in the Crown gold mine tailings. In the control soil, the weekly seedling emergence from coated *C. dactylon* seed was lower than the seedling emergence from uncoated seed at each count (Figure 4.52).

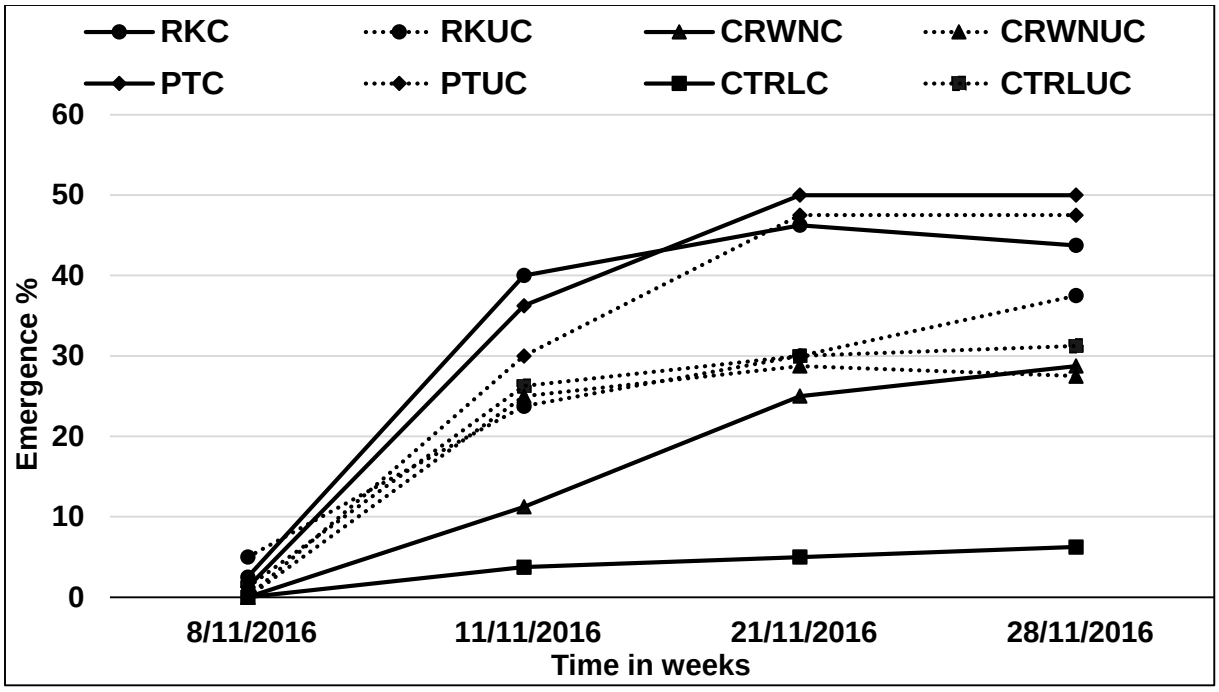

Figure 4.52: Weekly emergence of *C. dactylon* seedlings in November 2016 from coated and uncoated seed in the four growth mediums. (RKC –Rooikraal gold mine tailings coated seed; RKUC – Rooikraal gold mine tailings uncoated seed; CRWNC – Crown gold mine tailings coated seed; CRWNUC – Crown gold mine tailings uncoated seed; PTC – Platinum tailings coated seed; PTUC – Platinum tailings uncoated seed; CTRLC – Control coated seed; CTRLUC – Control uncoated seed).

- ***Digitaria eriantha***

According to the repeated measure ANOVA (Table 4.40), both the growth medium (p = 0,0002) and the seed type (coated and uncoated) (p = 0,01) had a significant influence on the weekly emergence of *D. eriantha* seedlings (Figure 4.53).

Seedlings from uncoated *D. eriantha* seed had a higher seedling emergence percentage than coated seed at every counted date in each of the growth mediums. The overall seedling emergence percentage was low (< 30%) for both the coated and uncoated seed (Figure 4.53). This could be attributed to the low viability of the seed of *D. eriantha* used in the trail (22% – Figure 3.11).

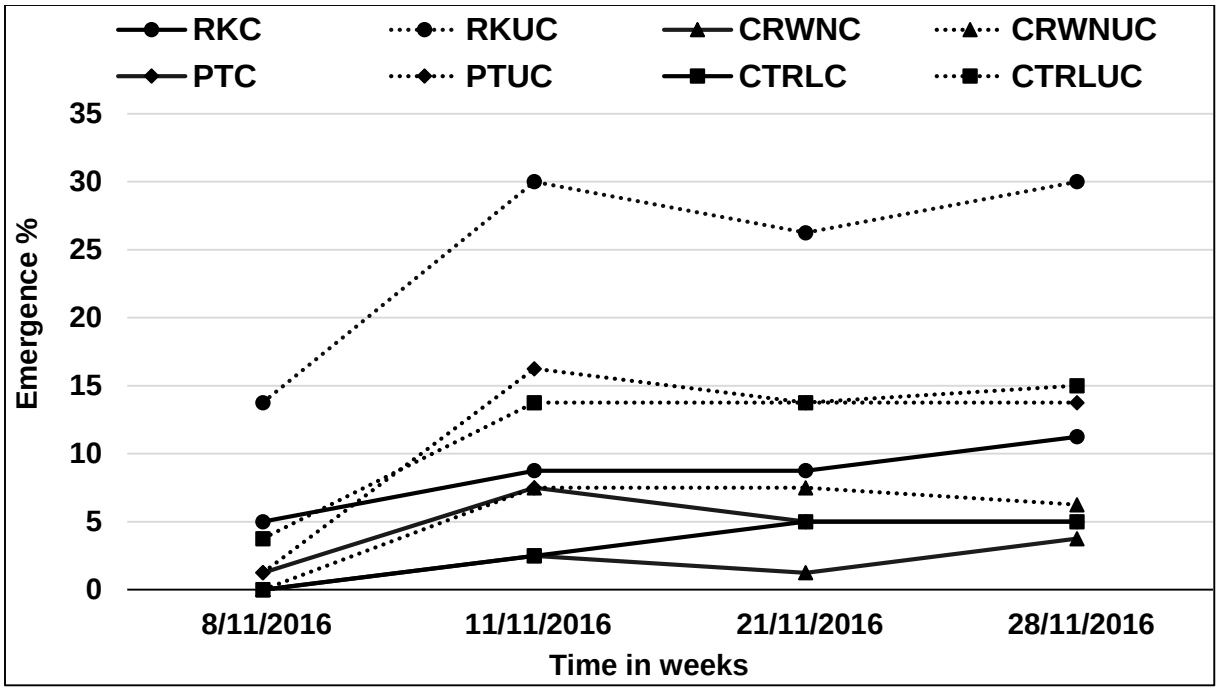

Figure 4.53: Weekly emergence of *D. eriantha* seedlings in November 2016 from coated and uncoated seed in the four growth mediums. (RKC – Rooikraal gold mine tailings coated seed; RKUC – Rooikraal gold mine tailings uncoated seed; CRWNC – Crown gold mine tailings coated seed; CRWNUC – Crown gold mine tailings uncoated seed; PTC – Platinum tailings coated seed; PTUC – Platinum tailings uncoated seed; CTRLC – Control coated seed; CTRLUC – Control uncoated seed).

- ***Eragrostis curvula***

According to the repeated measure ANOVA, the growth medium did not have a significant influence on the weekly emergence of *E. curvula* seedlings (p = 0,235), but the seed type (coated seed and uncoated seed) did (p = 0,027) (Figure 4.54 and Table 4.40).

One week after sowing (8 November 2016), a larger percentage of seedlings emerged from coated seed in the Rooikraal gold mine tailings, as well as the platinum tailings. The seedling emergence from uncoated seed was lower in the Rooikraal tailings, but there was not a significant difference between the emergence of *E. curvula* seedlings from coated and uncoated seed at the end of the trials on 28 November 2016 (Figure 4.54). The coated seed of *E. curvula* in the Crown gold mine tailings had the lowest seedling emergence throughout the trials in Figure 4.54. The seedling emergence from uncoated seed in the control soil was higher than coated seed throughout the trial (Figure 4.54).

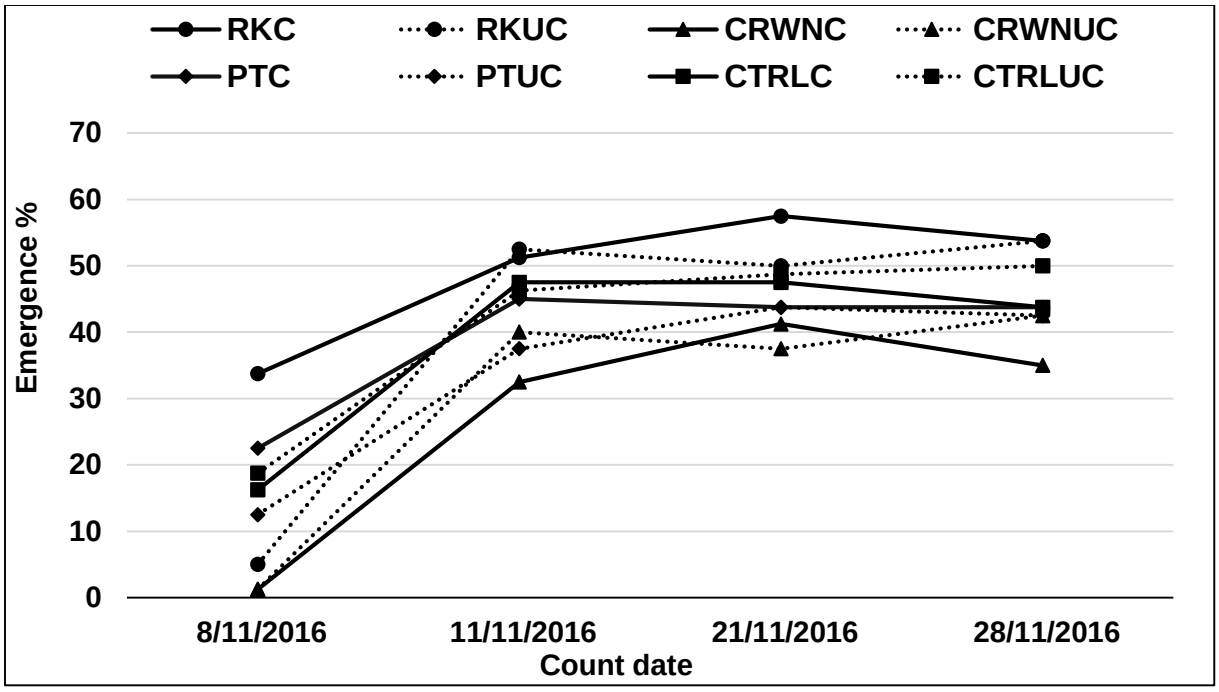

Figure 4.54: Weekly emergence of *D. eriantha* seedlings in November 2016 from coated and uncoated seed in the four growth mediums. (RKC – Rooikraal gold mine tailings coated seed; RKUC – Rooikraal gold mine tailings uncoated seed; CRWNC – Crown gold mine tailings coated seed; CRWNUC – Crown gold mine tailings uncoated seed; PTC – Platinum tailings coated seed; PTUC – Platinum tailings uncoated seed; CTRLC – Control coated seed; CTRLUC – Control uncoated seed).

- ***Eragrostis tef***

The repeated measure ANOVA revealed no significant influence (p = 0,902) of the growth mediums on the weekly emergence of *E. tef* seedlings (Figure 4.55 and Table 4.40).

E. tef had a uniform emergence in each of the growth mediums, with the seedling emergence remaining constant throughout the trial period (Figure 4.55). The highest emergence occurred in the Rooikraal gold mine tailings, while the lowest emergence occurred in the platinum tailings (Figure 4.55). This corresponded to the results obtained during Phase 1 trials (Section 4.3).

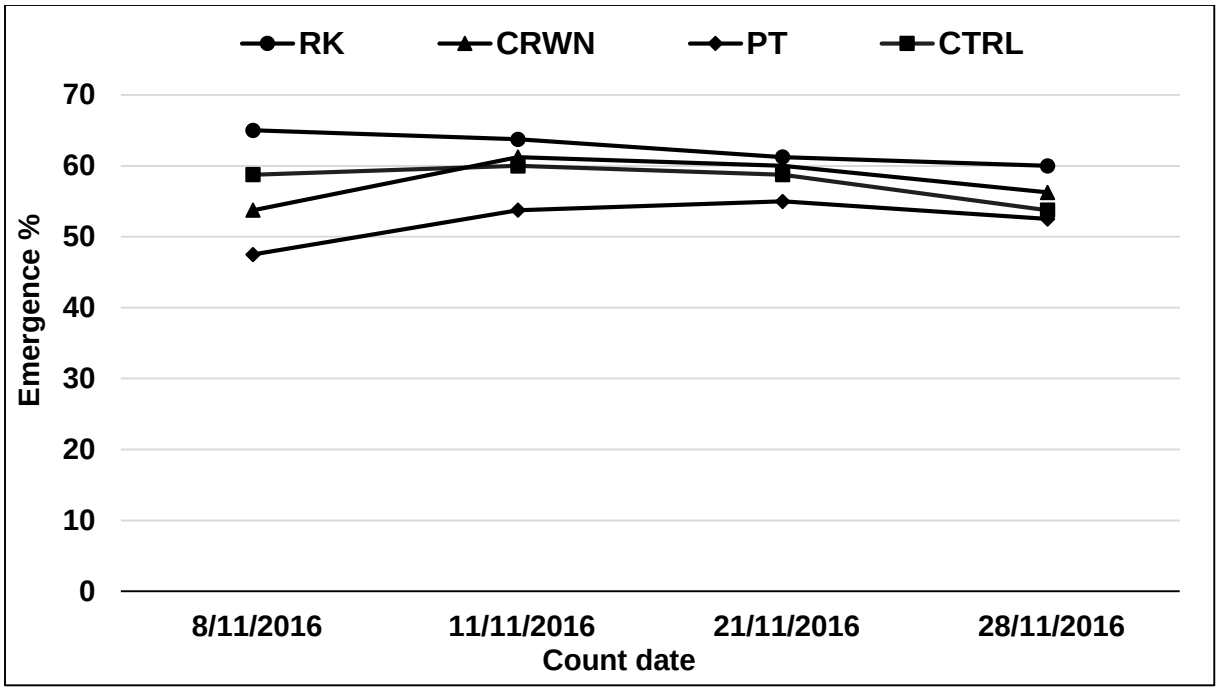

Figure 4.55: Weekly emergence of *E. tef* seedlings in four growth mediums. (RK – Rooikraal gold mine tailings; CRWN – Crown gold mine tailings; PT – Platinum tailings; CTRL – Control).

- **Summary**

To summarise the weekly emergence results obtained during the Phase 2 supporting pot trials.The daily temperature during the emergence period in November 2016 for the Potchefstroom area illustrated in Figure 3.9 of Section 3.2, averaged a minimum of 15,3°C and a maximum of 31,1°C. The aforementioned temperatures are above the cool daily average temperatures 15 to 19°C mentioned by to Evert *et al.* (2009:235) to restrict grass seed germination. The growth medium therefore did have a significant influence ($p < 0,05$) on the seedling emergence of *C. dactylon* and *D. eriantha* and the seed type (coated or uncoated) had a significant influence on the seedling emergence of *D. eriantha* and *E. curvula*.

D. eriantha seedlings had the lowest emergence percentage of the four species used in the pot trials species (< 20%) in each of the growth mediums using coated seed as well as uncoated seed, except in the Rooikraal gold mine tailings using uncoated seed (RKUC – 30%). The low emergence of *D. eriantha* seedlings corresponded with the low seedling densities obtained during the Phase 1 field trials (Section 4.3) and could be explained by the low seed viability of the seed batches used during the trials (22% presented in Figure 3.11 in Section 3.4.1).

The seedling emergence of *E. tef* was fast and uniform, with the largest portion of the seedlings emerging during the first week and at higher percentages than *C. dactylon*, *D. eriantha* and *E. curvula* within the same growth mediums.

4.4.2 Culm height and tuft width index

The repeated measures ANOVA illustrating whether the growth medium or the seed type (coated and uncoated) had a significant influence ($p < 0,05$) on the change in the height and width index of species is shown in Table 4.41.

It is indicated in Table 4.41 if the growth medium used during the trial or the seed type used to plant the species had an effect on the culm height and tuft width index of the species. If the $p < 0,05$ the factor in question that is either the growth medium or the seed type had a significant influence on the change of the culm height and tuft width index of emerged seedlings (Table 4.40).

Table 4.41: Repeated measures ANOVA results of height and width index for coated and uncoated seed of C. dactylon, D. eriantha, E. curvula and uncoated E. tef seed in the tailings growth mediums (RK – Rooikraal gold mine tailings; CRWN – Crown gold mine tailings; PT – Platinum tailings; CTRL – Control soil). The influence of the growth medium and the seed type was significant if p < 0,05.

Species	Variable	*P*
C. dactylon	Growth medium	0,048
	Seed type	0,026
D. eriantha	Growth medium	0,253
	Seedtype	0,14
E. curvula	Growth medium	0,037
	Seedtype	0,18
E. tef	Growth medium	0,33

The change in plant size (height and width index – see Section 3.4.2) of seedlings from coated and uncoated seed of *C. dactylon, D. eriantha*, *E. curvula* and uncoated seed of *E. tef* in the tailings and control soil over a period of three months is displayed in Figure 4.56 to 4.59 below.

- ***Cynodon dactylon***

According to the repeated measures ANOVA (Table 4.41), the growth medium ($p = 0,048$) and the seed type (coated and uncoated) ($p = 0,026$) had a significant influence on the height and width index of *C. dactylon.*

Plants grown from uncoated seed had a higher height and width index than coated seed in each of the four growth mediums throughout the trial (January to March 2017) (Figure 4.56).

Uncoated *C. dactylon* seed in the crown gold mine tailings provided the largest seedlings initially in January 2017 and at the end of the trial in March 2017. The lowest height and width index was observed for coated seed used in the control soil (Figure 4.56)

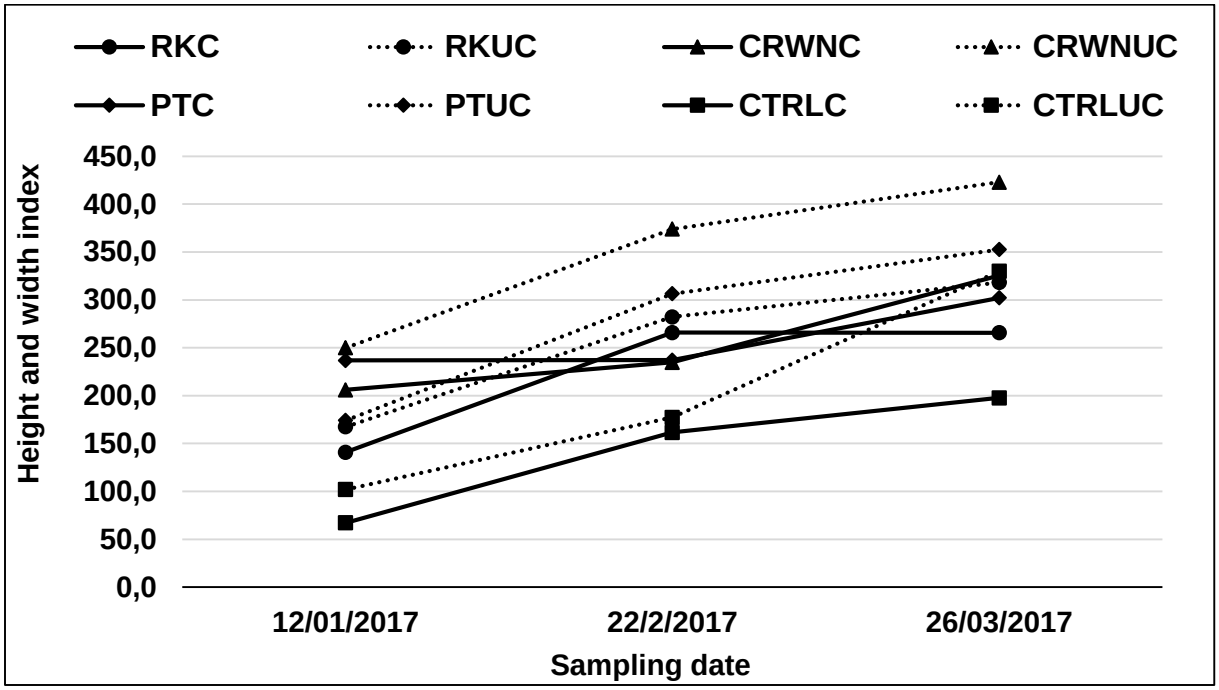

Figure 4.56: Change in grass height and width index of *C. dactylon* seedlings using coated and uncoated seed in the four growth mediums. (RKC – Rooikraal gold mine tailings coated seed; RKUC – Rooikraal gold mine tailings uncoated seed; CRWNC – Crown gold mine tailings coated seed; CRWNUC – Crown gold mine tailings uncoated seed; PTC – Platinum tailings coated seed; PTUC –– Platinum tailings uncoated seed; CTRLC – Control coated seed; CTRLUC – Control uncoated seed).

- ***Digitaria eriantha***

According to the repeated measure of the ANOVA (Table 4.41), neither the growth medium (p = 0,253) nor the seed type (coated and uncoated) (p = 0,14) had a significant influence on the growth of *D. eriantha* seedlings from January 2017 to March 2017.

In the first month, the height and width index of *D. eriantha* seedlings from coated and uncoated seed was higher in the platinum tailings compared to the Crown and Rooikraal gold mine tailings (Figure 4.57). Seedlings from uncoated seed of *D. eriantha* had a higher height and width index than seedlings from coated seed in the Rooikraal gold TSF, but the height and width index of seedlings from coated seed of this species was higher than seedlings from uncoated seed in the platinum tailings and control soil (Figure 4.57). In Figure 4.57 the Crown gold tailing, seedlings

from uncoated seed had a higher average height and width index in February 2017 before ending equal to the height and width index of seedlings grown from coated seed.

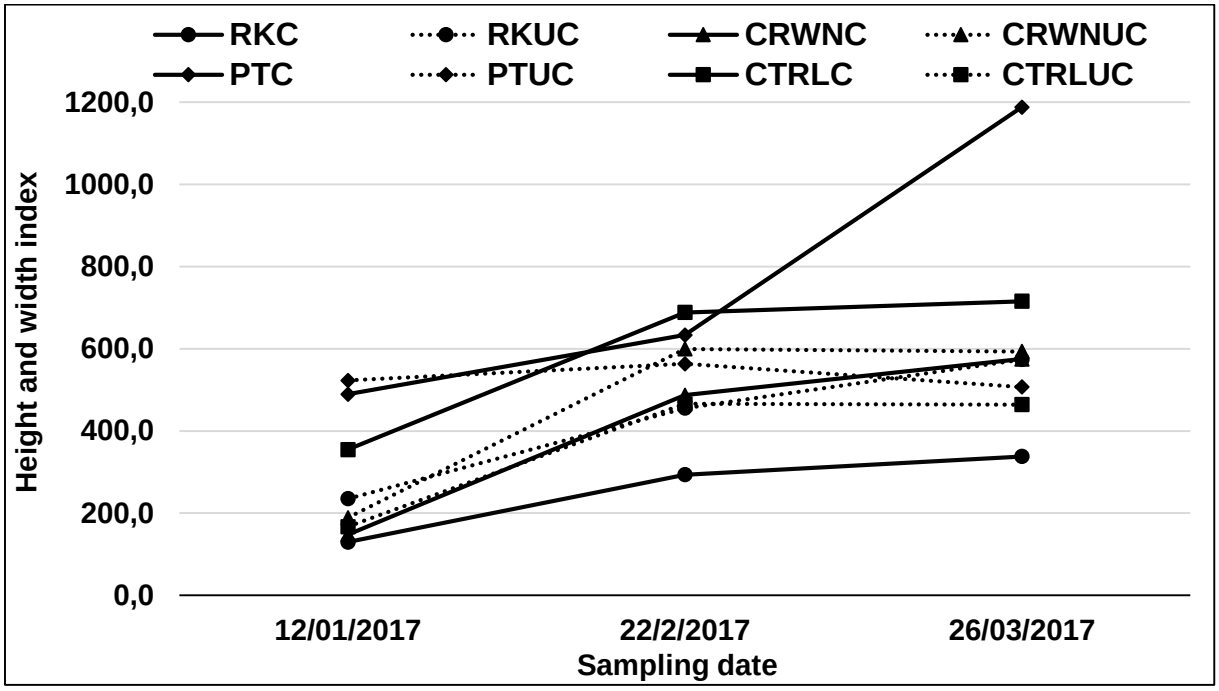

Figure 4.57: Change in grass height and width index of *D. eriantha* seedlings using coated and uncoated seed in the four growth mediums. (RKC – Rooikraal gold mine tailings coated seed; RKUC – Rooikraal gold mine tailings uncoated seed; CRWNC – Crown gold mine tailings coated seed; CRWNUC – Crown gold mine tailings uncoated seed; PTC – Platinum tailings coated seed; PTUC – Platinum tailings uncoated seed; CTRLC – Control coated seed; CTRLUC – Control uncoated seed).

- ***Eragrostis curvula***

The repeated measures ANOVA indicated a significant influence of the growth medium (p = 0,03) but not the seed type (coated and uncoated) on the change in the plant height and width index of *E. curvula* seedlings over three months (January 2017 to March 2017) (Table 4.41).

The seedlings from coated seed of *E. curvula* had a higher height and width index in the platinum tailings than seedlings from uncoated seed (Figure 4.58). The growth and height index of seedlings from the uncoated seed of *E. curvula* was higher in seedlings from coated seed in both the Crown and Rooikraal gold mine tailings and the control soil (Figure 4.58).

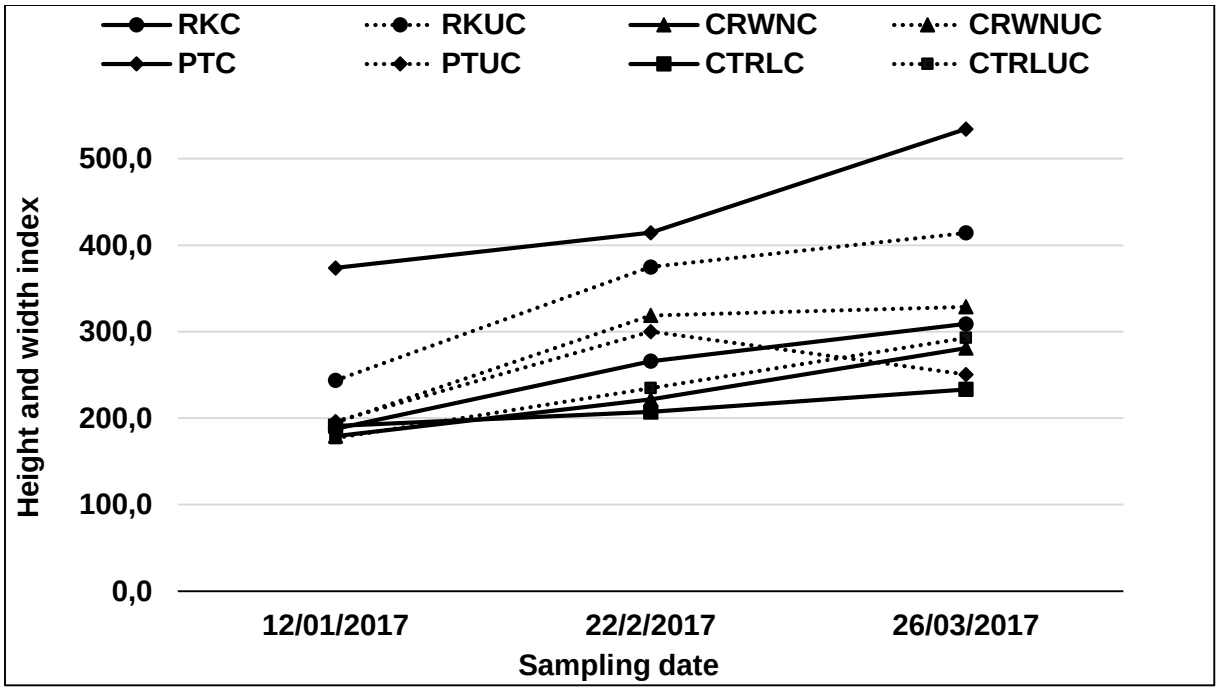

Figure 4.58: Change in grass height and width index of *D. eriantha* seedlings using coated and uncoated seed in the four growth mediums. (RKC – Rooikraal gold mine tailings coated seed; RKUC – Rooikraal gold mine tailings uncoated seed; CRWNC – Crown gold mine tailings coated seed; CRWNUC – Crown gold mine tailings uncoated seed; PTC – Platinum tailings coated seed; PTUC – Platinum tailings uncoated seed; CTRLC – Control coated seed; CTRLUC – Control uncoated seed).

- ***Eragrostis tef***

According to the repeated measurement ANOVA (Table 4.41), the growth medium did not have a significant influence on the change in the height and width index of *E. tef* seedlings during the pot trials (Figure 4.59). The height and width index of *E. tef* decreased in the control soil and platinum tailings towards the end of the trials and increased towards the end of the trial period in the Rooikraal and Crown gold mine tailings (Figure 4.59).

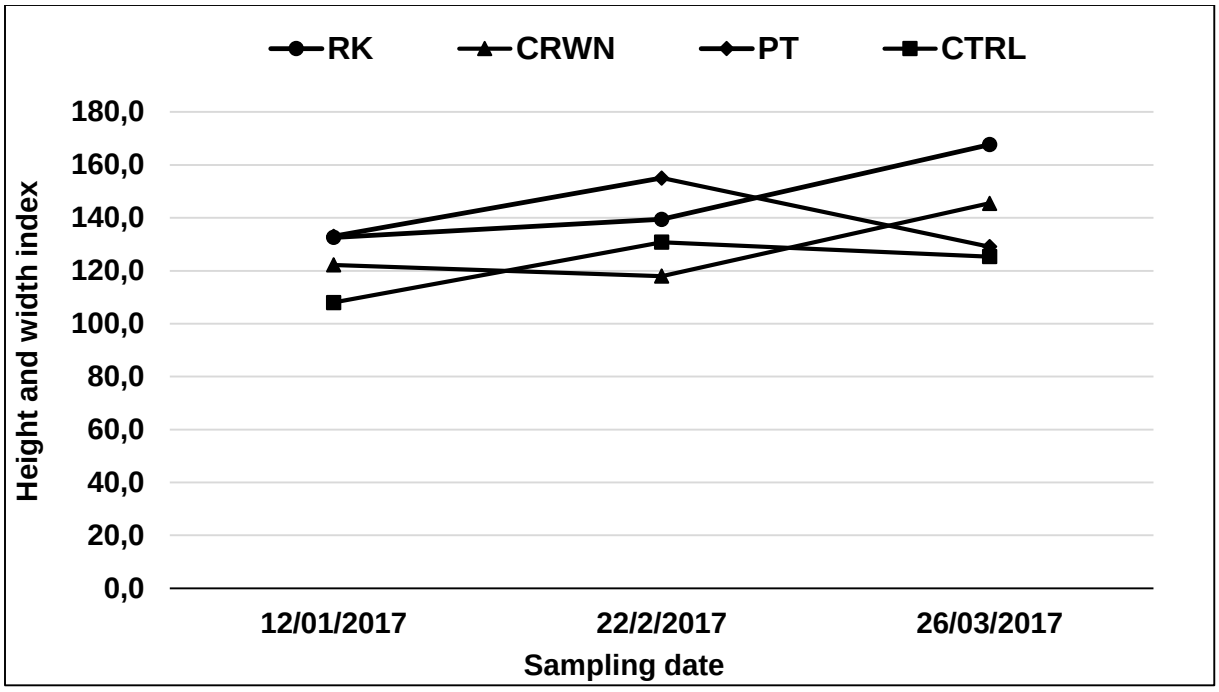

Figure 4.59: Change in height and width index of *E. tef* seedlings in tailings growth mediums. (RK – Rooikraal gold mine tailings; CRWN – Crown gold mine tailings; PT – Platinum tailings; CTRL – Control soil).

- **Summary**

To summarise the height and width index results obtained during the Phase 2 supporting pot trials: The growth medium had a significant influence on the change of the height and width index of *C. dactylon* and *E. curvula* seedlings. The seed type (coated and uncoated) only had a significant influence on the change in the height and width index of *C. dactylon* seedlings.

The highest height and width index for *C. dactylon* seedlings was obtained in the Crown gold mine tailings using uncoated seed (CRWNUC), and the lowest height and width index was obtained in the control soil using coated seed (CTRLC). *D. eriantha* obtained the highest height and width index in the platinum tailings using coated seed (PTC) and the lowest height and width index in the Rooikraal gold mine tailings using coated seed (RKC). Seedlings of *E. curvula* obtained the highest height and width index in platinum tailings using coated seed (PTC) and had the lowest height and width index in the control soil using coated seed (CTRLC).

The growth medium did not have a significant influence on the change of the height and width index of *E. tef* seedlings.

4.4.3 Dry matter (DM) production of seedlings from coated and uncoated seed

The average DM produced (g/m^2)) by seedlings from coated and uncoated seed of *C. dactylon, D. eriantha, E. curvula* and uncoated *E. tef* seed is displayed in Figure 4.60 to Figure 4.63 and discussed below.

According to the two-way ANOVA results (Table 4.42) on the average DM (g/m^2) produced by *C. dactylon* seedlings, the growth medium (p = 0,025) did have a significant influence on the average DM production by *C. dactylon* seedlings, but this was not significantly influenced by the seed type (p = 0,179).

Table 4.42: Two-way ANOVA results for the average DM (g/m^2) produced in March 2017 from coated and uncoated *C. dactylon* seed grown in the tailings and control soil growth mediums. The statistical significance of the variables (Growth medium and seed type) and the significant differences between trial groups (RKC – Rooikraal gold mine tailings coated seed; RKUC – Rooikraal gold mine tailings uncoated seed; CRWNC – Crown gold mine tailings coated seed; CRWNUC – Crown gold mine tailings uncoated seed; PTC – Platinum tailings coated seed; PTUC – Platinum tailings uncoated seed; CTRLC – Control coated seed; CTRLUC – Control uncoated seed) are displayed. The influence of the growth medium and the seed type was significant if p < 0,05.

Growth medium	Seed type	DM (g/m²)
RK	Uncoated	426,2 a
RK	Coated	465,0 a
CTRL	Uncoated	523,0 a
PT	Uncoated	523,0 a
CRWN	Coated	527,8 a
CTRL	Coated	551,9 ab
PT	Coated	551,9 ab
CRWN	Uncoated	1068,9 b
Growth medium	***p* = 0,03**	**Significant: Yes**
Seed type	***p* = 0,28**	**Significant: No**

The DM produced by seedlings from uncoated *C. dactylon* seed in the Crown gold mine tailings (1068,9 g/m^2) was notably higher than the DM produced by seedlings from coated seed 527,8 g/m^2 in the same Crown gold mine tailings growth medium (CRWNC). The DM of CRWNUC was also higher than the DM produced by uncoated seed in the platinum tailings (523,0 g/m^2 – PTUC), the control soil growth medium (523,0 g/m^2 – CTRLUC), Rooikraal gold mine tailings (426,2 g/m^2 – RKUC) and coated seed in the Rooikraal gold mine tailings (465,0 g/m^2 – RKUC) (Figure 4.60).

However, coated *C. dactylon* seed did produce higher average DM yields compared to uncoated seed in the Rooikraal gold mine tailings, the platinum tailings and the control soil. These results were however not significant (Figure 4.60).

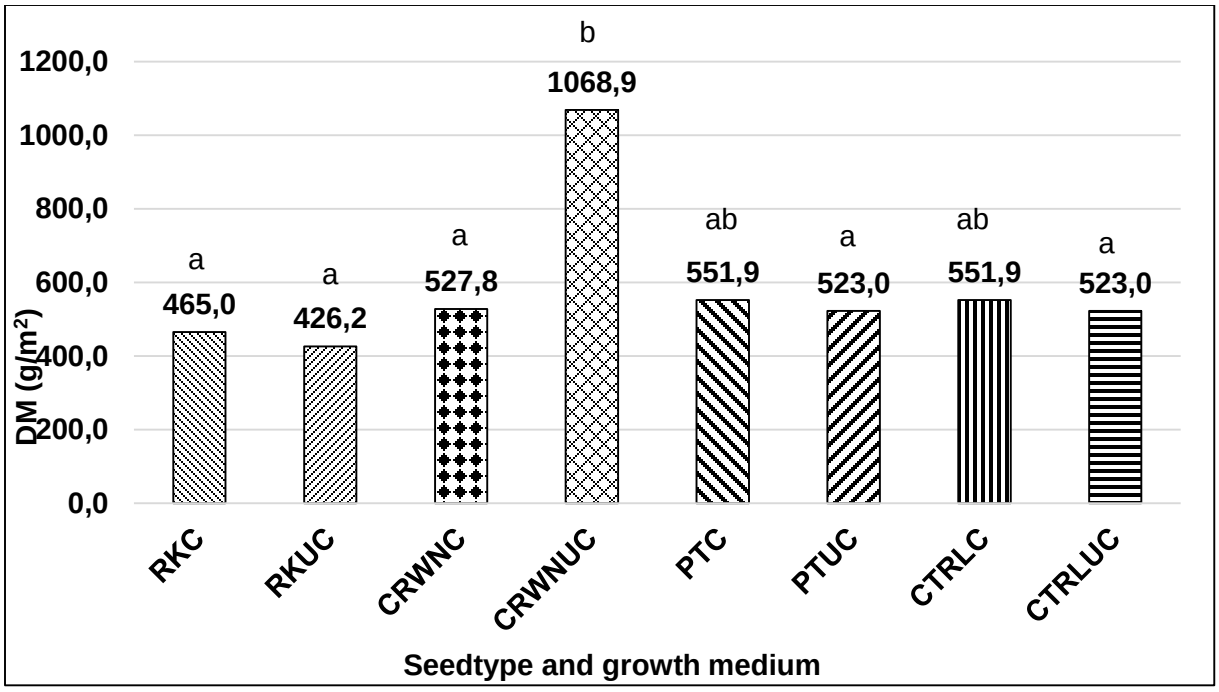

Figure 4.60: Average DM produced in g/ m^2 for coated and uncoated *C. dactylon* seed in tailing growth mediums in March 2017. (RKC – Rooikraal gold mine tailings coated seed; RKUC – Rooikraal gold mine tailings uncoated seed; CRWNC – Crown gold mine tailings coated seed; CRWNUC – Crown gold mine tailings uncoated seed; PTC – Platinum tailings coated seed; PTUC – Platinum tailings uncoated seed; CTRLC – Control coated seed; CTRLUC – Control uncoated seed). Statiscal significance between groups are indicated by letters above bars.

The average DM yield of seedlings from coated and uncoated *D. eriantha* seed is shown in Figure 4.61. According to the two-way ANOVA, there is no influence of the growth medium (p = 0,34) or the seed type (coated and uncoated) (p = 0,35) on the DM yield of *D. eriantha* (Table 4.43).

Although the difference between the DM yields of *D. eriantha* is not significant, uncoated seed did have a higher DM yield per m^2 than coated seed in each of the four growth mediums (Figure 4.61 and Table 4.43). The highest DM yield of *D. eriantha* was recorded for uncoated seed in platinum tailings providing an average of 542,5 g/m^2 and the lowest DM yield for *D. eriantha* was recorded using coated seed in Crown gold mine tailings and yielded and average of 233,6 g/m^2.

Table 4.43: Two-way ANOVA results for the average DM (g/m^2) produced in March from coated and uncoated *D. eriantha* seed grown in the tailings and control soil growth mediums. The statistical significance of the variables (Growth medium and seed type) and the significant differences between trial groups (RKC – Rooikraal gold mine tailings coated seed; RKUC – Rooikraal gold mine tailings uncoated seed; CRWNC – Crown gold mine tailings coated seed; CRWNUC – Crown gold mine tailings uncoated seed; PTC – Platinum tailings coated seed; PTUC – Platinum tailings uncoated seed; CTRLC – Control coated seed; CTRLUC – Control uncoated seed) are displayed. The influence of the growth medium and the seed type was significant if $p < 0,05$.

Growth medium	Seed type	DM (g/m²)
CRWN	Coated	233,6 a
RK	Coated	272,7 a
RK	Uncoated	344,8 a
CRWN	Uncoated	357,1 a
CTRL	Coated	380,4 a
PT	Coated	499,0 a
CTRL	Uncoated	523,7 a
PT	Uncoated	542,5 a
Growth medium	***p* = 0,03**	**Significant: Yes**
Seed type	***p* = 0,28**	**Significant: No**

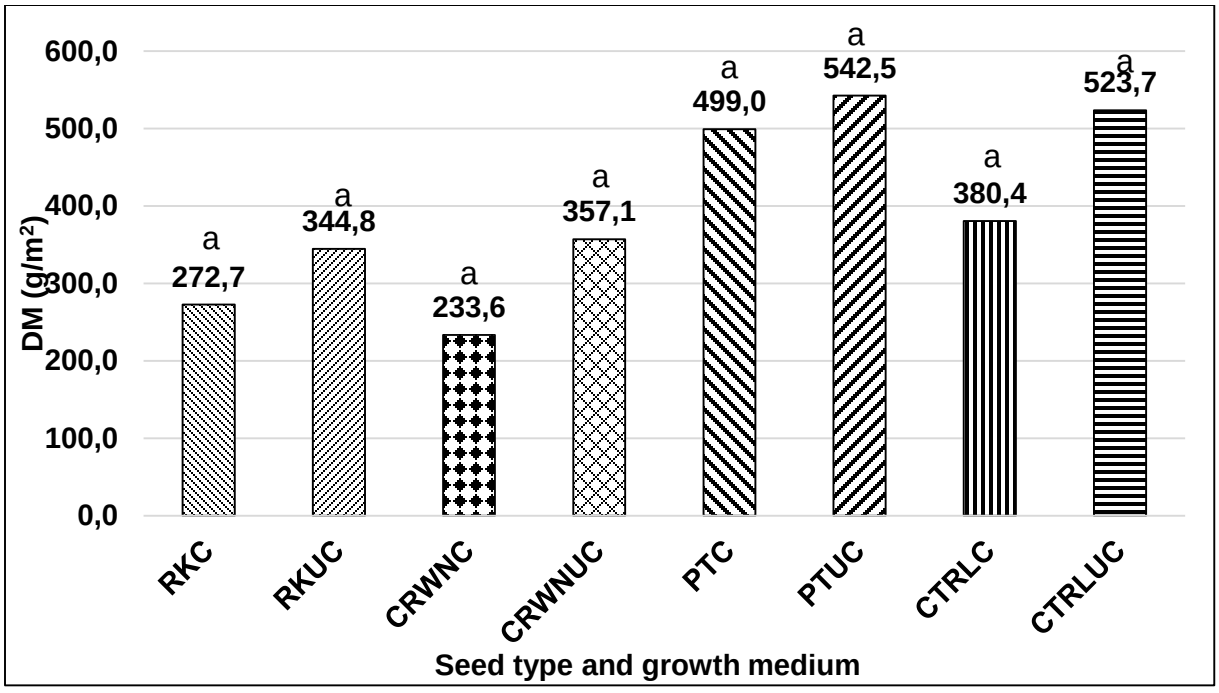

Figure 4.61: Average DM produced in g/m^2 for coated and uncoated *D. eriantha* seed in tailing growth mediums in March 2017. (RKC – Rooikraal gold mine tailings coated seed; RKUC – Rooikraal gold mine tailings uncoated seed; CRWNC – Crown gold mine tailings coated seed; CRWNUC – Crown gold mine tailings uncoated seed; PTC – Platinum tailings coated seed; PTUC – Platinum tailings uncoated seed; CTRLC – Control coated seed; CTRLUC – Control uncoated seed). Statistical significance between groups are indicated by letters above bars.

The average DM produced by *E. curvula* from coated and uncoated seed in the tailings growth mediums and control soil is displayed in Figure 4.62. According to the two-way ANOVA, the growth medium (p = 0,02) did have an influence on the DM produced by *E. curvula*, but the seed type (coated and uncoated) (p = 0,28) did not (Table 4.44).

The average DM produced by seedlings from coated *E. curvula* seed in platinum tailings (PTC) was higher at 996,9 g/m^2 than the DM produced by seedlings using uncoated seed in the platinum tailings at 416,0 g/m^2. The DM produced from uncoated seed in the Rooikraal gold mine tailings (RKUC) (468,5 g/m^2) and control soil (CTRLUC) (421,4 g/m^2) was lower than the DM yield of coated seed in platinum tailings. The DM produced by seedlings from coated *E. curvula* seed in Crown gold mine tailings (CRWNC) (283,1 g/m^2) and the control soil (CTRLC) (392,6 g/m^2) was also lower than the DM produced by seedlings from coated seed in the platinum tailings (Figure 4.62).

Table 4.44: Two-way ANOVA results for the average DM (g/m^2) produced in March 2017 from coated and uncoated *E. curvula* seed grown in the tailings and control soil growth mediums. The statistical significance of the variables (Growth medium and seed type) and the significant differences between trial groups (RKC – Rooikraal gold mine tailings coated seed; RKUC – Rooikraal gold mine tailings uncoated seed; CRWNC – Crown gold mine tailings coated seed; CRWNUC – Crown gold mine tailings uncoated seed; PTC – Platinum tailings coated seed; PTUC – Platinum tailings uncoated seed; CTRLC – Control coated seed; CTRLUC – Control uncoated seed) are displayed. The influence of the growth medium and the seed type was significant if p < 0,05.

Growth medium	Seed type	DM (g/m²)
CRWN	Coated	283,1 a
CTRL	Coated	392,6 a
PT	Uncoated	416,0 a
CTRL	Uncoated	421,4 a
RK	Uncoated	468,5 a
RK	Coated	471,7 ab
CRWN	Uncoated	482,4 ab
PT	Coated	996,9 b
Growth medium	***p* = 0,03**	**Significant: Yes**
Seed type	***p* = 0,28**	**Significant: No**

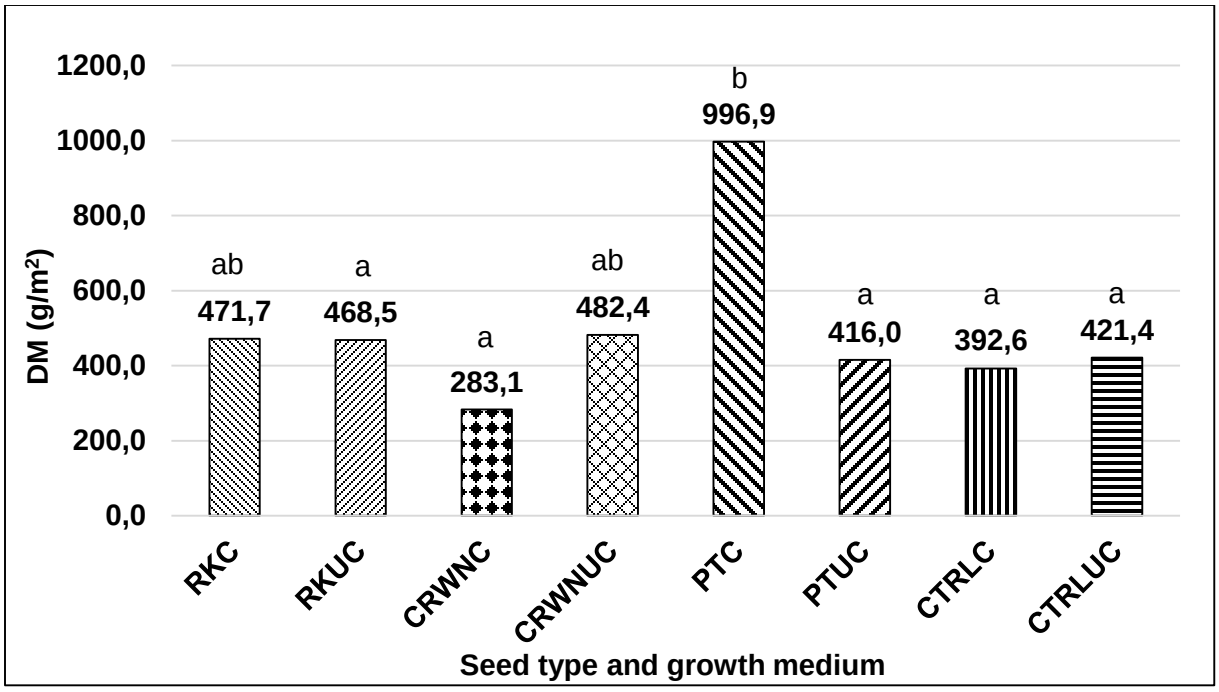

Figure 4.62: Average DM produced in g/m^2 for coated and uncoated *E. curvula* seed tailings growth mediums in March 2017. (RKC – Rooikraal gold mine tailings coated seed; RKUC – Rooikraal gold mine tailings uncoated seed; CRWNC – Crown gold mine tailings coated seed; CRWNUC – Crown gold mine tailings uncoated seed; PTC – Platinum tailings coated seed; PTUC – Platinum tailings uncoated seed; CTRLC – Control coated seed; CTRLUC – Control uncoated seed). Statistical significance between groups are indicated by letters above bars.

The DM produced by seedlings from uncoated *E. tef* seed at the end of the phase 2 pot trials in March 2017 are illustrated in Figure 4.63. According to the one-way ANOVA, there was not a statistical significant difference between the average DM produced from of *E. tef* seedlings in the growth mediums (p = 0,892) (Figure 4.63).

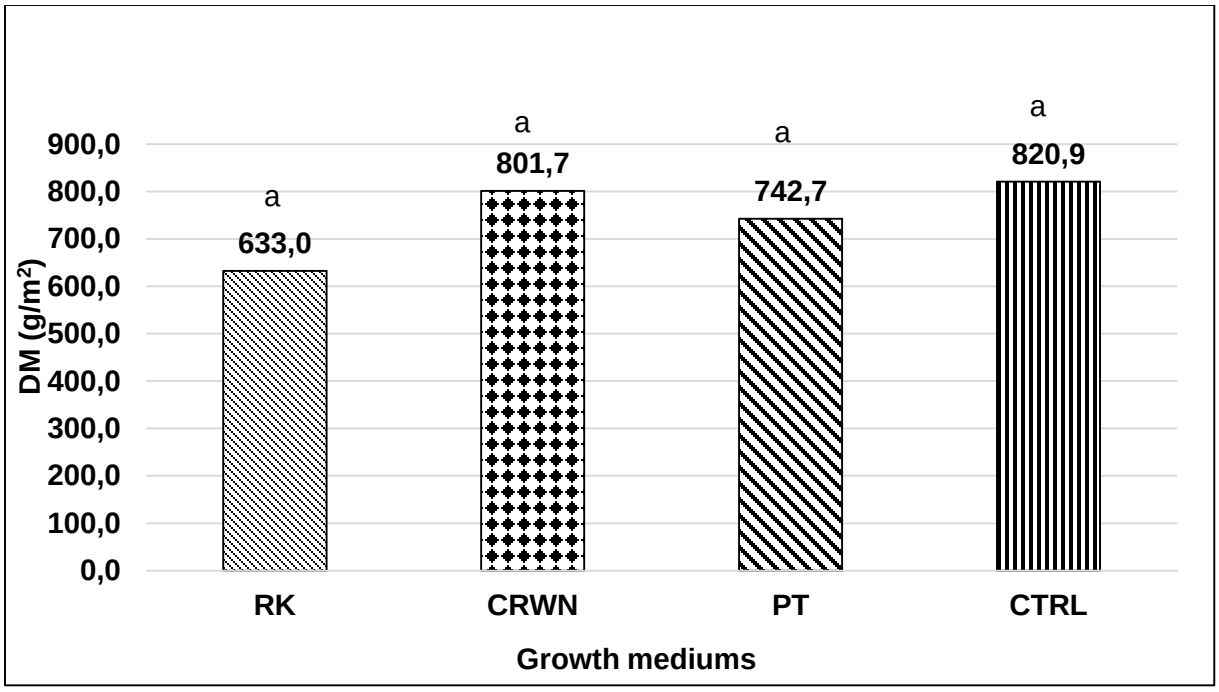

Figure 4.63: Average biomass produced per pot for *E. tef* seed grown in growth mediums (RK – Rooikraal gold mine tailings, CRWN – Crown gold mine tailings, PT – Platinum tailings, CTRL – Control).

- **Summary**

To summarise the DM produced seedlings from coated and uncoated seed of *C. dactylon, D. eriantha*, *E, curvula* and uncoated *E. tef* seed in the four growth mediums at the end of the Phase 2 pot trials in March 2017.

The highest DM produced of *C. dactylon* seedlings occurred in the Crown gold mine tailings using uncoated seed (CRWNUC). However, seedlings from coated *C. dactylon* seed yielded higher DM in each of the other three growth mediums (RKC, PTC and CTRLC), but the difference was not significant (Figure 4.60).

There was no statistically significant difference between the DM produced by seedlings of *D. eriantha* in either of the growth mediums or between coated and uncoated seed.

The DM yield of *E. curvula* was the highest in the platinum tailings using coated seed (Figure 4.62) however, according to the one-way ANOVA there were no statistically significant differences of the DM yields for *E. tef* in the various growth mediums. .

4.5 Phase 3 Additional field trials on Rooikraal gold TSF

The emergence of seed treatments sown in the lower density seed trials and the bio-stimulant trials is discussed in this Section.

4.5.1 Phase 3 lower seed density trials

During the Phase 1 field trials (Section 4.2), it was observed at the Crown gold, Rooikraal gold and platinum tailings that the coated seed treatment 1 sown at a seeding rate of 19 kg /ha (T1C in Figure 4.1 and Figure 4.3) had a higher seedling emergence percentage than uncoated seed treatment 2 sown at the same seeding rate (19 kg/ha) (T2UC in Figure 4.1). However, the seedling emergence densities of these seed treatments (T1C and T2UC in Figure 4.1) were not significantly different statistically according the one-way ANOVA. An increase in the seeding rate of coated seed (TC3 in Figure 4.2) did not result in a significant increase in the seedling emergence density and had a lower seedling emergence percentage than T1C at both the gold TSF sites and the platinum tailings (Figure 4.1 and Figure 4.3).

This prompted the setup of trials to evaluate when the seeding rate of the coated and uncoated seed treatments (T1C and T2UC) were decreased and whether this resulted in higher emergence percentages and lower seedling densities of seed.

The seedling emergence results including all the species in the seed treatments and excluding *E. tef* for the lower seeding density trials at the Rooikraal gold TSF site are illustrated in Figure 4.64.

According to the one-way ANOVA, there was a significant difference in the seedling emergence percentage of the lower density seed treatments both including ($p < 0,001$) and excluding *E. tef* ($p < 0,001$) (Figure 4.64 and Table 4.45).

The percentage seedlings that emerged from the 19 kg/ha coated lower density seed treatment (LD19C, 19,6%) was lower than the emergence percentage of the 5 kg/ha coated seed treatment (LD5C, 53,8%) (Figure 4.64 and Table 4.45).

When *E. tef* is excluded, the emergence percentage of the coated 19 kg/ha lower density seed treatment (LD19C, 4,9%) was lower than the seedling emergence percentage of the lower density 5 kg/ha coated seed treatment (LD5C, 9,5%) (Figure 4.64 and Table 4.45).

When considering the total seedling emergence percentage of the uncoated lower density seed treatments, the uncoated lower density 19 kg/ha seed treatment (LD19UC, 13%) did have a lower emergence percentage than the 12 kg/ha uncoated lower density seed treatment (LD12UC, 16,1%), but it was not significant. However, the uncoated 5 kg/ha seed treatment (LD5UC, 32,8%) did have a higher total seedling emergence percentage than LD19UC and LD12UC (Figure 4.64 and Table 4.45).

The seedling emergence percentage excluding *E. tef* was lower in LD19UC (1,9%) than LD5UC (8,4%) (Figure 4.64 and Table 4.45).

The total emergence percentage of the coated lower density seed treatments (LD19C, LD12C and LD5C) were higher than the uncoated lower density seed treatments (LD19UC, LD12UC and LD5UC) at each of the three seeding rates, but only the seedling emergence percentage of LD5C was higher than its uncoated equivalent LD5UC (Figure 4.64 and Table 4.45).

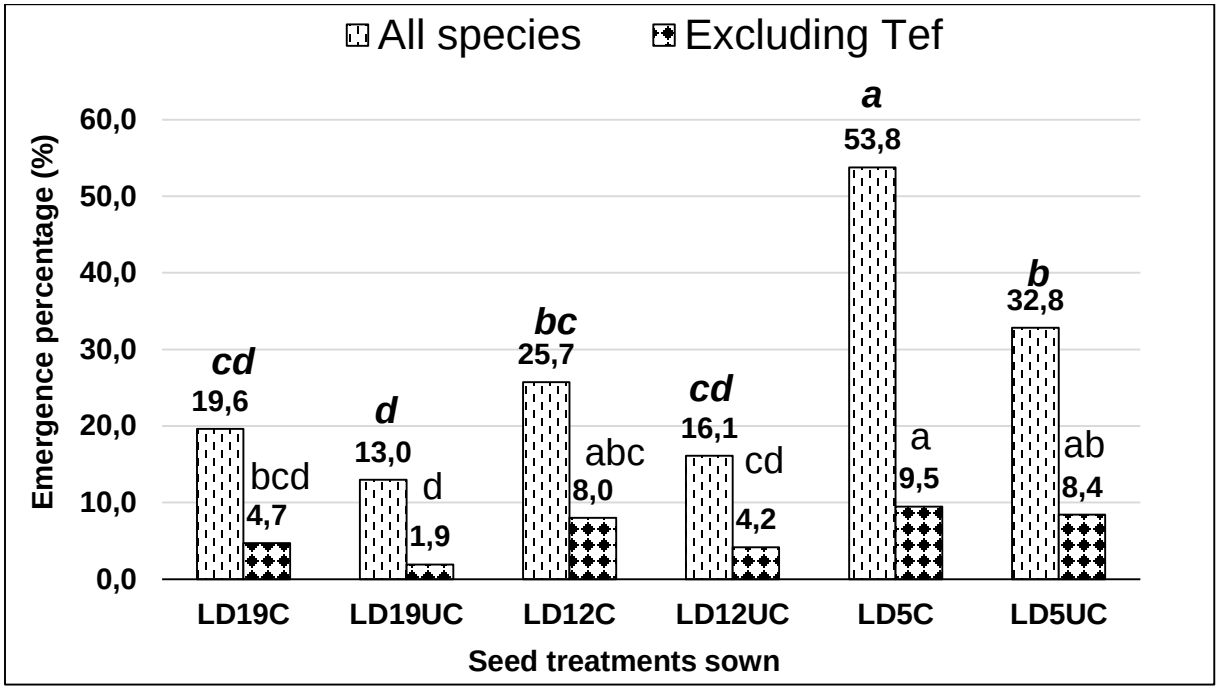

Figure 4.64: Average emergence percentage (%) for lower density seed treatment field trials during Phase 3 at the Rooikraal gold TSF site in March 2017 for all species and excluding *E. tef*. Statistical significance (p < 0,05) between average emergence percentage of seed treatments (LD19C, LD19UC, LD12C, LD12UC, LD5C, LD5UC see Section 3.4.3) are illustrated in bold italics i.e. "***a***". Statistical significance between emergence percentage of seed treatments excluding *E. tef* is displayed in normal font.

Table 4.45: Table displaying significant differences (p < 0,05) between the average emergence percentage of the lower seeding rate seed treatments (LD19C, LD19UC, LD12C, LD12UC, LD5C, LD5UC see Section 3.4.3) including and excluding E. tef seed (Figure 4.64) and the average emerged plant density (plants/m2) of the lower seeding rate seed treatments (LD19C, LD19UC, LD12C, LD12UC, LD5C, LD5UC Section 3.4.3) including and excluding E. tef seed (Figure 4.65).

Lower density seed trial emergence percentage in March 2017			
All species		Excluding *E. tef*	
Treatments	Emergence (%)	Treatments	Emergence (%)
LD5C	53,8 ***a***	LD5C	9,5 a
LD5UC	32,8 ***b***	LD5UC	8,4 ab
LD12C	25,7 ***bc***	LD12C	8,0 abc
LD19C	19,6 ***cd***	LD19C	4,7 bcd
LD12UC	16,1 ***cd***	LD12UC	4,2 cd
LD19UC	13,0 ***d***	LD19UC	1,9 d
p < 0,001		p < 0,0001	
Significant	Yes	Significant	Yes
Lower density seed trial seedling emergence density in March 2017			
All species		Excluding *E. tef*	
Treatments	Density (plants/m²)	Treatments	Density (plants/m²)
LD19UC	316,3 ***a***	LD12UC	56,3 a
LD19C	258,5 ***ab***	LD12C	51,8 a
LD12UC	247,8 ***ab***	LD19C	48,0 a
LD12C	213,8 ***b***	LD5UC	47,5 a
LD5UC	210,5 ***b***	LD19UC	41,0 a
LD5C	186,3 ***b***	LD5C	25,5 a
p = 0,0004		p = 0,1	
Significant	Yes	Significant	No

The total density of emerged seedlings including and excluding *E. tef* in plants/m^2 for the coated and uncoated seed lower seeding density trials at Rooikraal gold TSF site are illustrated and discussed below in (Figure 4.65 and Table 4.45).

According to the one-way ANOVA (Table 4.45), there was not a significant difference between the mean seedling emergence of seed treatments LD19C (258,5 plants/m^2), LD12C (213,8 plants/m^2), LD5C (183,3 plants/m^2), LD12UC (247,8 plants/m^2) and LD5UC (210,5 plants/m^2) when all species are considered, including *E. tef* (Figure 4.65 and Table 4.45). The uncoated seed treatment LD19UC (316 plants/m^2) had a higher emerged seedling density than LD12C (213,8 plants/m^2), LD5C (186,3 plants/m^2) and LD5UC (210,5 plants/m^2) when all species were considered (Figure 4.65 and Table 4.45). When *E. tef* was excluded, there was no statistical significance between the mean emerged seedling density of the lower density seed treatments (Figure 4.65 and Table 4.45).

A decrease in the seeding rate from 19 kg/ha to 5 kg/ha, did not result in a significant decrease in the emerged seedling density with coated seed and uncoated seed treatments (Figure 4.65 and Table 4.45). However, a decrease in seeding rate from 19 kg/ha to 5 kg/ha did result in an increase in the seedling emergence percentage. This indicates that similar emergence results could be obtained with lower seeding densities, and the coated seed treatments in Phase 1 have a higher emergence percentage because fewer seeds are being sown per weight unit compared to uncoated seed. Seedlings of *E. tef* emerged in densities exceeding the number of viable seed sown on the plots on the LD12C, LD12UC, LD5C and LD5UC seed plots. This can be explained by the dispersal of ungerminated seed by wind from adjacent lower density treatment plots. Also *E. tef* and *E. curvula* seedlings are difficult to distinguish from each other early on in the first growth season (Coetzee, 2014:76).

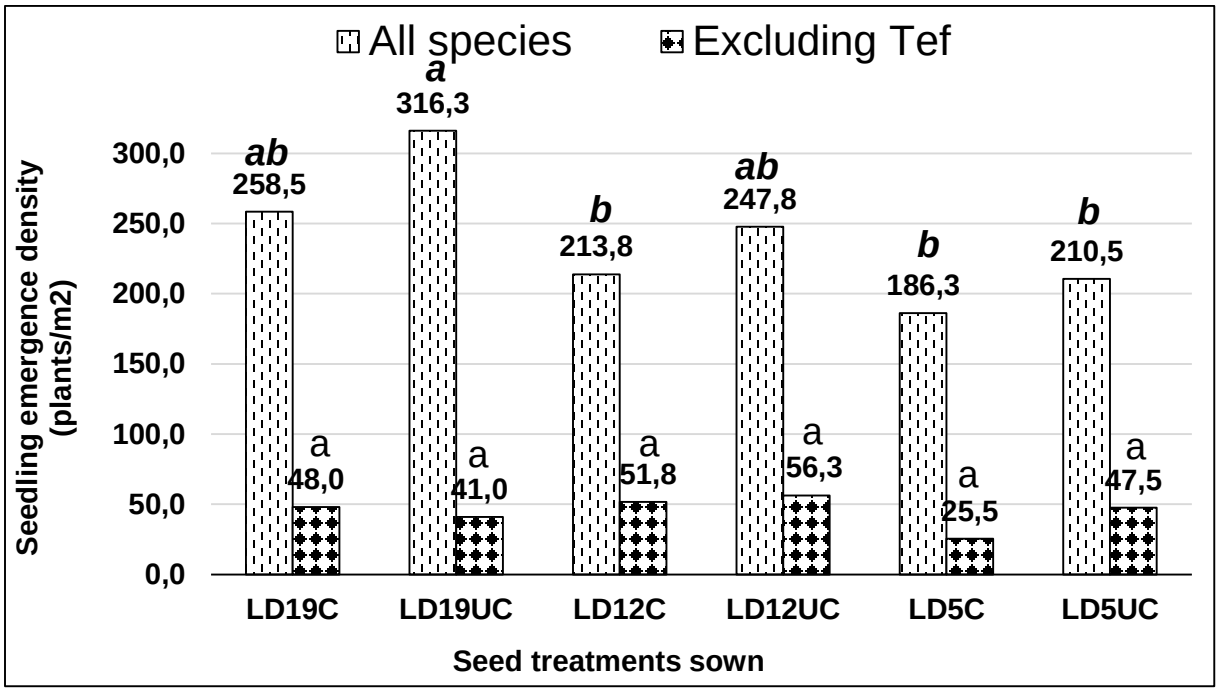

Figure 4.65: Average emergence density (plants/m^2) in March 2017 for lower density seed treatments field trials during Phase 3 at the Rooikraal gold TSF site in March 2017. Statistical significance (p < 0,05) between average emergence percentage of seed treatments (LD19C, LD19UC, LD12C, LD12UC, LD5C, LD5UC see Section 3.4.3) is illustrated in bold italics i.e. "***a***". Statistical significance between emergence percentage of seed treatments excluding *E. tef* is displayed in normal font.

4.5.2 Phase 3 bio-stimulant trials

The seedling emergence of seed treatments used in the bio-stimulant trials (LD5C, LD5UC, MA1, MA2, MA3 and MA4 see Section 3.4.3) are illustrated in Figure 4.66, Figure 4.67 and Table 4.66.

These trials were conducted to evaluate whether amelioration with bio-stimulants (*Trichoderma* fungi and bacteria) could increase the seedling emergence of seed treatments. Figure 4.66 illustrates the total emergence percentage and the seedling emergence percentage excluding *E. tef* in the bio-stimulant trial seed treatments (LD5C, LD5UC, MA1, MA2, MA3 and MA4).

The coated lower density seed treatment LD5C and LD5UC were not treated with bio-stimulants and treated as control seed treatments. The lower density seed treatment LD5C (53,8%) had a higher total seedling emergence percentage, including *E. tef* seed than the other seed treatments (LD5UC – 32,8%, MA1 – 34,7%, MA2 – 33,7%, MA3 – 38,6% and MA4 – 33,7%). This is due to the lower number of seed contained in LD5C compared to LD5UC, MA1, MA2, MA3 and MA4 as discussed in Section 4.5.1)

There was not a significant difference between the mean emergence percentages of LD5UC (32,8%), MA1 (34,7%), MA2 (33,7%), MA3 (38,6%) and MA4 (33,7%) (Figure 4.66 and Table 4.46).

If *E. tef* was not considered, then bio-stimulant seed MA3 – 6,8% had a lower seedling emergence percentage than MA2 – 11,7% (Figure 4.66 and Table 4.46).

According to the one-way ANOVA, there was not a significant difference between the mean seedling density of the bio-stimulant trial seed treatments ($p = 0,369$), but there was a significant difference ($p < 0,0001$) between the seedling density of seed treatments if *E. tef* seedlings were excluded (Figure 4.67 and Table 4.46).

Table 4.46: Table displaying significant differences ($p < 0,05$) between the average emergence percentages of the bio-stimulant seed treatments (LD5C, LD5UC, MA1, MA2, MA3 and MA4 see Section 3.4.3) including and excluding *E.* tef (Figure 4.66) and the significant difference between average emerged plant density (plants/m^2) of the bio-stimulant seed treatments (LD5C, LD5UC, MA1, MA2, MA3 and MA4 see Section 3.4.3) including and excluding *E. tef* (Figure 4.67).

Bio-stimulant trial emergence percentage in March 2017			
All species		Excluding *E. tef*	
Treatments	Emergence (%)	Treatments	Emergence (%)
LD5C	53,8 ***a***	MA2	11,7 a
MA3	38,6 ***b***	MA4	11,0 ab
MA1	34,7 ***b***	LD5C	9,5 ab
MA4	33,7 ***b***	MA1	8,7 ab
MA2	33,7 ***b***	LD5UC	8,4 ab
LD5UC	32,8 ***b***	MA3	6,8 b
$p = 0,0002$		$p = 0,046$	
Significant	No	Significant	Yes
Bio-stimulant seed trial emergence density in March 2017			
All species		Excluding *E. tef*	
Treatments	Density (plants/m^2)	Treatments	Density (plants/m^2)
MA3	247,3 ***a***	MA2	65,8 a
MA1	222,5 ***a***	MA4	62,0 ab
MA4	216,0 ***a***	MA1	49,0 abc
MA2	215,8 ***a***	LD5UC	47,5 abc
LD5UC	210,5 ***a***	MA3	38,3 bc
LD5C	186,3 ***a***	LD5C	25,5 c
$p = 0,359$		$p < 0,0001$	
Significant	No	Significant	Yes

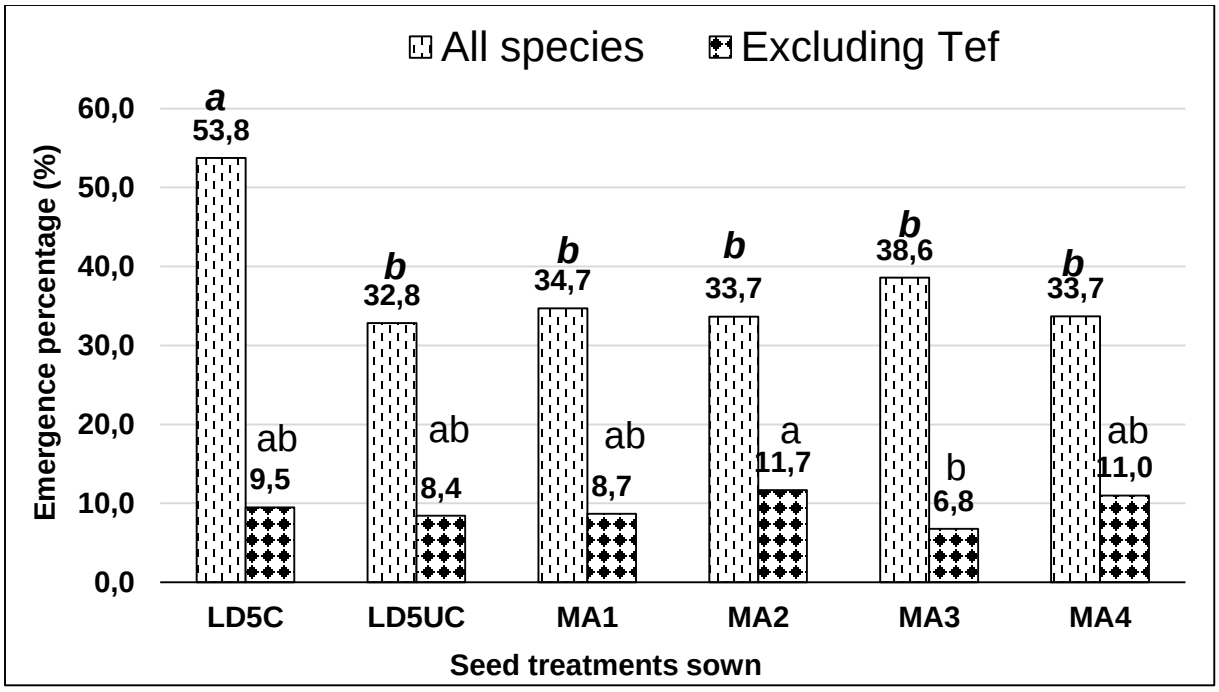

Figure 4.66: Average seedling emergence percentage (%) in March 2017for seed treatments (LD5C, LD5UC, MA1, MA2, MA3 and MA4 Section 3.4.3 used in the Phase 3 bio-stimulant trials at the Rooikraal gold TSF site considering all species used in the seed treatments and excluding *E. tef*. Statistical significance between average emergence percentage of seed treatments is indicated with bold italic letters i.e. "***a***", and normal letters are used to indicate significant differences between emergence percentage of seed treatments excluding *E. tef* seed.

Bio-stimulant MA2 – 65,8 plants/m^2 resulted in a higher emergence density of seedlings excluding *E. tef* compared to the coated treatment LD5C – 25,5 plants/m^2 and the bio-stimulant MA3 – 38,3 plants/m^2 (Figure 4.67 and Table 4.46).

Although not significantly higher than the LD5UC, both the seed treatments treated with *Trichoderma* fungi powder in combination with a food source (MA2 – 65,8 plants/m^2 and MA4 – 62 plants/m^2) had higher emergence densities excluding *E. tef* than the control treatments (LD5C – 25,5 plants/m^2 and LD5UC – 47,5 plants/m^2). The results suggest that the application of *Trichoderma* as a bio-stimulant to seed is capable of increasing the seedling emergence of specifically the species *C. dactylon*, *D. eriantha, E. curvula* and *S. bicolor* (Figure 4.67 and Table 4.46).

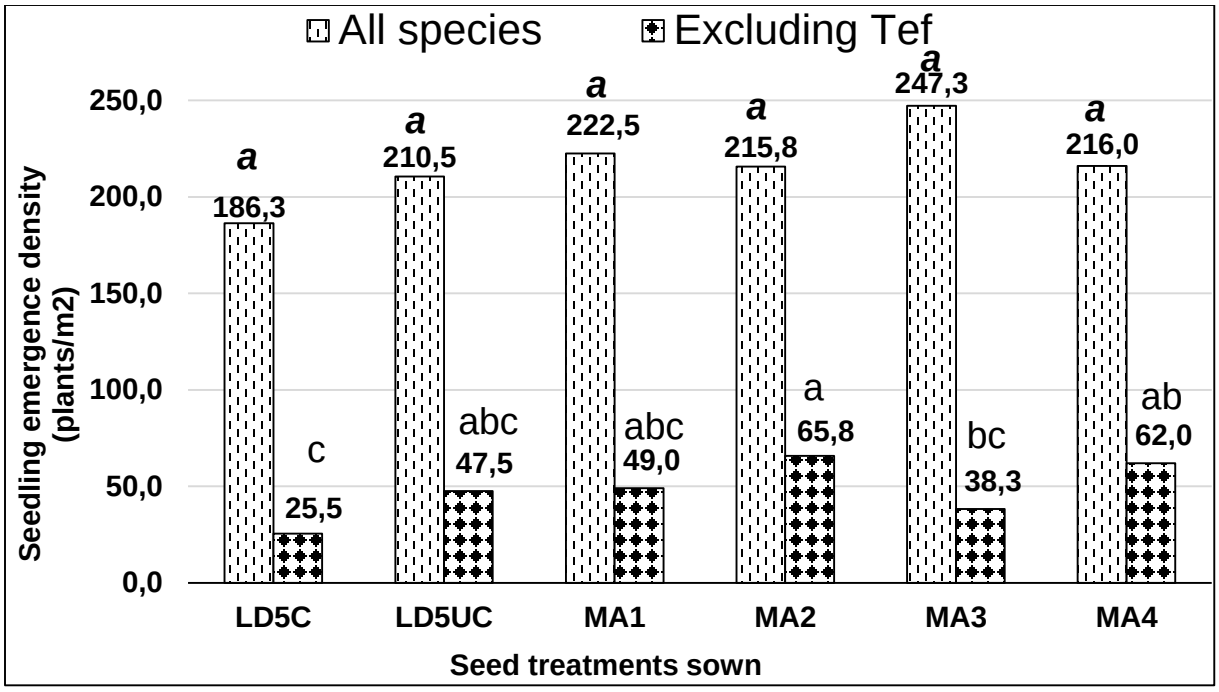

Figure 4.67: Average seedling emergence density (plants/m^2) in March 2017 for seed treatments (LD5C, LD5UC, MA1, MA2, MA3 and MA4 see Section 3.4.3) used in the Phase 3 bio-stimulant trials at the Rooikraal gold TSF site considering all species used in the seed treatments and excluding *E. tef*. Statistical significance between average emergence density of seed treatments is indicated with bold italic letters i.e. "***a***", and normal letters are used to indicate significant differences between emergence percentage of seed treatments excluding *E. tef* seed.

4.5.3 Dehydrogenase activity (DHA)

The dehydrogenase activity (DHA) (INF µg/g/2h) for the trials at Rooikraal gold TSF site in April 2016 of a natural soil (controls), non-ameliorated bare gold mine tailings and the bio-stimulant seed treatment trial plots are shown in Figure 4.68, Figure 4.69 and Table 4.47 and Table 4.48.

According to the one-way ANOVA (p < 0,0001) the DHA of the natural soil sampled a distance away from the Rooikraal gold TSF site boundary (351,02 INF µg/g/2h) was significantly higher statistically than the ameliorated bio-stimulant trial plots (LD5C, LD5UC, MA1T, MA2, MA3, MA4) and the non-ameliorated bare gold mine tailings, this is indicated by the symbol 'a' above the natural soil bar in Figure 4.68 and Table 4.47.

The significant difference between the DHA of natural soil compared to the bare tailings support the views of Mendez & Maier (2008a:278) and Petrisor *et al*. (2004:3) that the microbial populations present on mine tailings are reduced compared to natural soils.

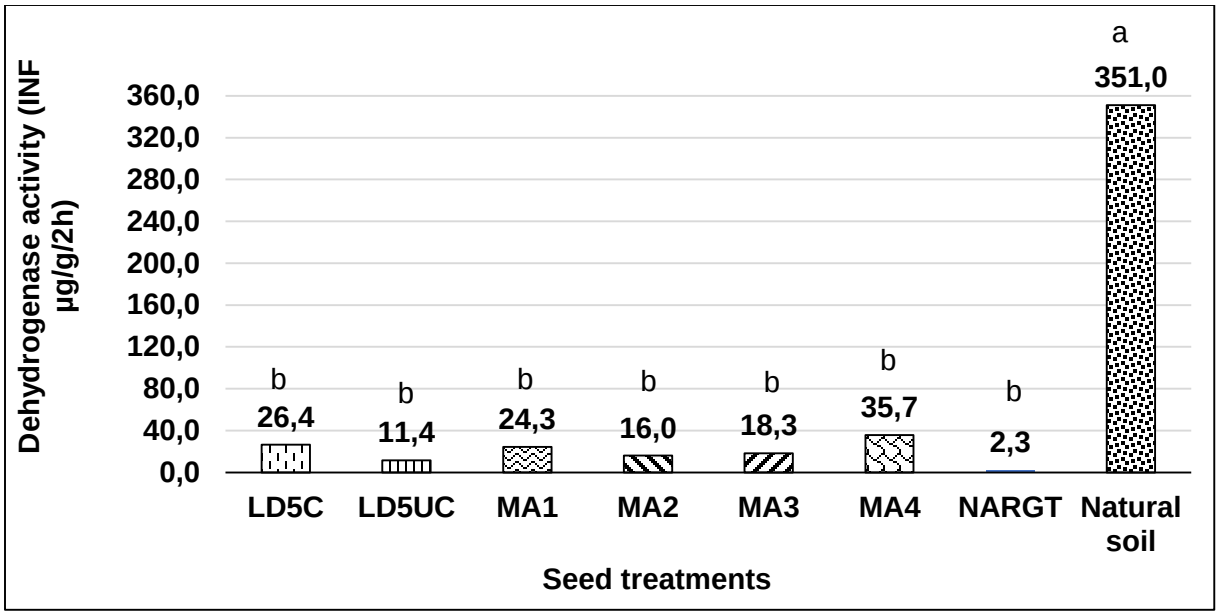

Figure 4.68: Dehydrogenase activity (INF µg/g/2h) of bio-stimulant trial seed treatment plots (LD5C, LD5UC, MA1, MA2, MA3, MA4) compared to non-ameliorated Rooikraal gold mine tailings (NARGT) and natural control soil (See dehydrogenase activity (DHA) under Section 3.4.5). Statistical significance (p < 0,05) between groups are indicated above bars.

Table 4.47: One-way ANOVA results of DHA (INF µg/g/2h) for bio-stimulant treatment plots (LD5C, LD5UC, MA1, MA2, MA3, MA), non-ameliorated Rooikraal gold mine tailings (NARGT) and natural soil background sample (control soil) near the Rooikraal gold TSF site (see dehydrogenase activity (DHA) under Section 3.4.5).

Sample	DHA (INF µg/g/2h)
Control soil	351,0 a
MA4	35,7 b
LD5C	26,4 b
MA1	24,3 b
MA3	18,3 b
MA2	16,0 b
LD5UC	11,4 b
NARGT	2,3 b
p < 0,0001	
Significant:	Yes

Figure 4.69 illustrates the DHA of non-ameliorated bare tailings to the DHA of the bio-stimulant trial plots (MA1, MA1, MA2 and MA4) and the 5 kg/ha lower density seed trial plots (LD5C and LD5UC).

The only significant difference was between the DHA of the non-ameliorated gold mine tailings (2,3 INF µg/g/2h) that was significantly lower than the DHA of the bio-stimulant seed treatment 4 (MA1 – 35,7 INF µg/g/2h) (Figure 4.69 and Table 4.48).

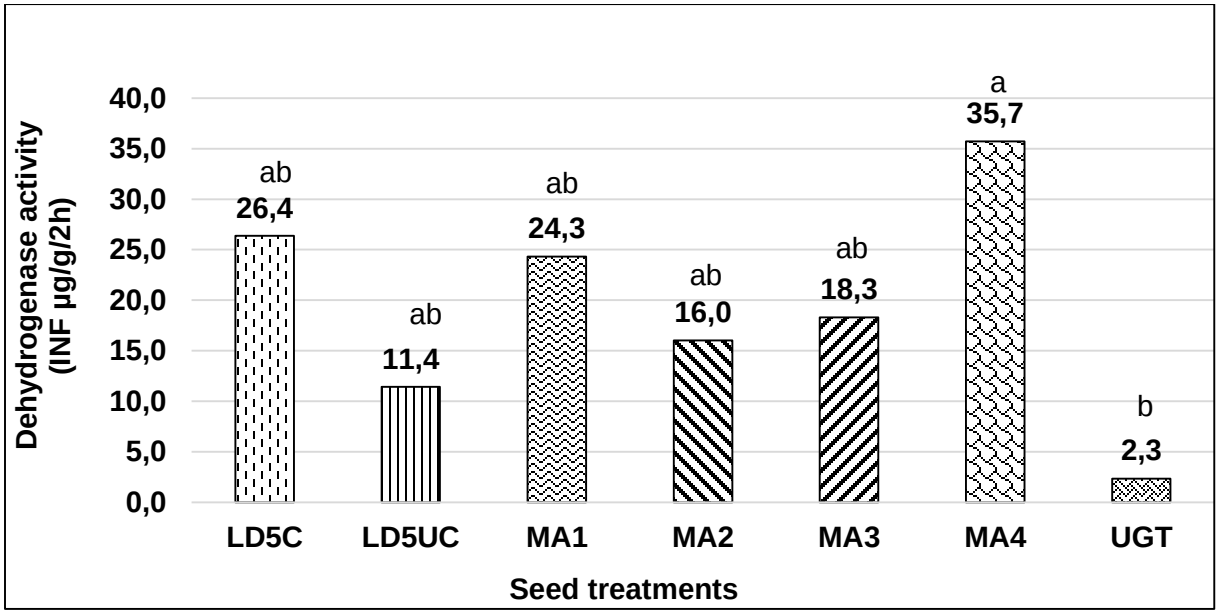

Figure 4.69: Dehydrogenase activity (INF µg/g/2h) of bio-stimulant trial seed treatments (LD5C, LD5UC, MA1, MA2, MA3, MA4) compared to non-ameliorated Rooikraal gold mine tailings (NARGT). Statistical significance ($p < 0,05$) between groups is indicated above bars.

Each of the seed trial plots did have a higher DHA than the non-ameliorated gold mine tailings, which indicates an improvement in the microbial population size after vegetation establishment. The DHA of the bio-stimulant seed treatment plots (MA1 – 24,3 INF µg/g/2h, MA2 – 16 INF µg/g/2h, MA3 – 18,3 INF µg/g/2h and MA4 – 35,7 INF µg/g/2h) all had a higher DHA measurement than the uncoated seed treatment LD5UC (11,4 INF µg/g/2h). However, the LD5C plots (26,4 INF µg/g/2h) had a higher DHA measurement than MA1, MA2 and MA3 (Figure 4.69 and Table 4.48).

Table 4.48: One-way ANOVA results of DHA (INF µg/g/2h) for bio-stimulant treatment plots (LD5C, LD5UC, MA1, MA2, MA3, MA4) and non-ameliorated Rooikraal gold mine tailings (NARGT). Statistical significance between groups ($p < 0,05$) is indicated by letters behind means.

Treatments	DHA (INF µg/g/2h)
MA4	35,4 a
LD5C	26,4 ab
MA1	24,3 ab
MA3	18,3 ab
MA2	16,0 ab
LD5UC	11,4 ab
NARGT	2,3 b
p = 0,035	
Significant:	Yes

In summary of the bio-stimulant trials, the microbial community is strained in non-ameliorated bare gold mine tailings, compared to natural soils. The establishment of vegetation did result in an increase in the microbial biomass of the treatment plots.

The bacterial bio-stimulant with a food source did not result in an increase in the seedling emergence, but both the seed treatments containing the *Trichoderma* bio-stimulant with a food source (MA2 and MA4) did result in a higher seedling emergence density compared to the untreated seed treatment LD5C and LD5UC if *E. tef* seedlings were excluded (Figure 4.67). This indicated that a *Trichoderma* bio-stimulant applied to the seed in conjunction with a food source, had the potential to increase grass establishment on gold TSFs.

Chapter 5: Conclusions and recommendations

5.1 Conclusions drawn from Phase 1 field trials

During the rehabilitation of mine TSF, seed from various tolerant grass species, namely, *E. tef, C. dactylon*, *D. eriantha*, *E. curvula* and *S. bicolor* in combination with a legume *M. sativa*, were sown to establish a vegetation cover on the TSF surface. Platinum and especially gold TSFs are hostile environments posing numerous obstacles to the establishment of a vegetation cover during rehabilitation. Coated seed that is enhanced by applying beneficial material such as growth stimulants, nutrients and pesticides to the seed exterior, offer a way to improve vegetation establishment on platinum and gold TSFs.

The trials conducted for this study evaluated the emergence and persistence of species used together within a seed treatment (T1C, T2UC, T3C, T4C and T5UC) when sown in mine TSF environments. Experiments were also carried out to test seedling emergence density when the seeding rate was decreased and whether the bio-stimulants applied to the seed will improve the establishment of vegetation.

5.1.1 The emergence and survival of seedlings when using coated and uncoated seed

The main aim of this study was to evaluate the establishment and growth of seedlings using coated and uncoated seed for the rehabilitation of gold and platinum TSFs.

The application of material to the seed exterior caused coated seed to be larger and heavier than uncoated seed. This resulted in fewer seed being sown in a seed treatment with coated seed than a treatment containing the same weight with uncoated seed. It was hypothesised that an increase in the seeding rate of coated seed would result in higher species emergence and survival.

In the Phase 1 field trials, there were no significant differences between the emerged seedling density (plants/m^2) of coated seed treatments T1C & T4C compared to uncoated seed treatments T2UC & T5UC using the same seeding rate, at the Crown gold TSF site, the Rooikraal gold TSF site and in the platinum tailings bulk bags. There was no significant increase in seedling emergence when sown with T3C, compared to the density of seedling emergence when sown with T3UC, even if the same amount of seed was used in both treatments at all three sites. However, when considering the percentage of seedlings that emerged from the uncoated seed

treatments T2UC & T5UC, as well as the increased seeding rate in the T3C, the seedling emergence percentages were lower than in the T1C & T4C at the three sites.

The density (plants/m^2) of surviving plants six months after seeding treatments were sown was not significantly different between coated and uncoated seed treatments T1C & T4C and T2UC & T5UC when using the same species and seeding rate. An increase in the seeding rate of T3C when the same number of seed as in the T2UC was sown, did not result in a higher surviving plant density at the two gold TSF sites and in the platinum tailings. However, the survival percentage of seedlings in the T1C and T4C was significantly higher compared to T2UC and T5UC. The surviving percentage of seedlings in T1C & T4C was also significantly higher at the Crown gold TSF site, the Rooikraal gold TSF site and in the platinum tailings than T3C, which contained the same amount of coated seed as T2UC.

Therefore, an increase in the seeding rate of coated seed did not result in higher seedling emergence, even though the coated seed treatments contained a smaller number of seed. When uncoated seed was used, it still provided the same emergence results as coated seed. This indicated that coated seed treatments were more efficient than uncoated seed treatments, since a higher percentage of the total seed sown was used while still providing the same establishment results.

5.1.2 Change in density and cover contribution of species used within the seed treatments

The change in the plant composition and cover contribution of species sown in the seed treatments at the different sites are discussed in this section.

- ***Cynodon dactylon***

Plants of *C. dactylon* were continuously present throughout the trial period on both the Crown and Rooikraal gold TSF sites and the platinum trials. This meant that this species is one of the most successful species contributing to the vegetation cover after *E. curvula.* The growth medium did have an influence on the emergence of *C. dactylon*, with lower emergence being recorded at the Crown gold mine tailings compared to the Rooikraal gold mine tailings and the platinum tailings. The growth medium and the seed type also did not have an influence on the growth (height and width index) of *C. dactylon* plants. *C. dactylon* was also one of the species that survived the burial that took place at the Rooikraal gold TSF site due to the heavy storm and erosion thereafter. This could be due to the creeping nature of the stolons and rhizomes enabling *C. dactylon* plants to

grow under soil and overhanging plant material to seek out opportunities and space to grow in adverse places and conditions.

- ***Digitaria eriantha***

At both the Crown and Rooikraal gold TSF sites, as well as the platinum trials, *D. eriantha* was the perennial grass species least present in the plant composition and therefore contributing the least vegetation cover when compared to the cover of *E. curvula* and *C. dactylon*. The pot trials also revealed a low seedling emergence of *D. eriantha* (< 20% in Crown gold and platinum tailings and ≤ 30% in Rooikraal gold mine tailings) with both coated and uncoated seed. The seedling emergence when using uncoated seed was also higher than when using coated seed in each of the growth mediums. Neither the growth medium nor the seed type had an influence on the growth (height and width index) of *D. eriantha*. The low emergence percentage during the pot and the field trials could be attributed to the low viable seed percentage of 22% normal seedlings; see Figure 3.11 in Section 3.4.1 of the coated and uncoated *D. eriantha* seed batches.

- ***Eragrostis curvula***

At both the Crown and Rooikraal gold TSF sites, as well as the platinum trials, *E. curvula* was the dominant and most successful species providing most of the vegetation cover throughout the trial period. The growth medium did not have an influence on *E. curvula* seedling emergence. The seedling emergence was higher when using coated seed during the first week of the trial. There was also no influence of the growth medium nor the seed type (coated and uncoated) on the growth (height and width index) of *E. curvula*. This indicated a possible benefit of coated seed for the emergence of *E. curvula* seedlings. *E. curvula* plants were also likely to survive due to the burial of the plants after the erosion caused by the heavy storm, as the culms and leaves were still growing and protruding from the tailing surface. This might indicate that the use of *E. curvula* would be an advantage when using it in rehabilitation of gold TSFs.

- ***Eragrostis tef***

In the Crown and Rooikraal gold TSF sites, the annual *E. tef* was present at its highest density in June and this species accounted for the largest part of the vegetation cover before decreasing and disappearing in November 2016. The Phase 2 pot trials confirm a rapid uniform emergence of *E. tef* with no influence of the tailings material. In the platinum bulk bags Phase 1 field trials, *E. tef* had a lower plant density and cover, which could be explained by the lower temperatures during May and June 2016 (Baskin & Baskin, 1998:41). The Phase 2 pot trials support this finding, since *E. tef* seed had an emergence above 55% (see Section 4.4.1, Figure 4.55) in the platinum tailings during November 2016. Again no influence was determined for the three different growth

mediums on the seedling emergence or growth (height and width index) of *E. tef*. The purpose of *E. tef* is to serve as a nurse crop, which emerges quickly to provide good cover in the first season before diminishing and allowing perennial species, such as *E. curvula*, *D. eriantha* and *C. dactylon* to establish and dominate in following seasons (Coaltech research association and Chamber of Mines, 2007:117). According to guideline set out by Coaltech research association and Chamber of Mines (2007:118), *E. tef* can easily outcompete other perennial species and negatively influence its establishment. This was however not the case in the seed treatments on the gold TSF sites. A seeding rate of 1 kg/ha was sufficient to provide good initial cover and seedling establishment without hindering the establishment of succeeding perennials such as *E. curvula* and *C. dactylon*.

- ***Sorghum bicolor***

S. bicolor had the highest establishment density at the Rooikraal gold TSF site but decreased at both the gold TSF sites over time. In the platinum bulk bags, this species did not establish well, most likely due to seeding that was only carried out in April 2016, as it was already past the warm temperature months that are needed for the germination and growth of *S. bicolor* (Jones, 1985:152).

- ***Medicago sativa***

Seedlings of *M. sativa* did not establish well at the Rooikraal and Crown gold TSF sites, especially not after November 2016. An alternative species to use for the rehabilitation of these more acid and low fertility situations could be *Lespedeza cuneate* (Coaltech research association and Chamber of Mines, 2007:118). In the platinum tailing bulk bag trials that were irrigated, *M. sativa* established and grew very well, even after being cut in November 2016. This species even showed a good regrowth within a period of two months after cutting, indicating that it could be used for hay making if established under irrigation conditions.

5.1.3 Lower seeding densities

A decrease in seeding rate did not result in a significantly lower density of emerged seedlings, but did result in a higher emergence percentage of seedlings when coated seed and uncoated seed were used. This denied the hypothesis that a decrease in the seeding rate would result in lower seedling establishment densities, which indicated that similar emergence results could be obtained using less seed for both coated and uncoated seed mixtures.

5.1.4 Bio-stimulant seed treatments

There is much literature supporting the application of bio-stimulants containing strains of beneficial bacteria and fungi to seed that will improve seedling establishment and plant growth (Burges *et al*., 2016:481; Du Jardin, 2015:7; Grandlic *et al.,* 2009:1740; Harmosa *et al.*, 2012:17; Khan, 2005:357, Ranasingh *et al*., 2006; Saba *et al*., 2012:525).

The findings by the numerous scientists referenced above were confirmed by measuring the dehydrogenase activity. The non-ameliorated Rooikraal gold mine tailings have a very strained microbial community when compared to a natural soil in the environment that was taken as the control. The application of bio-stimulants to the seeds and soil did not result in a significantly higher DHA measurement.

Although not significant, seed treatments containing the *Trichoderma* fungi with a food source did however result in an increased seedling density. This indicated that a *Trichoderma* fungal application in combination with a food source does have potential to increase the seedling emergence of perennial grass species such as *E. curvula, D. eriantha* and *C. dactylon.* However, further investigation is required to understand the species-specific association of beneficial fungi with grass species used within the seed treatment for rehabilitation of TSFs.

5.1.5 Summary of conclusions

- An increase in the seeding rate of coated seed did not result in higher seedling emergence.
- The creeping nature of the stolons and rhizomes of *C. dactylon* and the tall culms of *E. curvula* are traits which make these species very suitable for the rehabilitation of gold TSFs.
- A decrease in seeding rate did not result in a significantly lower density of emerged seedlings.
- Gold mine tailings have a very strained microbial community when compared to a natural soil.
- The application of bio-stimulants did not result in a signififcant increased seedling emergence.

5.2 Recommendations regarding the use of coated seed mixtures for gold and platinum TSF rehabilitation

Commercially available coated seed mixtures provide an advantage to uncoated seed mixtures for the rehabilitation of gold and platinum TSFs because a larger percentage of seed sown establishment occurs when compared to uncoated seed treatments. The research of this study therefore suggests that coated seed is more efficient than uncoated seed and it is recommended that this be researched in future studies.

E. tef is characterised by quick uniform emergence within one week of seeding in both gold and platinum tailings, therefore providing initial cover and protection for succeeding perennial species to establish. *E. curvula* and *C. dactylon* were the most successful perennial grass species persisting throughout the trial period and contributing the greatest portion vegetation cover in both gold and platinum tailings.

It is recommended that the seed viability of seed batches are verified by an International Seed Testing Association (ISTA) accredited seed testing laboratory before used for rehabilitation activities. *D. eriantha* had the lowest establishment rates of the perennial grass species due to the very low viable seed percentage of 22% compared to the other species that have viable seed percentages above 70%. This explains the necessity to test the seed viability by an ISTA-accredited laboratory.

M. sativa did not establish well on the gold TSFs, but had very good establishment on the growth medium from the platinum TSF. Proper irrigation methods should be implemented at the TSF site if *M. sativa* is to be propagated as a legume N fixating legume for rehabilitation. *M. sativa* may possibly also be harvested on platinum tailings and used as hay if grown under irrigation. Further research is suggested regarding the water.

Under dry land conditions (where no irrigation is used), climate plays a key role in the establishment of seed treatments on TSF sites for the purpose of rehabilitation. Seed treatments should be sown when climatic conditions are most favourable during or shortly after the first summer rains when temperatures are warm enough to drive seedling emergence and growth. Seeds sown in March 2016 at the Crown and Rooikraal gold TSF sites provided good establishment results. In the following year at the Rooikraal gold TSF site, seedling emergence densities sown in January 2017 exceeded seedling emergence densities of seeds sown in March 2016. On the growth medium from the platinum TSF, *E. tef* and *S. bicolor* seedlings had poor

emergence densities. It is therefore recommended to sow seed treatments during the warm summer months after October, but not later than March.

5.3 Suggestions for further research

During the seeding trials in the natural environment on the TSFs, the plots were ameliorated with lime, compost and fertiliser to improve the plant growth. The benefit of coated seed is that the seed is “enhanced” with its own supply of nutrients, growth stimulants and pesticides surrounding the seed exterior. This should enable the seed to establish better in the TSF material with fewer fertiliser inputs, which should cut down the costs of soil amelioration during rehabilitation. The extent to which the seed coating is able to substitute fertiliser inputs is not known. Therefore, vegetation trials evaluating the emergence and establishment of coated seed in trials where the amelioration of trial plots before seeding are restricted, i.e. lower fertiliser inputs should be investigated to be able to better understand the benefit of enhancing seed by coating.

Fungicides were added to the coating formula to protect seed from fungal attack (Weideman, 2012). There is much literature promoting the use of *Trichoderma* to act as a fungicide (Du Jardin, 2015:7; Kaveh *et al*., 2011:169; Ranasignh *et al*., 2006). The application of *Trichoderma* as a fungal bio-stimulant to the coating formula is potentially able to serve a double purpose as a pesticide and a plant growth promoter. The substitution of chemical fungicides with bio-fungicides warrants further investigation.

Chloris guayana commonly known as Rhodes grass is a weak perennial subclimax tufted grass that spreads by means of stolons and is a well regarded soil stabilising species (Van Oudtshoorn, 2004:230). It was not included in this study and it is recommended that future studies should include this species.

It is further recommended that the trials carried out for this research be done over a longer time span on more TSFs and to evaluate the persistence of the grass species for the rehabilitation of TSFs over the long term.

Chapter 6: Bibliography

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Chapter 7: Annexures

Annexure A: Seed analysis results of uncoated *Cynodon dactylon*

SEED TEST LABORATORY - Advance Seed
PO Box 414, Krugersdorp 1740
Tel: 011-762 5261
Fax: 011-762 4111

as registered with the **National Department of Agriculture's ISTA Accredited Seed Testing Station** in terms of the Plant Improvement Act, 1976 (Act 53 of 1976)
Reg. No. 94/01260/07
DIV. Seed Control Reg. No. 68050001

ANALYSIS OF SEED SAMPLE

INFORMATION AS STATED BY SENDER:

Kind and Variety: Cynodon **Code Number: 802**
Botanical Name: Cynodon dactylon **Reference Number**: A708/15
Date sample received: 2015/10/22
Weight of submitted sample(gram) 1264.0gr

PHYSICAL PURITY (% Calculated by Mass)				GERMINATION (% Calculated by Number)					
Pure seed	Inert matter	Other seeds	Other material (Total of 2 and 3)	Duration of test (days)	Normal seedlings	Hard seeds	Fresh seeds	Abnormal seedlings	Dead seeds
1	2	3	4	5	6	7	8	9	10
98.8	**1.2**	**0.0**	**1.2**	**21**	**79**	**0**	**0**	**7**	**14**

Inert matter: Broken seed, plant material

Germination method: TP:20<=>30°C

Remarks:

SEED TEST LABORATORY
P.O. BOX 414
KRUGERSDORP 1740
REGISTRATION No. 99650082
2016 -02- 19
SIG.:

Important: These results are applicable to the submitted sample and not necessarily to the seed lot from which the sample was taken. The percentage seed indicated in column 1 may include seeds from other species not visually distinguishable from the seed kind indicated against the botanical name.

Annexure B: Seed analysis results of coated *Cynodon dactylon* seed

SEED TEST LABORATORY - Advance Seed
PO Box 414, Krugersdorp 1740
Tel: 011-762 5261
Fax: 011-762 4111

as registered with the **National Department of Agriculture's ISTA Accredited Seed Testing Station** in terms of the Plant Improvement Act, 1976 (Act 53 of 1976)

Reg. No. 94/01260/07
DIV. Seed Control Reg. No. 68050001

ANALYSIS OF SEED SAMPLE

INFORMATION AS STATED BY SENDER:

Kind and Variety: Cynodon
Botanical Name: Cynodon dactylon
Date sample received: 2015/10/26
Weight of submitted sample(gram) 785.0gr

Code Number: 802E
Reference Number: 221015

PHYSICAL PURITY (% Calculated by Mass)				GERMINATION (% Calculated by Number)					
Pure seed	Inert matter	Other seeds	Other material (Total of 2 and 3)	Duration of test (days)	Normal seedlings	Hard seeds	Fresh seeds	Abnormal seedlings	Dead seeds
1	2	3	4	5	6	7	8	9	10
X	**X**	**X**	**X**	**21**	**-75-**	**-0-**	**-0-**	**-7-**	**-18-**

Inert matter:

Germination method: TP:20<=>30°C

Remarks: COATED SEED

SEED TEST LABORATORY
P.O. BOX 414
KRUGERSDORP 1740
REGISTRATION No. 09650082
2016 -02- 19
SIG.

Important: These results are applicable to the submitted sample and not necessarily to the seed lot from which the sample was taken. The percentage seed indicated in column 1 may include seeds from other species not visually distinguishable from the seed kind indicated against the botanical name.

Annexure C: Seed analysis results of uncoated *Digitaria eriantha* seed.

SEED TEST LABORATORY - Advance Seed
PO Box 414, Krugersdorp 1740
Tel: 011-762 5261
Fax: 011-762 4111

as registered with the **National Department of Agriculture's ISTA Accredited Seed Testing Station** in terms of the Plant Improvement Act, 1976 (Act 53 of 1976)

Reg. No. 94/01260/07
DIV. Seed Control Reg. No. 68050001

ANALYSIS OF SEED SAMPLE

INFORMATION AS STATED BY SENDER:

Kind and Variety: Smuts digitaria Irene
Botanical Name: Digitaria eriantha
Date sample received: 2016/02/11
Weight of submitted sample(gram) 454.3gr

Code Number: 873
Reference Number: A121/16

PHYSICAL PURITY (% Calculated by Mass)				GERMINATION (% Calculated by Number)					
Pure seed	Inert matter	Other seeds	Other material (Total of 2 and 3)	Duration of test (days)	Normal seedlings	Hard seeds	Fresh seeds	Abnormal seedlings	Dead seeds
1	2	3	4	5	6	7	8	9	10
96.5	3.0	0.5	3.5	14	22	0	0	1	77

Inert matter: Broken seed, plant material, stalks, sand

Weed: 0.5% 6 x Sporobolus sp. 5 x Urochloa sp. 6 x Verbena officinalis

Germination method: TP:20<=>30°C

Remarks:

SEED TEST LABORATORY
P.O. BOX 414
KRUGERSDORP 1740
REGISTRATION No. 09650062
2016 -02- 23
SIG.:

Important: These results are applicable to the submitted sample and not necessarily to the seed lot from which the sample was taken. The percentage seed indicated in column 1 may include seeds from other species not visually distinguishable from the seed kind indicated against the botanical name.

Annexure D: Seed analysis results of coated *Digitaria eriantha* seed.

SEED TEST LABORATORY - Advance Seed
PO Box 414, Krugersdorp 1740
Tel: 011-762 5251
Fax: 011-762 4111

as registered with the **National Department of Agriculture's ISTA Accredited Seed Testing Station** in terms of the Plant Improvement Act, 1976 (Act 53 of 1976)

Reg. No. 94/01260/07
DIV. Seed Control Reg. No. 68050001

ANALYSIS OF SEED SAMPLE

INFORMATION AS STATED BY SENDER:

Kind and Variety: Smuts digitaria Irene
Botanical Name: Digitaria eriantha
Date sample received: 2016/02/10
Weight of submitted sample(gram) 540.6gr

Code Number: 873E
Reference Number: 030216

PHYSICAL PURITY (% Calculated by Mass)				GERMINATION (% Calculated by Number)					
Pure seed	Inert matter	Other seeds	Other material (Total of 2 and 3)	Duration of test (days)	Normal seedlings	Hard seeds	Fresh seeds	Abnormal seedlings	Dead seeds
1	2	3	4	5	6	7	8	9	10
-X-	-X-	-X-	-X-	14	-22-	-0-	-0-	-1-	-77-

Inert matter:

Germination method: TP:20<=>30°C

Remarks: **COATED SEED**

SEED TEST LABORATORY
P.O. BOX 414
KRUGERSDORP 1740
REGISTRATION No. 09550082
2016 -02- 23
SIG.

Important: These results are applicable to the submitted sample and not necessarily to the seed lot from which the sample was taken. The percentage seed indicated in column 1 may include seeds from other species not visually distinguishable from the seed kind indicated against the botanical name.

Annexure E: Seed analysis results of coated *Sorghum bicolor* seed

SEED TEST LABORATORY - Advance Seed
PO Box 414, Krugersdorp 1740
Tel: 011-762 5261
Fax: 011-762 4111

as registered with the National Department of Agriculture's ISTA Accredited Seed Testing Station in terms of the Plant Improvement Act, 1976 (Act 53 of 1976)
Reg. No. 94/01260/07
DIV. Seed Control Reg. No. 68050001

ANALYSIS OF SEED SAMPLE

INFORMATION AS STATED BY SENDER:

Kind and Variety: Forage Sorghum Supergraze
Botanical Name: Sorghum spp.
Date sample received: 2016-02-12
Weight of submitted sample (gram) 1465 gram

Code Number: 787/32
Reference Number: A156/16

PHYSICAL PURITY (% Calculated by Mass)				GERMINATION (% Calculated by Number)					
Pure seed	Inert matter	Other seeds	Other material (Total of 2 and 3)	Duration of test (days)	Normal seedlings	Hard seeds	Fresh seeds	Abnormal seedlings	Dead seeds
1	2	3	4	5	6	7	8	9	10
99.9	0.1	0.0	0.1	10	88	0	0	7	5

Inert matter: Broken seed

Germination method: BP:20<=>30°C

Remarks:

SEED TEST LABORATORY
P.O. BOX 414
KRUGERSDORP 1740
REGISTRATION No. 09650082
2016 -02- 23
SIG.

Important: These results are applicable to the submitted sample and not necessarily to the seed lot from which the sample was taken. The percentage seed indicated in column 1 may include seeds from other species not visually distinguishable from the seed kind indicated against the botanical name.

Annexure F: Seed analysis results of coated *Medicago sativa* seed

SEED TEST LABORATORY - Advance Seed
PO Box 414, Krugersdorp 1740
Tel: 011-762 5261
Fax: 011-762 4111

as registered with the **National Department of Agriculture's ISTA Accredited Seed Testing Station** in terms of the Plant Improvement Act, 1976 (Act 53 of 1976)

Reg. No. 94/01260/07
DIV. Seed Control Reg. No. 68050001

ANALYSIS OF SEED SAMPLE

INFORMATION AS STATED BY SENDER:

Kind and Variety: Lucerne Supercuf
Botanical Name: Medicago sativa
Date sample received: 2016/02/16
Weight of submitted sample (gram) 1136gr

Code Number: 675E
Reference Number: 160216

PHYSICAL PURITY (% Calculated by Mass)				GERMINATION (% Calculated by Number)					
Pure seed	Inert matter	Other seeds	Other material (Total of 2 and 3)	Duration of test (days)	Normal seedlings	Hard seeds	Fresh seeds	Abnormal seedlings	Dead seeds
1	2	3	4	5	6	7	8	9	10
-X-	-X-	-X-	-X-	10	-76-	-7-	-0-	-9-	-8-

Inert matter:

Germination method: TP:20<=>30°C

Remarks: COATED

SEED TEST LABORATORY
P.O. BOX 414
KRUGERSDORP 1740
REGISTRATION No. 09650082
2016 -02- 29
SIG.

Important: These results are applicable to the submitted sample and not necessarily to the seed lot from which the sample was taken. The percentage seed indicated in column 1 may include seeds from other species not visually distinguishable from the seed kind indicated against the botanical name.

Annexure G: Seed analysis results of uncoated *Sorghum bicolor* seed

SEED TEST LABORATORY - Advance Seed
PO Box 414, Krugersdorp 1740
Tel: 011-762 5261
Fax: 011-762 4111

as registered with the **National Department of Agriculture's ISTA Accredited Seed Testing Station** in terms of the Plant Improvement Act, 1976 (Act 53 of 1976)

Reg. No. 94/01260/07
DIV. Seed Control Reg. No. 68050001

ANALYSIS OF SEED SAMPLE

INFORMATION AS STATED BY SENDER:

Kind and Variety: Forage Sorghum SSG1000
Botanical Name: Sorghum spp.
Date sample received: 2016-02-18
Weight of submitted sample (gram) 1333 gram

Code Number: 787/35
Reference Number: A164/16

PHYSICAL PURITY (% Calculated by Mass)				GERMINATION (% Calculated by Number)					
Pure seed	Inert matter	Other seeds	Other material (Total of 2 and 3)	Duration of test (days)	Normal seedlings	Hard seeds	Fresh seeds	Abnormal seedlings	Dead seeds
1	2	3	4	5	6	7	8	9	10
-100-	-TR-	-0.0-	-TR-	10	-82-	-0-	-0-	-10-	-8-

Inert matter: Broken seed

Germination method: BP:20<=>30°C

Remarks:

SEED TEST LABORATORY
P.O. BOX 414
KRUGERSDORP 1740
REGISTRATION No. 09650082
2016 -02- 29
SIG.:

Important: These results are applicable to the submitted sample and not necessarily to the seed lot from which the sample was taken. The percentage seed indicated in column 1 may include seeds from other species not visually distinguishable from the seed kind indicated against the botanical name.

Annexure H: Seed analysis results of coated *Eragrostis curvula* seed

SEED TEST LABORATORY - Advance Seed
PO Box 414, Krugersdorp 1740
Tel: 011-762 5261
Fax: 011-762 4111

as registered with the **National Department of Agriculture's ISTA Accredited Seed Testing Station** in terms of the Plant Improvement Act, 1976 (Act 53 of 1976)

Reg. No. 94/01260/07
DIV. Seed Control Reg. No. 68050001

ANALYSIS OF SEED SAMPLE

INFORMATION AS STATED BY SENDER:

Kind and Variety: Eragrostis Ermelo
Botanical Name: Eragrostis curvula
Date sample received: 2016-02-08
Weight of submitted sample(gram): 775.7 gram

Code Number: 769/1E
Reference Number:010216

PHYSICAL PURITY (% Calculated by Mass)				GERMINATION (% Calculated by Number)					
Pure seed	Inert matter	Other seeds	Other material (Total of 2 and 3)	Duration of test (days)	Normal seedlings	Hard seeds	Fresh seeds	Abnormal seedlings	Dead seeds
1	2	3	4	5	6	7	8	9	10
X	**X**	**X**	**X**	**10**	**-72-**	**-0-**	**-0-**	**-3-**	**-25-**

Inert matter:
Weed:

Germination method: TP:20<=>30°C

Remarks:

SEED TEST LABORATORY
P.O. BOX 414
KRUGERSDORP 1740
REGISTRATION No. 09650062

2016 -02- 19

SIG.:

Important: These results are applicable to the submitted sample and not necessarily to the seed lot from which the sample was taken. The percentage seed indicated in column 1 may include seeds from other species not visually distinguishable from the seed kind indicated against the botanical name.

Annexure I: Seed analysis results of uncoated *Eragrostis curvula* seed

SEED TEST LABORATORY - Advance Seed
PO Box 414, Krugersdorp 1740
Tel: 011-762 5281
Fax: 011-762 4111

as registered with the National Department of Agriculture's ISTA Accredited Seed Testing Station in terms of the Plant Improvement Act, 1976 (Act 53 of 1976)
Reg. No. 94/01260/07
DIV. Seed Control Reg. No. 68050001

ANALYSIS OF SEED SAMPLE

INFORMATION AS STATED BY SENDER:

Kind and Variety: Eragrostis Ermelo
Botanical Name: Eragrostis curvula
Date sample received: 2015-08-26
Weight of submitted sample(gram): 741.5 gram

Code Number: 769/1
Reference Number:A514/15

PHYSICAL PURITY (% Calculated by Mass)				GERMINATION (% Calculated by Number)					
Pure seed	Inert matter	Other seeds	Other material (Total of 2 and 3)	Duration of test (days)	Normal seedlings	Hard seeds	Fresh seeds	Abnormal seedlings	Dead seeds
1	2	3	4	5	6	7	8	9	10
-99.5-	**-0.4-**	**-0.1-**	**-0.5-**	**10**	**-78-**	**-0-**	**-0-**	**-2-**	**-20-**

Inert matter: Broken caryopsis, sand, chaff
Weed: 0.1% (4) Eragrostis plana

Germination method: TP:20<=>30°C

Remarks:

SEED TEST LABORATORY
P.O. BOX 414
KRUGERSDORP 1740
REGISTRATION No. 09650082
2015 -02- 19
SIG.:

Important: These results are applicable to the submitted sample and not necessarily to the seed lot from which the sample was taken. The percentage seed indicated in column 1 may include seeds from other species not visually distinguishable from the seed kind indicated against the botanical name.

Annexure J: Seed analysis results of uncoated *Eragrostis tef* seed

SEED TEST LABORATORY
AGT Foods Africa (Pty) Ltd
PO Box 414, Krugersdorp 1740
Tel: +2711-762 5261
Fax: +2711-762 4111

as registered with the National Department of Agriculture's ISTA Accredited Seed Testing Station in terms of the Plant Improvement Act, 1976 (Act 53 of 1976)
Reg. No. 1994/001269/07
DIV. Seed Control Reg. No. 68050001

ANALYSIS OF SEED SAMPLE

INFORMATION AS STATED BY SENDER:

Kind and Variety: Teff SA Brown
Botanical Name: Eragrostis tef
Date sample received: 2016-03-10
Weight of submitted sample (gram):1713gram

Code Number: 744
Reference Number: A359/15

PHYSICAL PURITY (% Calculated by Mass)				GERMINATION (% Calculated by Number)					
Pure seed	Inert matter	Other seeds	Other material (Total of 2 and 3)	Duration of test (days)	Normal seedlings	Hard seeds	Fresh seeds	Abnormal seedlings	Dead seeds
1	2	3	4	5	6	7	8	9	10
-99.4-	**-0.4-**	**-0.2-**	**-0.6-**	**10**	**-85-**	**-0-**	**-0-**	**-2-**	**-13-**

Inert matter: Broken caryopsis, soil
Weed: (4) Cyperus sp. (3) Amaranthus sp.

Germination method: TP: 20<=>30°C :KNO_3

Remarks:

SEED TEST LABORATORY
P.O. BOX 414
KRUGERSDORP 1740
REGISTRATION No. 09650082
2016 -04- 06
SIG.:

Important: These results are applicable to the submitted sample and not necessarily to the seed lot from which the sample was taken. The percentage seed indicated in column 1 may include seeds from other species not visually distinguishable from the seed kind indicated against the botanical name.

Annexure K: Summary of results obtained during Phase 2 pot trials

Seedling emergence percentage

Cynodon dactylon

Growth medium	Seed type	Sampling date 8/11/2016	11/11/2016	21/11/2016	28/11/2016
RK	Coated	2,5	40,0	46,3	43,8
RK	Uncoated	5,0	23,8	30,0	37,5
CRWN	Coated	0,0	11,3	25,0	28,8
CRWN	Uncoated	0,0	25,0	28,8	27,5
PT	Coated	1,3	36,3	50,0	50,0
PT	Uncoated	0,0	30,0	47,5	47,5
CTRL	Coated	0,0	3,8	5,0	6,3
CTRL	Uncoated	1,3	26,3	30,0	31,3

Digitaria eriantha

Growth medium	Seed type	Sampling date 8/11/2016	11/11/2016	21/11/2016	28/11/2016
RK	Coated	5,0	8,8	8,8	11,3
RK	Uncoated	13,8	30,0	26,3	30,0
CRWN	Coated	0,0	2,5	1,3	3,8
CRWN	Uncoated	0,0	7,5	7,5	6,3
PT	Coated	1,3	7,5	5,0	5,0
PT	Uncoated	1,3	16,3	13,8	13,8
CTRL	Coated	0,0	2,5	5,0	5,0
CTRL	Uncoated	3,8	13,8	13,8	15,0

Eragrostis curvula

Growth medium	Seed type	Sampling date 8/11/2016	11/11/2016	21/11/2016	28/11/2016
RK	Coated	33,8	51,3	57,5	53,8
RK	Uncoated	5,0	52,5	50,0	53,8
CRWN	Coated	1,3	32,5	41,3	35,0
CRWN	Uncoated	1,3	40,0	37,5	42,5
PT	Coated	22,5	45,0	43,8	43,8
PT	Uncoated	12,5	37,5	43,8	42,5
CTRL	Coated	16,3	47,5	47,5	43,8
CTRL	Uncoated	18,8	46,3	48,8	50,0

Eragrostis tef

Growth medium	sampling date 8/11/2016	11/11/2016	21/11/2016	28/11/2016
RK	65,0	63,8	61,3	60,0
CRWN	53,8	61,3	60,0	56,3
PT	47,5	53,8	55,0	52,5
CTRL	58,8	60,0	58,8	53,8

Height and width index

Cynodon dactylon

Growth medium	Seed type	Sampling date 12/01/2017	22/2/2017	26/03/2017
RK	Coated	140,9	265,8	265,7
RK	Uncoated	167,5	282,1	318,1
CRWN	Coated	234,8	206,1	325,3
CRWN	Uncoated	250,1	373,9	422,8
PT	Coated	236,7	237,1	302,2
PT	Uncoated	174,1	306,4	352,6
CTRL	Coated	67,2	161,7	197,6
CTRL	Uncoated	101,9	177,4	329,9

Digitaria eriantha

Growth medium	Seed type	Sampling date 12/01/2017	22/2/2017	26/03/2017
RK	Coated	129,4	293,5	337,8
RK	Uncoated	235,2	455,7	574,0
CRWN	Coated	148,0	487,1	575,2
CRWN	Uncoated	188,5	599,6	593,3
PT	Coated	489,4	633,5	1188,0
PT	Uncoated	522,8	563,3	507,0
CTRL	Coated	354,7	688,6	715,5
CTRL	Uncoated	167,0	465,9	464,2

Eragrostis curvula

Growth medium	Seed type	Sampling date 12/01/2017	22/2/2017	26/03/2017
RK	Coated	187,7	265,9	308,9
RK	Uncoated	243,6	374,8	414,2
CRWN	Coated	179,1	221,6	281,0
CRWN	Uncoated	194,8	318,7	328,8
PT	Coated	373,7	414,4	534,4
PT	Uncoated	196,0	300,1	250,5
CTRL	Coated	190,8	207,1	233,3
CTRL	Uncoated	176,7	234,8	292,8

Eragrostis tef

Growth medium	Sampling date 12/01/2017	22/2/2017	26/03/2017
RK	132,6	139,5	167,7
CRWN	122,2	118,0	145,5
PT	133,0	155,1	129,1
CTRL	108,0	130,8	125,4

Biomass

Cynodon dactylon

Growth medium	Seed type	DM (g/m²)
RK	Coated	465,0
RK	Uncoated	426,2
CRWN	Coated	527,8
CRWN	Uncoated	1068,9
PT	Coated	551,9
PT	Uncoated	523,0
CTRL	Coated	551,9
CTRL	Uncoated	523,0

Digitaria eriantha

Growth medium	Seed type	DM (g/m²)
RK	Coated	272,7
RK	Uncoated	344,8
CRWN	Coated	233,6
CRWN	Uncoated	357,1
PT	Coated	499,0
PT	Uncoated	542,5
CTRL	Coated	380,4
CTRL	Uncoated	523,7

Eragrostis curvula

Growth medium	Seed type	DM (g/m²)
RK	Coated	471,7
RK	Uncoated	468,5
CRWN	Coated	283,1
CRWN	Uncoated	482,4
PT	Coated	996,9
PT	Uncoated	416,0
CTRL	Coated	392,6
CTRL	Uncoated	421,4

Eragrostis tef

Growth medium	DM (g/m²)
RK	633,0
CRWN	801,7
PT	742,7
CTRL	820,9