The use of compost and recycled aggregates in the treatment of runoff pollutants in vegetated sustainable drainage devices such as swale

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Volume I

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Abstract

Urbanisation, a process associated with industrialisation and development has been characterised by unsustainable impacts such as increased impervious surfaces, increased air pollution, increased use of natural resources, increased volume of surface run-off, decreased quality of surface run-off, and depletion of biodiversity and habitats. The effects of these impacts on the environment include climate change, flooding, erosion, pollution of water bodies, and destruction of aquatic life and biodiversity. Studies have shown that sustainable designs such as Sustainable Drainage Systems (SuDS) would help mitigate some of these effects sustainably. SuDS are natural drainage systems that simulate the natural drainage of a site/catchment and work in harmony to achieve increase in ground infiltration and treatment of runoff; and reduction in flow rates and volume of surface runoff, thereby improving storm water quality, reducing erosion, recharging groundwater, improving biodiversity and ultimately improving sustainability. However, sustainability of SuDS devices are questionable because their component parts involve the use of natural resources i.e. topsoil and gravel.

The overall aim of this research was to evaluate the efficacy of the application of recycled/waste materials in performing at least as well as topsoil and gravel in vegetative SuDS, thereby improving water quality and overall sustainability. The materials applied were compost and recycled aggregates. In assessing their efficacy in vegetative SuDS, the risk these materials could pose to water quality was not overlooked but was considered in establishing an ideal model for the treatment of pollutants in vegetative SuDS.

Results of this research showed that overall compost and recycled aggregates were able to perform at least as well as gravel and topsoil in vegetative SuDS in terms of characterisation, biofilm and vegetative development, and remediation of runoff pollutants thereby improving the sustainability of vegetative SuDS. Compared to gravel and topsoil, characterisation of compost and recycled aggregates was shown to be less expensive, less time consuming (except for recycled aggregates) and more sustainable, in terms of conserving natural resources. It was deduced that compost would be able to biodegrade organic pollutants in vegetative SuDS in varying conditions, compared to topsoil, thereby improving water quality.

Vegetative growth in profiles containing compost were more prolific than those with topsoil alone, indicating that vegetative SuDS containing compost would attenuate stormwater and remediate pollutants by phytoremediation, better than topsoil. Results showed that compost and recycled aggregates performed as well as gravel and topsoil in remediating pollutants, with >98% of pollutants being retained mostly within the growth media, confirming that most pollutants are treated within the growth media of vegetative SuDS devices.

This research was able to establish that SuDS components can be as unsustainable as components of conventional drainage systems in terms of their social, economic and environmental impacts; and that recycled materials could perform just as well as conventional materials, whilst improving their sustainability. This research further established that compost and recycled aggregates can be used in vegetative SuDS, such as swales, as literature has shown that the use of compost and recycled aggregates in vegetative SuDS has been limited to compost blankets and socks and substrates for green roofs. Suggestions for other waste materials that can be used instead topsoil and gravel in vegetative SuDS were also made. Results from this research were applied in the development of a swale model for the treatment of pollutants in vegetative SuDS.

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List of Abbreviations

AES – Atomic Emission Spectroscopy

ASR – Alkali-silica reactions

ASTM – American Society for Testing and Materials

ATSDR – Agency for Toxic Substances and Disease Registry

BBC – British Broadcasting Corporation

BIOECODS – Bio-Ecological Drainage Systems

BIS – British Standard Institute

BOD – biological oxygen demand

BRP – British Recycled Products

CALU – Centre for Alternative Land Use

CAST – Council for Agricultural Science and Technology

CDEW – Construction, demolition and excavation wastes

CFU – Colony forming unit

CIRIA – Construction Industry Research and Information Association

CSO – Combined sewer overflows

CSUGE – Compost Specification for Use in Grass Establishment

DEFRA – Department for Environment, Food and Rural Affairs

DI – Deionised

EC – European Commission

EDTA – Ethylenediaminetetraacetic acid

ESA – Environmental Services Association

EU – European Union

FAO – Food and Agriculture Organisation of the United Nation

GI – Green Infrastructure

HCl – Hydrogen chloride

HF – Hydrogen fluoride

ICP – Inductively Coupled Plasma

ICRCL – Interdepartmental Committee on the Redevelopment of Contaminated Land values

IRGA – Infra-red gas analyser

ISM – Integrated stormwater management

LCCP – London Climate change partnership

LOI – Loss on ignition

MBTE – Methyl Tertiary-Butyl Ether

MPN – Most probable number

MSW – Municipal solid waste

NA – Nutrient Agar

NIEA – Northern Ireland Environment Agency

OECD – Organisation for Economic Co-operation and Development

PAH – Poly-aromatic hydrocarbon

PAS – Publicly Available Specification

PPS – Permeable paving system

RA – Recycled aggregates

RBC – Rose-Bengal Chloramphenicol agar

RCA – Recycled concrete aggregate

SEPA – Scottish Environment Protection Agency

SF – Surface flow

SSF – Sub-surface flow

SUDS – Sustainable urban drainage systems

SUDSnet – Sustainable Urban Drainage Systems Network

TDS – Total Driveway Services

TOC – Total organic carbon

TPH – Total petroleum hydrocarbon

UK – United Kingdom

UKCIP – UK Climate Impacts Programme

UKCP – UK climate projection

USDA – United States Department of Agriculture

USEPA – US Environmental Protection Agency

USGS – US Geological Survey

VOC – Volatile organic compound

WCED – World Commission on Education and Development

WERF – Water Environment Research Foundation

WHO – World Health Organisation

WRAP – Waste and Resource Action Programme

WHC – water holding capacity

Chapter One: Introduction

Urbanisation, as described by Wagner (2008: vii), is 'a process in which an increasing proportion of an entire population live in cities and the suburbs of cities' and this process is associated with industrialisation and development. The urbanisation process has occurred as a result of people migrating to cities in search of improved standards of living, better economic conditions, better services and amenities (Wagner, 2008). However, in as much as cities provide and offer various opportunities for improving the quality of life, they also create challenges and problems with unsustainable impacts characterised by increase in air pollution due to vehicular traffic and transport of resources, high rates of energy consumption, increased use of natural renewable and non-renewable resources such as water and rocks, increased volume of surface run-off, decreased quality of surface run-off and depletion of biodiversity and habitats (Garber, 2000; McGeough *et al*., 2004; Barrow, 2006). A combination of all these impacts has had devastating effects on the environment some of which include: climate change, global warming, flooding, pollution of water bodies, and destruction of aquatic life and biodiversity (Butler and Davies, 2004).

In order to address these issues, the World Commission on Environment and Development (WCED) or the 'Brundtland Commission' was commissioned in 1984 by the United Nations, to identify and develop strategies for attaining sustainable development within the international community (Elliott, 1994). In 1987, the Brundtland Report entitled "Our Common Future", was submitted to the United Nations. The commission defined sustainable development as "the development that meets the needs of the present without compromising the ability of the future generations to meet their own needs" (WCED, 1987: 43); and three main objectives were highlighted: social, economic and environmental objectives (OECD, 2002: 12). Sustainable development involves the maintenance of development over time with the partial and/or complete incorporation of the Brundtland Report objectives which are interdependent and complement each other. The social objectives refer to relationships, human values and institutions; the economic objectives refer to the allocation and distribution of natural resources; and the environmental objectives refer to the contribution of both the economy and the society and their effects on the environment and its resources (Baker, 2006).

Several approaches have been taken to promote sustainable urban development and one of such approaches is the 'self-reliant city approach'. This approach seeks to improve the unsustainable impacts of urbanisation on a city by reducing the overall natural resource consumption, minimising waste streams, use and re-use of local resources where feasible, increasing the implementation of renewable resources as against non-renewable resources, dealing with pollution *in-situ* rather than disposing of it externally, and adequate progress towards sustainable consumption (Morris, 1990; Satterwaite, 1997). One way of achieving these objectives is the incorporation of sustainable urban designs into urban development because urban design is directly linked to the use of energy, land and natural resources (McGeough *et al*., 2004). Sustainable designs can be applied as an integrated approach to landscape, buildings, roads and streets, and other aspects of city life and for these designs to be truly sustainable, land use has to be altered to require fewer resources and less maintenance (Clarke, 2003; McGeough *et al*., 2004).

In order to investigate various sustainable urban designs, a competition known as the 'International Competition for Sustainable Urban Design' was conducted by the International Gas Union in June 2003, to stimulate new ideas and practices in sustainable urban design. The competition involved design teams comprising of urban designers, architects and planners from nine nations namely Argentina, Canada, China, Germany, India, Japan, Russia, United States and Mexico (Krause *et al*., 2003). Highlights of the competition as shown below revealed remarkable similarities between independent designs:

- Utilisation of renewable forms of energy such as gas from landfills and sludge from municipal waste water treatment facilities.
- Integration of recyclable materials into building and construction projects.
- Establishment of 'green spaces' within urban boundaries which provides a means of replenishing ground water with vegetation, and forested areas acting as carbon dioxide sinks thereby mitigating greenhouse effects.

- Protection of groundwater recharge areas, replacement of impermeable road and roof surfaces with vegetation, stormwater re-use and establishment of natural wastewater filtration systems such as wetlands (Krause *et al.*, 2003).

In developing sustainable urban designs, several elements are considered such as water and land use. In considering sustainability in water supply, the main foci needs to be on reducing demand for water and the relationship of water quality to its intended use. Water supply can achieve sustainability if reduction in demand and intended water use are considered (Krause *et al.*, 2003). Potable water is often used for various domestic activities as shown in table 1.1:

Table 1.1: The percentages of national averages of typical present-day potable domestic water use in the UK

Source: Rawlings (1999)

Table 1.1 shows that in the UK, over a third of domestic water is used for toilet flushing and this can be considered unsustainable because a lower quality of water could be used whilst higher quality potable water can be reserved for activities such as cooking and drinking. To reduce demand for potable water, recycling of rainwater and grey water (wastewater from domestic appliances except toilets) is encouraged. The first choice is rainwater because it is generally cleaner than grey water and has less risk of infection. The rainwater is collected, filtered and stored for use. Recycled rainwater can be used for toilet flushing, clothes washing

and for gardening. In grey water recycling, grey water is collected, disinfected and recycled for toilet flushing (Rawlings, 1999).

Sustainability in land use is vital because land combines landscape, landforms and ecosystems which form the natural environment and this is important for sustainable development (van Borcke, 2003). One way sustainability in land can be attained is by the application of natural drainage systems which decreases the flow of surface runoff thereby reducing flooding and the associated costs and environmental implications of artificial flood control methods (e.g. pipes and sewers). By reducing the quantity of rainwater and surface runoff being channelled into municipal sewers, the amount of wastewater being treated by sewage plants is reduced, thereby conserving energy (van Borcke, 2003). Natural drainage systems simulate the natural drainage of a site/catchment and work in harmony to achieve reduction in flow rate, peak flow and volume of surface runoff, thereby reducing the quantity of runoff flowing into sewers, improving storm water quality, reducing erosion, recharging groundwater and improving biodiversity (CIRIA C523, 2001). These natural drainage systems are also known as sustainable urban drainage systems (SUDS). An example of SUDS is vegetative SUDS which are natural drainage systems that have vegetation growing on them (e.g. swales), which contributes to improving stormwater quality (CIRIA C523, 2001).

However, some of these natural systems may not be fully sustainable as they seem because though they drain and convey water sustainably, the components they comprise of may not be sustainable. This is because some of the materials used in the construction of SUDS deplete natural resources and increase pollution due to their transportation. One way of mitigating these unsustainable impacts is by sourcing for more sustainable materials, e.g. recycled materials such as compost and recycled aggregates, as a substitute for unsustainable materials, without compromising the performance of SUDS (BIS, 2008; WRAP, 2010). The purpose of this research was therefore to identify materials that are unsustainable for use in vegetative SUDS and suggest possible sustainable alternatives that could match and replace existing materials in terms of performance, and most importantly, sustainability.

Therefore, the main aims of this research were to:

1. Study the use of compost and recycled aggregates in vegetative SuDS
2. Study the use of swales comprising of compost and recycled aggregates in the treatment of runoff pollutants

Objectives in fulfilling the aims included:

a. Characterisation of the test materials i.e. compost and recycled aggregates.
b. Investigating the development of biofilms in compost under simulated swale conditions.
c. Investigating grass development in profiles comprising of compost and recycled aggregates under simulated swale conditions.
d. Investigating the efficacy of compost and RA in remediating pollutants in simulated swale conditions.
e. Studying the effect of recycled aggregates on oil pollution remediation.

Method applied in fulfilling the aims and objectives included:

i. Baseline analyses of test materials which include determination of moisture content, water holding capacity, bulk density, organic matter content, carbonate content, pH levels, heavy metal content, microbial enumeration and water quality assessment.
ii. Carbon dioxide monitoring for assessing microbial activity in test compost samples.
iii. Monitoring grass growth by biomass measurements in pot trials containing compost and recycled aggregates.
iv. Heavy metal and motor oil analyses of test samples and leachates derived from test samples dosed with heavy metals and motor oil in swale simulations.
v. Oil absorption studies on oil-dosed recycled aggregates and oil analysis of their leachates.

Thesis synopsis

Chapter Two provides a review on SUDS in comparison with conventional drainage systems, in terms of stormwater quality, sustainability of drainage components and the benefits of using vegetative SUDS compared to other SUDS devices, including relevant legislation. Materials that can make vegetative SUDS more sustainable in terms of stormwater quality and resource conservation were suggested. Chapter Three discusses experimental designs carried out to characterise and test the potentials of the materials suggested in Chapter Two, in order to determine their efficacy in treating stormwater compared to conventional materials. Chapter Four discusses the results obtained from experiments carried out on the test materials, based on the experimental designs described in chapter Three, including how these materials fared compared to conventional materials. Chapter Five discusses the application of the results to real-life scenarios, identifying if conventional materials were more sustainable than the tested alternatives or vice versa. Chapter Six highlights conclusions deduced from findings derived from this research and presents evidence of originality and design innovations of thesis.

Chapter Two: Literature review

2.1 Introduction

Urbanisation and development has led to an increase in impermeable surfaces globally leading to a drastic reduction in natural infiltration of runoff, thereby causing an increase in the volume of surface runoff to more than 80% of the total rainfall volume. Also, runoff flow rate has increased significantly thereby causing erosion of unsealed ground, flooding and pollution of water resources. A new site or catchment area can become impermeable due to developments (parking lots, roofs, pavements and roads) and natural infiltration becomes difficult; even available pervious ground cannot carry out infiltration normally, due to soil compaction and stripping of topsoil during construction (CIRIA C523, 2001). Also, paved surfaces have left little or no room for green infrastructure (GI) such as parks and other vegetated areas (Haase, 1986). As a result of these impacts, water bodies such as streams and rivers have to cope with larger volumes of stormwater than they would normally handle, hence constructed/artificial drainage systems are applied to reduce the risk of flooding, water logging, subsidence and stagnant pools. These conventional drainage systems consist of pipes and structures (drains, sewers, kerbs and gullies) that collect, channel and dispose of water (Butler and Davies, 2004; WRAP, 2011).

However, as the components of conventional drainage systems are designed to work independently of each other, they end up producing heterogeneous waste streams which make recovery of resources difficult and expensive, and could end up polluting water bodies (Butler and Davies, 2004). As a result, a more integrated approach to drainage is needed whereby drainage components interact with each other thereby reducing water pollution and flooding (Adams and Papa, 2000; Brown, 2005). Sustainable (Urban) Drainage Systems (SUDS) are integrated stormwater management systems which take into consideration the quantity, quality and amenity value as well as the biodiversity of stormwater, thereby reducing flooding and pollution of water resources. However, the source of some raw materials used in the construction of SUDS can be quite unsustainable, as some SUDS components require the use of natural resources (such as gravel) which leads to the depletion of natural resources, pollution, and the adverse effects of these impacts on the climate. As such, SUDS have to be

made more sustainable by using materials that would not adversely affect the environment (Woods-Ballard *et al*. (2007). Though the phrase 'sustainable urban drainage systems' implies that SUDS devices are only applicable in urban areas, various applications (as will be discussed later in this chapter) have shown that SUDS devices can be applied to any area including tropical regions; and so from now on, for the purpose of this study, the term Sustainable Drainage Systems (SuDS) is used.

This chapter discusses the advantages and disadvantages of conventional drainage systems and how integrated stormwater management systems, with emphasis on SuDS devices, are better able to deal with drainage problems such as flooding and pollution. The sustainability of SuDS components is discussed with emphasis on vegetative SuDS; and alternative materials that could make vegetative SuDS components more sustainable are suggested. Firstly however, conventional drainage and its specific problems are discussed so that comparison with the SuDS approach is given better context.

2.2 Conventional Drainage Systems

Urban drainage systems drain two types of water, wastewater and stormwater. Wastewater is the outcome of water used in everyday living and industrial use. It carries particles and chemicals which could cause pollution and create health risks. Stormwater is any form of precipitation that has fallen on a built up area and which, if not properly drained, could result in flooding (Butler and Davies, 2004). Conventional drainage systems consist of either combined sewers or separate sewers. Combined sewer systems convey both foul sewage and surface runoff through a single pipe to sewage treatment plants, which when overloaded, especially in periods of heavy rainfall, are allowed to overflow into water courses (USEPA, 2012[a]). In dry weather, the pipes convey mainly wastewater but during periods of rainfall, stormwater is added to the flow which can easily overwhelm the sewage treatment plants (Bell, 2011). Hence structures are constructed which divert flow above a certain level out of the sewers into water courses and these structures are called "combined sewer overflows" (CSOs). CSOs were constructed to retain as many solids as possible and were thought to be harmless because the diverted flow of sewage was diluted both by stormwater and receiving

water bodies. However, CSOs are serious sources of pollution to water bodies because the diverted flow of sewage from CSOs contain harmful pollutants such as heavy metals, motor oils and harmful chemicals from industrial sites, which when released into water bodies, lead to pollution and harm to aquatic ecosystems (CIRIA C523, 2001; Lau *et al*., 2002; Butler and Davies, 2004; Suarez and Puertas, 2005; Hoyer *et al*., 2011).

In separate sewer systems, surface runoff and sewage are conveyed in separate pipes usually laid side by side, the former to water courses and the latter to treatment plants. This system became common in the UK around the mid-1940s, due to pollution caused by CSOs (Butler and Davies, 2004). Separate sewer systems may seem better compared to the combined sewer system however, this may not be so because surface runoff becomes contaminated with pollutants such as oil, dust, organic matter, silt, nutrients, eroded soil particles and chemicals such as detergent and pesticides; which is then dumped directly into receiving water bodies. Also, there is the issue of cost when compared to combined sewer systems, due to the additional pipe and wider excavations required to accommodate both pipes (Pyzoha, 1994; CIRIA C523, 2001; Butler and Davies, 2004). The application of conventional drainage systems to stormwater drainage has led to major problems and these problems are discussed in the next section.

2.3 Problems associated with conventional drainage systems

One of the consequence of CSOs in combined sewer systems, and the application of an extra pipe that conveys runoff directly into water courses in separate sewer systems, is the pollution of receiving water bodies. The approach of conventional drainage systems to stormwater management focuses mainly on water quantity, much less on water quality and with little consideration for wildlife and amenity value, such as landscaping potentials and recreational opportunities (see figure 2.1). This means that the system focuses more on transporting stormwater as quickly as possible away from its source so as to prevent flooding, without considering pollutant concentration and its resultant effects on aquatic habitats and biodiversity. This method of conveying stormwater also ensures that infiltration of surface runoff is decreased thereby reducing groundwater recharge which leads to lowering of the

groundwater table, and depletion of groundwater can lead to water shortages and soil subsidence (CIRIA 522, 2000; Charlesworth, 2010).

Figure 2.1: Conventional drainage approach.

Source: Charlesworth (2010)

Due to increased pollution of runoff caused by urbanisation, pollutants such as organic matter, heavy metals, motor oil and grease end up in groundwater and water courses with adverse effects on aquatic ecosystems and human health. These pollutants, when assimilated by aquatic flora and fauna, move up the food chain to higher animals and when accumulation of pollutants reach toxic levels, they can lead to adverse effects on biodiversity, including humans, resulting in illnesses and sometimes death (Coupe *et al*., 2006[a]).The failure of existing systems to cope with runoff due to most of them exceeding their capacity has also led to an increase in wastewater flowing into water courses causing water bodies, such as streams and rivers, to swell and overflow their banks leading to flooding and erosion of aquatic habitats. Erosion causes the deposition of silt and sediments downstream where water flow is slower, further damaging aquatic habitats leading to loss of amenity and wildlife (CIRIA C523, 2001; Butler and Davies, 2004; Hoyer *et al*., 2011).

The various components of conventional drainage systems (sewerage system, wastewater treatment plants and receiving water bodies) have been designed to operate in such a way that each component meets the needs of its users, including the environment. However, because they are designed to work independently with little or no interaction between components (Butler and Schütze, 2005), they eventually produce heterogeneous waste streams which makes recovery of resources (such as water) difficult and treatment more complex as a higher level of expertise, energy, space and cost is required, thereby making treatment unsustainable (Balkema *et al.,* 2002).

Another major shortcoming of the conventional drainage system is the sustainability of materials used in their construction which include clay, iron, concrete or plastic for pipes, and aggregates for constructing pipe surrounds and pavements (WRAP, 2011). The extraction of gravel, an aggregate used in concrete production for concrete pipes, has a significant impact on the environment in terms of depletion of gravel deposits, dust pollution, poor visibility, increased soil erosion, silting up and pollution of water bodies (Paige-Green and Hongve, 2003; WRAP, 2011). In handling these problems, CIRIA C523 (2001) proposed two alternatives namely:

a) improvements in conventional drainage systems and engineering practices and
b) sustainable urban drainage.

In reality, these two approaches should be considered together rather than separately. Improvements in conventional drainage systems and engineering practices include construction of flood defences, new-generation screens on CSOs, end-of-pipe treatment and flow management within the sewerage network. However, flood defences require land-take and extensive construction works and due to restricted water flow, downstream developments and communities may become flooded. CSO screens are designed to remove coarse solids and will not solve the problem of pollution or flooding. End-of-pipe treatment is difficult because of the copious amounts of water and high pollution load involved, and as mentioned above, treatment is intensive and therefore not sustainable. Flow management involves the use of

flow-restricting devices or real-time control to manage sewage flow within a sewerage network. This method involves the mobilisation of sewage such that the storage capacity of sewerage networks are maximised thereby eliminating the need for major construction works to increase the capacity of sewers, however this system is cost intensive as it is a high-technology solution and does not encourage sustainability since it does not encourage stormwater re-use (CIRIA C523, 2001).

In addressing these problems associated with conventional drainage systems, Andoh (1994) proposed the separation of stormwater from wastewater and suggested that distributed systems provided an alternative preventative approach to urban drainage at reduced costs compared to conventional drainage systems. This alternative approach recommended the use of stormwater management techniques and/or natural drainage patterns as an option to the use of pipes in conveying stormwater (Butler and Davies, 2004). Neilson (1999) and Joos *et al*. (2007) also supported the decentralised approach to urban drainage suggested by Andoh (1994). They suggested the development of cost-effective integrated solutions which combined conventional 'hard' engineering schemes (e.g. pipes and sewers) with 'soft' engineering solutions (e.g. filter strips, swales and ponds) and watercourse improvements (e.g. river restoration); and hence the idea of an integrated management system to deal with stormwater was developed and this is discussed in the next section.

2.4 Integrated stormwater management

The significant impacts of urbanisation on drainage, as discussed in previous sections, have necessitated the development of stormwater management systems. Wanielista and Yousef (1993) defined stormwater management as the "knowledge used to understand, control and utilise water in its various forms within the hydrologic cycle", and the application of its designs, concepts and maintenance are evident in diverse areas such as lake management, agricultural drainage, flood control, forest management, urban runoff and ecological impact studies. Traditionally, the approach to stormwater management had been that of quantity control and drainage was designed to capture and convey stormwater away from places of residence as swiftly as possible. However, approaches to stormwater management now

include consideration of the quality of runoff and amenity value/biodiversity (Pyzoha, 1994; CIRIA C523, 2001). The main objectives of stormwater management are flood prevention, reduction of land loss due to erosion, preservation of environmental quality of receiving water bodies by the reduction of mass loading of chemicals, suspended solids and other pollutants which may cause physical, chemical and/or biological changes to receiving water bodies; reduction of peak flows to improve infiltration of runoff and recharge groundwater as well as encouraging passive recreation opportunities (Wanielista and Yousef, 1993).

The first step in an effective stormwater management programme is to identify the causes and effects of flooding in an area and this knowledge will go a long way in proffering drainage solutions (Wanielista and Yousef, 1993; Adams and Papa, 2000; Brown, 2005; Pitt and Clarke, 2008). However, some limitations to stormwater management identified by Wanielista and Yousef (1993) were: *cost* with emphasis being placed on cost effective systems which were not necessarily sustainable; *site feasibility* as some stormwater systems might have a substantial impact on land amenities and hence their application was not feasible; *environmental impact* on biodiversity and water quality; *potential re-use of stormwater* was lost when released directly into water bodies; *labour and maintenance* as some control methods were labour intensive and required regular maintenance. For stormwater management systems to overcome these limitations and become effective, an integrated approach has to be adopted in which a combination of a wide variety of individual stormwater controls interact interdependently with each other (Pitt and Clarke, 2008). This approach is known as 'integrated stormwater management' (ISM).

ISM is a criteria for sustainable drainage (Backstrom and Viklander, 2000) and an international survey conducted by Marsalek and Chocat (2002) clearly indicated a widespread interest in the holistic approach to stormwater management which promotes drainage systems that emphasises source control, transition from 'hard' infrastructure to GI, maintenance and rehabilitation of existing infrastructure and formation of integrated stormwater agencies; with participation of both the public and private sector in the planning, implementation and operation of stormwater management systems. The concept of ISM also stresses the

importance of involving the co-operation of practitioners (local authorities, architects, town planners and civil engineers) and stakeholders (developers and citizens) (Joos *et al*., 2007). However, Brown (2008) noted that despite significant advances in ISM techniques, its implementation in the UK has been quite limited and this limitation was attributed to the reluctance of practitioners and stakeholders to embrace change, and inertia in the public administration of stormwater management practices. This reluctance has been ascribed to lack of research and understanding of the long-term behaviour of ISM techniques as well as the long-term implication of unforeseen problems that may arise; hence practitioners and stakeholders would rather stick to what they know and what has worked for them (Doyle *et al*., 2003). This reluctance has therefore encouraged the adoption of traditional stormwater management practices at the implementation stage of a development rather than 'newer' more integrated drainage systems. Butler and Parkinson (1997), CIRIA (2001) and Balkema *et al.* (2002) all suggested that adopting a learning culture that values integration and participatory decision-making would provide the platform needed to improve adoption of ISM techniques. An integrated approach to stormwater management is exemplified in the application of sustainable drainage systems (SuDS) and the following sections detail its implementation.

2.5 Sustainable Drainage Systems

SuDS, rather than trying to modify nature, work in harmony with it by reducing the flow rate, peak flow and volume of surface runoff. SuDS, in most cases, simulate the natural drainage of a site/catchment thereby reducing the amount of runoff flowing into sewers, reducing erosion, improving the quality of surface runoff by treating pollutants, improving quality of water bodies, recharging groundwater and improving biodiversity (CIRIA, 2001; Casal-Campos *et al*., 2011). SuDS are devices that give equal consideration to water quantity, water quality and public amenity/biodiversity (see figure 2.2) in contrast to conventional drainage systems (see figure 2.1); and these three components are integrated, working together to reduce flood risk and pollution as well as improving the environment. SuDS do not function in isolation but as an integrated system and can either be used in conjunction with conventional drainage systems or other SuDS systems (CIRIA, 2001; Dickie *et al*., 2010).

Figure 2.2: The SuDS triangle.

Source: Woods-Ballard *et al*. (2007)

SuDS have been widely and successfully used in USA, Europe, Australia and Japan (Ghani *et al*., 2008) and their benefits are summarised below:

- SuDS help to identify and control flooding and pollution at source thereby encouraging easier prevention or containment measures, locally.
- As SuDS provide natural attenuation and temporary storage of surface runoff, flood risk is reduced in a catchment area and further downstream.
- Surface water retention in a development helps to recharge groundwater and maintain its balance by infiltration thereby preventing low river flows especially in summer periods.
- Stormwater treated by SuDS can be harvested and re-used for domestic uses such as toilet flushing and gardening.
- SuDS help to recharge groundwater and thereby maintaining natural vegetation.
- SuDS reduce the need and cost to enlarge and upgrade existing sewers to accommodate runoff.
- Runoff storage areas can serve as landscaping or amenity areas (CIRIA C523, 2001).

In addition to the benefits mentioned above, SuDS can also help mitigate the effects of climate change, for according to predictions by the United Nations, about 60% of the world's population will reside in urban areas by 2030. Urban environments are usually characterised by altered heat exchange (due to decreased evaporative cooling caused by decrease in vegetated surfaces) and increased surface runoff, would influence climate change (Kirkby, 2005). DEFRA (2010[a]) defined climate change as 'any change in climate over time, whether due to natural variability or as a result of human activity'. Observed climatic trends for UK have shown that climate change has brought about an increased contribution to winter precipitation and summer rainfall events, with an overall increase in the average annual precipitation (LCCP, 2002; UKCIP, 2011). Intense and more frequent rainfall events have increased the risk of summer flash flooding, winter flooding and river flooding (Arkell and Darch, 2006) and climate change will affect areas such as urban drainage and flood risk, water resources, and outdoor spaces (Schreider *et al.*, 2000; Gill *et al*., 2007; Wilby, 2007).

Key findings from future projections by UK climate projections 2009 (UKCP09) (DEFRA 2009[a]), showed that by 2080, average summer precipitations across the UK may decrease by 17%-23% and average UK winter precipitations may increase by 14%-23%. Therefore predicted climate change will be characterised by wetter, warmer winters; drier, hotter summers; heavy downpours of rain and further rise in sea levels and tidal surges (Hulme *et al.*, 2002; Arkell and Darch, 2006). Major effects of climate change on drainage and water resources identified by UKCIP (2011) include:

- increased risk of flooding and erosion,
- greater pressure on drainage systems,
- increased likelihood of winter storm damage,
- loss of habitat for wildlife,
- summer water shortages and low stream flows and
- increased risk of subsidence in susceptible areas.

Adapting to climate change involves a sustainable process of protection against negative environmental impacts, taking advantage of any benefits and costs minimisation. An adaptation suggested by DEFRA (2010[b]) was the adoption of SuDS which provides a sustainable way of draining surface water. Adoption of SuDS coupled with other adaptations such as provision of green spaces (e.g. gardens, trees and green roofs) which will improve biodiversity and act as carbon dioxide sinks, would reduce 'urban heat island effects' (increased temperatures in urban areas compared to rural areas), encourage rainwater and greywater re-use for potable water conservation, establish building designs that will keep buildings warmer in winter and cooler in summer months and encourage carbon capture and sequestration, which will all help to mitigate climate change. These adaptations were aptly illustrated by Charlesworth (2010), as shown in figure 2.3 below.

Figure 2.3: The SuDS rocket.

Source: Charlesworth (2010)

The impact of climate change on drainage and stormwater flow was demonstrated in two separate studies by Semadeni-Davies *et al.* (2008[a]) and Semadeni-Davies *et al.* (2008[b]). In Semadeni-Davies *et al.* (2008[a]), climate change impact assessment of combined sewer systems was carried out in the evolving urbanised area of Helsingborg, Sweden over a 10-year period, and results showed that climate change will exacerbate drainage problems due to the inability of existing conventional drainage systems to cope with increased surface runoff; except stormwater is disconnected from combined sewers to reduce the frequency of sewer overflow. Similarly, a study by Semadeni-Davies *et al.* (2008[b]) on the same area in Sweden over a 15-month period to assess the potential impacts of climate change on stormwater flows to a suburban stream showed that increased rainfalls will raise runoff peak flow and increase flood risks. Both studies agreed that the adoption of SuDS will have a positive effect on the urban environment by reducing the release of pollutants into water bodies, peak flow and total volumes, thereby minimising the adverse effects of climate change (Semadeni-Davies *et al.*, 2008[a]; Semadeni-Davies *et al.*, 2008[b]).

However, the success of the benefits of SuDS depends on identification and implementation of clear design and maintenance objectives tailored to suit local conditions. The implementation of SuDS requires developers to work in conjunction with other disciplines and agencies, such as architects, planners, drainage engineers, landscape architects and hydrologists, right from the onset of the development process (Kirkby, 2005; NIEA, 2009). Before establishing and implementing SuDS, some basic requirements should be met. Ideally, SuDS should be incorporated into the original water resource management and control design of developments so as to provide enough space and resources for stormwater control. Nevertheless, SuDS can also be incorporated into existing developments as retrofit systems. Runoff from developments should not exceed runoff prior to development so as to encourage natural infiltration, groundwater recharge and prevent flooding. Also runoff generated within an area/development should be infiltrated onsite before discharge so as to protect river and groundwater quality, because runoff should not compromise the quality of downstream watercourses and habitat (CIRIA, 2001; Dickie *et al*., 2010). Other requirements of SuDS are avoidance of degradation of the environment, minimisation of the utilisation of natural

resources and long term reliability and adaptability to future requirements (Butler and Parkinson, 1997).

Nonetheless, as laudable as SuDS are, there are some constraints associated with their implementation. Maintenance of SuDS could increase construction costs and take up more space compared to conventional systems, although SuDS infrastructure can be integrated into surrounding landscapes. SuDS can be difficult to retrofit into an existing development and should therefore be considered at the early stages of planning of a development. Though adequate care has to be taken to ensure that proposed SuDS devices are capable of handling runoff from proposed sites and in extreme events such as increased intensity/duration of rainfall, contingency measures have to be in place to cater for any excesses. SuDS devices, like conventional systems, can become overwhelmed when runoff far exceeds what they have been designed for (CIRIA C523, 2001; Pratt, 2003). Also, the disposal of accumulated waste products from SuDS devices have to be carefully considered as such wastes products can only be localised and mitigated but not eradicated (Heal, McLean and D'Arcy, 2004; NIEA, 2009). However, some of these problems can be mitigated with proper operational and maintenance schedules.

The development of operational and maintenance schedules for SuDS during initial design and construction can help to reveal deficiencies and shortcomings in the original design and installation of SuDS, in addition to providing effective SuDS performance and site after-care, long term administration and management (Ellis *et al.*, 2003[a]). Studies by Heal, McLean and D'Arcy (2004) and Schlüter and Jefferies (2005) also confirmed that regular maintenance is vital for the longevity of SuDS. Therefore, failure to establish and implement operational and maintenance schedules could lead to poor performances and failures of many SuDS devices, hence operation and maintenance has become a major issue with potential SuDS stakeholders (McKissock *et al*. 1999). For example, an assessment of infiltration devices and filter drains in central and eastern Scotland reported unsatisfactory performance of some of the systems as a result of inadequate maintenance which led to the blockage of roadway inlets to the filter

drains and inlet pipes to infiltration trenches with sediments, and so treatment of road runoff by these devices were truncated (Heal, McLean and D'Arcy, 2004).

The operation and maintenance regime should put into consideration flow, quality control and amenity functions, and its main objective should be the maximisation of SuDS to function at or near its original design. It should also distinguish between regular maintenance and crisis maintenance as well as intervals for each maintenance regime varying on monthly, seasonal, annual or less frequent intervals. Some basic regular operational and maintenance schedules for SuDS, according to Ellis *et al.* (2003[a]) and Shaffer *et al*., (2009) should include:

- monitoring levels of sediments and solids accumulation to avoid inlet and flow blockages,
- checking for erosion,
- removal of sediments, litter and solids when necessary,
- monitoring inlet and outlet structures,
- maintenance of vegetation and landscaped areas and
- regular application of replaceable SuDS components such as gravel.

2.6 Types of Sustainable Drainage Systems

There are several SuDS devices and each one is designed to fulfil the three objectives of sustainable drainage as described in figure 2.2. They manage surface water by processes of attenuation, infiltration and detention, and each device can be applied in a sequence such that it mimics the natural drainage of a site before development (Woods-Ballard *et al.,* 2007). According to Woods-Ballard *et al. (*2007), SuDS devices are grouped into four main techniques:

a) permeable surfaces and filter drains,
b) infiltration devices,
c) basins, ponds and wetlands and
d) filter strips and swales.

Permeable surfaces are designed to allow surface water drain from permeable paved surfaces to the sub–base (layer below the surface) faster than rainfall intensity so as to prevent flooding (Coupe *et al*. 2006[b]; Woods-Ballard *et al.*, 2007), and are therefore necessary for stormwater management (Gomez-Ullate *et al*., 2011). The sub-base is of the open-grade type which means that it is comprised of large aggregate spaces which are porous to allow the infiltration of water into the ground, into an underground storage or into the next SuDS management stage, depending on the permeability of the existing ground/soil.

Permeability of surrounding soil is vital as this will determine what type of sub-base is installed. Permeable soils will allow total infiltration of treated water into the ground and therefore does not need to be diverted into other drainage systems such as sewers or water courses. Semi-permeable soils allow the partial infiltration of water and a fixed amount of water is allowed to infiltrate into the ground and excess water is drained via a series of perforated pipes into storage tanks or other drainage devices. Impermeable soils will not allow the infiltration of water and hence the sub-base is lined with impermeable flexible membranes to capture water and divert it into other drainage devices, a method which is also currently applied to contaminated sites to prevent contamination of groundwater (Interpave, 2006; Interpave, 2008). The sub-base filters out particles and sediments, and organic matter is reduced by microorganisms present on the sub-base material, unlike conventional drainage surfaces such as concrete pavements, which convey surface water quickly to overloaded drains and water courses through pipes, thereby leading to water pollution and flooding. However, permeable surfaces are prone to clogging by stormwater sediments which could affect infiltration rates and water quality (Siriwardene *et al*., 2007) but unlike conventional piped systems for which underground inspection required, clogging of permeable surfaces can easily be identified and rectified with visual inspection (Woods-Ballard *et al.*, 2007; Interpave, 2008; Shaffer *et al*., 2009).

Permeable surfaces include permeable block paving, porous asphalt, reinforced grass and gravel systems, and gravel (Shaffer *et al*., 2009). Studies by Brattebo and Booth (2003) and Gomez-Ullate *et al*. (2011) showed that water storage by these surfaces was not significantly

different and, according to Brattebo and Booth (2003), water quality was significantly better than impermeable surfaces. Figures 2.4 and 2.5 below illustrate the various types and uses of permeable drainage systems.

a) Block permeable pavements usually consist of clay/concrete paving blocks with gaps between blocks, or porous concrete blocks with holes in them. They are commonly applied in car parks, walkways, cycleways, residential roads and container terminals due to their skid and slip resistance, durability and strength (Interpave, 2008) as shown in figure 2.4a, 2.4b and 2.4c.

Figure 2.4a: Permeable pavement with concrete blocks.

Source: Ecofriend (2007)

Figure 2.4b: Permeable pavement with clay blocks showing water infiltration.

Source: Interpave (2011)

Figure 2.4c: Permeable paving in a car park.

Source: WERF (2009)

b) Reinforced grass consists of laying down a network of grids made up of synthetic fibres over the upper layer of topsoil to which grass carpets have been established. The grids help to support the grass tufts, bear the load of vehicles and are porous enough to allow water through (see figure 2.5a and 2.5b). The grids could also be replaced by concrete paving blocks with pores in them as seen in figure in 2.5c. Reinforced gravel is similar to reinforced grass only that grasses are replaced by gravel (see figure 2.5d) (Kazda and Caves, 2007).

Figure 2.5a: Reinforced grass paving with composite plastic grids that allow grass/moss to grow through openings in the grid squares.
Source: Smith (2009)

Figure 2.5b: A plastic grid used in permable paving.

Source: BRP (2011)

Figure 2.5c: Reinforced grass paving with concrete blocks pores that allow grass to grow.
Source: TDS paving and Landscaping (2011)

Figure 2.5d: Reinforced gravel paving with concrete blocks that allow infiltration of water.
Source: The Enhance Companies (2011)

c) Porous asphalt (also known as pervious macadam) consists of aggregates bound with bituminous material with open voids to allow water to pass through and they are used to make the upper layer of sub-bases permeable (Pratt *et al*., 2002).

d) Gravel is used to provide a porous surface for water to infiltrate into sub-bases and is usually used for the construction of filter drains. However they have a shorter life-span structurally compared to permeable blocks or asphalt pavements as they are easily displaced by vehicles (Woods-Ballard *et al.*, 2007; Shaffer *et al*., 2009).

Filter drains are narrow strips of permeable surface (e.g. gravel) running along road sides which allows the movement of water through the sub-base to a discharge point or to the ground and can be used in conjunction with other permeable surfaces as shown in figure 2.6 (Woods-Ballard *et al.*, 2007).

Figure 2.6: Filter drains with permeable block paving.

Source: SUDSnet (2008[a])

Infiltration devices are devices that make use of the ability of the soil and underlying geology to absorb water. This is achieved by the provision of a large surface for rain water to drain through and some storage capacity to detain runoff. Infiltration through these surfaces encourages filtration of runoff thereby reducing sediment and organic pollutants (Pratt 2003; Woods-Ballard *et al.*, 2007). Examples include:

a) *Soakaways and infiltration trenches:* which store water in underground storages and soaks into the ground through the sides and base of the storage (Pratt, 2003). Figure 2.7 shows the cross-section of a soakaway, with pore spaces between the gravel backfill storing water long enough for it to infiltrate into surrounding soil. Figure 2.8 shows the top of an infiltration trench filled with rocks and surrounded by vegetated cover.

Figure 2.7: Cross section of a soakaway showing the flow of water.

Source: Septic tank installation (2010)

Figure 2.8: An infiltration trench.

Source: Department of Ecology State of Washington (2011)

b) *Infiltration basins* which detain water above the ground and soaks into the ground through the base of the basin.
c) *Green roofs* which are systems that cover the roof a building or platform structure with a vegetative cover over a drainage layer. They are designed to capture and retain

stormwater thereby reducing the volume and flow rates of runoff thus relieving other drainage structures of the pressure of stormwater draining (Nicholaus *et al*., 2005). They also have the capability of removing atmospherically deposited urban pollutants such as suspended solids thereby improving air quality (Woods-Ballard *et al.*, 2007). Rainwater treated by green roofs can also be recycled for domestic use (e.g. watering plants and flushing toilets) without any additional treatment (Scholz, 2004) as they are generally considered to be cleaner than road runoff (Ellis *et al*., 2002). The vegetative cover of green roofs has been shown to reduce summer air temperatures (Niachou *et al*., 2011) and urban heat island effects (Booth, 2006). Green roofs can be used on elevated buildings or at ground level, usually above underground car parks (usually called podium roofs) (Woods-Ballard *et al.*, 2007). Green roofs are of four types as described by Woods-Ballard *et al.*, (2007):

i. Extensive green roofs: These are roofs with covered low growing, low maintenance plants such as mosses and grasses and are intended to be self-sustaining (see figure 2.9a). They are only accessed for maintenance purposes. They are light weight, cost effective and can therefore be used in a wide variety of locations. Other names by which they are known include sedum roofs, eco-roofs or vegetated roof covers.
ii. Intensive green roofs: (also known as roof gardens) are accessible landscaped environments with amenity benefits such as trees (see figure 2.9b). They may include water storage facilities of rainwater reuse. However, this type of green roof impose greater loads on roof structure and require significant on-going maintenance
iii. Simple intensive green roofs: These are roofs cultivated with lawns or ground growing vegetation and require regular maintenance. They are occasionally accessible, demand on roof structure is moderate and its construction is less expensive
iv. Biodiverse or wildlife roof: This type of green roof is becoming more popular due to increased awareness in biodiversity and conservation issues. They are

designed to replicate a specific habitat for a limited number of species or create a range of habitats with diverse species within the roof.

Figure 2.9a: A green bus shelter roof, an example of an extensive green roof.

Source: SUDSnet (2008[b])

Figure 2.9b: An intensive green roof.

Source: Eco Home Resource (2008)

Basins are designed to retain surface runoff and allow it percolate slowly through a filter layer comprising of a porous material such as gravel. The infiltrated water is then directed either into a pond for further treatment or allowed to percolate further into groundwater depending on the level of runoff pollution. They are effective in removing suspended solids and reducing heavy metal concentrations but may not significantly reduce soluble pollutants. They are usually dry except during and after rainfall events (CIRIA C523, 2001; Highways Agency, 2006; Woods-Ballard *et al*., 2007). Examples of basins include:

a) *detention basins* which are usually vegetated and can also be used as recreational facilities such as playgrounds when dry (see figure 2.10), and
b) *flood plains* which are land adjacent to a water course and are subject to repeated flooding under natural conditions (Apostolaski and Jefferies, 2005)**.**

Figure 2.10: A detention basin in Brookfields Park, Rustington.
Source: Apostolaski and Jefferies (2005)

Ponds are designed to either accept, attenuate and slowly discharge stormwater flow (known as balancing ponds) or alternatively treat runoff by allowing suspended solids to settle out (known as sedimentation ponds), although in practice, they perform both functions to a certain extent (see figure 2.11). They also provide aesthetic, amenity and wildlife benefits. Ponds are

of two types: *retention or wet ponds* which retains water thereby allowing treatment and *detention or dry ponds* which are designed to be dry for extended periods or to empty after a rainfall event (CIRIA C523, 2001; Apostolaski and Jefferies, 2005; Highways Agency, 2006; Woods-Ballard *et al.*, 2007).

Figure 2.11: A retention pond in Brookfields Park, Rustington.
Source: Apostolaski and Jefferies (2005)

Wetlands are areas that are permanently saturated either by ground or surface water and are able to support aquatic vegetation. Natural wetlands are relatively a rare occurrence and are generally of high nature conservation value and should therefore not be used in treatment of surface runoff. Wetlands required for runoff treatments are usually of the constructed type (see figure 2.12). Constructed wetlands are usually of two types: sub-surface flow (SSF) wetlands and surface flow (SF) wetlands. Constructed SSF wetlands are basins filled with porous materials through which water flows. The porous material which is a growth medium is usually saturated and planted with reed swamp vegetation. The flow of water is usually through the growth media and plant root zone. They are effective in removing nutrients (nitrates and phosphates) and heavy metals from municipal waste water and domestic sewage by processes of adsorption, biodegradation and biological uptake, as long as the runoff residence time is relatively long i.e. 24 hours or more.

Constructed SF wetlands are similar to constructed SSF wetlands but in this case the flow of water is across or close to the surface of the growth media and through the above-ground vegetative parts of the plants. They are effective in removing suspended solids and heavy metals through processes of settlement, sedimentation and filtration in addition to adsorption, biodegradation and biological uptake of metals and nutrients, within the recommended residence time of 24 hours or more (Ellis, Shutes and Revitt, 2003[b]; Highways Agency, 2006; Scholz, 2011).

Figure 2.12: A constructed wetland.

Source: University of Edinburgh (2008)

Filter strips are vegetated strips of gentle, sloping land that lie between an impermeable area and a receiving system, which could drain runoff into any of the other SuDS techniques already discussed. They are often used as a pre-treatment of runoff. They receive runoff as a sheet flow from impermeable surfaces (e.g. roads) and are treated by vegetative filtering, sedimentation of particles and infiltration (Woods-Ballard *et al.*, 2007). Figure 2.13 shows a filter strip leading to a filter drain where storm water is further treated.

Figure 2.13: A filter strip incorporating a filter drain

Source: Engineering Nature's Way (2011)

Swales are 'wide, shallow, gently sloping depressions designed to convey water' (Highways Agency, 2006: 2/1). Swales are a type of SuDS which can reduce the problems associated with increased run-off by encouraging attenuation, infiltration and evaporation of runoff thereby reducing considerably, the amount of pollutants entering into water courses. Swales are best suited for areas where roads are located on gently sloping embankments as this helps to convey runoff away from the road (see figure 2.14). They are also most effective when flow rate is low and can be applied as the first form of treatment in a stormwater management train. Swales are capable of reducing suspended solids by over 50% but may not significantly reduce soluble pollutants (Highways Agency, 2006). The fact that swales replace the 'out of sight, out of mind' concept of piped drainage systems, by bringing drainage systems to the surface means that pollutants and pollution are easily identified (MacDonald and Jefferies, 2003[a]). In assessing the effectiveness of swales in water quality improvement, two swale sites (Emmock Wood and West Grange) were monitored, and runoff entering the swales from the road and runoff emanating from the swales were compared. Results showed an overall improvement in water quality of runoff output from the swales compared to the road runoff. Observations included removal of suspended solids and sediments, and reduction in pollutants concentrations such as hydrocarbons and heavy metals (Bryce, 2001).

Figure 2.14: Roadside swale; West Grange, Dundee.

Source: Apostolaski and Jefferies (2005)

According to Woods-Ballard *et al.* (2007), swales are of three types:

a) *dry swales* which includes a filter bed of prepared soil thereby providing additional treatment, remaining dry most of the time,
b) *wet swales* usually applied to poorly drained soils, are usually wet and marshy and encourages biodegradation treatment and
c) *standard conveyance swales* which are applied to convey runoff to the next stormwater treatment stage.

All the SuDS devices described in this section can be connected together to form a cascade or management train which treats stormwater serially, such that its flow rate and volume is reduced and the resultant water is less polluted and this is discussed in the next section.

2.7 SuDS stormwater management train

As discussed in section 2.5, the philosophy behind SuDS is to mimic the natural drainage of a site/catchment area and for this to be effectively achieved a SuDS stormwater management train is required. The application of SuDS stormwater management train is cost effective and beneficial in improving water quality compared to end-of-pipe solutions (Heal *et al*., 2008; Bastien *et al*., 2010). This concept involves the serial use of drainage techniques to reduce flow rates, flow volume and pollution in rural and even developed areas as these drainage techniques can be retrofitted. However, the management train can be limited by individual site constraints, such as lack of space and poor drainage of site due to impermeable soils and this can ensure that the overall benefit of the management train is not fully achieved. SuDS, as an integrated approach to stormwater and surface water management, creates opportunities for stormwater control, flood risk management, groundwater recharge, water conservation and re-use (Wilson *et al*., 2005; Woods-Ballard *et al.,* 2007; Bastien *et al*., 2010). The flow chart in figure 2.15 illustrates the hierarchy of stormwater management train as stormwater moves from source to point of discharge.

Figure 2.15: The conveyance of stormwater from source to point of discharge.
Source: modified from CIRIA, (2005)

In establishing a stormwater management plan, good housekeeping is essential. Good housekeeping helps to prevent excessive runoff and mitigate pollution. Some good housekeeping techniques include enlightening programmes for stakeholders and maintenance of pollution sources e.g. by sweeping, rainwater harvesting/re-use (D'Arcy and Frost, 2001; Wilson *et al*., 2005; Woods-Ballard *et al.,* 2007). Source control of runoff treats water at, or

very near, the runoff source and examples include green roofs and permeable pavements. Site control involves the management of runoff in a particular site or local area and it involves steering water from areas around the source to areas where runoff can be treated. For example, water from roofs and car parks can be conveyed to infiltration devices and detention basins for treatment. Regional control involves the treatment of runoff from several sites e.g. the use of wetlands.

Conveyance of water between each hierarchy of control should include the use of natural systems such as swales and filter strips, although the use of pipes may be unavoidable especially in cases of space constraints. Generally, it is best for runoff to be treated at/near the source and should only be conveyed elsewhere if runoff cannot be managed on site. Conveyance of runoff may occur if it exceeds the capacity of natural drainage of the site or if the pollutant concentration is high thereby necessitating additional treatment than that which natural drainage can provide (Ellis *et al*., 2002; Ellis *et al.*, 2004). Factors that need considering when establishing a site's stormwater management train include flood risk and drainage characteristics of the site, cost-effectiveness, maintenance frequency, local land use, involvement of local people and overall effect of the management train on the hydrological cycle. In making an active decision, compromise has to be reached between different stakeholders and the risk associated with the various design options (CIRIA, 2005; Wilson *et al*., 2005; Woods-Ballard *et al.,* 2007). In addition to improving water quality through stormwater management trains, the various SuDS devices discussed have to be truly sustainable in terms of their component materials because sustainability in SuDS has been limited to costs, water conservation and water quality alone (Shaffer *et al*., 2009) and this is discussed in the next section.

2.8 Sustainability of SuDS components

Sustainability in SuDS has been limited to arguments and concerns over:

(a) maintenance requirements because of the need for regular inspections and upkeep of SuDS,

(b) the fate of contaminants especially accumulation of potentially toxic non-biodegradable compounds with the associated risks of groundwater pollution due to leaching, and polluted sediments/soil that needs to be landfilled,

(c) impoverished ecology due to the introduction of alien, invasive plant and animal species during SuDS construction which makes ecosystems unstable and

(d) field evidence of SuDS failures (Heal, McLean and D'Arcy, 2004; Jefferies *et al*., 2009; Casal-Campos, Jefferies and Momparler, 2011).

Other criteria for measuring sustainability in SuDS include life cycle costs (Ellis *et al*., 2004; Ellis, Lundy and Revitt, 2011) and catchment dynamics (Scholz, 2006) which includes rainfall and infiltration characteristics, runoff quality and flood protection (Kellagher and Udale-Clarke, 2008).

However, sustainability in SuDS should not only be limited to costs, water conservation and water quality alone but also to its overall structure and components. When considering the sustainability of materials employed in the construction of SuDS devices, SuDS components may be as unsustainable as components of conventional drainage systems (Shaffer *et al*., 2009). In fact, SuDS may actually have more significant environmental impacts (e.g. resource depletion, release of emission and waste generation), social impacts (e.g. noise pollution and traffic associated with transportation of materials) and economic impacts (e.g. cost implications of consumption of water and energy), compared to conventional drainage systems (Shaffer *et al*., 2009; WRAP 2010). This is because SuDS devices sometimes require slightly more materials in their construction (e.g. the sub-base of permeable pavements) compared to conventional systems, and this involves the utilisation of more natural resources thereby increasing unsustainability. For example, the use of thicker sub-bases in SuDS required for water storage implies that more aggregates are needed and larger volumes of soil

excavated and transported for disposal or re-used elsewhere thereby increasing their overall environmental impact compared to conventional drainage systems (Shaffer *et al*., 2009). Also 'hard' materials such as concrete and gravel used in SuDS are similar to materials used in conventional drainage systems and therefore the impact of the manufacture and transportation of these materials on the environment, from source to site, may also be similar in both systems (Interpave 2008; Shaffer *et al*., 2009). Table 2.1 shows the different materials used in various SuDS devices and their application.

Materials used in SuDS devices	Uses	Application
Cement	To increase the structural capacity of sub-bases	Permeable pavements, soakaways
Impermeable liners	To aid water retention and harvesting, and to prevent groundwater contamination by pollutants from contaminated sites	Permeable pavements, swales, ponds
Concrete paving blocks (with enlarged joints on the sides of each unit)	Used for the construction of permeable surfaces to encourage infiltration of runoff	Permeable pavements, filter drains
Geotextiles	Acts as filter for particles and pollutants, allowing infiltration into the ground or storage	Permeable pavements, filter drains, swales
Rough crushed rock	Used for sub-base construction to increase strength, permeability and storage	Swales, wetlands, permeable pavements
Gravel	Used for sub-base construction and as a porous surface to aid runoff infiltration	Filter drains, swales, permeable pavement
Topsoil	Sustains vegetative cover which aids infiltration and attenuation	Swales, filter strips, green roofs, wetlands
Lattice plastic cellular units	Used for sub-base construction and as a substitute to crushed aggregates	Permeable pavements

Materials used in SuDS devices	Uses	Application
Dense bitumen macadam	Used to protect the upper layer of permeable sub-bases during permeable pavements construction, in order to prevent the contamination and blockage of the sub-base layer by construction debris and dust	Permeable pavements
Asphalt	Mixed with aggregates to create porous surfaces for water infiltration	Permeable pavements

Table 2.1: Uses and application of materials used in the construction of SuDS Devices.
Source: Interpave (2006); Interpave (2008).

In order to address the issue of sustainability in construction, the UK government launched an initiative known as the 'Strategy for Sustainable Construction' on the 11th of June 2008 (BIS, 2008). The Strategy was initiated because it recognised the significant impact of the construction industry on natural resources and the environment as a whole, due to its processes and outputs, and the need for radical change. It stated that construction processes should be environmentally, socially and economically sustainable and proposes that this can be achieved by resource efficiency (and hence profitability), use of sustainable products thereby cutting down energy consumption and environmental impacts due to sourcing of raw materials, manufacture or transport, and energy efficiency and waste minimisation (BIS, 2008).

In view of this initiative, one way to improve the sustainability of SuDS components is to source for more sustainable construction materials including recycled materials (BIS, 2008; WRAP, 2010). However, for the purpose of this research, sustainability of vegetative SuDS components will be the focus because of the reasons discussed in the next section.

2.9 Vegetative SuDS

Vegetative SuDS are a group of drainage devices that can effectively attenuate runoff and remove pollutants such as motor oil and heavy metals. They are designed to complement or replace conventional drainage systems and usually contribute to the landscape and amenity value of a given area (Revitt and Ellis, 2001; Highways Agency, 2006; European Commission, 2012). Examples of vegetative SuDS include swales and filter strips which convey and treat runoff, constructed wetlands, green roofs, detention and retention basins which treat runoff as it flows slowly through the system, and ponds which treat water at rest (Revitt and Ellis, 2001; HighwAltenays Agency, 2006). These devices have been discussed in section 2.6. These systems are usually more effective when combined as part of a stormwater management train (section 2.7). For example, a swale which reduces the load of suspended solids in surface runoff can be followed by a constructed wetland to treat soluble pollutants (Highways Agency, 2006; Woods-Ballard *et al*., 2007).

Vegetative SuDS usually employ vegetation and/or topsoil as primary treatment elements (CIRIA C523, 2001; Highways Agency, 2006) and these components take up pollutants from stormwater thereby improving water quality (Woods-Ballard *et al*., 2007). They also help to sequester carbon thereby contributing to the mitigation of climate change effects (discussed in section 2.5) (Pan *et al*., 2004; Tratalos *et al.,* 2007; Charlesworth, 2010; Warwick and Charlesworth, 2013). In addition to vegetation and topsoil, some vegetative SuDS devices incorporate gravel drain beds (e.g. swales and infiltration basins) for retaining and treating runoff and as an underlay for stability (e.g. filter strips) (American Rivers, 2004). For these reasons, MacDonald and Jefferies (2003[b]) confirmed that it is best practice to incorporate gravel below topsoil as shown in the figure 2.16.

Figure 2.16 Schematic representation of a swale, an example of a vegetative SuDS device.
Source: SuDS Wales (2013)

Figure 2.16 shows the illustration of a swale comprising of a vegetative layer made up of vegetation and soil, gravel drain bed for water storage and an under drain pipe system for conveying water, that does not get infiltrated into native soil, into storm sewer systems, reservoirs for water harvesting purposes or other SuDS devices as part of a stormwater management train (Woods-Ballard *et al*., 2007).

Vegetative SuDS components can increase the permeability of an area as topsoil soaks up rainfall allowing water to slowly migrate into the nearest water body or groundwater, and the vegetation reduces the flow rate of surface runoff. As a result, the overall effect of rainfall has little impact on the flow rates of surface runoff and these effects are spread out over a period of time (CIRIA C523, 2001). A case study on London by Wilby and Perry (2006) showed that the incorporation of green spaces in the form of vegetative SuDS (which are core elements of green infrastructure) and improved building designs into city planning would greatly reduce urban heat island effects. The latter is caused by the absorption of heat by impermeable surfaces thus increasing surrounding air temperature (Ellis, 2012; European Commission, 2012). The study also demonstrated that vegetative SuDS improve flood control and water quality compared to other SuDS devices (Wilby, 2007). Table 2.2 shows the degree of

impermeability of different land uses in urban areas, showing that land covered with vegetation are more permeable to stormwater than paved surfaces and built-up areas.

Table 2.2: The degree of impermeability for different land use.

Source: Haase (1986)

Table 2.2 showed that inner urban areas (such as city centres), dense residential areas and a combination of both have a high degree of impermeable surfaces, whilst areas possessing permeable surfaces, such as gardens, had a lower degree of impermeability. Table 2.3 also shows that roofing and paving materials were the cause of increased impermeable surfaces in urban areas thereby hindering natural infiltration, while permeable surfaces such as vegetative surfaces reduced impermeability. This indicates that the presence of vegetated surfaces increases the permeability of an area reducing runoff flow, encouraging natural infiltration and controlling flooding.

Table 2.3: Degree of impermeability for different construction materials.

Source: Haase (1986)

Vital treatment processes that take place in vegetative SuDS include sedimentation and filtration of particle, adsorption of organic pollutants (hydrocarbons, pesticides and organic matter) and heavy metals by vegetation and soils, biodegradation of organic pollutants, phytoremediation by vegetation, and precipitation of dissolved pollutants, and these processes are discussed further in section 2.16. Table 2.4 summarises the various treatment processes carried out in different vegetative SuDS devices while table 2.5 summarises the pollutant removal performance of vegetative SuDS devices.

N. B. Bold type indicates dominant processes.

Table 2.4: Principal treatment processes in vegetative SuDS.

Source: Highways Agency (2006)

Table 2.5: Pollutant removal performance of vegetated SuDS.

Highways Agency (2006)

In constructing vegetative SuDS, stability is of immense importance and this includes the way soil is handled, stored and used on site. Great care must be taken during the excavation and placement of soil during construction so as to prevent the compaction of soil and subsoil, as well as damage to soil structure (Highways Agency, 2006; Woods-Ballard *et al*., 2007). Soil type is also vital in determining the stability of vegetative systems especially for devices which experience greater flow of water such as swales, compared to ponds and basins. For the system to be stable, flow velocities must either be low enough to prevent erosion or the soil must be supplemented to decrease input velocities of runoff. For example, soils consisting of a mixture of sand, clay and gravel may be considered as erosion resistant and ideal whilst soils comprising of fine sand and silt are erosion susceptible (Highways Agency, 2006).

Other factors for consideration in vegetative SuDS construction includes the rate of soil permeability, the area of permeable and impermeable (paved) ground to be drained, duration and intensity of rainfall and storms, runoff quality and quantity, availability of land and characteristics of receiving waters (van Borcke, 2003; Highways Agency, 2006). The types of vegetation commonly used in vegetative SuDS are grasses, reeds, rushes and shrubs, and their growth characteristics are important to the effectiveness of vegetative SuDS in treating pollutants. The plant species must have a combination of rapid establishment and recovery, and tolerance to salt, wet conditions and inundation (Highways Agency, 2006).

Vegetated SuDS devices need to have certain specifications to enhance natural drainage and table 2.6 shows the required specifications in conjunction with evaluation of construction costs and environmental ratings. The environmental ratings show the sustainability of these devices on a scale of low to high, with the most sustainable device having a high environmental rating (van Borcke, 2003).

Table 2.6: Retention systems used to enhance natural drainage.

Source: van Borcke (2003)

2.10 Case Studies on Vegetative SuDS

To understand the application of vegetative SuDS devices, the following sections discuss four case studies which highlight the advantages and disadvantages of various vegetative SuDS devices in relation to their real-life application.

2.10.1 Case Study 1: Vegetative SuDS as source control of pollution (Jefferies *et al*., 2008)

According to Jefferies *et al*. (2008), source control in SuDS has provided an avenue for the retention and degradation of pollution close to their source. However, the risk of this method of pollution control to groundwater quality has been unclear due to insufficient evidence. As a result, research was carried out on vegetative SuDS with the aim of determining the behaviour and fate of pollutants within them; and the efficacy of soil-based SuDS in treating these pollutants compared to conventional drainage systems, which in this case was an end-of-pipe pond solution. The devices applied in the study were a filter strip, filter drain, swale and detention basin, and the pollutants analysed for were total petroleum hydrocarbons (TPH), polycyclic aromatic hydrocarbon (PAH), heavy metals as well as nutrients. The research included artificial dosing experiments and field monitoring experiments and was carried out in four phases:

- Desk-top nutrient study linking nutrients from agricultural activities to urban areas.
- A semi-field study to determine the movement and degradation of pollutants.
- A laboratory-based degradation study.
- A field study to measure actual concentrations of pollutants.

Research objectives included monitoring the movement of pollutants through soil to groundwater, measuring the degradation of pollutants, identifying degradation products, determining optimal conditions for pollutant's degradation and determining the fate of nutrients. Findings from the study showed that:

1. the risk to groundwater by pollutants in runoff was low due to their low vertical migration rates,

2. soil/vegetation in SuDS were effective in the attenuation of pollutants,
3. the majority of pollutants were retained in the top 10cm of soil, and pollutant concentrations in pond sediments (conventional drainage system) were generally higher than in the soil-based devices,
4. TPH and PAH concentration were higher in the receiving detention basin soil than in filter drain soil suggesting their accumulation over time,
5. vegetation on filter strips and swales were effective in retaining pollutants,
6. heavy metals will accumulate on vegetation and surfaces of soil layers of infiltration-based SuDS, thereby reducing the risk of groundwater pollution and
7. sediments from 75% of the conventional ponds examined contained high concentrations of pollutants including TPH and could be classed as hazardous waste.

It was concluded that the risk of groundwater pollution by pollutants passing through the soil was very low. The research also highlighted the benefits of using soil/vegetation-based systems in treatment trains when treating surface runoff, as compared to conventional end-of-pipe solutions (Jefferies *et al*., 2008).

2.10.2 Case Study 2: Vegetative SuDS as a retrofit device (Macer-Wright *et al.*, 2003)

This case study involved the evaluation of the adoption of swales for road edge channels in the 23km long A120 Stansted to Braintree dual carriageway project (an upgrade of an existing single carriageway route) compared to conventional drainage systems. The evaluation, carried out by Macer-Wright *et al.* (2003), showed that there were greater environmental benefits in applying vegetative SuDS compared to conventional drainage systems. There was less demand for concrete throughout the project, thereby reducing the demand for aggregates and the amount paid on aggregate tax, hence reducing the depletion of natural resources and improving sustainability. Other benefits to the environment included reduction in fuel consumption and engine emissions as well as reduction in soil compaction (which are all characteristic of construction sites) due to the reduction in lorry movement to and from the quarry, batching plant and site. Initial pollutant deposition and removal within swale vegetation and soil by processes of enhanced immobilisation, sedimentation and natural

degradation of pollutants helped to reduce maintenance requirements of component parts (e.g. oil separators in swales), compared to concrete drains. Increased storage of runoff as a result of the lower flow rate, encouraged by a vegetated channel, provided more time for mitigation measures in cases of emergency such as spillages, compared to a concrete channel. In terms of amenity value, there was improved natural aesthetics due to green vegetation around the verges and reduction of earthworks excavation as swales do not require extensive excavations, compared to that of concrete channels. Economically, the application of swales provided better cost savings because fewer materials were utilised (such as concrete and aggregates) and there was a reduction in the employment of specialised staff such as engineers (Macer-Wright *et al.*, 2003).

However as laudable as the benefits were, there were some limitations in the adoption of swales over conventional drainage system in the dual carriageway project. Swale constructions were delayed till after the completion of construction works in the autumn with delays due to rain. This was because swale construction is weather-sensitive and so prolonged heavy rainfall can affect the consistency of the swale materials rendering them unusable for days whilst, concrete constructions are usually not affected by wet weather conditions. There was also a great risk that the swales would be subject to its full working flow of runoff and rainwater before the establishment of vegetation, and hence re-working and re-seeding of the swale may have to be carried out. Also, spillages of hazardous and flammable substances into swales pose a greater risk of ignition than a pipe drain due to increased storage of runoff. It was, however, concluded that the environmental benefits of incorporating swales into retrofit schemes far outweighed its disadvantages (Macer-Wright *et al.*, 2003).

2.10.3 Case Study 3: Biodegradation in Vegetative SuDS (Napier *et al*., 2008)

A lysimeter soil core study by Napier *et al*. (2008) was undertaken to measure the leaching potential of different soil types, representative of soils predominant in vegetative SuDS, comprising sand, silt, clay and a specially constructed SuDS core profile consisting of layers of gravel, sand and a top layer of biologically active topsoil. The different soil profiles were exposed to concentrations of oil relating to traffic conditions.

Table 2.7: Oil mass balance for SUDS, sand, silt and clay soil core lysimeters.

Source: Napier *et al*. (2008)

Results, as shown in table 2.7, showed little leaching of oil through the four cores samples (≤0.07%) but 71-81% of the oil applied was degraded in all the samples except for sand. The SuDS core profile had the highest oil degradation rate and lowest leaching rates which was attributed to the presence of the biologically active topsoil.

2.10.4 Case Study 4: Vegetative SuDS as stormwater management train (Zakaria *et al*., 2007; Ghani *et al*., 2008)

In Malaysia, a middle income country in a tropical region, stormwater runoff especially in urban areas was managed by conventional drainage systems but these systems were challenged by flash floods, water pollution and water scarcity. As a result, vegetative SuDS techniques were recommended and applied as part of a management train to reduce rates and volume of runoff and pollutant loads. The techniques applied were termed 'BIOECODS' i.e. Bio-Ecological Drainage Systems. BIOECODS consists of three main components namely ecological swales, sub-surface bio-filtration storage and ecological ponds, comprised of a wet pond, a detention pond, a constructed wetland, a wading river and a recreational pond (Zakaria *et al*., 2007; Ghani *et al*., 2008).

This approach was adopted because surface water features, which are common in temperate regions, pose risks of creating environments that favour the breeding of mosquitoes and other vectors of diseases in tropical regions. The application of sub-surface bio-filtration storage systems all through the BIOECODS train created anaerobic environments which is not conducive for mosquitoes, thereby making these types of vegetative SuDS suitable for tropical

regions (Charlesworth and Warwick, 2012). The swale (figure 2.17a) and bio-filtration underground storage were designed to carry out pre-treatment processes of infiltration and settlement of runoff debris. The wet and detention ponds allowed for sedimentation, flocculation and biological treatment before being discharged via a wading river (figure 2.17b) into the constructed wetland for further treatment, and finally into the River Kerian (Zakaria *et al*., 2007; Ghani *et al*., 2008).

Figure 2.17a: Ecological swale with gravel lining.

Source: Ghani *et al*., (2008)

Figure 2.17b: A wading river.

Source: Ghani *et al*., (2008)

Overall these case studies showed that there are sustainable benefits in applying vegetative SuDS to the drainage of stormwater because of their ability as:

a) a source control of pollution,
b) retrofit drainage devices,
c) treatment for stormwater pollutants and
d) a management train.

However, in addition to the limitations of vegetative SuDS discussed in section 2.10.2, other limitations of vegetative SuDS include impermeable soils (e.g. clay) which does not encourage infiltration of surface runoff; shallow water table which encourages pollution of groundwater as infiltration does not take place long enough for polluted runoff to be treated; pollution of groundwater if runoff is heavily polluted for vegetative SuDS devices to cope with; the use of liners such as clay or geotextiles at the base of vegetative SuDS such as swales has been used to protect groundwater pollution but there is a risk of vegetation dying off in dry conditions; adverse effect to ground stability especially if infiltration is constrained to a limited area and not spread over a wider area (Woods-Ballard *et al*., 2007; NIEA, 2009). Another major limitation of vegetative SuDS is the sustainability of materials used in constructing them i.e. topsoil and gravel (as discussed in section 2.8). These materials are natural resources and in order to conserve them, suitable sustainable alternatives, preferably waste or recycled materials, are required to either replace or substitute these elements (BIS, 2008), but before this can be achieved an understanding of legislation governing waste and recycling is required and this is discussed in the following section.

2.11 Waste Legislation

Over 80 million tonnes of waste are generated every year from households, commerce and industry. 37% of household waste was recycled and 50% of recycled household waste was landfilled in 2008/2009. In 2008, the waste management sector in the UK accounted for 3.6% of the UK's total estimated emissions of greenhouse gases with 89% arising from landfill, 9% from waste-water handling and 2% from waste incineration. As a result, policies and

regulatory restrictions have been put in place to combat these problems and improve environmental quality; one of which includes waste recovery (DEFRA, 2011[a]).

A major driver for waste recovery is the imposition of stringent legislation such as the EU Waste Framework Directive which provides the main legislative framework for the collection, transport, recovery and disposal of waste without endangering human health or causing harm to the environment (DEFRA, 2012[a]). As described by the waste hierarchy in figure 2.18, measures are taken to encourage firstly, the prevention or reduction of waste production and its effects; and secondly waste recovery by means of re-use/reclamation, recycling, or the use of waste as a source of energy e.g. by processes of anaerobic digestion. Waste disposal to landfills is to be considered as a last resort (DEFRA, 2010[c]).

Figure 2.18: The waste hierarchy.
(DEFRA, 2010[c])

The EU Waste Framework Directive's main requirements are supplemented by other Directives for specific waste streams such as the Landfill Directive (DEFRA, 2012[a]). The

Landfill Directive represents a shift in the way waste is disposed with emphasis on waste minimisation which encourages waste re-use, recycling and energy recovery. The Directive's overall aim, according to DEFRA (2011[a]), is "to prevent or reduce as far as possible negative effects on the environment, in particular the pollution of surface water, groundwater, soil and air, and on the global environment, including the greenhouse effect, as well as any resulting risk to human health, from the landfilling of waste, during the whole life-cycle of the landfill". This directive has encouraged the adoption of cheaper alternatives to waste disposal such as recycling of biodegradable wastes in composting processes. Aspects of the directive cover leachate management, water control, soil and water protection, and methane emissions control. In achieving the aims of the Directive, stringent technical requirements for waste and landfills were introduced and targets were set to reduce the amount of biodegradable municipal waste being disposed into landfill, since biodegradable waste in landfill produces methane, a greenhouse gas which increases greenhouse emissions and contributes to climate change. One of such requirements is the landfill tax which is the tax imposed on waste disposed at designated landfill sites, with the view of encouraging the reduction of waste being disposed into landfill. The recent increase in landfill tax from £56 per tonne to £64 per tonne in April 2012, and its subsequent increase by £8 every year till at least April 2014, will further encourage organisations to seek alternatives to landfill (Sequi, 1996; HM Revenue and Customs, 2012).

One way that vegetative SuDS can be made more sustainable, thereby fulfilling The EU Waste Framework Directive and The Landfill Directive as well as mitigating the depletion of natural resources, is by replacing the natural resources that constitute these systems with more sustainable materials, i.e. waste and/or recycled materials. These replacement materials have to perform at least as well as the original materials, but must have less significant environmental, social and economic impacts compared to conventional materials. Topsoil and gravel are natural resources which constitute vegetative SuDS components and recycled materials such as compost and recycled aggregates can be used as their replacements respectively thereby making vegetative SuDS more sustainable (Lazarus, 2005). Compost can

be derived from municipal wastes and recycled aggregates can be derived from construction wastes and this will be discussed in the next four sections.

2.12 Compost

Municipal solid wastes (MSW) are heterogeneous solid wastes generated by domestic households, businesses and light industries, and can have a direct impact on environmental quality and natural resources (Golueke and Diaz, 1996). About 65% of civic-amenity waste is organic in nature and low in contaminants, and therefore recycling should be encouraged especially with current legislation on waste, as discussed in section 2.11 (Keeling *et al*., 1995). MSW can be divided into two components – organic and inorganic. The organic components can be further divided into three broad categories:

a) putrescible wastes – which decompose quickly and if not controlled, undergoes putrefaction,
b) fermentable wastes – which also decompose rapidly but without putrefaction, and
c) non-fermentable wastes – which decompose very slowly (UNEP, 2005).

Resource recovery is a major element in waste management and as such the organic component of MSW can be recycled in commercial composting processes (Diaz *et al*., 2005; Defra, 2011[a]). Composting is commonly used as a method for disposing, utilising and managing organic wastes with the product being compost (Goyal *et al*., 2005; Narkhede *et al*., 2010). Compost is applied to soil as amendments to increase soil organic matter (Evanylo and Daniels, 1999; Ozores-Hampton and Obreza, 1999), to improve crop yield (Litterick, 2008; Copetta, 2011), to inoculate biodegrading microbial populations into soils (Atlas and Barthar, 1993; Ros *et al*., 2011), and improve the aggregate stability of soil by at least 45% (Arthur *et al*., 2011). Studies have shown that the application of compost to soil as an amendment helps to improve soil structure, increase soil moisture, modify and stabilise pH, provide nutrients, bind contaminants, increase soil organic matter content, increase soil biodegrading organisms and hence biodegradation, and improve soil resistance to erosion (Reganold *et al*., 1987; Beyer *et al*., 1999; Ibekwe *et al*., 2001; Faucette *et al*., 2004) although Edwards *et al*. (2000) and Arthur *et al.* (2011) indicated that compost had no effect on erosion.

Compost quality is closely related to the stability and maturity of compost (Wang *et al*., 2004), which have effects on organic matter decomposition and potential phytotoxicity levels respectively (Laviv and Lieth, 2008; Epstein, 2011). Compost stability was defined by Haug (1993: 602) as the 'microbial degradation rate of organic matter under aerobic conditions' and hence a lower degradation rate corresponds to a higher level of compost stability. The degradation of compost high in soluble organic carbon, such as green compost and animal manure, produces high concentrations of CO_2 immediately after their addition to soil due to increased microbial activities (Marstorp, 1996), and this can lead to decreased O_2 concentrations and increased anaerobic conditions in the soil. Some of the intermediate products of biodegradation, such as alcohols, phenols and acids, may be toxic to plants and low levels of O_2 may stimulate the solubilisation of heavy metals in soil by reduction processes. Apart from phytotoxicity, the application of non-composted or non-stabilised materials to soil can lead to the immobilisation of nutrients either within soil microbial populations during their metabolism or incorporated into developing humus (a stable organic material), rendering them unavailable to plants (Keeling *et al*., 1994[a]; Butler *et al*., 2001; Cambardella *et al*., 2003; Laviv and Lieth, 2008). As a result compost is usually allowed to 'mature' and stabilise before being applied for agricultural purposes.

Maturation will allow degradation of labile organic matter by soil microorganisms thereby reducing phytotoxicity and release nutrients as well as attaining equilibrium on soil microbial activity (Garcia *et al*., 1991; Keeling *et al*., 1995; Bernal *et al*., 1998[a]). The benefit of applying mature compost to soil was reported by Bernal *et al*. (1998[b]). It was reported that mature compost had a low degree of organic carbon degradation in soil and that degradation of more than 25% of total organic carbon (TOC) was indicative of 'immature' compost. The only exception to this observation is in the decomposition of slowly degradable wastes such as maize straw, in which case TOC may be more than 25% as these materials require longer composting times. It was then concluded that maturation of compost was necessary for carbon stabilisation which is essential in soil conservation and reclamation (Bernal *et al*., 1998[b]). However, Keeling *et al*., (1991) and Keeling *et al*., (1994[b]) reported that unstable compost especially, refuse-derived compost, possess long-term nutrient-releasing benefits and

beneficial plant growth properties. A further study by Keeling *et al*., (1995) showed that less stable compost gave higher plant biomass, microbial biomass and nutrient yield over the one year period of study, indicating that unstable compost can attain stability as soil amendments over an extended period of time.

2.13 The Composting process

Composting is a biological process which involves the aerobic decomposition of labile organic matter into carbon dioxide (CO_2), water vapour, inorganic nutrients, ammonia and humus (compost) (Senesi, 1989; Beck-Friis, 2001; Goyal *et al*., 2005; Insam and de Bertoldi, 2007). Composting was defined by Insam and de Bertoldi (2007: 26), as 'a biodegradation process of a mixture of substrates carried out by a microbial community composed of various populations in aerobic conditions and in the solid state'. It includes an exothermic process that produces energy in the form of heat resulting in increased temperature within the substrate due to the presence of fresh organic matter, which stimulates microbial activities. Decomposition depends on the nutrient content, quantity and biodegradation rates of the carbon compounds they contain i.e. carbohydrates, fatty acids, amino acids and lignin (Ajwa and Tabatabai, 1994; Insam and de Bertoldi, 2007).

The composting process is carried out mainly to avoid phytotoxicity associated with un-stabilised organic matter which is the result of incomplete composting, to reduce the number of organisms that could be pathogenic to plants and animals to numbers that does not pose a health risk, and to produce organic fertilisers from recycled organic wastes and biomass (Insam and de Bertoldi, 2007). The composting process is essentially in four phases:

1) *Initial mesophilic phase* with temperature ranging between 25^{o}C - 40^{o}C. In this phase, also known as the decomposition phase, easily degradable compounds such as proteins and sugars are degraded rapidly by primary decomposers e.g. bacteria, actinobacteria and fungi, and are sometimes aided by organisms such as worms, millipedes and mites, which act as catalysts. The activities of primary decomposers lead to an increase in temperature giving rise to the thermophilic phase.

2) *Thermophilic phase* with temperature ranging between 35^{o}C - 65^{o}C (optimum at 62^{o}C). As temperature rises, microorganisms which are better adapted to higher temperatures colonise the substrate thereby phasing out the mesophilic organisms almost entirely. Thermophilic microorganisms then continue decomposition of easily degradable compounds including dead mesophilic microorganisms. Although temperature can rise to about 80^{o}C, most microorganisms are destroyed at temperatures exceeding 65^{o}C. However, thermophilic bacteria, thermotolerant bacteria and actinobacteria can remain active at higher temperatures. The disadvantage of composting temperatures exceeding 70^{o}C is that most mesophilic microorganisms are killed and hence their re-colonisation may be quite slow after temperature decreases. This phase is vital for the sanitisation of compost as human and plant pathogens (*E. coli* and *Fusarium spp.*) as well as weed seeds and insect larvae are killed off by the heat. The production of antibiotics by actinobacteria also helps in sanitisation.
3) *Second mesophilic phase* where temperature falls back to between 25^{o}C - 40^{o}C. This marks the end of the bio-oxidative phase where biodegradation reduces drastically due to the exhaustion of easily degradable substrates. The activities of thermophilic microorganisms cease and temperature falls to almost ambient temperatures. Mesophilic microorganisms re-colonise the substrate either from surviving spores or from external inoculation. This phase is characterised by mesophiles (bacteria and fungi) that can easily degrade starch and cellulose.
4) *The maturation phase* is a stabilisation period which involves the humification of composting substrate producing highly stabilised and mature compost free of phytotoxic compounds and suitable for plant growth. Mature compost consists predominantly of compounds that are not biodegradable such as lignin-humus complexes, and the proportion of fungi increases in relation to bacteria (Garcia *et al*., 1991, Bernal *et al*., 1998[a]; Ishii *et al*., 2000; Beck-Friis, 2001; Insam and de Bertoldi, 2007).

2.14 Standardisation of compost

Most compost produced is used for agricultural purposes and their application has to be managed and regulated to prevent environmental pollution. One of such regulatory standards is PAS 100 which is a Publicly Available Specification for Composted Materials, 2011 published by the British Standards Institute (BSI). This standard specifies the minimum quality that compost must possess such that any risk associated with its handling and application is limited to acceptable levels. The aim of PAS 100 was to produce baseline specifications for compost thereby informing compost producers of what consumers require from compost products (BSI, 2011). Compost quality requirements are the key emphasis of PAS 100 and these are requirements that compost should meet in order to comply with PAS 100. The quality requirements cover elements such as human pathogens (e.g. *E. coli*), potentially toxic elements (e.g. heavy metals), physical contaminants (e.g. glass, metal and plastic), phytotoxins (alkaloids and phenolics) and weed propagules (ESA, 2002; WRAP, 2003; BSI, 2011).

In addition to the PAS 100 specifications, further specifications are required depending on the specific use of compost and an example of such specifications is the Compost Specification for Use in Grass Establishment (CSUGE). This specification sets out the requirements needed for the use of compost in grass establishment such as on golf courses, pitches, swales and filter strips. CSUGE has been designed to be used in conjunction with the PAS 100 specifications (WRAP, 2003). Tables 2.8 and 2.9 summarises the specifications for PAS 100 and CSUGE respectively.

Table 2.8: BSI PAS 100 specifications for composted materials.

Source: WRAP (2003); BSI (2011)

Table 2.9: CSUGE specifications for composted materials.

Source: WRAP (2003)

2.15 Recycled aggregates

Aggregates, such as gravel, are known to improve the performance of vegetative SuDS when applied as a layer beneath the growth medium (MacDonald and Jefferies, 2003[b]). An alternative to gravel (which is a natural resource) in vegetative SuDS is construction, demolition and excavation wastes (CDEW) which is one of the largest waste streams in the UK (Lamb, 2006; WRAP, 2007[a]). This inert waste accounts for over 60% of UK total waste (approximately 110 million tonnes) and accounts for approximately 55% of primary aggregates used in the UK construction industry each year (Dhir and Paine, 2007). CDEW such as rocks, crushed bricks and crushed concrete are disposed in landfills (Poon and Chan, 2007). However, the increasing distance between demolition sites and disposal areas due to urbanisation has led to increasing transportation and energy costs (Gupta, 2009), depletion of landfill spaces, contamination of soil and groundwater by landfill leachate and current legislation on landfill (discussed in section 2.11). Therefore, it has become important to find viable sustainable ways of recycling CDEW (Poon and Chan, 2007). One way of achieving this is the re-use of CDEW in construction materials (such as concrete) (Dhir and Paine, 2007; Algin and Turgut, 2008) and road construction (such as concrete pavements) (Chen *et al*., 2011). Paige-Green and Hongve (2003) also reported that the use of local materials (which

consisted of aggregates such as crushed rocks; Overby, 1999) for road constructions was a more sustainable alternative to the use of gravel

Recycled aggregates (RA), according to Dhir and Paine (2007:7), are 'aggregates resulting from the processing of inorganic material previously used in construction, e.g. crushed concrete, masonry and brick'. Recycled concrete aggregate (RCA), which is RA predominantly derived from crushed concrete, is considered better than RA derived from other CDEW based on their performance characteristics and hence, is commonly re-used in concrete production for construction purposes (Dhir and Paine, 2007). RCAs are currently being re-used as fill materials in land reclamation, as foundations for buildings and in the production of concrete, concrete bricks and blocks (Dhir *et al*., 1999; Poon *et al*., 2002). The other CDEW mentioned above can also be re-used for construction purposes, however, their uses can be limited because of their deteriorating effect on concrete over time, due to the presence some harmful substances they may contain (Dhir and Paine, 2007). For example, studies by Khalaf and DeVenny (2004) and Poon and Chan (2007) showed that crushed clay bricks can be re-used for construction purposes and Khalaf and DeVenny (2004) further showed that coarse crushed bricks were better suited as a substitute for coarse RCA in concrete production compared to fine crushed bricks.

However, Dhir and Paine (2007) showed that the presence of certain levels of compounds such as sulphates and chlorides in crushed brick aggregates may cause the expansive disruption of concrete leading to its deterioration. Also, Olotuah (2002), Dhir *et al*. (2005[a]) and Poon and Chan (2006) all reported that the use of fine crushed bricks (<5mm) was not suitable in the production of concrete paving blocks as it decreased the compressive strength and density, and increased the water absorption of the resulting paving blocks. Brick aggregates may be high in sulphur if the bricks were originally made with sulphate–rich clay or if the aggregates contained gypsum (calcium sulphate) which is present in the interior walls of buildings or plaster work; and its application to concrete production may lead to sulphate expansion of concrete which could be detrimental to construction (Dhir and Paine 2007).

Nevertheless, sulphate studies by Dhir *et al*. (2005[b]) on five types of bricks including two from demolition rubble showed that a maximum sulphate content of 0.1% by mass was obtained which was well below the BS 8500-2:2002 (BSI, 2002) limit of 1.0% by mass for acid-soluble sulphate content. This showed that there were no concerns with expansion resulting from the use of sulphate–rich brick aggregates. Chlorides in concrete could also lead to its deterioration (Shi *et al*., 2010) however further studies by Dhir *et al*. (2005[b]) showed that bricks had zero chloride content and therefore had no detrimental effect on concrete. The presence of alkali (sodium oxide usually from cement) and silica in aggregates can lead to expansive alkali-silica reactions (ASR) leading to deterioration of concrete and care should be taken to limit the alkali content of concrete. For example, when RCA with residual cement pastes on them are used in concrete production, it could increase the alkali content of concrete putting it at risk of ASR (Dhir and Paine, 2003). But studies by Dhir *et al*. (2005[b]) showed that RCA had a lower alkali content compared to brick aggregates. However, in considering the use of recycled aggregates in vegetative SuDS, stability is quite important because the sub-bases of these systems are porous and less compact in structure compared to conventional systems which have a more compact structure, because the aggregates they contain provide more stability and durability (Shaffer *et al*., 2009).

In order to assess the suitability of RA application in vegetative SuDS, they have to be assessed for contaminants that may likely compromise water quality. The Interdepartmental Committee on the Redevelopment of Contaminated Land values (ICRCL 59/83) and the Kelly indices guideline values are standard guidelines used for assessing contaminated land based on its use (ContaminatedLand, 2000). These guidelines can be used to assess contaminants that may be present in aggregates as they give threshold values for contaminants such as heavy metals and hydrocarbons.

In order to determine if compost and RA will be suitable to replace topsoil and gravel in vegetative SuDS devices, they must be able to carry out at least all the processes discussed in section 2.9 and table 2.4. However, to make this assessment, the types of pollutants being treated by these devices and the role of microorganisms in pollutant removal must first be

identified, as knowledge of these two factors is necessary to determine the effectiveness of vegetative SuDS devices. The next two sections discuss the types of pollutants present in runoff, their sources, processes of removal and the role of microorganisms in pollutant removal.

2.16 Pollutants treated by vegetative SuDS

In line with the objectives of SuDS in improving the quality of surface runoff, sources of pollution and types of pollutants have to be identified. Pollution can either be from point or non-point sources. Point source pollution is pollution discharged from a particular, usually traceable source, and discharges include sewage and process effluents from particular industries; and because their polluting constituents are usually known, their discharge can therefore be regulated to prevent pollution of receiving water bodies. Non-point source pollution usually does not have a single discharge point and contamination may occur as a result of diffuse pollution from urban runoff and nutrient release from farmlands (D'Arcy and Frost**,** 2001). Novotny (2003:33) defined diffuse pollution as 'pollution arising from land-use activities (urban and rural) that are dispersed across a catchment or sub-catchment, and do not arise as a process industrial effluent, municipal sewage effluent, deep mine or farm effluent discharge'. Diffuse pollution could arise as sheet runoff from fields, seepage into groundwater and collection of effluents from different minor point sources such as urban surface water drains (D'Arcy and Frost**,** 2001). Wilson *et al*. (2005) quantified the impact of diffuse urban pollution in a survey of nine streams across Scotland, receiving runoff from urban areas and results identified hydrocarbons as a major urban pollutant followed by heavy metals, with Pb, Cr, Ni, Zn and Cu being the most predominant heavy metals with concentrations exceeding sediment quality standards across the nine streams. This result was also corroborated by Beasley and Kneale (2004) and Poleto *et al*. (2009).

As the use of SuDS has developed over the years, it has become apparent that SuDS implementation has been applied mainly to new developments and this is quite understandable as new developments provide opportunities for SuDS consideration at early planning and design stages (D'Arcy and Frost**,** 2001; Ferrier, 2005). However, most diffuse pollution of

surface waters emanate from existing developments and diffuse pollution sources have to be quantified and controlled according to the EU Water Framework Directive (Environment Agency, 2006). Areas prone to diffuse pollution will benefit from retrofitted SuDS devices, such as vegetative SuDS, as part of a management train in order to reduce the impact of diffuse pollution. Retrofitting could be challenging though, as existing developments place physical and design constraints on SuDS implementation (Heal *et al*., 2005; Mitchell, 2005; Wilson *et al*., 2005); but evaluations carried out by Ellis (1998), Makropoulos *et al.* (1998), Sieker and Klein (1998), and D'Arcy and Frost (2001) have shown that there are many opportunities available for installing SuDS retrofits and the attendant positive effects on runoff. Other ways of effectively controlling diffuse pollution include the reduction of persistent pollution at the source (e.g. construction and motor industries), enforcing good housekeeping such as reducing litter and preventing the illegal dumping of oil and solvents, and raising awareness to curb polluting practices (Heal *et al*., 2005; Wilson *et al*., 2005). Table 2.10 below highlights some urban pollutants and their sources.

Pollutants	Sources of pollutant
Sediments	Winter de-icing operations, motor vehicles exhaust, road surface materials, construction work stockpiles and spillages, litter, pet droppings, vegetation, sewage, roofing materials, atmospheric deposition (Butler and Clark, 1995; D'Arcy and Frost**,** 2001).
Heavy metals	Atmospheric deposition, wear and tear of motor parts, corrosion of metallic surfaces, roofing materials, cleaning detergents, incinerator ash and fumes, oil, street dust (Monna 1997; Charlesworth *et al*., 2003; Beasley and Kneale, 2004; Wilson *et al*., 2005; Wander *et al.*, 2010; Charlesworth *et al*., 2011).
Nutrients	Pet droppings, fertilisers from farmlands and golf courses (Wander *et al*., 2010).

Pollutants	Sources of pollutant
Hydrocarbons	Coal combustion, motor oil and drips from vehicles in parking bays and vehicle service areas, grease from machinery, spillages from stores and fuelling sites, improper disposal of oil-related wastes (Menichin, 1992; Pitt *et al.,* 1995; Beasley and Kneale, 2004; Wilson *et al*., 2005; Ellis and Chatfield, 2006).
Organic matter	Farmlands, vegetation, sewage (Fernandez *et al*., 2000; D'Arcy and Frost, 2001).
Pesticides	Farmlands (SEPA, 1996).
Nitrates	Fertilisers (Barrett *et al*., 1999).
Ammonia	Sewage (Barrett *et al*. 1999; D'Arcy and Frost**,** 2001).
Chlorides	Pesticides (Davies *et al*., 1998).
Cyanide	Industrial processes involving bleaching and dyeing (Barrett *et al*., 1999).
Pathogenic organism	Sewage (Barrett *et al*., 1999).

Table 2.10: Urban pollutants and their sources.

In treating pollutants in vegetative SuDS, two major processes are carried out: quantity and quality control processes, and these processes help to mitigate the effect of urban pollutants on receiving water bodies (Woods-Ballard *et al.,* 2007). Quantity control processes involves the controlled conveyance of runoff, infiltration, detention or attenuation of runoff and water harvesting. *Controlled conveyance of runoff* from place to place through a network of pipes, open trenches and channels is considered an essential tool for managing flow rates and volumes, and also for linking SuDS components together (Tang, Yue and Ku, 2007). *Infiltration*, which is the soaking of water into the ground, reduces surface runoff and can be a means of recharging groundwater. Infiltration, is however, limited by soil type and condition and can pose significant risks to groundwater if not monitored and controlled. *Detention or attenuation of runoff* slows down runoff flow and is achieved by the temporary storage and

constrained release of runoff. This process reduces peak flow rate but does not reduce total flow volume (NCHRP, 2006; Woods-Ballard *et al.,* 2007). *Water harvesting* is the direct capture, storage and use of runoff for purposes such as toilet flushing and irrigation (Environment Agency, 2013). However, checks must be put in place to ensure that the system does not overflow when floods occur (Woods-Ballard *et al.,* 2007).

Quality control treatment processes carried out in vegetative SuDS involve sedimentation, filtration, adsorption, biodegradation, phytoremediation, precipitation and volatilisation (Woods-Ballard *et al.,* 2007). *Sedimentation* occurs when sediments in runoff fall out of suspension due to reduced flow velocities. However, there is a risk of re-suspension when extreme rainfall events occur. *Filtration* is a process whereby pollutants, sometimes in association with sediments, is removed from percolating waters and can occur either on vegetation, soil or aggregate matrix. *Adsorption* occurs when pollutants bind to surfaces of soil or aggregate particles by various chemical reactions. However, the effectiveness of this process depends on the acidity of the runoff as a change in acidity can either increase or decrease adsorption of pollutants. The limitation to this process is that adsorbing materials will eventually get saturated and the treatment becomes ineffective. *Biodegradation* involves the microbial degradation of organic pollutants such as oil and grease by microorganisms present in vegetative SuDS devices. Biodegradation is, however, limited by physical and environmental conditions present in the system such as temperature and oxygen levels, and availability of nutrients (Pitt *et al.,* 1995; Campbell *et al*., 2004). *Phytoremediation* involves the uptake of nutrients and heavy metals present in runoff by plants. However, appropriate maintenance of the plants is required to prevent the metals from returning into runoff or groundwater when the plants die. *Precipitation* is a process whereby chemical reactions between dissolved pollutants and soil/aggregate leads to the formation of insoluble particles (called precipitates) in suspension. Precipitation is highly influenced by the pH level of runoff. *Volatilisation* involves the conversion of volatile organic compounds (VOC) present in runoff to vapour or gas but the rate of volatilisation depends on environmental temperature and pressure. This process is applicable to runoff containing petroleum products and pesticides. Another process includes *nitrification* which is the oxidation of ammonia and ammonia

compounds by bacteria present in vegetative SuDS devices into nitrates, which are a source of nutrients for plants (Wilson *et al.,* 2004; Woods-Ballard *et al.,* 2007; Rivett, Sadler and Barnes, 2011). Table 2.11 summarises the removal mechanism suitable for each pollutant type present in surface runoff.

Pollutant	Removal mechanism in SuDS
Nutrients Phosphorus and nitrogen	Sedimentation, biodegradation, precipitation and de-nitrification
Sediments Total suspended solids	Sedimentation, filtration
Hydrocarbons TPH, PAH, VOCs, Methyl Tertiary-Butyl Ether (MTBE)	Biodegradation, filtration and adsorption
Metals Lead, Copper, Cadmium, Mercury, Zinc, Chromium, Aluminium	Sedimentation, adsorption, filtration, precipitation, plant uptake
Pesticides	Biodegradation, adsorption, volatilisation
Chlorides	Prevention
Cyanides	Volatilisation
Litter	Trapping, removal during routine maintenance
Organic matter, BOD	Filtration, sedimentation, biodegradation

Table 2.11: Removal mechanisms of pollutants in SuDS.

Source: Wilson *et al.,* (2004); Woods-Ballard *et al.,* (2007)

2.17 Role of microorganisms in pollutant removal in Vegetative SuDS

Management of stormwater pollutants is one of the priorities of SuDS and most SuDS are designed to trap and remove contaminants by processes described in section 2.16. However, in order to prevent the accumulation and over-saturation of these contaminants in topsoil thereby jeopardising the quality of groundwater, it is necessary to encourage the application of

vegetative SuDS that not only remove pollutants but also degrade them whenever and wherever possible (Horner *et al*., 1994; Napier *et al*., 2008). Organic pollutants such as oil, grease, organic matter and sediments are susceptible to biodegradation under favourable conditions (Singh and Ward, 2004).

Microorganisms have the tendency to create their own microenvironments known as biofilms. Biofilms are organised microbial systems comprising of microbial cell layers associated with surfaces. The growth and reproduction of these biofilms are dependent on factors such as readily available nutrients and light (Beyenal and Lewandowski, 2002; Prescott *et al.,* 2002). As biofilms grow and reproduce by metabolic processes, they are capable of transforming polluting materials into less harmful metabolites which are friendlier to the ecosystem, a process known as biodegradation (Wackett and Hershberger, 2001; Bradley and Chapelle, 2010). Biodegradation is usually carried out by a combination of organisms comprising of bacteria, fungi, protists and animals, and their different levels of feeding and metabolism are described in a 'microbial loop' (Coupe *et al.*, 2006[a]). For example, during oil biodegradation, hydrocarbons (which are a source of organic carbon) are metabolised by bacteria and fungi (decomposers) into less harmful metabolites such as glucose and carbon dioxide. The decomposers in turn are fed upon by protists and small animals, releasing metabolites such as vitamins and nitrogenous compounds. These nutrients are then utilised by the decomposers thereby encouraging their multiplication; hence the loop (see figure 2.19). This loop ensures that microbial biofilms are constantly renewed and dead materials are either consumed or degraded by other microbes and recycled (Coupe *et al*., 2006[a]).

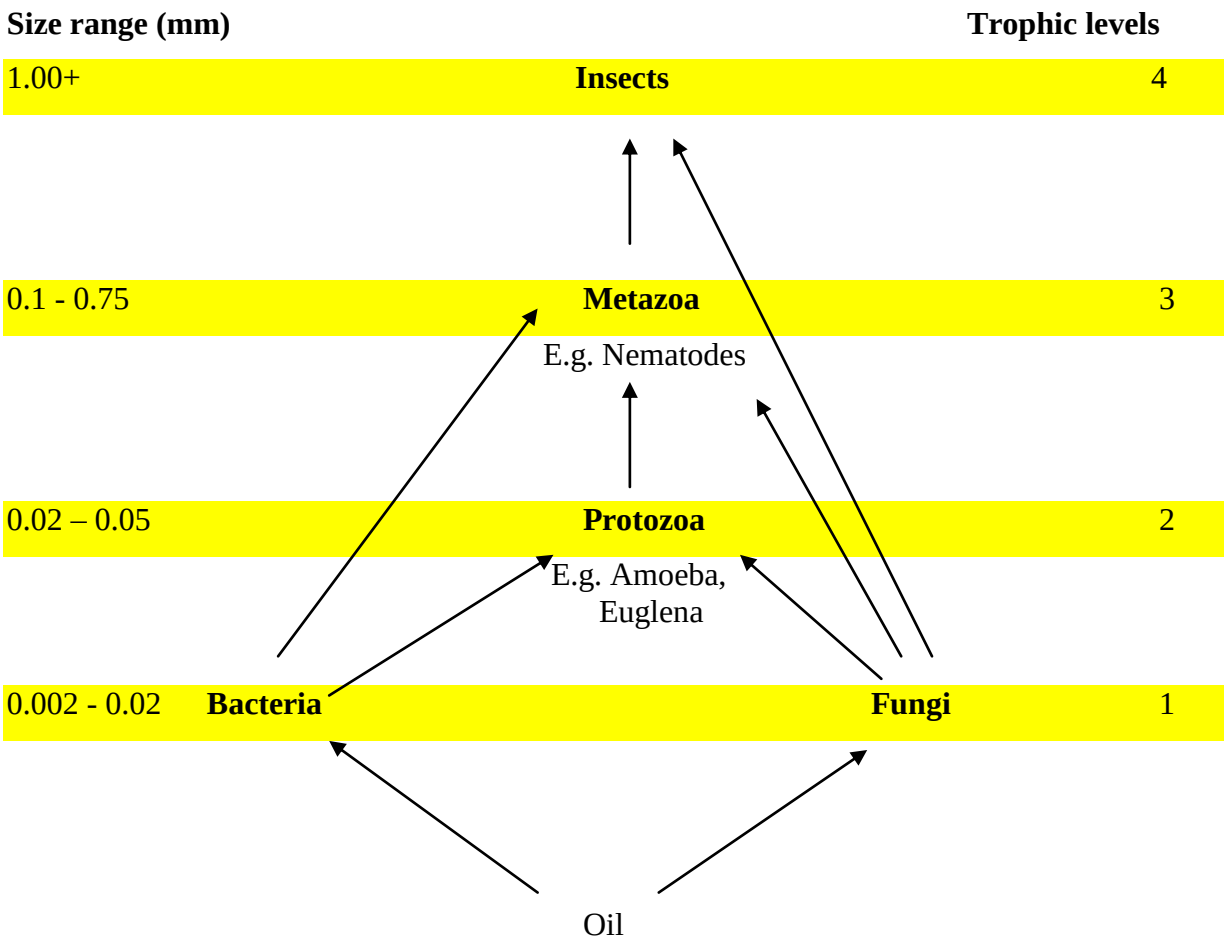


Figure 2.19: The structure and composition of a simplified microbial food web showing four levels of complexity.

(Modified from Coupe *et al*., 2006[a])

Biodegradation in SuDS has been extensively studied and it was reported by Brownstein (1998) that biofilms were found to develop on geotextiles, which were used to line permeable pavement systems (PPS), and on which motor oil pollutants were mostly immobilised. Further studies by Puehmeier *et al*. (2005) and Coupe *et al.* (2006[a]) showed that the presence of microbial biofilms on geotextiles indicated that motor oil present on the geotextile was undergoing biodegradation. Another study by Newman *et al.* (2002) showed that experimental PPS were able to retain 99.6% of the motor oil and grease applied to them, a slight improvement on Bond (1999) whose oil retention level was over 98%. They also discovered

that biodegradation of oil and grease trapped within the PPS was quite successful over a 1220 day period, after the application of a horticultural fertiliser (Osmocote Plus) and inoculation of a bacterial seed. It was estimated that microbial activity, and hence biodegradation, could be maintained at an acceptable level for at least a year, for a single fertiliser application, and that PPS would, at least in the short term, be able to retain motor oil and grease effectively without being oversaturated (Bond, 1999; Newman *et al.*, 2002).

As biodegradation is linked to microbial activity in SuDS, monitoring microbial activity can provide a means of assessing the rate of biodegradation, and this can be achieved by monitoring microbial respiration (also known as carbon dioxide monitoring) (Puehmeier *et al*., 2005). In applying this method of monitoring microbial activity, Pratt *et al.* (1999), Puehmeier *et al*. (2005) and Coupe *et al.*(2006[a]) proved that biodegradation occurred in PPS containing accumulated motor oil. Initial studies on microbial communities in SuDS by Newman *et al.* (2002) showed that there was an increase in microbial diversity and complexity in experimental PPS over time. This increase was partly responsible for the stability of microbes and biodegradation within the PPS. Studies by Coupe *et al.* (2006[a]) showed that the biofilms in PPS consisted of many morphological types of bacteria and fungi as viewed by scanning electron microscopy. Bacterial species believed to be oil degraders, which have been isolated from SuDS, include *Achromobacter putrefaciens, Acinetobacter iwoffi, Bacillus cereus, Pseudomonas putida, Pseudomonas vesicularis* and *Vibrio fluvialis* (Coupe, 2004). Other oil degrading bacteria genera in literature include *Rhodococcus, Arthrobacter* (Sorkhoh *et al*., 1985; Bouchez *et al.*, 1999; Ueno *et al*., 2007) and *Alcaligenes* (Andreoni *et al.*, 1998). Oil-degrading fungi include *Penicillium* and *Aspergillus* (Sorkhoh *et al*., 1985; Adenuga *et al.*, 1992). Some of these microorganisms are found in soil either in their vegetative forms or present as spores (Vecchioli *et al*., 1990; Adenuga *et al.*, 1992).

Now that pollutants treated by vegetative SuDS and the role of microorganism in biodegradation processes have been discussed, the next section discusses some applications of compost and recycled aggregates in erosion control and slope stabilisation as well as in vegetative SuDS devices.

2.18 Applications of Compost and Aggregates in vegetative SuDS

Edwards *et al*. (2010) reported the application of BSI PAS 100 certified compost as compost socks and blankets, and as substrates for green roof. Compost socks are biodegradable tubular geotextile mesh filled with compost and can either be vegetated or non-vegetated. They are used for slope stabilisation, erosion prevention and soil creation. These custom-made socks of varying length are placed on sloppy, erosion-prone areas or they can be cut open and the compost spread on the land to mitigate erosion. Compost socks can be filled on-site if the site is accessible or off-site if access to the site is limited. Figure 2.20 shows the application of compost socks and loose compost in Centenary Riverside Nature Reserve in Rotherham South Yorkshire. The compost socks were used to stabilise the river bank and protect it from erosion. The socks were sown with fast-growing seeds to provide vegetative cover so as to prevent compost erosion (Edwards *et al*., 2010).

Figure 2.20: Centenary Riverside compost socks.

Source: Edwards *et al*. (2010)

Apart from erosion prevention and soil creation, compost socks also improve runoff quality as seen in the Drylaw Hill Farm Recycling Facility in East Lothian, Scotland (Zero Waste Scotland, 2010). This facility manufactures BSI PAS 100 certified compost products including

soil conditioners and mulch. Composting processes on the site produces leachate which is nutrient-rich and potentially polluting, therefore treatment was required to mitigate the effect of this leachate on surface and ground water. Treatment involved filtering systems consisting of compost socks and reed beds. The system was installed in such a way that the leachate was first allowed to pass through the compost socks, filtering off silt and sediment, before being channelled through a drainage channel lined with compost socks (for stability) to the constructed reed beds for further treatment. The reed beds also consisted of compost socks sown with reeds to ensure the complete removal of silt, sediment or soil (see figure 2.21). Benefits derived from the application of compost socks to leachate from the composting site included improvement of run off quality, quick establishment of vegetation, erosion control and slope stability of channel (Zero Waste Scotland, 2010).

Figure 2.21: Compost socks and reed beds lined with compost socks at Drylaw Hill Farm Recycling Facility in East Lothian, Scotland.

Source: Zero Waste Scotland (2010)

Compost blankets consist of a loose layer of compost applied over existing soil such that it acts as a covering for soil. Compost blankets can be applied to both sloppy and flat land in order to absorb water and encourage vegetative growth (see figure 2.22 and 2.23).

Figure 2.22: An Ecoblanket: A type of compost blanket employed on slopes.

Source: Denbow (2011)

Figure 2.23: A stormwater blanket: A type of compost blanket employed on flat land.

Source: Denbow (2011)

Compost blankets can be used in conjunction with compost socks and can help to reduce runoff by up to 100% especially on slopes. When applied to severe slopes, a lock down netting can be applied over the compost which allows the compost particles to mat together thereby creating a blanket which holds without slipping. They are used for sheet-flow runoff management and the inclusion of compost socks provides protection against peak flow of runoff (Faucette, 2006; Denbow, 2011).

Compost and crushed bricks have been used as components of green roofs substrates (Oberndorfer *et al*., 2007; Townshend, 2007; Molineux, Fentiman and Gange, 2009; WRAP, 2012). These two components were applied in the installation of a green roof system installed in a mixed use apartment and business development in Sheffield city centre in 2007 (see figure 2.24).

Figure 2.24: Green roof installed on The Cube, a business and housing development in Sheffield.

Source: The Green Roof Centre (2011)

The green roof substrate consisted of 70% crushed bricks and 30% compost which was spread unevenly across the roof space. Vegetation comprised of grasses, perennials and alpine

species which were chosen to suit the Sheffield climate and enhance biodiversity. Within the first year of installation, 60% of vegetation cover was achieved with the establishment of various habitats (The Green Roof Centre (2011).

Sharrow School in Sheffield also employed the use of compost and RA in the installation of its green roof also in 2007 (see figure 2.25).

Figure 2.25: Green roof on Sharrow School, Sheffield.
Source: The Green Roof Centre (2010)

The school building was designed to integrate with the surrounding landscape and enhance sustainability. The substrate used in the green roof installation consisted of a mixture of crushed bricks, limestone and green compost. Green compost was probably applied because they possess high quality organic matter which aids nutrient and moisture retention as well as adsorption and degradation of pollutants (Alexander, 2004).

These case studies demonstrate the application of compost and recycled aggregates in maintaining water quantity, quality and amenity/biodiversity; as well as improving the sustainability of SuDS components. However, the successful application of these materials to vegetative SuDS has been limited to green roofs.

2.19 Conclusion: Research in context

Flooding has become a major challenge in society today due to urbanisation which has increased the number of impermeable surfaces particularly in urban areas, thereby leading to increased surface runoff (CIRIA C523, 2001). Also, increased rainfall and water bodies overflowing their banks, attributable to changes in climatic conditions due to anthropogenic activities, have also contributed to flooding (DEFRA, 2010[a]; Charlesworth, 2010). The quality of water bodies have been greatly compromised by the increase in contaminants being carried into them (Butler and Davies, 2004; Hoyer *et al*., 2011). Measures to mitigate flooding at local, national and global levels have necessitated taking an inward look at our drainage systems and discovering more sustainable means of controlling flooding. As sustainability involves the preservation and maintenance of natural resources available to society (OECD, 2002), and in accordance with the Flood and Water Management Act 2010 (which aims to reduce both the flood risks associated with extreme weather and water scarcity (DEFRA, 2012[b])), drainage systems have to be sustainably modified to take into account the quality and quantity of surface runoff, and the amenity value/biodiversity of surface water (Charlesworth, 2010). These systems, whether retrofitted or brand-new, are now known as Sustainable Drainage Systems (SuDS). SuDS mimic the natural drainage of a particular site or region thereby reducing surface runoff and flooding. SuDS are currently being implemented alongside upgrading conventional drainage systems, but implementation of some SuDS devices has been ineffective both in the short and long term due to cost implications, reluctance of practitioners and stakeholders to implement these systems and inadequate research (Woods-Ballard *et al*., 2007).

Vegetative SuDS are systems that attenuate the flow of surface runoff through vegetation, thereby encouraging infiltration and mimicking natural drainage patterns. However, some components of vegetative SuDS, such as topsoil and gravel, are unsustainable due to resource depletion and costs (both to the environment and to humans) of transporting these resources to where they are needed. The use of sustainable alternatives, such as compost and recycled aggregates, to topsoil and gravel should make vegetative SuDS more sustainable as topsoil and gravel are both natural resources that need to be conserved. Compost can be a suitable

available alternative to topsoil as biodegradable wastes, which usually end up in landfill, are currently being composted due to the implementation of the Landfill Directive (DEFRA (2011[a]). RA which is limited to concrete production (Dhir and Piaine, 2007) can also be used as a suitable available alternative to gravel in vegetative SuDS, thereby helping to save landfill space and conserve natural resources (Alexander, 2004; The Green Roof Centre, 2010). However, the efficacy of compost and RA in mitigating pollution and improving water quality in vegetative SuDS has not been extensively studied in vegetative SuDS such as swales. The following chapter details experiments carried out on these materials in order to determine their individual properties and how well they can improve water quality in vegetative SuDS.

For the purpose of this study, the swale structure will be simulated. This is because swales are effective at treating runoff pollutants by most of the processes described in section 2.16, thereby improving water quality, (Woods-Ballard *et al*., 2007). Also according to Jefferies and Napier (2008), swales are effective at treating runoff pollutants compared to other vegetative SuDS devices because they are periodically dry and wet and so are more effective at treating runoff pollutants such as oil, hydrocarbons and sediments compared to ponds and wetlands which are constantly wet. Jefferies *et al*. (2008) showed that soil and vegetation in swales retained heavy metals, oil and hydrocarbons, thereby reducing the risk of groundwater contamination. Swales are also cheaper to construct and take up less space (van Borcke, 2003).

Knowledge gained from the extensive literature reviewed in this chapter has facilitated the development of various experimental designs aimed at maximising the use of compost and recycled aggregates in vegetative SuDS, in comparison to the original materials (i.e. topsoil and gravel), and these designs will be discussed in the next chapter.

Chapter Three: Methodology

3.1 Introduction

In the construction of vegetative SuDS, certain properties are considered when determining which materials would be most suitable and sustainable for use. However, as discussed in chapter two, the application of topsoil and gravel in vegetative SuDS raised the question of sustainability and it was suggested that compost can either replace or complement topsoil as a growth medium, and recycled aggregates (RA) can replace gravel as sub-base aggregates in vegetative SuDS.

In assessing the practicality of applying compost in vegetative SuDS, compost must possess the following qualities:

a) suitable bulk density i.e. it is porous enough to encourage infiltration and plant uptake of soil water/moisture (Celik *et al*., 2004; VanDerZanden and Cook, 2011), but not too porous so as to avoid subsidence, which is the sinking of land due to low bulk densities, which could affect the overall structural stability of vegetative SuDS devices (Arthur *et al*., 2011),
b) retain water for attenuation of surface runoff and sedimentation of particles (Markham, 2006; Thakur, 2006),
c) contain acceptable levels of organic matter as a source of carbon for microbial nutrition (Marinari *et al*., 2000; Vidal-Beaudet and Charpentier, 2000) and for the removal of pollutants such as heavy metals and motor oils from runoff (Huang *et al*., 2005; Seelsaen *et al*., 2007),
d) have the capability to sustain microbial activity by containing adequate nutrients (such as carbon, nitrogen, phosphorus and potassium) and moisture, in order to carry out biodegradation processes (Marinari *et al*., 2000),
e) be free from contamination which would otherwise have an adverse effect on water quality (preferably PAS 100 compliant) (Amir, 2005) and
f) be relatively cheap and easy to obtain.

Recycled aggregates must be inert such that they do not introduce pollutants such as heavy metals into groundwater and they must be structurally stable (Poon *et al*., 2002; Dhir and Paine, 2007).

In assessing the qualities of compost mentioned above, physical, geochemical and microbiological tests were carried out to ensure compliance with the PAS 100 specification for composted materials (BSI, 2011) and the Compost Specification for Use in Grass Establishment (CSUGE) (WRAP, 2003), which has been discussed in detail in section 2.14. Physical analysis of compost included the determination of moisture content, water holding capacity, bulk density, organic matter content, carbonate content and pH. Geochemical tests on compost included heavy metal analysis, and microbiological tests included microbial enumeration, monitoring of microbial activity and microbial water quality assessment of compost for microorganisms indicative of faecal contamination. Other analyses carried out on compost were leaching experiments with heavy metals and motor oil, and plant trials to assess the ability of compost to sustain the growth of vegetation. Analyses carried out on RA were heavy metal analysis and leaching tests and values obtained were compared to ICRCL 59/83 values and Kelly indices guideline values (ContaminatedLand, 2000), as mentioned in section 2.15.

The methods were divided into two segments: the first segment included baseline characterisation of the test samples (i.e. compost and RA) comprising of determination of moisture content (for the purpose of this study, water and moisture were used interchangeably), organic matter and organic carbon content, water holding capacity, bulk density, pH, heavy metal content, bacterial and fungal enumeration and water quality assessments. The second segment comprised of experiments carried out on the test samples to determine their ability to mitigate pollution and analyses carried out included leaching experiments with pollutants such as heavy metals and motor oil, biomass determination and microbiological assessments. This chapter describes and justifies the experimental designs used to analyse the test materials, thereby providing information on their constituents and potentials, which will be used to determine their practical use in vegetative SuDS.

3.2 Description of test materials

The recycling of garden waste comprised of grass hedge and tree clippings and weeds produces green compost, which until now has been mostly applied in agriculture either as a soil amendment, mulch or growth media depending on their grade i.e. particle size (Keeling *et al*., 1995; Peatering Out Ltd., 2005; Lamb, 2006; BSI, 2011). Green compost of grade size <10mm are used as garden compost, grade sizes between 10mm and 25mm is used as mulch on farmlands to prevent erosion and evaporation of soil moisture, and grade sizes >25mm are limited in use as it is mostly used in grow bags for growing vegetables (BBC, 2011; Vital Earth, 2011) or otherwise ends up in landfill (Defra, 2011[a]). The recycling of a mixture of kitchen and garden waste produces mixed compost, and grade sizes of between 10mm to 25mm are used mainly as plant grow bags or as mulch on farmland, and the rest ends up in landfill (BBC, 2011 and Vital Earth, 2011). There is therefore a greater need for the use of coarser grades of compost of grade sizes >10mm. Also for recycled aggregates, the increasing distance between demolition sites and disposal areas due to urbanisation has led to increasing transportation and energy costs (Gupta, 2009), depletion of landfill spaces, contamination of soil and groundwater by landfill leachate and current legislation on landfill (discussed in section 2.11). Therefore, it has become important to find viable sustainable ways of re-using aggregates derived from construction demolition and excavation wastes (CDEW) (Poon and Chan, 2007). One way of recycling and re-using compost and CDEW is in the incorporation of these materials into vegetative SuDS, subject to the various tests described in this chapter.

The growth media test samples comprised of compost and topsoil while the sub-base aggregates test samples were RA and gravel. The compost samples analysed were of two types: mixed compost (MC) and green compost (GC), and topsoil (T) was the control. Compost samples of particle size >25mm were used because these sizes are limited in use (as discussed in section 2.12) and mostly ends up in landfill (DEFRA 2011[a]).

GC, also known as garden waste, consisted of tree clippings, leaves, grass cuttings and weeds, and was obtained from a farm in Coventry that uses GC as mulch. They were moist and dark brown in colour with particle sizes >25mm. Compost of this particle size are predominantly

used in agriculture as surface mulch to suppress weeds and prevent erosion (Epstein, 2011). GC also enhances the biodegradation of solid wastes such as biodegradable plastic carrier bags which would ordinarily have ended up in landfill (Unmar and Mohee, 2008).

MC consisted of a mixture of kitchen and garden waste, and a moist, coarse grade (>25mm) of this compost was used. MC of this grade is used mainly in plant grow bags or as mulch on farmlands. The MC samples were supplied by Vital Earth Ltd. which is a composting company that processes kitchen and garden waste into peat-free compost (BBC, 2011; Vital Earth, 2011).

T, which is a primary component of vegetative SuDS such as swales (Highways Agency, 2006; Woods-Ballard *et al*., 2007), was used as the control, to which performances by the compost samples were compared. T, obtained from a garden in Warwick, was a moist clayey loam soil (Plaster, 2013: 74) and was passed through a 2mm sieve to homogenise it. This soil texture is recommended for use in swales because it holds water long enough for infiltration and other treatment processes, described in section 2.16, to occur without being waterlogged (Highways Agency, 2006; Pittner and Allerton, 2009; Plaster, 2013).

RA comprised of crushed old bricks (OB), crushed new bricks (NB) and recycled limestone aggregates (L) with gravel (G) as control. OB was obtained from the rubbles of a demolished building in Coventry, NB was obtained from a construction site in Coventry, L was obtained from a railway yard at Coventry train station, and G was obtained from the Engineering and Computing department at Coventry University. G and L were approximately 10mm in size, and OB and NB were crushed to approximately the same size so as to provide uniformity with the aggregates before being analysed.

3.3 Background analysis of growth media test samples

3.3.1 Moisture content determination

Water is vital for the effective functioning of compost in vegetative SuDS both as bio-treatment systems and as media for plant growth, as it is necessary for the existence and activity of microorganisms (i.e. bacteria, fungi, protozoa and nematodes) living within compost (Schnürer, 1986). Water is required for microbial metabolism, mineralisation (decomposition of organic matter), bio-fertilisation (increased supply of mineral nutrients to plants) (Ingham, 1985; Lugtenberg *et al*., 1991), and biodegradation (breakdown of complex materials into simpler ones). The products and by-products of these processes provide the macro-nutrients and micro-nutrients required for plant growth. Hence, moisture levels have a direct impact on the types, number and activity of organisms in compost, as well as the nutrients available for plant growth (Wiant, 1967; Davidson *et al*., 2000).

Liang *et al*. (2003) reported that a minimum moisture content of 50% was adequate for sustaining microbial activities in compost because Vallini *et al.* (2002) and Liang *et al.* (2003) showed that biodegradation of organic matter slowed down dramatically at vaues below 40%. Lin (2008) and Mohee *et al*. (2008) also supported this value by reporting that 50% - 60% moisture content was efficient for microbial activities. The CSUGE specification of compost moisture content is between 35-55% (WRAP, 2003).

As a result of the factors discussed above, it was therefore necessary to determine the initial moisture present in compost in order to establish their behaviour and reactions to different treatments.

Moisture content is defined as the ratio of the mass of water removed from a wet material after drying at a particular temperature to the mass of the wet material (Woodcock and Mason 1995; Maail *et al*., 2004). The direct method of oven-drying is a standard procedure for determining soil moisture in the laboratory (Head, 1992). The standard drying temperature is 105 - 110^{o}C and this method measures the total moisture present in the samples including, bound water which is unavailable to plants (Razumova and Verigo, 1966). However, Ackroyd

(1957), Jarrett (1983) and ASTM D2216 (1998) recommended that soils containing organic matter (such as compost) be dried at 60^{o}C to prevent the oxidation and decomposition of organic matter leading to charring, which could affect the value of the derived moisture content. Charring would further decrease the weight of the drying samples thereby overestimating the moisture content value which leads to inaccuracy (O'Kelly, 2004).

However, MacFarlane and Allen (1965) observed that there was no evidence of charring of samples containing organic matter at temperatures less than 85^{o}C, and therefore it can be deduced that temperatures between 60^{o}C and 80^{o}C will give the most accurate result for determining plant available moisture content. Therefore the temperature employed for the moisture content determination of the test samples in this study was 80^{o}C. The procedure was carried out as stated by Head (1992).

Apparatus used included:

- A fan assisted oven
- Desiccator containing anhydrous silica gel
- Foil trays for drying samples
- Weighing balance with accuracy to 0.01g

Procedure:

a) 200cm^{3} each of green compost (GC), mixed compost (MC) and topsoil (T) was weighed in its moist condition to 0.01g (WWK), in foil trays of known weight (K).
b) The trays were placed in the oven at 80^{o}C and dried for three days.
c) The trays were then transferred into a desiccator to cool to room temperature (for about 30 minutes).
d) The tray and their contents were then weighed and recorded (DWK).
e) This process was carried out in five replicates for each sample.

The percentage moisture content of the samples was calculated thus:

$$M = \frac{(WWK-K) - (DWK-K)}{(WWK-K)} * 100$$

Where: M moisture content

WWK = weight of wet samples + tray in grams

DWK = weight of oven-dried samples at 80oC + tray in grams

K = weight of tray in grams

3.3.2 Water Holding Capacity

Water is required for the many processes that are carried out in vegetative SuDS as discussed in section 2.16 and therefore, knowledge of the ability of the vegetative growth media to retain moisture is necessary. The media should be able to hold water without being water-logged (as this could lead to anaerobic conditions which could slow down microbial activity and biodegradation), and should not infiltrate quickly so as to encourage vegetative growth, runoff attenuation and treatment processes (Woods-Ballard *et al.*, 2007).

Gershuny (2011) reported that WHC of compost should be at least 100%. The method used in this study was a modification of the WHC procedure carried out by Sloan *et al*. (2008). In Sloan *et al*. (2008), wet weight measurements of samples were carried out to determine the water retained by the samples. Nevertheless for this study, leachate volumes relative to initial water added to the test samples were measured because continuous measurements of WHC was required to see how well each test sample would retain water over time, and displacing samples to measure their wet weight might affect water retention capabilities thereby leading to inaccuracy.

Apparatus used included:

- Polyvinyl chloride rigs
- De-ionised water
- Plastic bottles

- Cling film

Procedure:

a) Each test sample, i.e. GC, MC and T, were placed in polyvinyl chloride rigs in triplicate (see figure 3.1), to fill about two-thirds of the container (about 3 litres of each sample).
b) The samples were saturated with de-ionised water (approximately 100ml) for a period 24 hours.
c) Leachates were collected in bottles and their volume measured after 24 hours.
d) The rigs were sealed with cling film to prevent evaporation of water.
e) The experiment was repeated until water holding capacity was almost zero in all the samples which was after about 20 days.

Water holding capacity was calculated as:

$$\frac{\text{Initial volume of added water (100ml)} - \text{volume of leachate}}{\text{Initial volume of added water (100ml)}} * 100$$

Figure 3.1: The experimental set up for determining water holding capacity

Source: Author's own

3.3.3 Organic Matter Content

Organic matter, which is abundant in compost, is a complex mixture of plant and animal residue at varying stages of decomposition and its presence in soil affects soil fertility and behaviour (Rosell *et al*., 2001). The application of compost to soil increases its organic matter content which stimulates soil microbial activities (Marinari *et al*., 2000; FAO, 2005) and affects its bulk density as organic matter increases the porosity of soils (Zeytin and Baran, 2003; Celik *et al*., 2004), lowers its bulk density and decreases soil compaction (Zhang *et al*., 1997), thereby increasing infiltration rates of runoff (Froehlich *et al*., 1985; Rawls and Brakensiek, 1989; Arvidsson, 1998). Furthermore, organic matter provides sorption sites for certain compounds and therefore the higher the organic matter content of compost, the more the sorption sites available for adsorbing and absorbing runoff pollutants such as heavy metals, thereby immobilising them (Kaschl *et al*., 2002).

All the beneficial properties of compost in compost-amended soil discussed above are required in the effective functioning of vegetative SuDS and hence determination of organic

matter content is vital. Gershuny, (2011) reported that mature compost should have an organic matter content ranging from 45% - 65%, while the CSUGE specification for compost organic matter content is >25% (WRAP, 2003).

One method for estimating organic matter content is the technique of loss on ignition (LOI). This involves the dry combustion of test samples and measuring the weight lost, which is synonymous with the amount of degraded organic matter. The LOI method was based on the theory that organic matter was decomposed at a temperature range in which mineral decomposition (e.g. carbonates) is negligible and errors are minimised, usually between 450^{o}C and 550^{o}C (Galle and Runnels, 1960; Ball, 1964; Dean, 1974; Rosell *et al*., 2001). However, Schulte and Hopkins (1996) discovered that temperatures above 500^{o}C led to errors and inaccuracies in determining organic matter content due to decomposition of hydrated salts, loss of CO_2 from carbonates, loss of structural water from clay minerals and oxidation of Fe^{2+}. Gallardo *et al.* (1987) suggested that temperatures below 500^{o}C should eliminate these errors though incomplete decomposition of organic matter may occur. However, Davies (1974) and Giovanni *et al*. (1975) have recorded complete organic matter decomposition without any loss of carbonate at temperatures between 430^{o}C and 500^{o}C. Therefore the temperature used for organic matter determination in this study was 500^{o}C and methods applied by Davies (1974) and Giovanni *et al.* (1975) were adopted.

Apparatus used included:

- A fan assisted oven
- Tongs with a pair of asbestos gloves
- Porcelain crucibles
- 2mm sieve
- A muffle furnace
- A desiccator

Procedure:

a) Test samples were oven-dried at 50^{o}C overnight (to avoid degradation of organic fractions), cooled, sieved and homogenised.
b) Crucibles (K) for each of the samples were placed in an ignited muffle furnace using a pair of asbestos gloves and tongs, and heated for 1hr at 500^{o}C. This was allowed to cool in a desiccator for about 10mins and weighed.
c) 2g of each of the oven-dried samples were added to the cooled crucibles (AK) and re-weighed.
d) The crucibles with the samples were placed in the ignited muffle furnace at 500^{o}C for 2hrs after which the crucibles were removed and allowed to cool in a desiccator. The crucibles with their contents (BK) were then re-weighed.
e) The experiment was carried out in five replicates.

% organic matter content was then calculated thus:

$$LOI_{(500)} = \frac{(AK-K) - (BK-K)}{(AK-K)} * 100$$

Where AK = weight of oven dried samples at 50^{o}C + crucible

BK = weight of dried samples at 500^{o}C + crucible

K = weight of crucible

3.3.4 Bulk Density

The bulk density of compost has a significant effect on the availability of water to SuDS vegetation, as higher bulk densities, which are indicative of more compact soil structures, retard plant growth and root elongation rates compared to less dense soils (Hanks and Thorp 1956; Grable and Siemer, 1968; Rawls and Brakensiek, 1989). Amendment of soil by compost reduces the bulk density of soil (especially compacted soils) thereby decreasing root restriction which encourages the establishment of vegetation (Tester, 1990; Celik *et al*., 2004; VanDerZanden and Cook, 2011). Similarly, bulk density affects water movement and retention (Rawls, 1983) as less dense soils have a higher porosity, thereby aiding infiltration of surface runoff (Froehlich *et al*., 1985). Soils that are high in organic matter (loamy and

compost-amended soils) usually have bulk densities of <1.0gcm^{-3}, clayey soils have bulk densities of between 0.9 - 1.4gcm^{-3} and sandy soils have the highest bulk densities ranging from 1.4 - 1.9gcm^{-3} (Rawls, 1983; Froehlich *et al*., 1985; Poon and Chan, 2006). The CSUGE specification for compost bulk density is 1.0gcm^{-3} (WRAP, 2003).

Bulk density is the mass of dry soil contained in a unit volume. It is basically a measure of soil porosity (pore spaces) and soils with higher soil-pore spaces have lower bulk densities and vice versa (Head, 1992). The method applied in determining bulk density was as described by Diaz *et al*. (2005).

Apparatus used included:

- Beakers
- Weighing scale
- Fan assisted oven

Procedure:

a) The oven dried test samples at 80^{o}C were tightly packed into beakers of known volume (200cm^{3}).
b) The test samples were then weighed and the formula below was applied:

$$\text{Dry bulk density, } \rho \text{ (gcm}^{-3}\text{)} = \frac{M}{V}$$

Where M = weight of oven dried samples in grams

V = volume of samples in cm^{3}

c) The experiment was carried out in five replicates.

3.3.5 Carbonate Content

Studies by de Matos *et al*., (2001) and Lafuente *et al*., (2008) showed that carbonate content was one of the principal soil characteristics that determined the retention and mobility of pollutants such as heavy metals. They reported that carbonates provided binding sites for heavy metals in solution making them unavailable to plants or groundwater thereby improving stormwater quality. According to Bengtsson and Enell (1986), carbonate content can be determined by measuring the amount of inorganic carbon oxidised due to the degradation of carbonates at high temperatures multiplied by a factor of 1.36 (this is the CO_3:CO_2 ratio, where the molecular weight of carbonate (CO_3) (60) is divided by the molecular weight of CO_2 (44)). When the carbonate compounds are degraded, they give rise to carbonate ions which act as binding sites for heavy metals (Huang *et al*., 2005). Galle and Runnels (1960) and Dean (1974) discovered that the decomposition of carbonates begins at a temperature of 800^{o}C to about 1000^{o}C with an optimum temperature of 950^{o}C. Carbonate content was therefore measured at 950^{o}C in order determine how well the test samples would retain pollutants such as heavy metals according to methods by Heiri *et al.* (2001) and Santisteban *et al*. (2004).

Apparatus used included:

- Tongs with a pair of asbestos gloves
- Porcelain crucibles
- A muffle furnace
- A desiccator

Procedure:

a) Crucibles containing ignited samples at 500^{o}C (BK) (see organic matter content determination in section 3.3.3) were placed in a muffle furnace and were heated at 950^{o}C for 4hrs.
b) These were allowed to cool in a desiccator and the ash weight plus the crucibles (CK) were weighed.

c) The experiment was carried out in five replicates.

Carbonate content of the samples was calculated thus:

$$LOI_{(950)} = \frac{(BK-K) - (CK-K)}{(AK-K)} * 100 * 1.36$$

Where AK = weight of oven dried samples at 50^{o}C + crucible

BK = weight of ignited samples at 500^{o}C + crucible

CK = weight of ignited samples at 950^{o}C + crucible

K = weight of crucible

3.3.6 Hydrogen ion level (pH)

The pH of soil refers to how acidic or alkaline soil is as referenced on the scale of 0 – 14 with 0 being the most acidic, 14 being the most alkaline and 7 being neutral (Harpstead, Sauer and Bennett, 2001; Slattery, Conyers and Aitken, 2001; Patiram *et al.*, 2007). The addition of compost to soils can modify the ambient pH of soil by acting as a buffer thereby stabilising it (Tester, 1990). Soil pH measures the activity and concentration of hydrogen ions in soil (Harpstead, Sauer and Bennett, 2001) and as most plants and microorganisms thrive well at a pH between 6 and 7, soils and/or compost within this pH range should be utilised in vegetative SuDS. The CSUGE specification recommended for compost pH is in the range of 7.0 - 8.7 (WRAP, 2003).

Hydrogen ion level can be measured in an aqueous matrix in two ways: either in water or in dilute salt solution (0.01M $CaCl_2$). Measurement in water is closest to the value obtainable in field conditions but this method is mostly suitable for soils that are not fertilised. The advantage of measuring pH in salt solutions is that the measurement is less dependent on fertiliser history, although Schofield and Taylor (1955) found that the addition of salt lowers pH by about 0.5pH units compared to soil pH in water (Hendershot *et al.*, 2008). Therefore, method applied in this study was the measurement of pH in water as described by Amir *et al*. (2005), due to its accuracy.

Apparatus used included:

- pH meter with electrodes
- glass rod for stirring
- buffer solutions of pH 4 and pH 7
- beakers for electrode immersion

Procedure:

- Air-dried compost and topsoil samples were crushed and sieved with a 2mm sieve.
- 10grams of each sample were placed in beakers to which 20ml of de-ionised water was added and this was done in five replicates.
- The suspensions were stirred intermittently for 30mins and allowed to stand for an hour.
- The pH meter was calibrated with two standard buffer solutions pH 4 and pH 7.
- The suspensions were re-stirred and the electrodes of the pH meter were inserted into individual samples paying attention not to touch the sides or bottom of the beaker.
- The meter readings were then recorded when stable.

3.3.7 Heavy metal analysis

In environmental studies, heavy metal analysis helps to measure total heavy metal concentration and bioavailable metals; relating to mobility of metals within soil and compost matrices, phytotoxicity in plants and potential groundwater contamination (Quevauviller, 1998). According to Quevauviller (1998), analytical tests employed in analysing for heavy metals in soil and other such matrices such as sludge and compost include:

1. Extraction of immobile fractions with aqua regia by acid digestion e.g. hydrogen fluoride or hydrogen chloride (HF or HCL). This method is usually employed in environmental risk assessments and gives the total concentration of heavy metals present in the soil.
2. Extraction of easily mobilisable fractions with water in leaching column tests. This method is also employed in environmental risk assessments and gives the concentration of available heavy metals present in soil.

3. Extraction of slowly mobilisable fractions with ethylenediaminetetraacetic acid (EDTA) for studies of trace metal mobility such as in soil-plant transmissions.

Methods 1 and 2 were employed in this study to measure the concentration of immobile and mobile heavy metal fractions present in the test samples, because this study seeks to ascertain the presence and availability of heavy metals in the test samples to plants, microorganisms and groundwater in relation to the specified standards; and not necessarily mobility of trace metals between these living components. Data obtained from method 1 were compared to the PAS 100 specification for heavy metal concentrations in compost (BSI, 2011) described in table 2.8, and ICRCL 59/83 and Kelly indices specifications (ContaminatedLand, 2000) described in table 3.6;while data obtained from method 2 were compared to WHO standards (WHO, 2011). For the purpose of this study, the heavy metals analysed included a range of metals commonly found in surface runoff and their sources are described in table 3.1:

Heavy metals	Sources
Al	Mining, incinerator ash and emissions, corrosion of vehicle body panels, engine and aircraft components (Health Protection Agency, 2010[a]), air emissions and waste effluents from aluminium ore processing (ATSDR, 2008).
Cd	Street dust (Nazzal, Rosen and Al-Rawabdeh, 2012), fertilisers (Romero-Puertas *et al*., 2012).
Cr	Brake lining emissions, air conditioning coolants, emissions from cement-producing plants, leather tanneries (USEPA, 2007[a]).
Cu	Combustion of oils and fuel, emissions from waste incineration, coal combustion, metal production (ATSDR, 2004; Health Protection Agency, 2010[b]), street dust (Charlesworth *et al*., 2003; Nazzal, Rosen and Al-Rawabdeh, 2012).
Fe	Street dust, corrosion of automobile body parts (Nazzal, Rosen and Al-Rawabdeh, 2012), fly ash from coal combustion (Chen *et al*., 2012).
Mn	Combustion of fossil fuels and fuel additives, mining processes, fertilisers (WHO, 2004).

Heavy metals	Sources
Ni	Street dust (Nazzal, Rosen and Al-Rawabdeh, 2012), waste incineration, emissions from nickel mining (ATSDR, 2005).
Pb	Emission from mining, metal smelting, waste incineration, manufacturing of batteries and electronic equipment, pesticides, leaded paints, pipes and fuel, vehicle radiators (Health Protection Agency, 2007; Department of Health, New York, 2010).
Zn	Tire-wear (Councell *et al*., 2004), street dust (Charlesworth *et al*., 2003; Nazzal, Rosen and Al-Rawabdeh, 2012), runoff from zinc roofs (Shirley *et al*., 2008).

Table 3.1: Common heavy metals in runoff and their sources.

In preparing the test samples for baseline immobile heavy metal analysis, the samples were first pulverised using a hammer and mill before acid digestion was carried out. The samples were then subjected to high pressure microwave acid digestion in order to destroy organic matter and other compounds thereby releasing the heavy metals they contained, making them available for analysis (Guven and Akinci, 2011). Organic matter needs to be destroyed before heavy metal analysis as its presence may cause interference leading to false results (Falcina *et al*., 2000). During digestion, the organo-metallic compounds are converted to their inorganic forms (Abu-Samra *et al*., 1975; Hwang and Wang, 1995), thereby eradicating any organic matter interference. This procedure was followed by analysis on the Inductively Coupled Plasma – Atomic Emission Spectrometer (ICP - AES). The procedure used was as described by Page *et al*. (1982), Nadkani (2005) and Lomonte *et al*., (2008).

Total heavy metal analysis

a) 0.5g of each of the samples was weighed out and put into perfluoroalkoxy (PFA) vessels, after which a reverse aqua regia mixture of 3ml nitric acid (HNO_3) and 1ml hydrochloric acid (HCl) was added to each vessel.

b) Spikes of known heavy metal concentrations to be analysed were made by adding 5ppm of the known elements and reverse aqua regia mixture into vessels. This process was carried out to determine how accurate the experimental procedure was.
c) Acid blanks were also prepared with the aqua regia mixture only and placed in vessels. This was carried to determine the concentration of impurities present in the acid.
d) The vessels were sealed, placed in a carousel and subjected to microwave heating for about 20minutes. The microwave provides the required heat and agitation whilst the sealed vessels elevate pressure thereby enabling the acid mixture reach its boiling point, which destroyed the soil and compost matrices thereby releasing heavy metals into solution.
e) After the microwave treatment, the vessels were cooled and their contents were filtered into 25ml volumetric flasks, rinsing out the vessels severally. The flasks were then made up to volume with distilled water.
f) Standard solutions containing known concentrations of the heavy metals to be analysed were prepared for calibration purposes.
g) The treated samples were then aspirated in the ICP-AES in order to determine the heavy metal content. The samples analysed for were aluminium (Al), cadmium (Cd), chromium (Cr), copper (Cu), iron (Fe), manganese (Mn), nickel (Ni), lead (Pb) and zinc (Zn).
h) Each analysis was carried out in triplicate.
i) The values derived were compared with the PAS 100 (BSI, 2011) standards described in table 2.8.

In analysing for baseline mobile (bioavailable) heavy metal fractions, leaching experiments were carried out on the test samples to determine the background concentrations and trends of leaching of available heavy metals present in the samples, as described by Quevauviller (1998). The procedure was however modified for this study as described below:

Available heavy metal analysis

a) Approximately 3 litres of each sample was saturated with 100ml of de-ionised water in duplicate and the leachates were collected after 24hours with a similar experimental setup to the one shown in figure 3.1 but without the cling film.
b) The effluents were then analysed using the ICP – AES as described above.
c) This process was repeated once a week until the concentration of heavy metals in leachate were below limits of detection which was between one to five weeks
d) The results were compared with WHO drinking water guidelines for heavy metals (WHO, 2011)

3.3.8 Microbial enumeration and identification

Effective biodegradation of pollutants in vegetative SuDS have been attributed to the presence of microbial biofilms as discussed in section 2.17. As a result, microbial enumeration of test samples will give an indication of microbial population, activity, microbial response to the vegetative SuDS environment and biodegradation rates (Sessitsch *et al*., 2001); and microbial identification will identify pollutant-degrading species essential for the degradation of pollutants in vegetative SuDS (Coupe *et al*., 2006). Methods applied included viable cell count for bacterial and fungal enumeration, identification of oil-degrading bacteria and fungi, and water quality assessments by analysing for faecal indicator organisms.

In order to ensure that test samples were not contaminated or compromised by other microorganisms present in the laboratory atmosphere, materials and in media, aseptic techniques and sterilisation methods were employed. Aseptic technique involves the sterilisation of all instruments and media before use and the subsequent avoidance of their re-contamination by non-sterile objects and/or environment. Before opening any vessel such as sterile media, the rim of the vessel was passed over a Bunsen flame to kill off any microbial contaminants, thereby preventing them from falling into the sterile media when the cap was removed. The rim of the vessel was also flamed before replacing the cap. Aseptic techniques also involve disinfecting bench tops and carrying out microbiological work in a safety cabinet which filters air to remove cells and spores of micro-organisms. Sterilisation involves the

complete destruction of all microorganisms including endospores and this technique is usually quick, efficient, cheap and applicable to a wide range of materials (Singleton, 1999; Prescott *et al*., 2002). There are several sterilisation techniques but the ones applied in this study were steaming under pressure, direct heat and disinfection.

Steaming under pressure (also known as autoclaving) involves the application of steam under pressure to items needing sterilisation. It applies the principle that almost all microorganisms are destroyed by moist heat, even heat-resistant forms such as endospores. Under normal atmospheric pressure, steam has a temperature of 100^{o}C but when under pressure, its temperature increases to levels acceptable for sterilisation. The procedure was carried out in a strong metal gas-tight chamber known as an *autoclave* and all culture media and aqueous solutions used in this experiment were 'autoclaved' at a typical sterilising temperature of 121^{o}C, at a pressure of 1.02 bar for 15-20 minutes (this timing started when the sterilising temperature was reached) (Prescott *et al*., 2002 and Singleton, 2004).

Direct heat was used for the rapid sterilisation of instruments such as inoculating loops and spreaders, which were passed over a flame from a Bunsen burner (a process known as 'flaming') and cooled before use. This process was carried out in-between isolation and inoculation of microbial colonies. Disinfection with alcohol (70% or 95% ethanol in water) inactivates or destroys microorganisms and was used in disinfecting work surfaces. They are effective on susceptible microorganisms such as vegetative bacteria and fungi but not on endospores (Collins and Lyne, 1984; Prescott *et al*., 2002 and Singleton, 2004).

Viable cell count for bacterial enumeration

This method was carried out to enumerate viable bacterial cells present in the test samples and the procedure applied was as described by Singleton (2004).

Apparatus used included:

- Media – saline and nutrient agar (NA)
- Weighing scales and boats, spatula, universal bottles, pipettes,
- Sterile Petri dishes, glass spreaders

- 70% ethanol
- Bunsen burner
- Water bath
- Incubator set to 25^{o}C (This temperature was used because the microorganisms to be enumerated were environmental strains)

Procedure:

a) Saline was prepared by adding 8.5g of sodium chloride to a litre of deionised (DI) water and 28g of powdered NA was added to a litre of DI water.

b) The media were autoclaved at 121^{o}C for 15 minutes, cooled to about 50^{o}C in a water bath and poured aseptically into Petri dishes.

c) 1g of each original sample was weighed out and introduced into universal bottles containing 9ml of sterile saline and shaken thoroughly.

d) From this mixture, 1ml was extracted and introduced into another universal bottle containing 9 ml of sterile saline. This process was repeated five more times to produce a set of serially diluted samples at concentrations of 10^{-1}, 10^{-2}, 10^{-3}, 10^{-4}, 10^{-5} and 10^{-6}.

e) From each dilution, 100µl (0.1ml) was extracted using a pipette and inoculated onto NA plates and spread evenly using a sterile glass spreader (spread plate method). Each inoculation was carried out in duplicates.

f) In-between inoculation of plates, the spreader was rinsed in 70% ethanol and flamed on a Bunsen flame to burn off the alcohol thereby killing off any cells and preventing cross-contamination. The plates were then incubated at 25^{o}C for 48 hours. The whole process was carried out aseptically in a safety cabinet.

g) After incubation, individual colonies on each plate was counted and recorded. Colonies exceeding 300 were discarded. The number of colony forming unit per gram ($CFUg^{-1}$) was determined by dividing the colony count by the original mass of the sample, multiplied by the dilution factor (which is the inverse of the dilution).

Viable cell count for fungal enumeration

This method was carried out to enumerate viable fungal cells present in the test samples and the procedure applied was as described by Singleton (2004).

Apparatus used was the same as that of bacterial enumeration listed above.

Procedure:

a) Rose-Bengal Chloramphenicol agar (RBC) was prepared by adding 32g of Rose-Bengal agar to a litre of DI water.
b) The media were autoclaved at 121^{o}C for 15minutes, cooled to about 50^{o}C in a water bath.
c) 0.05g of Chloramphenicol in 10ml of DI water was added to the sterile RBC to inhibit the growth of bacteria (Otitow and Glathe, 1968) and the media was poured aseptically into Petri dishes.
d) Steps c-g described in bacterial enumeration above were repeated but the RBC plates were however incubated in the dark for 3 days at 25^{o}C.
e) The RBC plates were incubated in the dark because exposure of the medium to light could lead to the photo-degradation of Rose-Bengal agar thereby producing compounds which are toxic to fungi (Tassou, Drosinos and Nychas, 1996).

Oil-degrading bacteria identification

After enumeration of the bacterial cells described above, pure cultures were obtained from the mixture of organisms present on the agar plates for further identification by several confirmatory tests. Due to a broad spectrum of microorganisms present in compost and topsoil and for the purpose of this study, microorganisms identified as oil degraders (mentioned in section 2.17) were analysed for. The microorganisms included the genera *Pseudomonas, Bacillus, Acinetobacter, Alcaligenes, Aspergillus and Penicillium* (Sorkhoh *et al*., 1985; Adenuga *et al.*, 1992; Andreoni *et al.*, 1998 and Coupe, 2004). Preliminary and biochemical tests were carried out to identify these organisms by procedures described by Collins and Lyne (1984) and Singleton (2004).

Apparatus used was the same as that of bacterial enumeration listed above.

Procedure:

a) Individual colonies derived from bacterial enumeration were sub-cultured onto fresh sterile NA plates to ensure that a pure culture of each colony was obtained. To achieve this, the surface of each well-separated colony in the plates was lightly touched with a sterile inoculating loop so that a bit of the colony adhered to it and this was then streaked onto fresh sterile NA plates. Streaking involves moving the inoculating loop across the agar in such a way that the inoculum is thinned out so that well-separated individual colonies are obtained as a pure culture.
b) After streaking, the plates were incubated at 25^{o}C for 48hours. The results produced were pure cultures of each colony present in the initial mixed culture and these were examined for identification purposes as described below.

Preliminary tests for microbial identification

a) Morphology: This test helps to identify the shape of the cells whether they are spherical (cocci), rod-shaped (bacilli), curved rod-shaped (vibrios) or spiral-shaped (spirilli). A heat-fixed smear of the colonies (described in Gram staining below) was stained for a minute in methylene blue, rinsed and examined under an oil immersion microscope with a total magnification of about x1000.
b) Motility test: This test determined the motility of the bacterial cells and was carried out by the 'hanging loop' method. A drop of the culture was placed on a clean cover-slip with the aid of an inoculating loop and a small drop of water was placed at each corner of the slip. A glass slide with a central depression was inverted over the cover-slip and the glass slide was then re-inverted with the cover-slip and culture adhering to the slide. The slide was viewed under a phase contrast microscope for motility.
c) Gram staining: This method is used to identify the thickness, structure and composition of bacterial cell walls. When cells are stained with Gram stain (crystal violet and iodine), their ability to retain the dye even after being treated with solvents such as ethanol, makes them Gram-positive but if they get decolorised by the solvent

then they are Gram-negative. A drop of water was placed on a clean slide and using a loop, a speck of the test colony was emulsified with the water drop to form cells suspension. The suspension was spread over a small area of the slide and allowed to dry to form a 'smear'. The smear was fixed by quickly passing the slide over a Bunsen flame twice. The heat-fixed smear was then stained for a minute with a drop of crystal violet (a dye), briefly rinsed under running water, treated for a minute with Lugol's iodine and briefly rinsed again. Drops of 95% ethanol (solvent) were then added to try to decolourise the stained smear. The slide was tilted as the solvent was added so that it runs over the smear for about 1-3 seconds. The smear was then immediately rinsed in running water. Gram-positive cells will be stained violet while Gram-negative cells will be colourless. The smear was then counter-stained with dilute carbolfuchsin (a dye) for 30 seconds and any Gram-negative smear will be stained red. After briefly rinsing the slide, the smear was blotted dry and observed under the oil-immersion objective lens of a microscope with a final magnification of x1000.

d) Endospore formation: This test detects the formation of spores within the cells in response to starvation or shortage of nutrient such as carbon and it is highly resistant to factors such as extremes of temperatures, desiccation and various chemical agents. The test cultures in NA were incubated for a week at 25^{o}C and a heat-fixed smear of the growth was treated with a concentrated solution of carbolfuchsin and the slide heated until the solution steamed. The slide was left hot for about 5 minutes and allowed to cool, then rinsed in running water. Decolourisation of the dye was then attempted by passing the slide through several changes of 95% ethanol. Vegetative cells are decolourised but endospores retain the red dye as seen under the microscope.

e) Catalase test: Catalase is an enzyme produced by most aerobic bacteria and it helps to decompose hydrogen peroxide, produced during aerobic metabolism, into oxygen and water. This test detects the presence of catalase in the bacteria species. A small portion of the bacterial growth was placed in a clean Petri dish. Two drops of hydrogen peroxide was added to the Petri dish, a short distance from the growth. The dish was covered and tilted so that the hydrogen peroxide ran over the growth. Effervescence of oxygen (appearing as bubbles) showed a positive result for catalase.

Biochemical confirmatory tests for microbial identification

a) Oxidase test: This procedure tests for the presence of the oxidase, a respiratory enzyme which helps the organism utilize oxygen for energy production. The enzyme oxidises Kovac's oxidase reagent to give an intense violet coloration. A small amount of the bacterial growth was smeared onto filter paper that had been moistened with a few drops of Kovac's oxidase reagent (1% tetramethyl-ρ-phenylenediamine dihydrochloride). A violet coloration gives a positive result while a light pink to no coloration at all signifies the absence of oxidase.

b) Nitrate reduction test: This detects the ability of the microorganism to reduce nitrate by the presence of the enzyme *nitratase* in anaerobic conditions. The test culture was inoculated into nitrate broth (peptone water + 0.1% w/v potassium nitrate) overnight and examined for nitrite, which is indicative of nitrate reduction. To test for nitrate reduction, 0.5ml of nitrite reagent was added to the broth culture and nitrite, if present, combines with the reagent to give a red coloration. In the absence of a red coloration, a trace amount of zinc dust was added to the medium to enhance the reduction of nitrate to nitrite thereby giving a red coloration (positive result). If there isn't any red coloration after this, then the test is negative.

c) Oxidation-fermentation test (Hugh and Leifson test): This test determines whether bacteria use respiratory or fermentative metabolism for the utilisation of its carbon source, usually glucose. Two tubes of Hugh and Leifson medium (peptone-agar medium + glucose + bromthymol blue – a pH indicator) were half-filled and one of the tubes was steamed to remove dissolved oxygen, cooled and both tubes were inoculated. In the 'steamed' tube, the surface of the medium was sealed with a layer of sterile liquid paraffin, about 1cm deep, to give anaerobic conditions. Both tubes were incubated and examined after 1-14 days and a yellow coloration gives a positive result. Glucose fermenters produce acid in both tubes while glucose oxidisers produce acid only in the unsealed aerobic tube.

d) Arginine test: This test detects the bacteria's ability to hydrolyse the amino acid arginine. The culture was inoculated into arginine broth and incubated for 24-48 hours

after which a few drops of Nessler reagent was added. A brown coloration indicates a positive result of arginine hydrolysis.

e) Gelatine liquefaction: MacConkey agar was inoculated and incubated for five days. Gelatine liquefaction gives a positive result.

f) Urease test: This procedure detects the presence of the urease enzyme. Culture was inoculated into urea agar containing the pH indicator phenol red, and incubated for 3-12 hours at 25^{o}C. A positive result indicates the presence of the enzyme urease which the organism produces to facilitate the conversion of urea to ammonia and the colour of the indicator changes from yellow colour to red.

Oil-degrading fungal Identification

After enumeration, pure cultures were obtained from the mixture of organisms present on the agar plates for further identification. Pure fungal cultures were obtained by procedures similar to those described in oil-degrading bacterial identification described above. Oil-degrading fungi to be identified were *Aspergillus spp* and *Penicillium spp* as described by (Collins and Lyne, 1984).

Apparatus used was the same as that of bacterial enumeration.

Procedure:

- Morphology of the individual growths on RBC plates were recorded and sub-cultured onto fresh RBC plates to obtain pure growths as described in bacterial identification above.
- Colony colours are very useful in preliminary identification of fungal species (Kornerup and Wanscher, 1978) and therefore Individual colonies were examined under a lens and light microscope (magnification up to x1000) to identify vegetative parts and spores. Spores were examined after incubating plates for up to 5 days and compared with results from literature.

- The fungi species were then cultured by inoculating their mycelia growths directly onto Czapek-Dox agar for *Aspergillus spp*. identification and Czapek-Dox agar + 20% sucrose for *Penicillium* identification; and incubated at 25^{o}C for about 3 – 5 days (Collins and Lyne, 1984). Identifications were made based on colour of colonies.

Coliform identification

Bacteriological tests were carried out on compost to detect the presence of indicator organisms, whose presence signify some form of contamination. Coliforms, some of which are commensals of intestinal origin, are examples of indicators organisms. They are lactose-fermenting gram-negative bacilli and their presence is indicative of faecal contamination from birds, animals or humans. Coliforms could be 'typical' (faecal) or 'atypical'. Typical coliforms are naturally found in the intestine and their presence is indicative of faecal contamination e.g. *Escherichia coli* and *Streptococcus faecalis*. Atypical coliforms are mainly saprophytes (obtains food from dead or decaying organic matter); these may grow on vegetation or in the soil and their presence in compost is not indicative of faecal contamination e.g. *Klebsiella aerogenes*. In detecting faecal contamination, it is therefore necessary to differentiate between the typical and atypical strains (Cruickshank *et al.*, 1975). For the purpose of this study, typical (faecal) coliforms in compost were of interest because their presence is directly linked to the quality of water derived from the compost effluents. There are two methods of enumerating coliforms which are viable cell count and presumptive coliform tests. The former gives the number of viable cells in compost while the presumptive tests give an estimation of the cells present in compost leachate.

Viable cell count for coliforms

The procedure for enumerating atypical coliforms and *E. coli* was similar to bacterial enumeration.

Apparatus used was the same as that of bacterial enumeration listed above.

Procedure:

- Media used were MacConkey agar (52g of MacConkey powder in 1liter of DI water) and saline
- The procedure for bacterial enumeration was repeated but incubation temperatures used were 25^{o}C for coliform count and 37^{o}C for *E. coli* count, both for 24 hours.
- Colonies which showed a red colour were indicative of coliforms.

Presumptive coliform count

The multiple tube test technique gives an estimation of coliform bacilli present per 100ml of water sample. The test is said to be 'presumptive positive' for coliforms if there is acid and gas formation. This test gives the most probable number (MPN) of the coliforms, which is an estimation of the number of coliforms present in the sample effluents as described by Cruickshank *et al.,* (1975), Ashbolt, Grabow and Snozzi (2001) and Puehmeier *et al*., (2005)

Apparatus used included:

- Media used were MacConkey broth and saline.
- In addition to apparatus listed in bacterial enumeration, Durham tubes and measuring cylinders were used.

Procedure:

a) Double strength MacConkey broth was prepared by adding 52g of the media to 1 litre of DI water.
b) Single strength MacConkey broth was prepared by adding 26g of the media was added to 1 litre DI water.
c) With sterile measuring cylinders, effluents from the growth media test samples were added to the broth as follows:
    i) One 50ml quantity of effluent + 50ml double strength medium MacConkey broth.
    ii) Five 10ml quantity of effluent + five 10ml double strength medium MacConkey broth.

iii) Five 1ml quantity of effluent + five 5ml single strength medium MacConkey broth.

d) The bottles were incubated at 37^{o}C for 18-24 hours.

e) All the bottles had Durham tubes in them to show gas formation which will fill up the concave part of the tube and the broth shows acid formation by turning from purple to yellow. This is indicative of a positive result.

f) After 24 hours, all negative bottles were re-incubated at the same temperature and those that developed acid and gas were positive to the test.

g) In completing this test, reference was made to the McCrady probability tables to determine coliform numbers/100ml of effluent (Cruickshank *et al,*. 1975; Tillett, 1987).

h) Each of the samples was tested in triplicates.

Differential coliform test

To ascertain if the coliforms in the presumptive test were the enteric bacteria, *Escherichia coli*, the Eijkman test was employed. This test is based on the fact that *E. coli* is able to produce gas at a temperature of 44^{o}C which the atypical strains cannot do (Cruickshank *et al.*, (1975).

Apparatus used included:

Medium used was single strength MacConkey broth and apparatus was the same as that of the presumptive test.

Procedure:

a) Subcultures were made from all the bottles that showed positive results in the presumptive test into bottles of fresh single strength MacConkey broth. Prior to this, the bottles of broth were heated in a water bath at 37^{o}C.

b) The inoculated bottles were then incubated at 44^{o}C and examined after 24 hours.

c) Tubes containing *E. coli* produced gas which was visible in the Durham tube after incubation.

3.4 Experiments on growth media test samples

This section describes the different experiments carried out on the test samples to assess their behaviour in terms of microbial activity, plant growth, biomass production, and contamination with heavy metals and motor oil.

3.4.1 Monitoring of Microbial Respiration

Experiments on growth media samples to monitor the rate of evolution of carbon dioxide (CO_2) by the microorganisms they contained, which is directly related to microbial activity and biodegradation (Puehmeier *et al*., 2005; Coupe *et al.*, 2006[a]), was carried out. The experiments were carried out in low moisture, restricted oxygen conditions in the presence of light, and in saturated aerobic conditions in the absence and presence of light. The test was carried out in Perspex rigs with dimensions of 11.6cm by 11.8cm by 28.7cm. Into each of these rigs were placed 3 litres of test materials (i.e. GC, MC and T as control) in triplicates. The samples were measured as a function of their volume rather than masses because their bulk densities varied, with topsoil having the highest density and mixed compost having the lowest density.

The first phase of the test was carried out to determine microbial respiration in low moisture conditions simulating periods of low or no rainfall/moisture; and microaerophilic conditions simulating conditions of restricted oxygen (such as compacted soils and bases of vegetative SuDS) that may occur in vegetative SuDS (Iijima *et al*., 2003; David and Sousa, 2008). The rigs were covered in cling film to concentrate the CO_2 given off (set up similar to that of figure 3.1) and gas samples were extracted once a week with a 1ml syringe and analysed by an infra-red gas analyser (IRGA; ADC-225-MK3, UK). The IRGA was calibrated with 3% ppm CO_2 at a standard volume of 0.2ml. Measurements were made and recorded to measure CO_2 production by microorganisms present in the samples.

In the second phase, CO_2 monitoring tests were carried out in saturated aerobic conditions (a) in the absence of light (figure 3.2) simulating aerobic regions within vegetative SuDS with access to little or no light (e.g. the sub-base layer); and (b) in the presence of light (figure 3.1) simulating aerobic regions within vegetative SuDS accessible to light (e.g. the layer closer to

the surface). The procedure described above was repeated but this time the samples were saturated with de-ionised water and aerated once a week. These tests were carried out in triplicates and carried out over a period of nine weeks.

Figure 3.2: The experimental set-up for monitoring microbial activity in the absence of light showing rigs covered in plastic bags.

Source: Author's own

3.4.2 Plant Trials - grass biomass determination and leachate analysis

The presence of vegetation on impermeable surfaces improve surface water quality by increasing infiltration and decreasing flow rate of run-off, thereby enabling the infiltration of water into the soil, where processes such as biodegradation, absorption, adsorption, sedimentation and flocculation of pollutants (e.g. organic matter, motor oil, heavy metals, dust, dirt etc.) can take place. This ultimately helps to reduce the quantity and pollutant load present in runoff. Vegetative SuDS such as swales and filter strips operate under this principle

(Revitt and Ellis, 2001; Woods-Ballard *et al*., 2007). The plant trials were carried out in plant pots to assess the fate of pollutants and biomass yield under swale conditions, such as the addition of water (to simulate rainfall events) and pollutants (to simulate pollutants present in surface runoff). Small plant pots were used so as to provide a high number of replicates, as opposed to using larger vessels.

The compost samples were used as growth media in different combinations as described below: green compost (GC), mixed compost (MC), 1:1 blend of green compost and topsoil (GCT), 1:1 blend of mixed compost and topsoil (MCT) and topsoil (T) as control. The four types of test aggregates were used as the sub-base i.e. old bricks (OB), new bricks (NB), limestone (L) and gravel (G) as control. The four types of aggregates with the five types of growth media, each with four replicates made a total of eighty pots. Mixtures of four types of grasses which are usually used in vegetative SuDS were sown. They were perennial rye grass (*Lolium perenne*), red fescue (*Festuca rubra*), tall fescue (*Festuca arundinacea*) and creeping bent (*Agrostis stolonifera*) (Wilson *et al*., 2004; Highways Agency, 2006), and the grass seeds were obtained from Rothamsted Research, UK. The eighty pots were arranged in a randomised block design as described in tables 3.2, 3.3, 3.4 and 3.5. The method carried out was a modification of Sloan *et al*., (2008).

Old bricks

	Column a	Column b	Column c	Column d
Row 1	OBT_1	$OBMC_4$	$OBGCT_4$	$OBMC_2$
Row 2	$OBMCT_1$	$OBMC_3$	$OBGC_3$	$OBGC_1$
Row 3	$OBGC_4$	$OBMCT_2$	$OBMCT_3$	$OBGCT_2$
Row 4	OBT_2	$OBGCT_3$	$OBGCT_1$	$OBMC_1$
Row 5	OBT_4	$OBMCT_4$	$OBGC_2$	OBT_3

Table 3.2: Randomised block design of plant trials showing a combination of green compost (GC), mixed compost (MC), green compost + topsoil (GCT), mixed compost + topsoil (MCT) and topsoil (T) underlaid with crushed old bricks.

New bricks

	Column a	Column b	Column c	Column d
Row 1	NBT_1	$NBMC_4$	$NBGCT_4$	$NBMC_2$
Row 2	$NBMCT_1$	$NBMC_3$	$NBGC_3$	$NBGC_1$
Row 3	$NBGC_4$	$NBMCT_2$	$NBMCT_3$	$NBGCT_2$
Row 4	NBT_2	$NBGCT_3$	$NBGCT_1$	$NBMC_1$
Row 5	NBT_4	$NBMCT_4$	$NBGC_2$	NBT_3

Table 3.3: Randomised block design of plant trials showing a combination of green compost (GC), mixed compost (MC), green compost + topsoil (GCT), mixed compost + topsoil (MCT) and topsoil (T) underlaid with crushed new bricks.

Recycled limestone aggregates

	Column a	Column b	Column c	Column d
Row 1	LT_1	LMC_4	$LGCT_4$	LMC_2
Row 2	$LMCT_1$	LMC_3	LGC_3	LGC_1
Row 3	LGC_4	$LMCT_2$	$LMCT_3$	$LGCT_2$
Row 4	LT_2	$LGCT_3$	$LGCT_1$	LMC_1
Row 5	LT_4	$LMCT_4$	LGC_2	LT_3

Table 3.4: Randomised block design of plant trials showing a combination of green compost (GC), mixed compost (MC), green compost + topsoil (GCT), mixed compost + topsoil (MCT) and topsoil (T) underlaid with recycled limestone aggregates.

Gravel

	Column a	Column b	Column c	Column d
Row 1	GT_1	GMC_4	$GGCT_4$	GMC_2
Row 2	$GMCT_1$	GMC_3	GGC_3	GGC_1
Row 3	GGC_4	$GMCT_2$	$GMCT_3$	$GGCT_2$
Row 4	GT_2	$GGCT_3$	$GGCT_1$	GMC_1
Row 5	GT_4	$GMCT_4$	GGC_2	GT_3

Table 3.5: Randomised block design of plant trials showing a combination of green compost (GC), mixed compost (MC), green compost + topsoil (GCT), mixed compost + topsoil (MCT) and topsoil (T) underlaid with gravel.

Apparatus used included:

- Plants pots with saucers to collect leachates
- Filter paper
- Forced-air oven
- Watering can with rose showerhead
- Weighing scales

Procedure:

a) The four types of aggregates with the five types of growth media mentioned above were placed in 13.3cm diameter pots layered with Whatman filter paper to filter the leachate. The pots were placed on saucers to collect the ensuing leachate. There were four replicates for each combination, making a total of eighty pots (see tables 3.2 – 3.5).

b) The control was the topsoil and gravel combination (GT).

c) The four grass types were planted in each pot. The seeds were sown at the rate of 32g per m^2 (McKenzie and Hill, 1990) and therefore each pot contained approximately 0.1g of each grass type.

d) The plants were cultivated in a greenhouse at Coventry University with a temperature of 19-22^{o}C throughout the experiment. Light was provided by daylight as well as supplementary light from 400W high pressure sodium lamps providing a minimum of 16-hour photoperiod and photon flux of 180-210µmol$m^{-1}s^{-1}$ at bench level (see figure 3.3).

e) The eighty pots were arranged in a randomised block design.

f) The pots were saturated with DI water, three times a week using a watering can fitted with a rose showerhead.

g) After four weeks of germination, the grasses were harvested by cutting them with a scissors at 2cm above the growth medium.

h) Wet and dry weights of the harvested grasses were measured to determine biomass production (Tackenberg, 2007). The dry weight was determined by drying the harvested grasses in a forced-air oven at 80^{o}C for 3 days and re-weighing them.

i) After the 1st grass harvest (which was analysed for background heavy metal concentrations in both grasses and leachate), 10ml of an aqueous solution containing 250µg (25mg/L) of aluminium (Al), cadmium (Cd), chromium (Cr), copper (Cu), iron (Fe), manganese (Mn), nickel (Ni), lead (Pb) and zinc (Zn) was added to each pot once a week for four weeks, after which the grasses were harvested and analysed for heavy metals. This process was repeated the next month giving a total of eight weeks metal addition by the 3rd harvest in the third month of plant growth. The 4th and 5th harvests/months had no heavy metals added, and was also analysed for heavy metals to determine the concentration of residual metals.

j) The pots were leached with 250ml of DI water, a day after each metal addition. A total of 200mg/L of each heavy metal was added over the eight-week test period (i.e. 25mg/L*8weeks).

k) All liquids, i.e. heavy metals and DI water, were carefully added to the centre of the plant pots so as to prevent the liquids from running down the sides of the pots.

l) The leachate volume was measured and analysed for Al, Cd, Cr, Cu, Fe, Mn, Ni, Pb and Zn by inductively coupled plasma atomic emission spectroscopy (ICP-AES).

m) The results were compared with WHO drinking water guidelines for heavy metals (WHO, 2011).

n) Steps g and h were repeated every month for five months and grasses were analysed for heavy metal content as described in section 3.3.7.

Figure 3.3 shows a cross section of the plant trial experiment.

Figure 3.3: Cross-section of plant trial pots.

Source: Author's own

3.4.3 Oil retention experiments on test profiles

In vegetative SuDS, organic pollutants such as motor oil can either be adsorbed or absorbed by components such as vegetation, aggregates and especially growth media which will help to improve water quality by making these pollutants unavailable in water (Highways Agency, 2006; Woods-Ballard *et al*., 2007). Oil retention tests on the test profiles were carried out to ascertain the fate of clean and used motor oil in vegetated SuDS and to determine the ability of the profiles to mitigate oil pollution. The experimental set-up similar to that of figure 3.3 was used to examine the fate of motor oil in growth media in vegetative SuDS, as this is where motor oils are mostly retained for treatment (Napier *et al*., 2008). As a result there was no vegetation on the growth media during the motor oil experiment. The experiment involved the addition of used and clean oil to the test profiles weekly with application rates of 25ml/m^2, which is equivalent to a month's worth of oil loading in a typical urban environment (Wilson *et al*., 2003). The application of motor oils was applied to simulate a year's worth of oil loading in a typical urban environment because vegetative SuDS are to be monitored, at most, yearly for motor oil contamination and accumulation (Woods-Ballard *et al*, 2007; Pittner and Allerton, 2009). Therefore this experiment will help to ascertain which of the test profiles will be best suited for mitigating oil pollution long term.

The volume of oil applied was calculated thus:

Radius of pot = 0.067m

Surface area of pot = $\pi r^2 = 0.014m^2$

Standard oil volume per unit area = $25ml/m^2$

Oil volume/pot/week = $\frac{25ml * 0.014m^2}{1m^2}$

= 0.4ml which is equivalent to a month's worth of oil loading in a typical urban environment (Wilson *et al*., 2003).

Therefore for one year's worth of oil loading in a typical urban environment, the total volume of oil required for addition into each pot = 0.4ml*12weeks = 4.8ml, considering that 1 weekly addition of motor oil to the test profiles was equivalent to 1 months' worth of oil loading in a typical urban environment (Wilson et al., 2003).

Clean motor oil has been used in previous oil studies as a standard contaminant in laboratory tests because of its consistent viscosity and chemical compositions (Bond, 1999; Coupe, 2004; Puehemier *et al*., 2004). However for the purpose of this study, used motor oil was applied because it is the one of the major sources of hydrocarbon contaminants in surface runoff and will eventually be treated in vegetative SuDS (Wilson *et al*., 2005; Ellis and Chatfield, 2006). However, due to variations in viscosity, chemical and biological compositions of used oil compared to clean oil, as a result of the processes used oil undergo in vehicle engines (Coupe *et al*., 2005), clean oil was used as the control

Experimental procedure:

Oil volume of 0.4ml per pot was added using a 1ml syringe and the pots were leached 24 hours later with 200ml of DI water. This process was repeated once a week for 12 weeks (which is equivalent to 12 months' worth of oil added to each pot for the 12-week test period). The leachates were collected and analysed for oil content using the OCMA – 310 Horiba oil content analyser. The oil analyser measured the concentration of oil in water by infra-red absorption of various wavelengths emitted from hydrocarbons in oil. The oil present in the

samples were extracted using a solvent known as S-316 which is a double chlorotrifluoroethylene obtained from a dimerization (doubling) reaction of chlorotrifluoroethylene monomer. After extraction, the machine measured the oil concentration in the samples from the changes in the amount of infra-red absorption in the 3.4 – 3.5µm wavelength range of the extracted liquid (Horiba Ltd., 2004).

Before analysis, a zero calibration was carried out by inserting 10ml of the pure solvent (S-316) into the machine, followed by one drop of hydrochloric acid and 20ml of de-ionised water. The mixture was extracted pressing the 'extract' button and the oil concentration was measured by pressing the 'measure' button. Standard calibration was made using the method for the zero calibration but in this case, a standard solution of 50mg/L of the used and clean motor was used. After calibration, the test sample leachates were analysed using the procedure stated above. The machine was re-calibrated after the analysis of every ten samples to prevent the drift of data (Horiba Ltd., 2004).

3.5 Analysis on aggregates test samples

3.5.1 Background analysis of aggregates test samples

Background heavy metal analysis was carried out on RA, i.e. OB, NB, L and G, to measure the concentration of immobile and mobile heavy metals they may contain. The aggregates were pulverised using a mill before analysing for total heavy metal concentrations by ICP analysis as described in section 3.3.7. Leaching experiments were carried out by washing the RA with 100ml of distilled water and the leachates were analysed for heavy metals by ICP analysis. Data derived from total heavy metal analysis on the aggregates were compared to the Interdepartmental Committee on the Redevelopment of Contaminated Land values (ICRCL 59/83) and the Kelly indices guideline values, as these standards give guidelines for contaminated land based on its use (ContaminatedLand, 2000). These values are shown in table 3.6:

Table 3.6: Heavy metal concentrations based on ICRCL 59/83 and Kelly indices specifications.

Source: ContaminatedLand (2000).

Data derived from heavy metal analysis on the leachates from the aggregates were compared to the WHO (2011) potable water guideline. These values are shown in table 3.7:

Table 3.7: WHO (2011) potable water guideline.

Source: WHO (2011)

3.5.2 Experiments carried out on aggregates test samples

Oil absorption experiments on aggregates

Oil retention tests were carried out to determine the ability of RA in the retention of motor oil (used and clean oil from car parks, mechanic workshops and spillages), because motor oil is a common pollutant found in surface runoff (Highways Agency, 2006; Woods-Ballard *et al*., 2007). This test will suggest which of the aggregates will be suitable for use as a sub-base component in vegetative SuDS due to their retention capabilities.

Apparatus used included:

- Tin foils
- Used and clean oil
- Weighing scales

Procedure:

a) Aggregates of varying known weights were placed in tin foils, labelled accordingly (see appendix 33) and then saturated with clean and used motor oil.
b) The aggregates were allowed to soak in the oil for two days after which they were removed and allowed to sit for another two days in order to allow excess oil drain off, after which they were weighed.
c) The % weight of oil absorbed was calculated thus

$$\frac{(\text{weight of aggregates + oil}) - \text{initial weight of aggregates}}{\text{Initial weight of aggregates}} * 100$$

e) The drained aggregates were then put back into the oil. This process was repeated until the weights of the aggregates became fairly constant.

Oil analysis of leachates from aggregates

To further determine how well RA would retain oil when flushed with water e.g. during infiltration of surface runoff and during movement of water into the water table, leaching experiments were carried out on RA containing motor oil, because if aggregates release their oil content easily when flushed with water, it can jeopardise water quality.

Apparatus used included:

- One litre plastic bottles
- 1ml syringe
- Used and clean oil
- Weighing scales

Procedure:

a) Approximately 250grams of each aggregate i.e. OB, NB, L and G was placed in inverted bottomless one litre plastic bottles in triplicates (figure 3.4)
b) 1ml (or 0.83g) of clean and used motor oil (equivalent to 8 months' worth of oil loading in a typical urban environment as calculated below) were applied to the aggregates using a 1ml syringe.

Average radius of bottle = 0.0398m

Surface area of bottle = πr^2 = 0.00498m^2

Standard oil volume per unit area = 25ml/m^2 (Wilson *et al*., 2003)

Oil volume/bottle = $\frac{25ml * 0.00498m^2}{1\ m^2}$

= 0.125ml ≈ a month's worth of oil loading in a typical urban environment

Therefore 1ml (or 0.83g) of oil ≈ eight months' worth of oil loading in a typical urban environment

c) After 24 hours, the aggregates were leached with 100ml of deionised (DI) water and the effluent collected was analysed for oil content using the Horiba OCMA – 310 oil content analyser (see section 3.4.3). This process was repeated once a week for three weeks

d) Total concentration of oil added by the end of the experiment in mg/L was calculated thus:

Weight of clean/used oil added to aggregates = 0.83g = 830mg

Total volume was water added aggregates = 100ml * 3weeks = 300ml

Weight of oil added per 1L (1000ml) = $\frac{830mg * 1000ml}{300ml}$

Total concentration of used/clean motor oil added to aggregates = **2767mg/L**

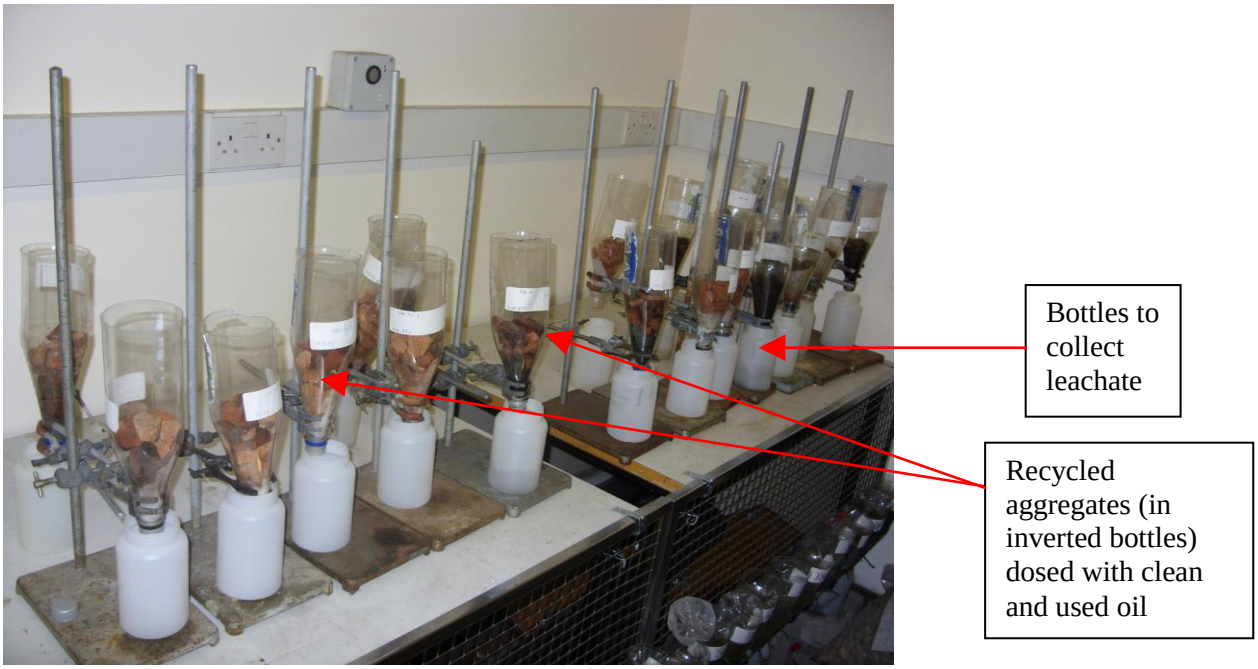


Figure 3.4: The experimental set-up for determining oil retention capabilities of recycled aggregate flushed with water.

Source: Author's own

3.6 Conclusion

The overall purpose of carrying out experiments on the test samples was to determine the potential of compost in replacing or complementing topsoil, as well as the replacement of gravel with RA, in vegetative SuDS in order to further improve the sustainability of these devices. Compost has been applied as compost socks and blankets in vegetative SuDS for erosion control and improvement of water quality, while RA has been applied as substrate to vegetative SuDS such as green roofs for improving water quality. However, the direct application of compost and RA to other vegetative SuDS devices such as swales has not been carried out and this study was carried out to assess the performance of the test materials in swales.

The test samples individually have some promising qualities as discussed in chapter two, which when applied will improve the sustainability of vegetative SuDS. However, it is

expected that the samples themselves may contain some compounds and possess some qualities which may further jeopardise water quality and quantity, and as a result, experimental designs were carried out over a period of time to discover their capabilities and shortcomings.

The first objective of this study was to characterise the test materials i.e. compost and recycled aggregates. To fulfill this aim, analysis on moisture content, organic matter and carbonate content, water holding capacity, bulk density, pH, total heavy metal content, dissolved heavy metal content, bacterial and fungal enumeration and identification were carried out. Data derived from baseline analyses were compared to PAS 100:2011 (BSI, 2011), CSUGE (WRAP, 2003), ICRCL 59/83 values and Kelly indices guideline values (ContaminatedLand, 2000). It was expected that these analyses would give a clear picture of test materials' characteristics and how they would affect treatment and quality of runoff in vegetative SuDS such as swales.

The second objective was to investigate the development of biofilms in compost in simulated swale conditions, as this is necessary for the biodegradation of organic pollutants within vegetative SuDS such as swales. This aim was achieved by monitoring microbial activity over time, in varying moisture and light conditions simulating various swale conditions. The tests were carried out by measuring the CO_2 evolved during metabolic activities of microorganisms, and would give indications of how microorganisms biodegrade pollutants in vegetative SuDS (such as swales) under varying conditions.

The third objective was to investigate biomass development in profiles comprising of compost and recycled aggregates under simulated swale conditions. This was carried out by plant trials which involved the harvesting of grasses every month over time, and dry weight measurements were taken in order to ascertain which of the profiles were able to sustain the most vegetative growth over time. This assessment is important because biomass quality is necessary for the effective functioning of vegetative SuDS, in phytoremediation and

increasing soil permeability thereby aiding infiltration and flood attenuation (Woods-Ballard *et al*., 2007).

The fourth objective was to investigate the efficacy of compost and recycled aggregates in remediating pollutants in simulated swale conditions. This was achieved by heavy metal and motor oil analyses of test profiles and their leachates, dosed with heavy metals and motor oil in swale simulations. Heavy metal and oil analyses were carried out to determine the concentration of total heavy metals and motor oil retained within the samples; and leaching tests were carried to determine the concentrations of mobile heavy metals and motor oil in ensuing leachate. Deliberate spiking of profiles with heavy metals and motor oil, commonly found in runoff was carried out to determine how well components of the test profiles would treat these pollutants.

The fifth objective which was to study the effect of RA on oil pollution mitigation was carried out by oil absorption studies on oil-dosed RA and oil analysis of their leachates. Results of this analysis would give an indication of which aggregate type would be most suitable for replacing or supplementing gravel as aggregate beds in vegetative SuDS. In the following chapter, results from the tests discussed in this chapter will be analysed and compared with existing data and specifications, to see the suitability of each test sample in maintaining water quality on vegetative SuDS.

Chapter Four: Results

4.1 Introduction

This chapter comprises of results derived from applying the methods and experimental designs described in chapter three to the following test samples: growth media samples which included mixed compost (MC), green compost (GC) with topsoil (T) as the control; and recycled sub-base aggregates which included crushed old bricks (OB), crushed new bricks (NB), limestone aggregates (L) with gravel (G) as control. These test samples were also combined together as described in section 3.4.2 making a total of twenty test profiles each with four replicates.

The chapter is divided into two main sections:

1) ***Baseline data derived from the test samples*** which included moisture content, organic matter and carbonate content, water holding capacity, bulk density, pH, total heavy metal content, bioavailable heavy metal content, bacterial and fungal enumeration and identification. Background analyses were carried out to ascertain if the test samples were within specified standards, as well as giving a first-hand view of the potentials of the samples before they were actually applied. This section contains data in fulfilment of the first aim and objective of this study.

2) ***Data derived from experiments carried out on the test samples*** which includes monitoring microbial activity, monitoring of grass biomass yield, heavy metal monitoring of test growth media by leaching experiments; heavy metal monitoring of grass biomass and test growth media; monitoring of motor oil in effluents by leaching experiments; and monitoring of motor oil in test profiles by absorption studies. This section contains data in fulfilment of the second to fifth aims and objectives of this study.

Characterisation of the test samples was the first objective of the study and this was carried out to establish baseline physical, geochemical and biological properties of the samples which provided bases for further studies. Baseline data on growth media was compared to PAS

100:2011, a British Standards Institute (BSI) Publicly Available Specification (PAS) standard for composted materials (BSI, 2011); as well as the Compost Specification for Use in Grass Establishment (CSUGE) (WRAP, 2003), which consists of additional requirements to the PAS 100 standards (discussed in section 2.14). Baseline data on recycled aggregates were compared to the ICRCL 59/83 values, the Kelly indices guideline values (ContaminatedLand, 2000) – see table 3.6, and WHO (2011) potable water guidelines – see table 3.7

4.2 First objective: Baseline data derived from test samples

4.2.1 Moisture content in growth media

Determination of moisture content was carried out to measure the amount of moisture present in the growth media test samples, as this is a standard requirement for composted materials as stipulated by PAS 100:2011 (BSI, 2011). Figure 4.1 shows that GC had the highest moisture content of 42%, falling within the CSUGE specification of between 35-55% (WRAP, 2003). MC and T had moisture contents of 28% and 14% respectively, both below the standard values. Statistical analysis carried out by one-way analysis of variance (ANOVA) (see appendix 1) showed that moisture contents of GC followed by MC were significantly higher than T.

Figure 4.1: Percentage moisture content of green and mixed compost and topsoil compared to the Compost Specification for Use in Grass Establishment (CSUGE). (WRAP, 2003)

4.2.2 Water holding capacity of growth media

In as much as initial moisture of growth media is important for the reasons stated above, the ability of these media to retain water is equally vital to microbial activity and biodegradation. Water retention abilities of the test growth media were determined by measuring their water holding capacity (WHC), because this factor is important for microbial activity (associated with biodegradation) and growth of vegetation (Mamo *et al*., 2000; Sloan *et al*., 2008) in vegetative SuDS. Figure 4.2 shows the trend of water holding capacities of MC, GC and T over time.

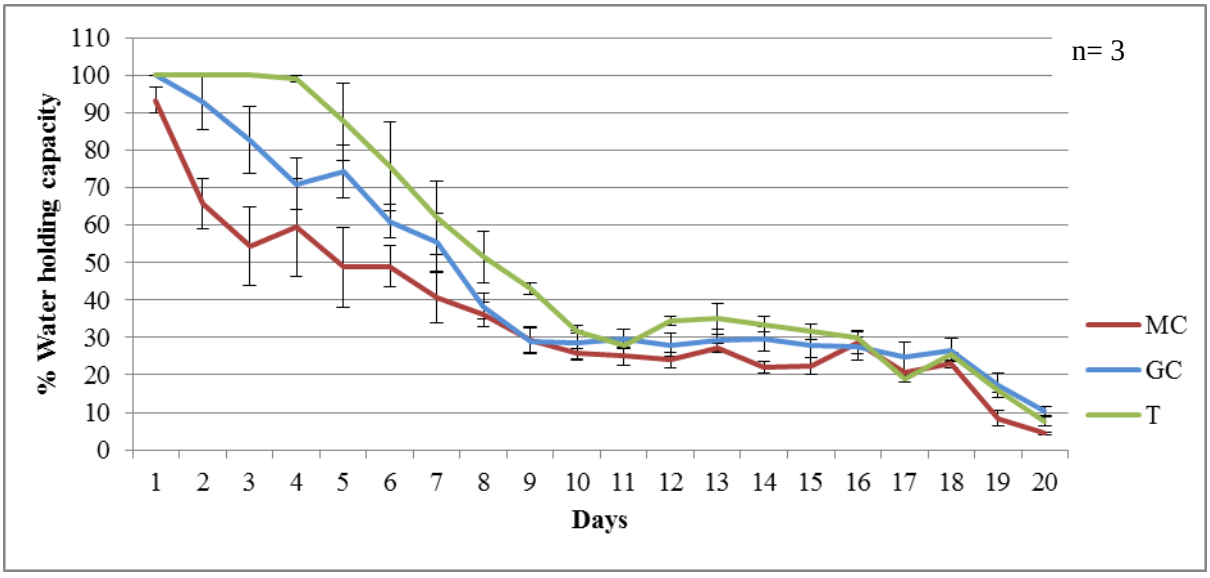


Figure 4.2: Percentage water holding capacity of green and mixed compost and topsoil.

Maximum water retention occurred in T for 3 days before becoming saturated, after which WHC began to decline. Maximum water retention occurred in GC for 24hours before reaching saturation while MC reached saturation in less than 24hours (retaining 93% of the added volume of water), and WHC continued to decrease until the end of the experiment. The error bars and one-way ANOVA analysis showed that statistically, WHC was not significantly different ($p>0.05$) in the three samples from day 7 till the end of the experiment indicating that water retention in the three samples declined similarly from day 7 till the end of the

experiment (see appendix 2). WHC on the last day of the experiment was highest in GC at 10% closely followed by T at 8% and lastly, MC at 4%.

4.2.3 Organic matter content of growth media

In the determination of organic matter content in the test growth media, MC contained the highest organic matter at 67%, followed by GC at 42% (see figure 4.3). T had the least at 10%, which was below the CSUGE specification of >25% (WRAP, 2003). Statistical analysis carried out by one-way ANOVA showed that organic matter contents of MC followed by GC were significantly higher than T (see appendix 3).

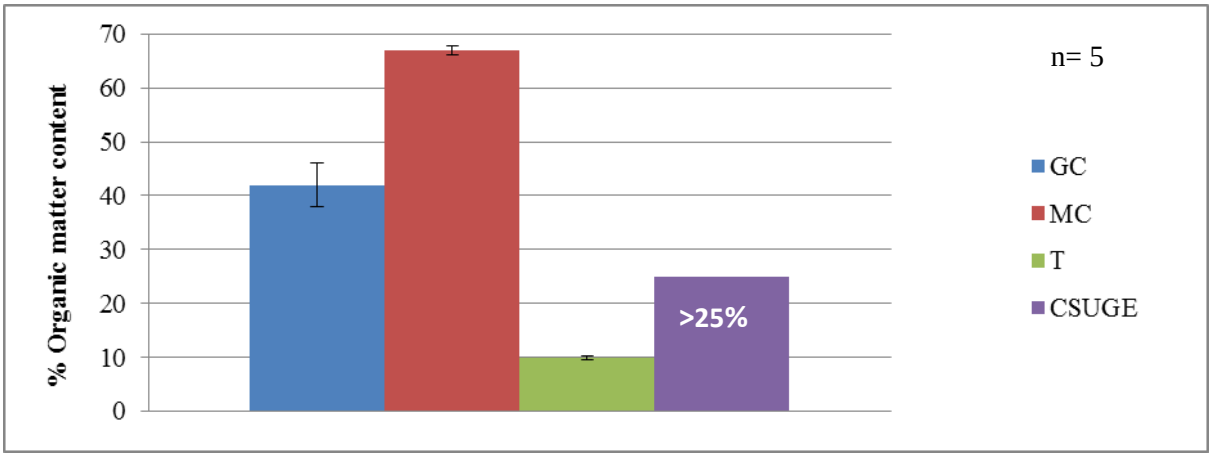


Figure 4.3: Percentage organic matter content of green and mixed compost and topsoil compared to the Compost Specification for Use in Grass Establishment (CSUGE).
Source: WRAP, 2003

4.2.4 Bulk density of growth media

Soils that are high in organic matter have bulk densities of $\leq 1.0 gcm^{-3}$ and such soils will encourage the biodegradation of pollutants and improve soil structure in vegetative SuDS and so the higher the organic matter content, the lower the bulk density and vice versa (Chan, 2006). The bulk densities of GC and MC fell within the CSUGE specification value of $1.0gcm^{-3}$ at $0.5gcm^{-3}$ and $0.2gcm^{-3}$ respectively (figure 4.4). T had the highest bulk density of $1.02gcm^{-3}$ indicating that it was denser than GC and MC. Statistical analysis carried out by

one-way ANOVA showed that bulk densities of T followed by GC were significantly higher than MC (see appendix 4).

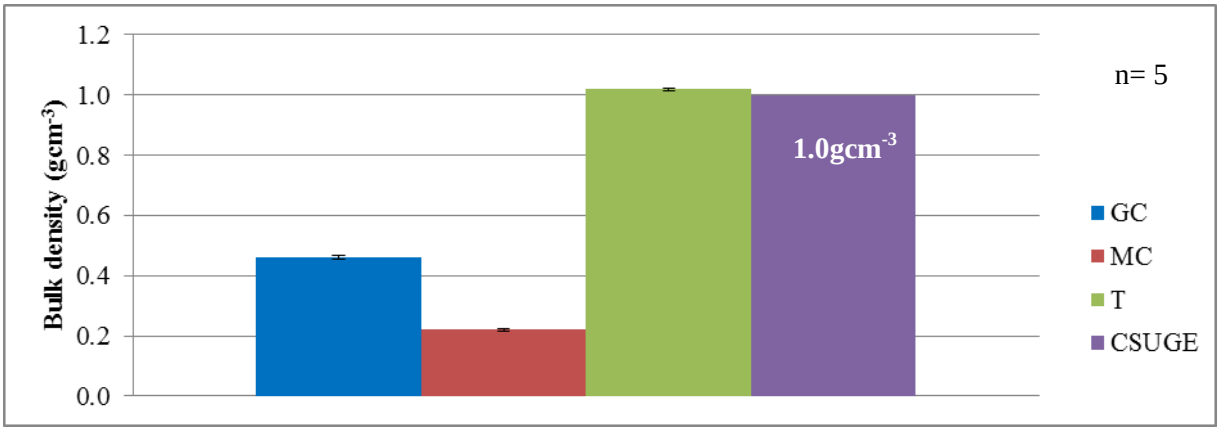


Figure 4.4: Bulk density of green and mixed compost and topsoil compared to the Compost Specification for Use in Grass Establishment (CSUGE).

Source: WRAP, 2003

4.2.5 Carbonate Content of Growth Media

As well as having the highest organic matter, MC also contained the highest carbonate at 6% followed by T at 4%, GC had the lowest carbonate content at 3% (see figure 4.5). Statistical analysis carried out by one-way ANOVA showed that carbonate contents of MC followed by T were significantly higher than GC (see appendix 5).

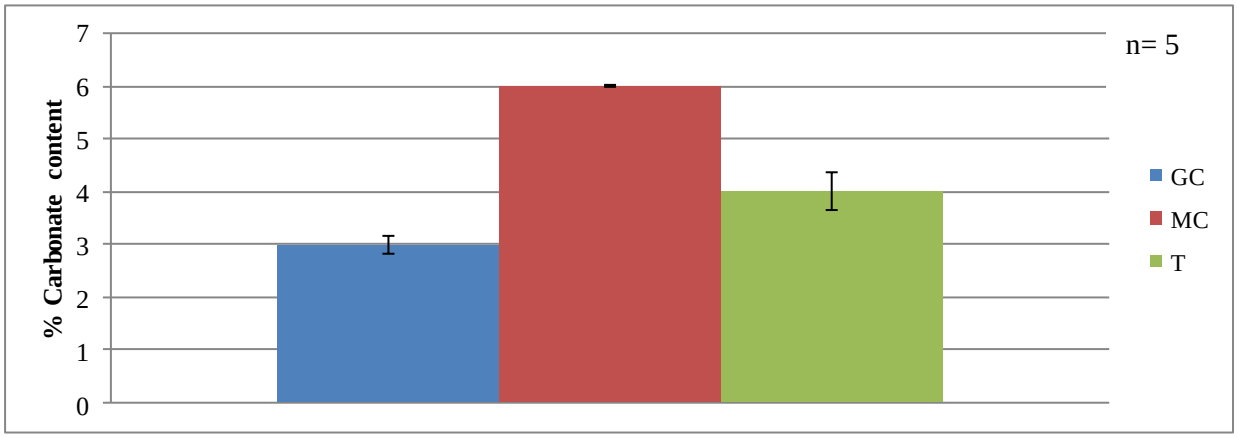


Figure 4.5: Percentage carbonate content of green and mixed compost and topsoil.

4.2.6 Hydrogen ion level (pH) of growth media

The pH level of GC, MC and T were close to neutral at 7.3, 7.6 and 6.8 respectively, with GC and MC falling within the CSUGE specification pH range of 7.0-8.7 (WRAP, 2003), as seen in figure 4.6 below. Statistical analysis carried out by one-way ANOVA showed that pH levels of MC followed by GC were significantly higher than T (see appendix 6).

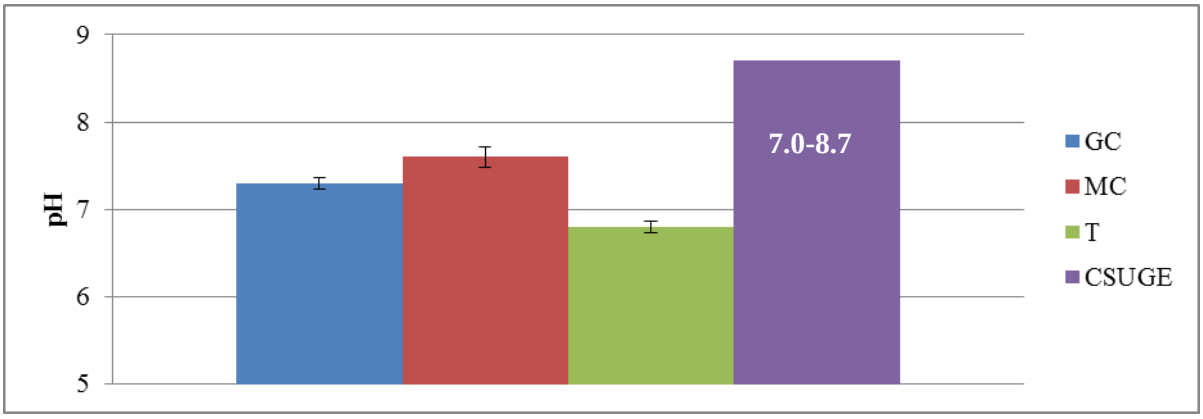


Figure 4.6: pH of green and mixed compost and topsoil compared to the Compost Specification for Use in Grass Establishment (CSUGE).

Source: WRAP, 2003

4.2.7 Heavy metal analysis of growth media and aggregates

In analysing for background heavy metal concentrations in the test samples (see table 3.1 for heavy metals analysed), two heavy metal fractions were analysed: total heavy metals by inductively coupled plasma-atomic emission spectroscopy (ICP-AES) and mobile heavy metals by leaching experiments (Quevauviller, 1998). Table 4.1 shows the mean total heavy metal concentrations present in the growth media for Al, Cd, Cr, Cu, Fe, Mn, Ni, Pb and Zn, and these metals were compared with the PAS 100 specification, although values were only available for Cd, Cr, Cu, Ni, Pb and Zn (BSI, 2011). Metals that had no PAS 100 values (i.e. Al, Fe, and Mn) were compared with soil typical values in literature and with Kelly indices values for uncontaminated soils.

Samples (n=3)	**mgkg^{-1}**								
	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
GC	4899±3.15	0.9±0.00	**104**±0.19	65±0.07	10796±7.77	322±0.07	23±0.05	131±0.10	247.6±0.23
MC	2228±1.92	1.3±.002	18±0.02	33±0.01	4793±5.21	223±0.14	9±0.00	62.5±0.04	125.9±0.09
T	8333±14.94	0.3±0.00	29±0.09	194±0.10	13359±12.51	395±0.14	22±0.00	**2945**±5.60	171.3±0.06
PAS 100 (BSI, 2011)	–	1.5	100	200	–	–	50	200	400
Kelly indices (ContaminatedLand, 2000)	–	≤1	≤100	≤100	–	≤500	≤20	≤500	≤250
Soil typical values for Al (ATSDR, 2008[a])	7,000-100,000	–	–	–	–	–	–	–	–
Soil typical values for Fe (Jankiewicz, Ptaszyński and Turek, 2002)	–	–	–	–	2,000- 48,000	–	–	–	–

Table 4.1: Total heavy metal content of green compost, mixed compost and topsoil.

Table 4.1 shows that total heavy metal concentrations in the samples were within the PAS 100 values except for Cr, which was slightly over the Cr PAS 100 and Kelly indices limits in GC at 104 $mgkg^{-1}$, and Pb which was extremely high in T at 2945$mgkg^{-1}$. Mn concentrations for growth media test samples fell within the Mn Kelly indices guideline value of ≤500 $mgkg^{-1}$for uncontaminated soils (ContaminatedLand 2000). Al concentrations for GC, MC and T were 4,899 $mgkg^{-1}$, 2,228 $mgkg^{-1}$ and 8,333 $mgkg^{-1}$ respectively (or 0.5%, 0.2%, 0.8% respectively) and these values were well below typical soil aluminium range of 0.7% - 10% (i.e. 7,000 $mgkg^{-1}$ - 100,000 $mgkg^{-1}$) (ATSDR, 2008[a]; Verstraeten, Aimo and Oteiza, 2008). Likewise Fe concentrations in all the growth media test samples were well below the soil typical values of 0.2% to 4.8% (2,000 $mgkg^{-1}$ - 48,000 $mgkg^{-1}$) (Jankiewicz, Ptaszyński and Turek, 2002; Payne *et al*., 2007).

Table 4.2 shows the mean total heavy metal concentration present in RA samples for Al, Cd, Cr, Cu, Fe, Mn, Ni, Pb and Zn compared with ICRCL 59/83 values and Kelly indices guideline values (ContaminatedLand, 2000). Like the PAS 100 specification, ICRCL 59/83 had no values for Al, Fe and Mn while the Kelly indices values had no values for Al and Fe. Heavy metals that had no guideline values (i.e. Al and Fe) were compared to values in literature.

Samples (n=3)	mgkg^{-1}								
	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
NB	10263±11.56	0.02±0.00	37±0.03	6±0.01	7829±22.23	218±0.33	9±0.01	38±0.27	6±0.01
OB	16405±26.05	0.00±0.00	2±0.15	12±0.02	13543±15.40	**562**±0.73	20±0.04	8±0.02	37±0.09
G	410±1.26	0.02±0.00	2±0.01	2±0.01	4066±17.47	47±0.33	2±0.01	3±0.01	4±0.02
L	2939±9.41	0.27±0.00	17±0.04	6±0.03	12346±2.46	370±0.16	13±0.05	7±0.01	9±0.03
ICRCL 59/83	–	3	600	130	–	–	70	500	300
Kelly indices	–	≤1	≤100	≤100	–	≤500	≤20	≤500	≤250
Soil typical values for Al (ATSDR, 2008[a])	7000-100,000	–	–	–	–	–	–	–	–
Soil typical values for Fe (Jankiewicz, Ptaszyński and Turek, 2002)	–	–	–	–	2,000-48,000	–	–	–	–

Table 4.2: Total heavy metal content of new bricks, old bricks, gravel and limestone.

Table 4.2 shows that concentrations of Cd, Cr, Cu, Ni, Pb and Zn in the test aggregates were well below the ICRCL 59/83 and Kelly indices values. Al and Fe concentrations in all the aggregate samples were either below or within typical soil values for Al (0.7% - 10% or 7,000 $mgkg^{-1}$-100,000 $mgkg^{-1}$ (ATSDR, 2008)) and Fe (0.2% - 4.8% or 2,000 $mgkg^{-1}$ - 48,000 $mgkg^{-1}$ (Jankiewicz, Ptaszyński and Turek, 2002; Payne *et al.*, 2007)). Mn concentrations in the aggregates were compared with the Kelly values only because it had no ICRCL 59/83 specification. Mn concentrations in NB, G and L fell below the Kelly values of ≤500 $mgkg^{-1}$ while OB exceeded this value at ≤562 $mgkg^{-1}$. In order to determine the availability of backround heavy metals present in compost and RA to water and plants, leaching experiments were carried out and will be discussed in the next section

4.2.8 Leaching experiments on growth media and aggregates

Background leaching experiments were carried out to determine the background concentration of mobile heavy metals present in the test samples and the trend of leaching of these heavy metals. This procedure was necessary because bioavailability of heavy metals could affect stormwater and ground water quality in vegetative SuDS, and aquatic life (Dudka and Miller, 1999; Seelsaen *et al*., 2007).

Background concentrations obtained were compared with the WHO drinking water guidelines for heavy metals (WHO, 2011) and toxicity levels for freshwater organisms. In order to test the toxicity of pollutants such as heavy metals in freshwater, fresh water organisms such as water fleas (e.g. *Daphnia spp*.) are mostly used for monitoring water quality (Vesela and Vijverberg 2007; Offem and Ayotunde, 2008). This is because of their sensitivity to poor water conditions (Grosell, Nielsen and Bianchini 2002; Bossuyt and Janssen 2005). The comparison between these two parameters were made because if heavy metal content of leachates were comparable to potable water standards then the leachates would pose no threat to water bodies, groundwater and the aquatic ecosystems they sustain. However in reality, leachates from the test samples would not be used for potable purposes but would be most likely discharged into water courses or reused for domestic purposes.

The graphs below (figures 4.7a-g) show the baseline mobile heavy metals present in the test growth media samples compared to the WHO (2011) heavy metal drinking water guidelines. The graphs only show those heavy metal concentrations that were detectable and where they are omitted, concentrations were below limits of detection. Throughout the five-week experimental period, Al concentrations in leachates were not detectable indicating that Al was held within the matrices of the test samples (Banks, Schwab and Henderson, 2006), also Cd concentrations were below limits of detection for the experimental duration and so results were reported here for Cr, Cu, Fe, Mn, Ni, Pb and Zn only.

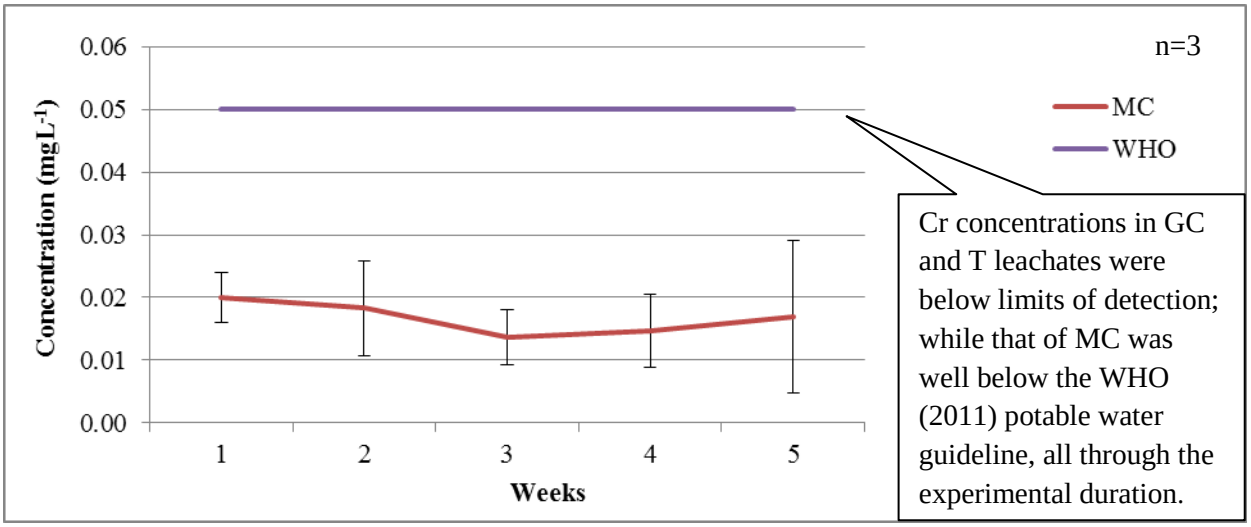


Figure 4.7a: Chromium concentration in leachates with Chromium WHO (2011) potable water guideline.

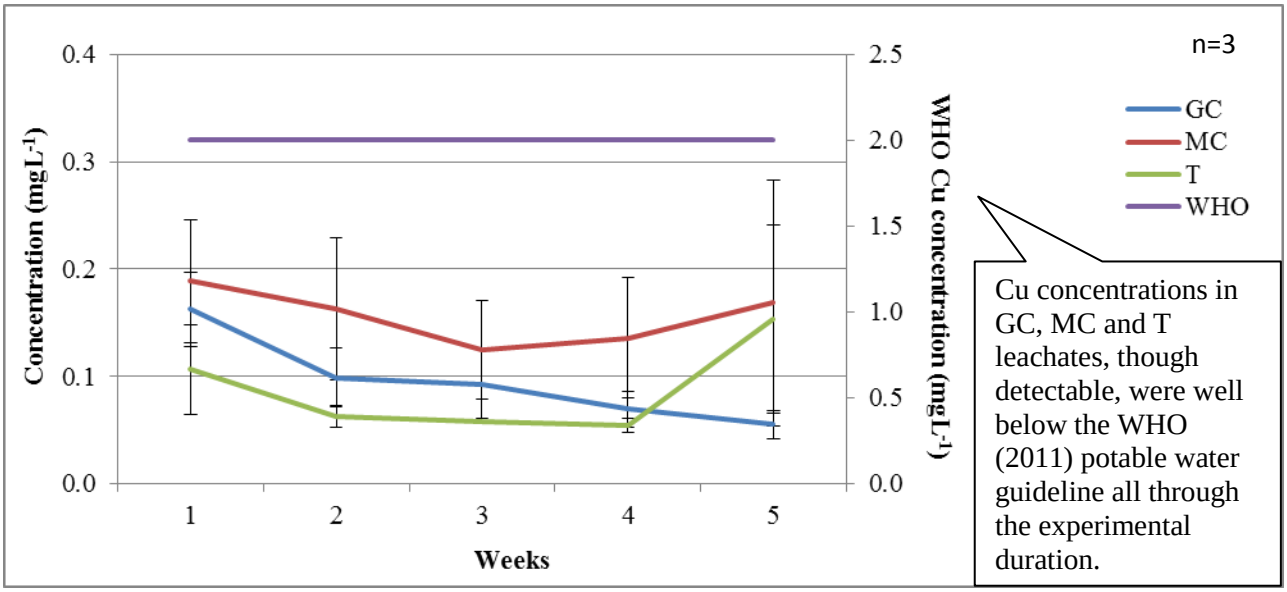


Figure 4.7b: Copper concentration in leachates with Copper WHO (2011) potable water guideline.

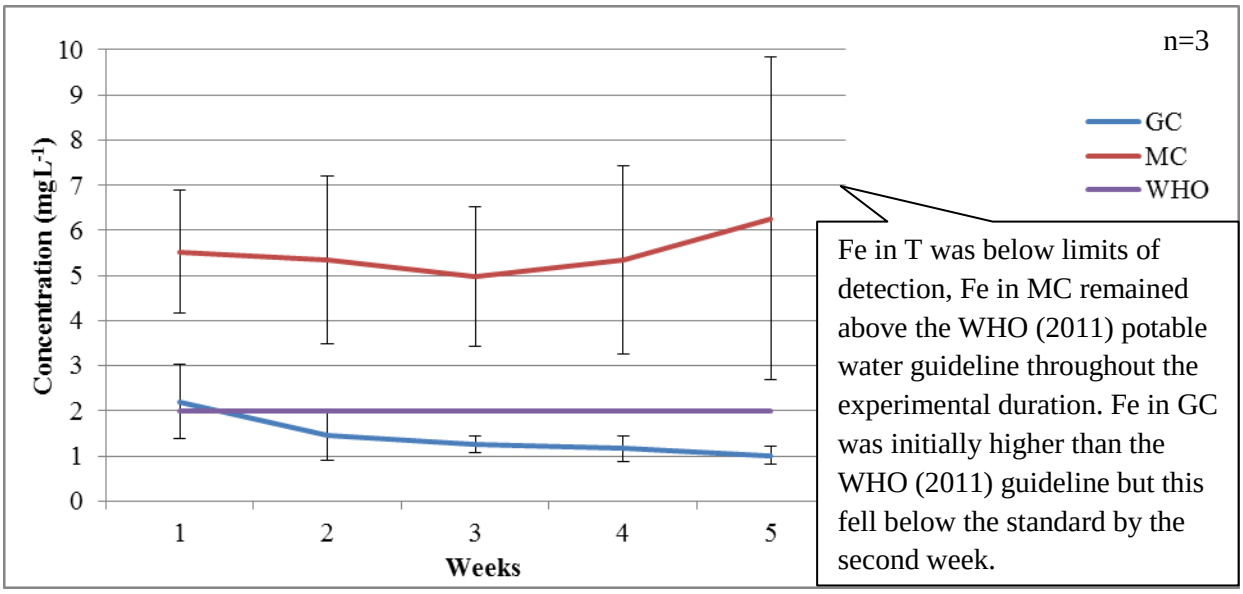


Figure 4.7c: Iron concentration in leachates with Iron WHO (2011) potable water guideline.

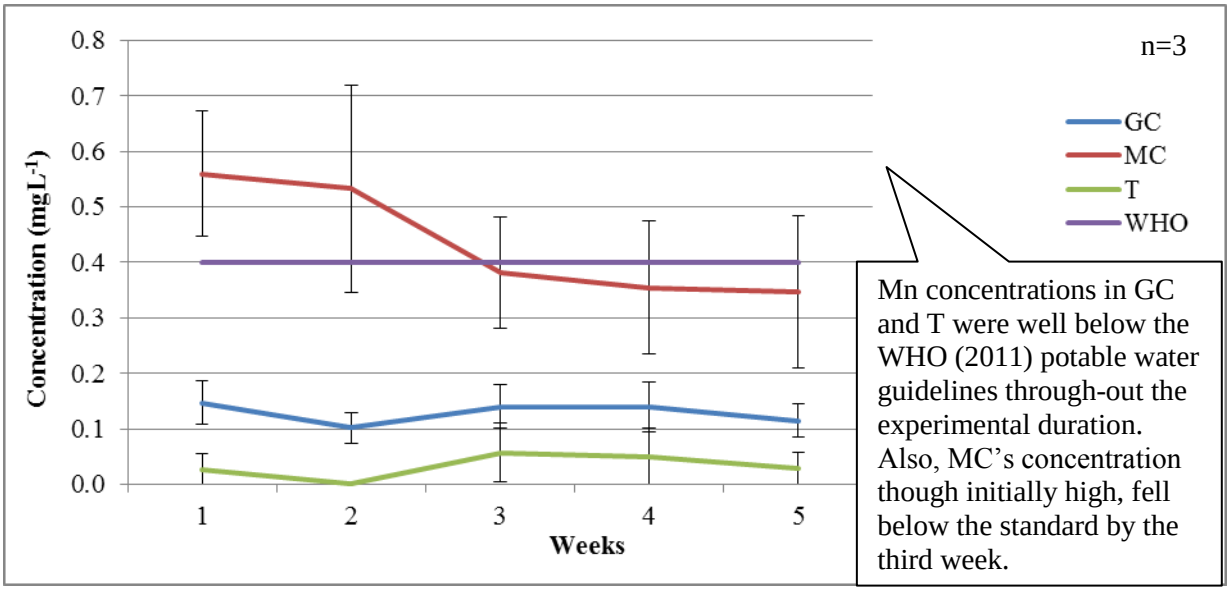


Figure 4.7d: Manganese concentration in leachates with Manganese WHO (2011) potable water guideline.

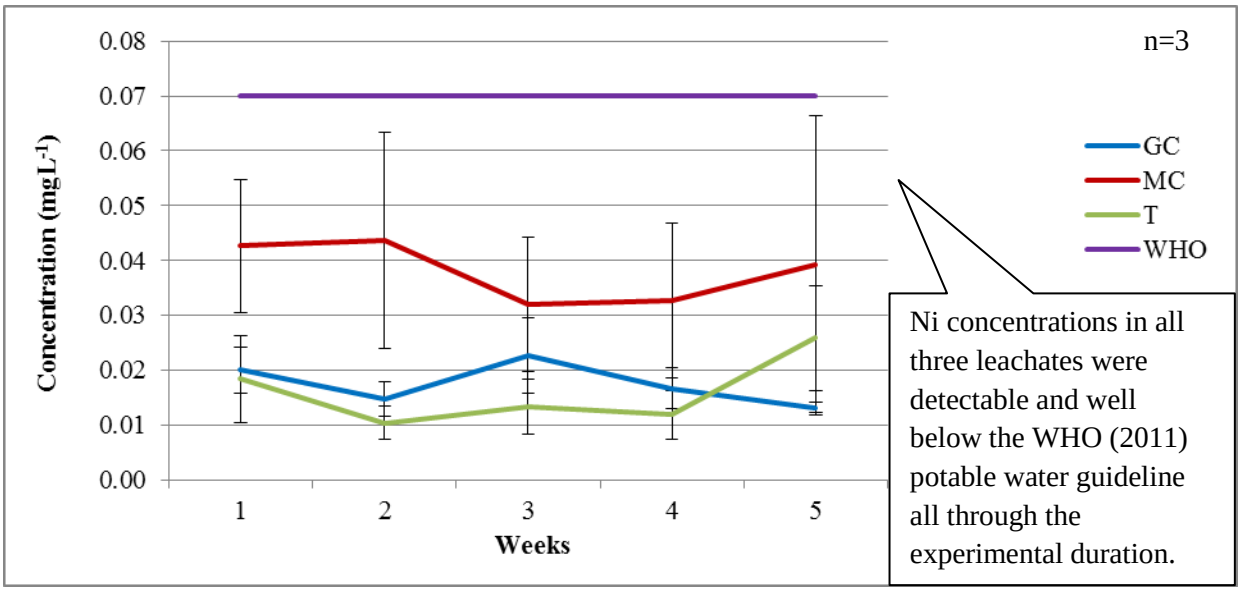


Figure 4.7e: Nickel concentration in leachates with Nickel WHO (2011) potable water guideline.

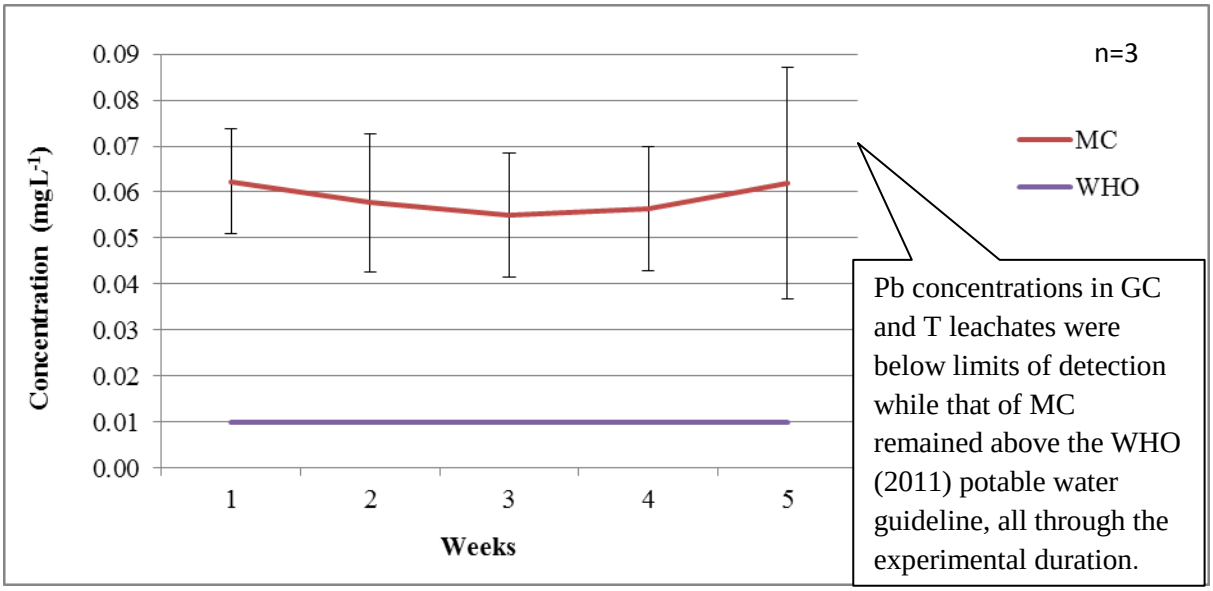


Figure 4.7f: Lead concentration in leachates with Lead WHO (2011) potable water guideline.

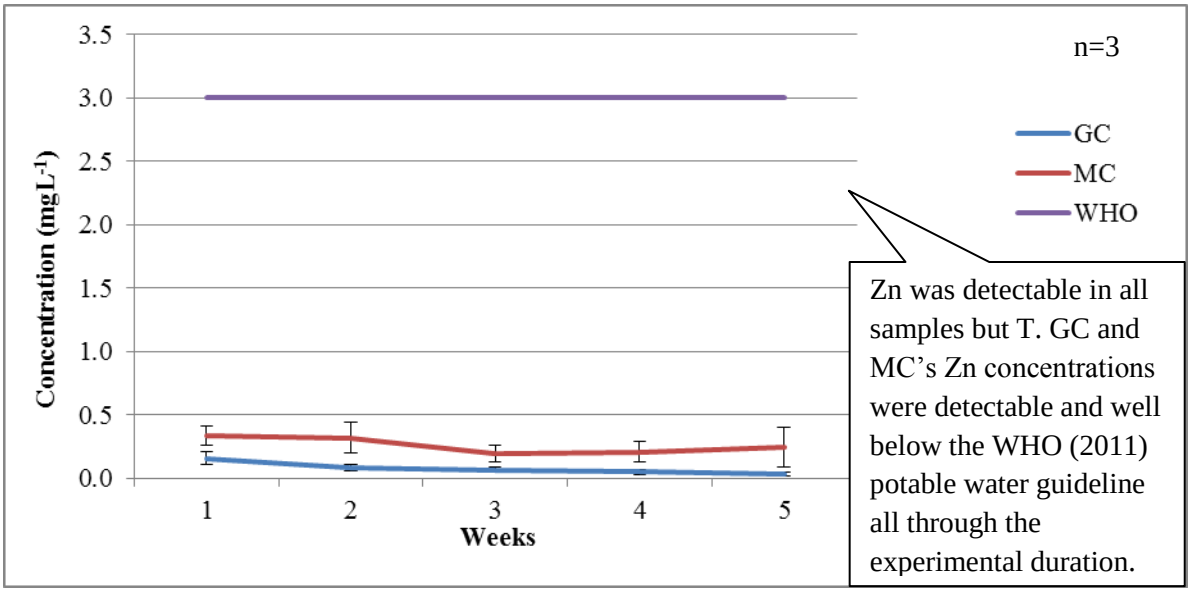
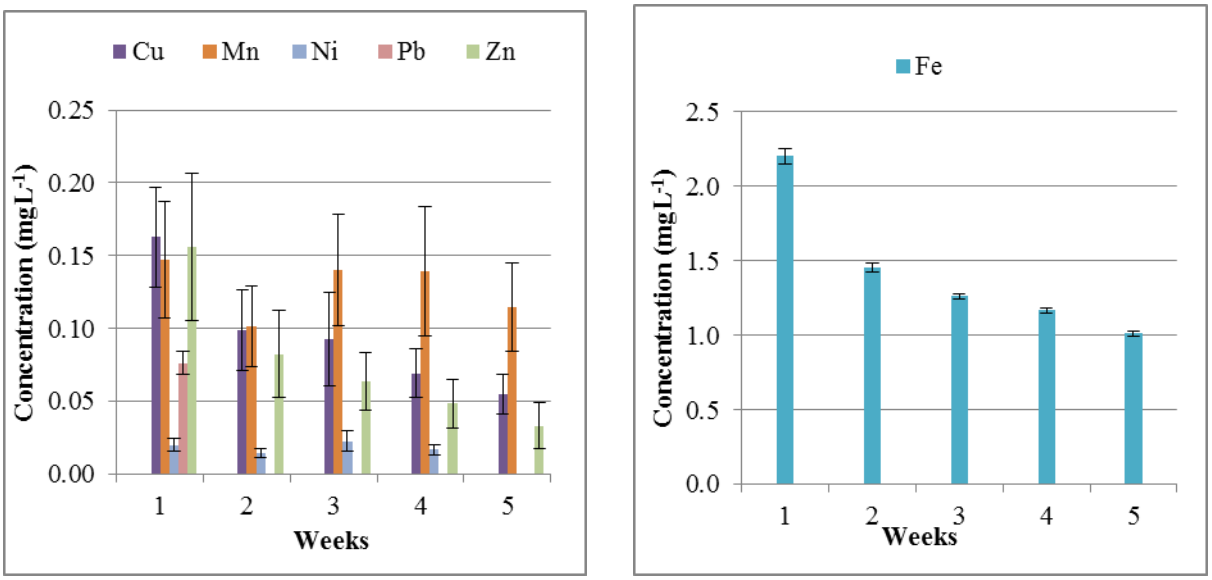


Figure 4.7g: Zinc concentration in leachates with Zinc WHO (2011) potable water guideline.

Overall, the graphs show that while background leachate concentrations of Cr, Cu, Ni and Zn were below the WHO (2011) potable water guidelines for all the test samples, Mn concentrations in MC leachate remained above the standard for two weeks while Fe and Pb concentrations in MC leachates remained well above the WHO (2011) guidelines all through the test period. However, the concentrations of these three metals in MC leachate were much lower than that of toxicity levels for freshwater organisms found in literature, as discussed in section 5.2. It must be noted here that though total Cr concentration in GC and total Pb concentration in T exceeded the PAS 100 standard (BSI, 2011) as shown in table 4.1, both metals remained non-detectable in leachate all through the five–week test period further emphasising the strong adsorption of these heavy metals to organic matter and topsoil particles (Banks, Schwab and Henderson, 2006; Boni and Sbaffoni, 2009).

Apart from observing background heavy metal concentrations in leachates, trends of leaching by the test samples were also observed as shown in figures 4.8a-c below, as the persistent presence of mobile and hence bioavailable heavy metals in leachate is indicative of less sorption of these metals by the test samples (Quevauviller *et al*., 1996; Huang *et al*., 2005). The graphs only show heavy metal concentrations that were above limits of detection and where omitted, the metals were either absent or below limits of detection.

According to figure 4.8ai-ii, six out of the nine heavy metals analysed for (listed in table 3.1) were detected in GC leachate in the first week (i.e. Cu, Mn, Ni, Pb, Zn and Fe) after which they decreased to four (i.e. Cu, Mn, Zn and Fe) by the fifth week. Please note that Fe concentrations had to be plotted on a separate graph because it had higher concentrations, and hence scales, compared to the other heavy metals.

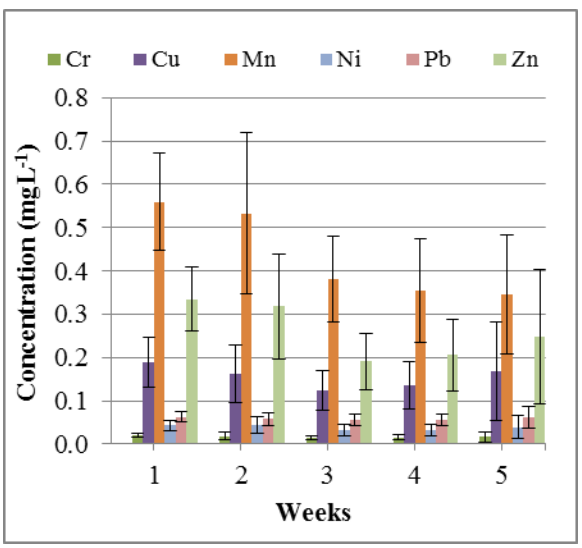


i ii

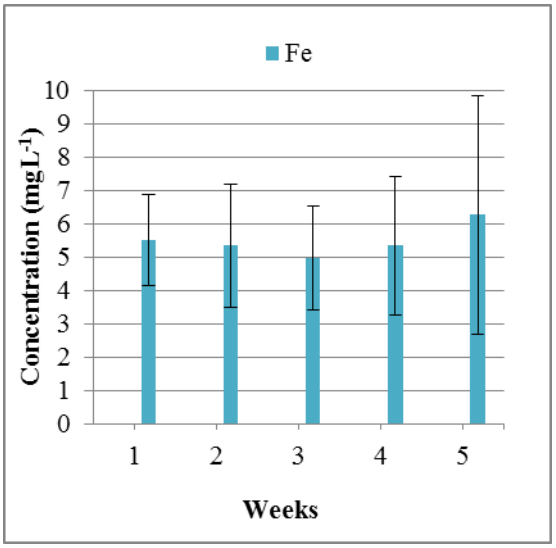
Figure 4.8a: Leaching trend of background heavy metals in green compost.

Of all the heavy metals analysed for in GC leachate, Fe had the highest concentration in leachate all through the experimental period compared to the other heavy metals, with a decrease over time (see figure 4.8aii). Cu and Zn showed a decrease in concentration over time while Mn ad Ni showed no particular trend. Pb concentrations were only detectable in the first week after which concentrations fell below limits of detection. One-way ANOVA analysis showed that the leaching of Fe was significantly higher compared to the other heavy metals in GC leachate (see appendix 7).

As shown in figure 4.8bi-ii, seven out of the nine heavy metals analysed for were detected in MC leachate in the first week and this remained so till the fifth week.



i



ii

Figure 4.8b: Leaching trend of background heavy metals in mixed compost.

Like GC leachates, Fe concentrations were highest in MC leachates compared to the other heavy metals tested for, but unlike GC, Fe concentrations in MC leachate increased with time, decreasing slightly in week 2 and 3 (see figure 4.8bii). This same trend was also observed in Cu and Zn. Mn concentrations decreased with time as mobile ions were depleted, and Cr, Ni and Pb concentrations in leachate remained fairly constant. One-way ANOVA analysis showed that the leaching of Mn, Zn and Fe were significantly higher compared to the other heavy metals in MC leachate (see appendix 7).

Figure 4.8c showed that, five out of the nine heavy metals analysed for were detected in T leachate in the first week and this decreased to three by the fifth week.

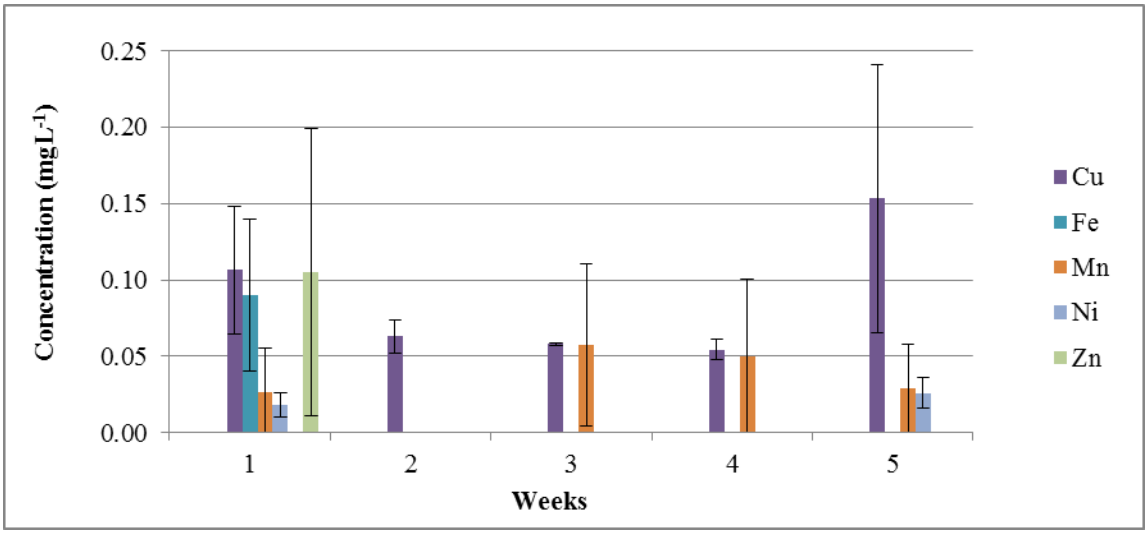


Figure 4.8c: Leaching trend of background heavy metals in Topsoil.

In T, only Cu concentrations in leachate showed a particular trend of decreasing with time, increasing on the fifth week. Fe and Zn were detectable only in the first week of the experiment. Mn leachate concentrations began to decrease from the third week while Ni was detected in the first and last week of the experiment. One-way ANOVA analysis showed that the leaching of Cu was significantly higher compared to the other heavy metals in T leachate (see appendix 7).

Overall, the graphs show that MC followed by GC contained the highest number of mobile heavy metals which persisted in leachate over time, while T contained the least heavy metals in leachate.

Leaching experiments on the RA samples and gravel were also carried out over a period of three weeks and heavy metal concentrations in leachates mostly remained non-detectable except for Mn and Zn in all the samples, which were detected in small concentrations and were well below the WHO (2011) potable water guidelines (see appendix 7).

4.2.9 Microbial enumeration in growth media

Apart from sorption of pollutants by growth media, the presence of microbial populations in vegetative SuDS will also positively influence pollutant biodegradation (Singleton, 1999; Prescott *et al*., 2002) and as such microbial enumeration and identification is necessary. Knowledge of microbial populations and diversity is required to understand and evaluate the function of soil amendments in vegetative SuDS as they are significant to the biodegradation of pollutants in vegetative SuDS (Wackett and Hershberger, 2001; Bradley and Chapelle, 2010). Therefore bacteria and fungi in growth media samples were enumerated and identified. Table 4.3 shows the number of viable bacterial and fungal cells present in 1g of each test growth media sample. Microbial enumeration showed that MC had the highest bacterial and fungal count followed by GC. T had the least number of bacterial and fungal cells.

Samples	Bacterial count *($CFUg^{-1}$)	Fungal count *($CFUg^{-1}$)
Mixed compost	3×10^8	4×10^4
Green compost	4×10^7	8×10^3
Topsoil	7×10^5	1×10^3

*CFU=colony forming units

Table 4.3: Number of viable bacterial and fungal cells in compost samples and topsoil.

4.2.10 Microbial identification in growth media

a) Identification of oil-degrading bacteria

Identification of oil-degrading bacteria was carried out by comparing results from preliminary and biochemical tests with results from previous studies and advice from the laboratory microbiologist. Table 4.4 shows the results of bacterial preliminary and biochemical tests carried out on the test growth media samples for identifying oil-degrading bacteria.

Tests	*Pseudomonas aeruginosa*	*Pseudomonas putida*	*Bacillus cereus*	*Acinetobacter iwoffi*	*Alcaligenes faecalis*
Morphology	• Large, flat, spreading colonies, greenish in colour • Cells were rod-shaped	• Large, flat colonies, florescent green in colour • Cells were rod-shaped	• Large, flat irregular colonies • Cells were rod-shaped	• Large colonies on MacConkey agar and NA • Cells were rod-shaped	• White colonies on MacConkey agar • Cells were rod-shaped
Motility	Motile	Motile	Motile	Non-motile	Motile
Gram staining	–	–	+	–	–
Endospore Formation	–	–	+	–	–
Catalase test	+	+	+	+	+
Oxidase test	+	+	–	–	+
Nitrate reduction test	–	–	–	–	–
Oxidation-fermentation test (Hugh and Leifson test)	Glucose oxidisers	Glucose oxidisers	Both glucose oxidisers and fermenters	No reaction (Neither glucose oxidisers nor fermenters)	No reaction (Neither glucose oxidisers nor fermenters)
Arginine test	+	+	–	–	–
Gelatin liquefaction	+	–	+	–	–
Urease test	–	–	–	–	–

Table 4.4: Results of bacterial preliminary and biochemical tests carried out on compost samples and topsoil.

Results showed that the test growth media samples contained the following bacterial oil-degraders: *Pseudomonas aeruginosa*, *Pseudomonas putida, Bacillus cereus, Acinetobacter iwoffi and Alcaligenes faecalis* (Sorkhoh *et al*., 1985; Adenuga *et al.*, 1992; Andreoni *et al.*, 1998 and Coupe, 2004) as shown in table 4.5.

Oil degrading bacteria	Mixed Compost	Green compost	Topsoil
Pseudomonas aeruginosa	Present	Present	Present
Pseudomonas putida	Present	Absent	Present
Bacillus cereus	Present	Present	Present
Acinetobacter iwoffi	Absent	Absent	Present
Alcaligenes faecalis	Present	Absent	Present

Table 4.5: Occurrence of oil-degrading bacteria in compost samples and topsoil.

Table 4.5 showed that T contained all of the oil-degrading bacteria identified. MC contained all of the oil-degrading bacteria except *Acinetobacter iwoffi* and GC contained the least number of oil-degrading bacteria i.e. *Pseudomonas aeruginosa* and *Bacillus cereus*.

b) Identification of oil-degrading fungi

Results showed that some of the test growth media samples contained the following fungal oil-degraders: *Aspergillus and Penicillium*, and table 4.6 shows the species of oil-degrading fungi identified in the growth media test samples:

Tests	*A. fumigatus*	*A. niger*	*Penicillium spp*
Morphology	Green colonies	Black colonies	Greenish yellow
Microscopy	• Columnar spore heads • Well-developed foot cells	• Round spore heads • Well-developed foot cells	• Vegetative parts are finger-like • No foot cell
Media selection	Czapek-Dox agar	Czapek-Dox agar	Czapek-Dox agar + 20% sucrose

Table 4.6: Results of fungal identification tests carried out on compost samples and topsoil

As seen from table 4.7, MC contained all three fungal oil-degraders followed by GC which contained two of the fungal oil-degraders. T contained no oil-degrading fungi at all.

Oil degrading fungi	Mixed Compost	Green compost	Topsoil
Aspergillus fumigatus	Present	Present	Absent
Aspergillus niger	Present	Absent	Absent
Penicillium	Present	Present	Absent

Table 4.7: Occurrence of oil-degrading fungi in compost samples and topsoil.

Overall, MC contained the highest species of oil degrading microorganisms followed by T and lastly GC.

c) Identification of coliforms

The presence of coliforms especially *Escherichia coli* (an indicator organism for faecal contamination) in compost can compromise the quality of stormwater and groundwater (Ramos *et al*., 2006; WHO, 2008) and hence their identification was carried out to determine the effect of the test samples on water quality in terms of pathogenicity. Assessments were carried out by two methods: viable cell count and presumptive coliform count.

i. Viable coliform cell count

A viable coliform cell count showed that GC contained no viable coliforms but MC and T contained a high number of viable coliform cells as seen in table 4.8. All the test samples contained *E. coli* cells with GC containing the lowest number of viable cells and T containing the, highest, however *E. coli* cells in all the samples were below the PAS 100 standard of 1000CFUg^{-1} (BSI, 2005; BSI, 2011).

Samples	Coliforms ($CFUg^{-1}$)	*E. coli* ($CFUg^{-1}$)
Green compost	-	100
Mixed compost	1200	240
Topsoil	5000	560
PAS 100	-	1000

Table 4.8: Number of viable coliforms and *E. coli* cells present in compost samples and topsoil.

ii. Presumptive coliform test

This test was carried out to estimate the number of coliforms present in effluents derived from the test growth media samples. Table 4.9 shows the number of positive and negative tubes for coliforms after incubating for 18-24 hours at 37^{o}C, including the most probable number (MPN) of coliforms present in 100ml of effluents, with reference to McCrady's probability tables. These probability tables give a direct relation between the number of positive and negative tubes (identified by colour change from purple to yellow, and gas formation), and the number of coliforms present in 100ml of test sample effluents (Cruickshank *et al.,* 1975; Tillett, 1987; Ashbolt, Grabow and Snozzi, 2001). According to table 4.9, MC contained the highest estimated number of coliforms in effluent. GC and T, while containing the same number of coliform cells, had lower MPNs compared to MC.

Quantity of water	50ml	10ml	1ml	MPN/100ml of coliforms present according to McCrady's values
Number of samples of each quantity tested	1	5	5	
Green Compost	0	1	0	1
Mixed Compost	1	1	0	3
Topsoil	0	1	0	1

Table 4.9: Most probable number of coliforms present in compost and topsoil effluents according to McCrady's values.

d) Identification of E. coli

The test known as the differential coliform test was carried out to ascertain the presence of *E. coli* in the samples tested in the presumptive test. Table 4.10 shows the number of tubes producing gas after 24 hours at 44^{o}C including the MPN for *E. coli* present in 100ml of effluent, with reference to McCrady's probability tables. Results showed that T had the highest MPN for *E. coli* followed by MC while GC had the lowest MPN.

Quantity of water	**50ml**	**10ml**	**1ml**	**MPN/100ml of *E. coli* cells present according to McCrady's values**
Number of samples of each quantity tested	1	5	5	
Green Compost	1	4	1	17
Mixed Compost	1	4	2	20
Topsoil	1	5	1	35

Table 4.10: Most probable number of *E. coli* present in compost and topsoil effluent according to McCrady's values.

4.3 Data derived from analysis on test samples

4.3.1 Second objective: Monitoring of microbial activity in growth media

This experiment was carried out to assess microbial activity in simulated swale conditions and to determine which of the test samples will be more versatile in its application in vegetative SuDS under varying conditions, similar to those which occur in vegetative SuDS**.** Figure 4.9 shows microbial respiration in the test samples over a period of nine weeks in low moisture and microaerophilic conditions as described in section 3.4.1. As microbial respiration is directly associated with microbial activity (Pratt *et al.*, 1999; Puehmeier *et al*., 2005; Coupe *et al.*, 2006[a]), both terms will be used interchangeably for this study.

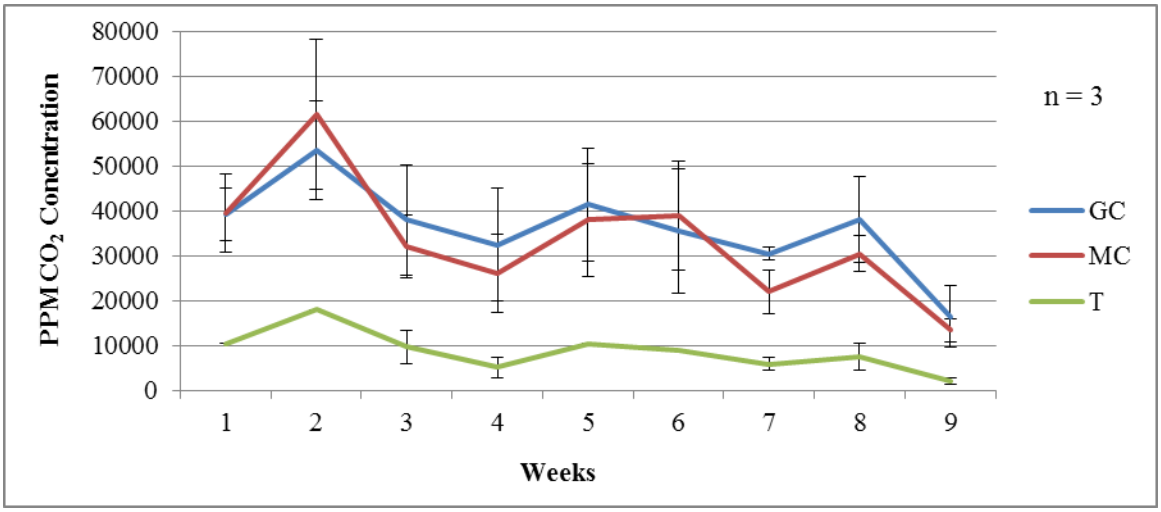


Figure 4.9: Microbial respiration in mixed and green compost and topsoil under low moisture and restricted oxygen conditions.

Results showed a gradual decline in microbial respiration and hence microbial activity over the nine-week period as oxygen and moisture were depleted by microorganisms. Maximum activity was achieved in the 2nd week for all samples with GC having the highest activity and T having the lowest activity at the end of the experimental period. Overall, there was no significant difference in microbial activity between GC and MC as shown by the overlapping error bars and confirmed by one-way ANOVA test (see appendix 8). Therefore, microbial activity in GC and MC were similar and both significantly higher in these conditions than T, which was significantly the lowest all through the nine-week test period.

Figure 4.10 below shows microbial respiration in the test samples over a period of nine weeks in the absence of light and presence of oxygen and moisture, simulating aerobic, wet conditions in regions within vegetative SuDS devices that are not accessible to light such as the sub-base.

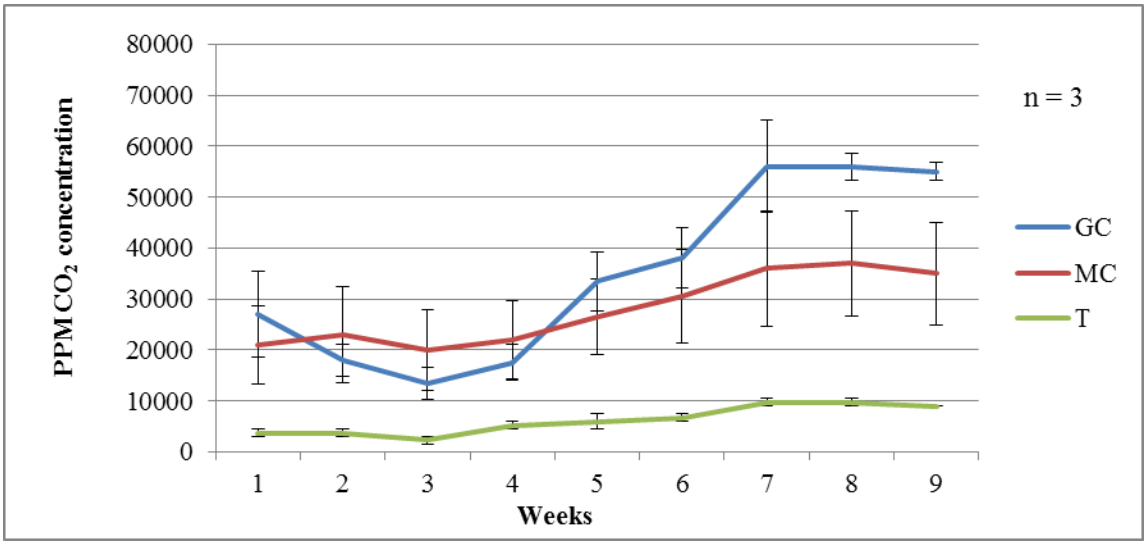


Figure 4.10: Microbial respiration in mixed and green compost and topsoil in the absence of light, aerobic, wet conditions.

The graph shows that microbial activity increased gradually peaking at the 7th week for all test samples after which it decreased slightly by the 9th week. GC had the highest microbial activity at the end of nine weeks, followed by MC. Microbial activity in T was significantly low all through the nine weeks compared to GC and MC. Just as in low moisture and microaerophilic conditions, there was no significant difference in microbial activities between GC and MC as shown by one-way ANOVA testing (see appendix 8).

Figure 4.11 below shows microbial respiration in the test samples over a period of nine weeks in the presence of light, oxygen and moisture simulating aerobic, wet conditions in regions accessible to light within vegetative SuDS devices such as the top part of the vegetative layer.

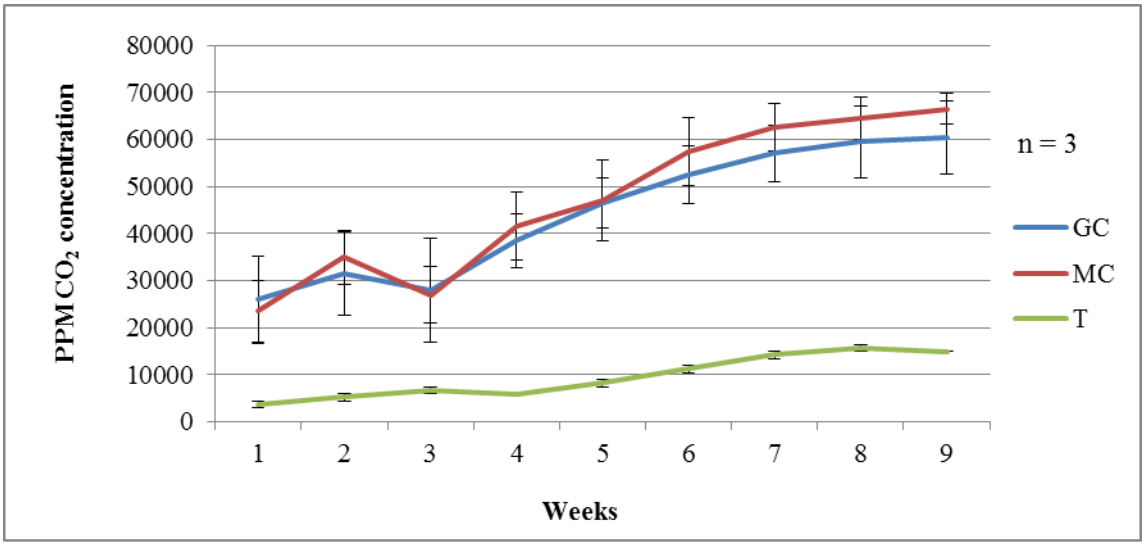


Figure 4.11: Microbial respiration in mixed and green compost and topsoil in the presence of light, aerobic, wet conditions.

Microbial activity was highest in MC closely followed by GC, with T being the lowest. Microbial respiration, and hence activity in GC and MC, showed no significant difference during the nine-week test period (see one-way ANOVA test in appendix 8), however, there was a significant difference in microbial activity in T compared to GC and MC as microbial activity was low all through the nine weeks. It was also observed that microbial activities were more prolific in the light than in the absence of light for all the samples.

After monitoring biofilm development, the test samples were put to work by growing grasses on them to determine their potentials in the production of grass biomass, as this is a necessary component of vegetative SuDS.

4.3.2 Third objective: Grass biomass development

The role of vegetation in vegetative SuDS include attenuation of stormwater, entrapment of particulate pollutants, phytoremediation, carbon sequestration, and provision of amenity and aesthetic value to surrounding landscape (Woods-Ballard *et al.*, 2007). Therefore, plant trials

were carried out to determine grass biomass yield in profiles comprising of the test growth media and aggregates, thereby simulating conditions in real vegetative SuDS.

The following results show the production of grass biomass by GC, MC, GCT (green compost + topsoil), MCT (mixed compost + topsoil) with T as control, each with sub-bases of OB (old bricks), NB (new bricks), L (limestone aggregates) with G (gravel) as control, making a total of twenty test profiles (described in section 3.4.2). Figure 4.12a shows the grass biomass yield of GC each with sub-bases of OB, NB, G and L over five months.

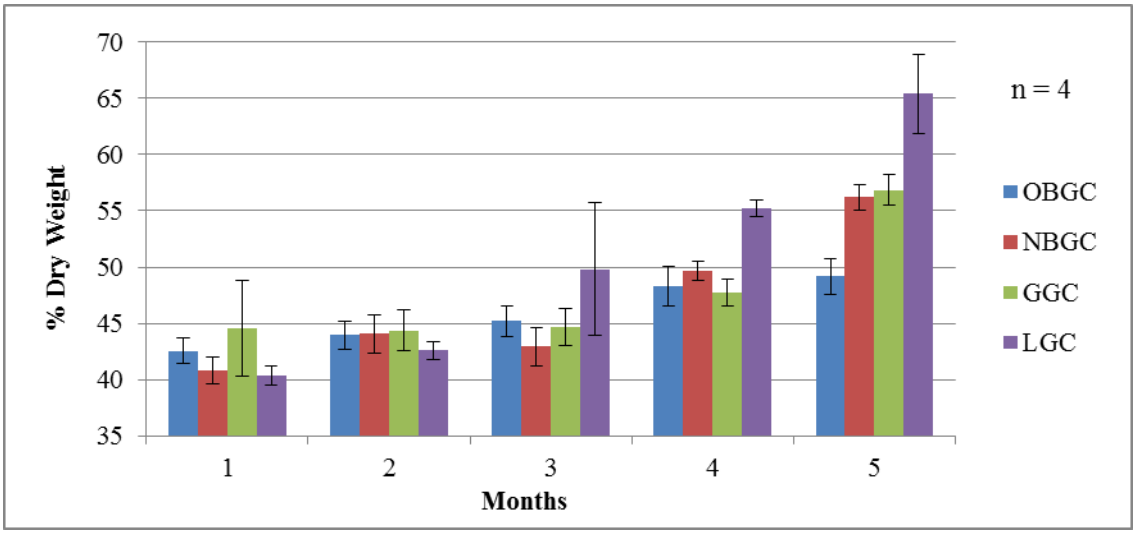


Figure 4.12a: Grass biomass yield obtained from green compost combined with sub-base aggregates of crushed old and new bricks, gravel and recycled limestone aggregates.

On average, grass biomass in GC profiles increased from about 42% in the first month to 57% by the fifth month, with LGC producing the highest biomass and OBGC producing the least biomass by the fifth month. One-way ANOVA testing showed that there was no significant difference in biomass yield in GC profiles over the five month period (see appendix 9).

Figure 4.12b below shows the grass biomass yield of MC with sub-bases of OB, NB, G and L. On average, grass biomass in MC profiles increased from about 48% in the first month to 54% by the fifth month with, LMC producing the highest biomass and OBMC and NBMC

producing the least biomasses by the fifth month. Statistically, there was no significant difference in biomass yield in MC profiles over the five month period (see appendix 10).

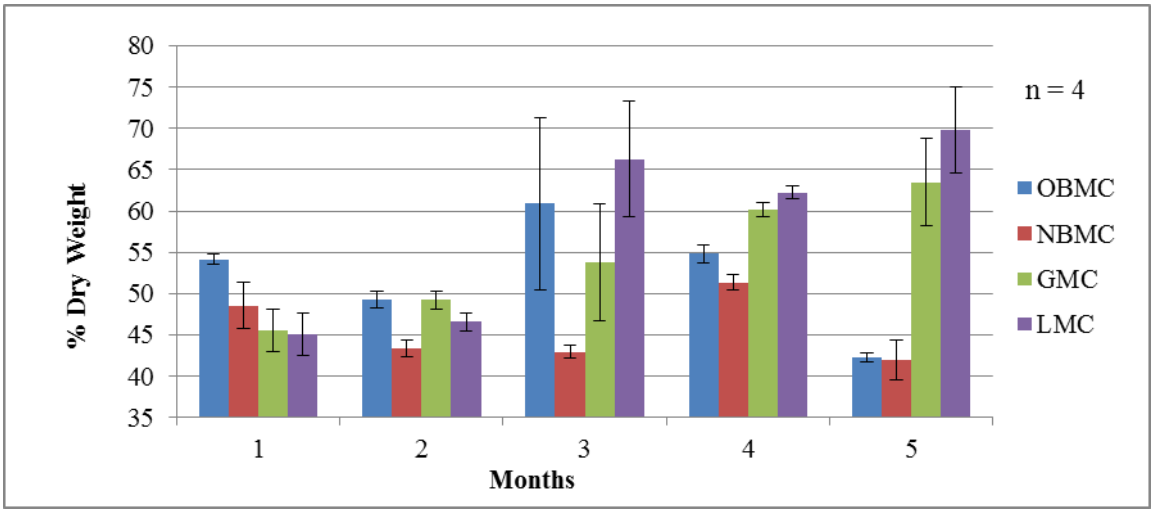


Figure 4.12b: Grass biomass yield obtained from mixed compost combined with sub-base aggregates of crushed old and new bricks, gravel and recycled limestone aggregates.

On average, grass biomass in GCT profiles increased from about 52% in the first month to 59% in the fifth month and as shown in figure 4.12c below, the LGCT combination produced the highest biomass while OBGCT produced the least biomass by the fifth month. There was no significant difference in biomass yield in GCT profiles over the five month period (see appendix 11).

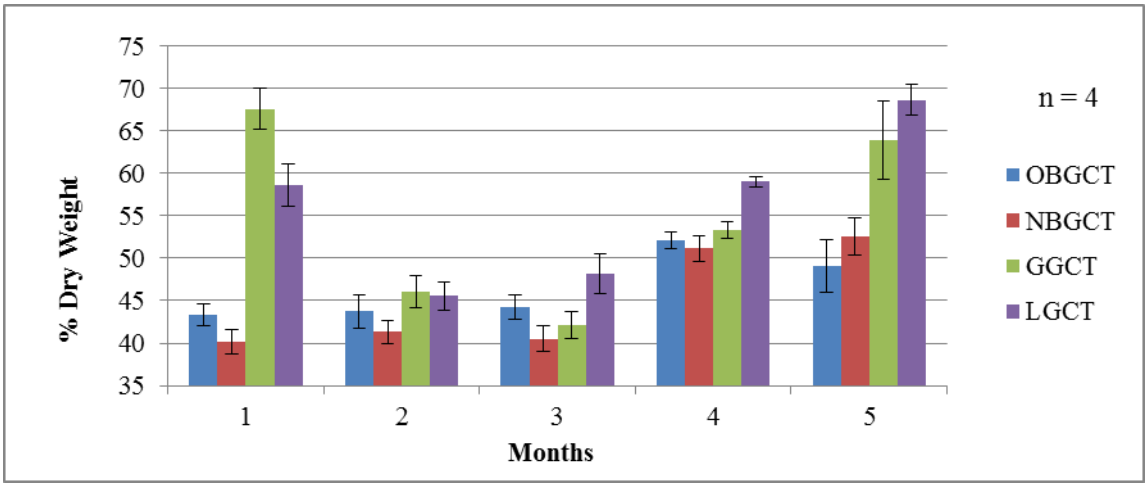


Figure 4.12c: Grass biomass yield obtained from a combination of green compost + topsoil and sub-base aggregates of crushed old and new bricks, gravel and recycled limestone aggregates.

On average, grass biomass increased in MCT profiles from about 47% in the first month to 61% in the fifth month. Figure 4.12d below shows that by the fifth month, LMCT produced the highest biomass while OBMCT produced the least biomass and there was no significant difference in biomass yield in MCT profiles over the five month period (see appendix 12).

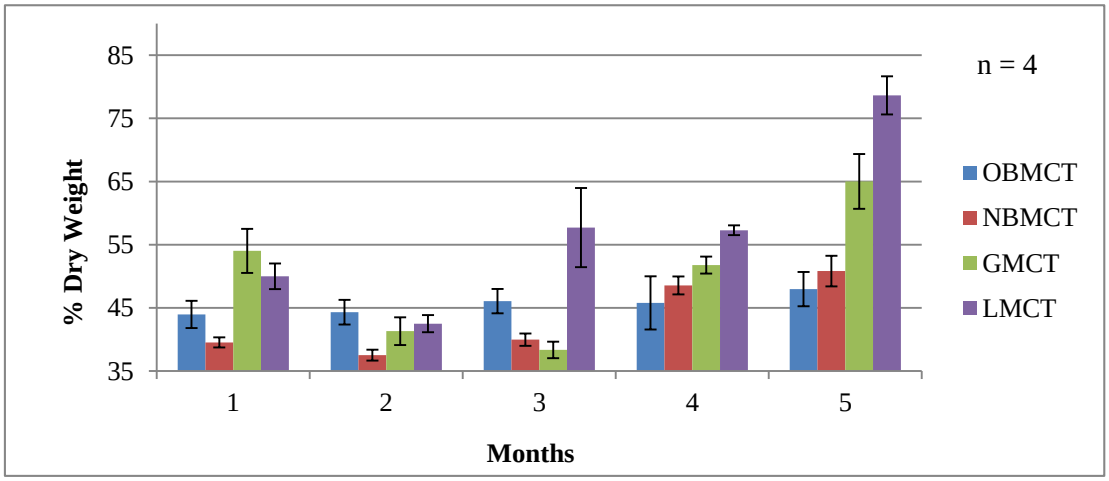


Figure 4.12d: Grass biomass yield obtained from a combination of mixed compost + topsoil and sub-base aggregates of crushed old and new bricks, gravel and recycled limestone aggregates.

On average, grass biomass in T profiles increased from about 55% in the first month to 66% in the fifth month. According to figure 4.12e below, LT produced the highest biomass while NBT produced the least biomass by the fifth month. Statistically, there was no significant difference in biomass yield in T profiles over the five month period (see appendix 13).

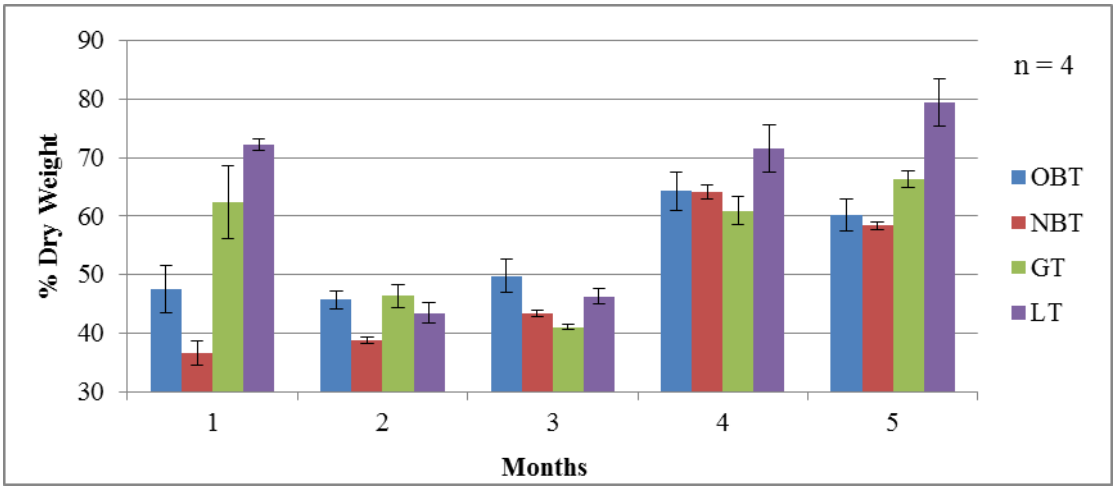


Figure 4.12e: Grass biomass yield obtained from topsoil combined with sub-base aggregates of crushed old and new bricks, gravel and recycled limestone aggregates.

Overall, percentage increase in grass biomass for GC, MC, GCT, MCT and T were 36%, 13%, 14%, 30% and 20% respectively. Also, all growth media containing L consistently produced the highest grass biomass yield especially from the third to the fifth month, with LT and LMCT producing the highest biomass and LGC producing the least biomass (see table 4.11a). In contrast, profiles containing OB predominantly produced the least grass biomass by the fifth month as summarised in table 4.11b. Statistically, profiles containing GC, MC, GCT, MCT and T produced grass biomass consistently each month for five month, with GC and MCT being the highest producers. This implies that any of these growth media would consistently produce the dense vegetation required for treatment of runoff in vegetative SuDS, with GC and MCT being the highest grass producers.

Growth media combination	Highest % Biomass yield
LT	79
LMCT	78
LMC	68
LGCT	68
LGC	65

Table 4.11a: Highest grass biomass yield by test profiles by the fifth month.

Growth media combination	Lowest % Biomass yield
NBT	58
OBGC	49
OBGCT	49
OBMCT	47
OBMC	42
NBMC	42

Table 4.11b: Lowest grass biomass yield by test profiles by the fifth month.

To further determine the effects of test growth media, aggregates and their interaction on grass biomass compared to the controls i.e. T and G, two-way ANOVA analysis was carried out on the total grass biomass yield data for the five-month test period (see appendix 14). Results showed that there were highly significant interactions between growth media and aggregates as highlighted in table 4.12 and therefore the two factors could not be treated independently.

Total biomass for five months	Growth media	Aggregates	Interactions
p-values	.000	.000	**.001**

Where p <0.05 = significant; p<0.01 = very significant; p<0.001 = highly significant

Table 4.12: Results of Two-way ANOVA analysis carried out on total grass biomass data for five months.

A post-hoc test was carried out to further investigate the relationship between growth media and aggregates in grass biomass yield and to identify which factors were significant (see appendix 14). Post-hoc results in table 4.13 showed that for growth media, GC, GCT and MCT differed significantly from T, with GC having the highest significance followed by MCT. For the aggregates, OB and NB differed significantly from G with NB having the highest significance. This result implies that GC, GCT and MCT interacted with aggregates OB and NB to significantly influence grass biomass yield compared to the interactions between G and T, with NBGC having the highest influence on biomass yield.

Post hoc test			
Growth media	p-values	Aggregates	p-values
GC	**.000**	OB	.008
MCT	.001	**NB**	**.000**
GCT	.036	-	-

Where p <0.05 = significant; p<0.01 = very significant; p<0.001 = highly significant

Table 4.13: Results of post-hoc test carried out on total grass biomass data for five months indicating significant biomass development.

Applying these statistical results for OB to results derived from biomass measurements as shown in table 4.11b, it was confirmed that profiles OBGC, OBGCT and OBMCT would significantly produce the least biomass compared to the controls GT. Also applying these results for NB to figures 4.12a, 4.12c and 4.12d, NBGC, NBGCT and NBMCT would also

significantly produce biomass lower than GT, with NBGC producing the lowest biomass. Applying these results to the growth media, percentage biomass increase in GC and MCT (36% and 30% respectively) were significantly higher than that of T (20%). Though profiles producing the highest biomass consisted of L, its biomass production was non-significant and therefore performed just as well as G.

4.3.3 Fourth objective: Monitoring of heavy metals and motor oils in test profiles and their leachates

After assessing the effects of growth media and aggregates on grass biomass yield, the fate of heavy metals, and clean and used motor oils in the test profiles, their components and their leachates were also determined and results were statistically analysed, thereby simulating conditions in real vegetative SuDS. In order to assess the mobility of heavy metals in the test profiles, total heavy metal concentrations were analysed both in plant parts and leachates derived from test profiles and this is discussed below.

4.3.3.1 Monitoring of heavy metal concentrations in leachates from test profiles

As described in section 3.4.2, background heavy metal concentrations of leachates were first measured before introducing the heavy metals (200mg/L) into the test profiles for eight weeks, and analysing their leachate every week for heavy metal concentrations. The results were compared with WHO drinking water guidelines for heavy metals (WHO, 2011) and in cases where concentrations were higher than the specified standard, heavy metal toxicity levels for freshwater organisms were used as explained in section 4.2.8. The figures below show heavy metal concentrations in leachates derived from test profiles for eight weeks, including background concentrations. Profiles that contained no heavy metals or had concentrations below limits of detection in both background and spiked leachates were omitted from the graphs.

Aluminium

As seen in figure 4.13Ai, only Al concentrations in LGCT and GGCT exceeded the Al WHO (2011) potable water guideline of 0.2mg/L in the first background leachate (BK1). In the second background leachate (BK2), Al concentrations in GGCT leachate had fallen below the specified Al standard and only LGCT remained above the standard.

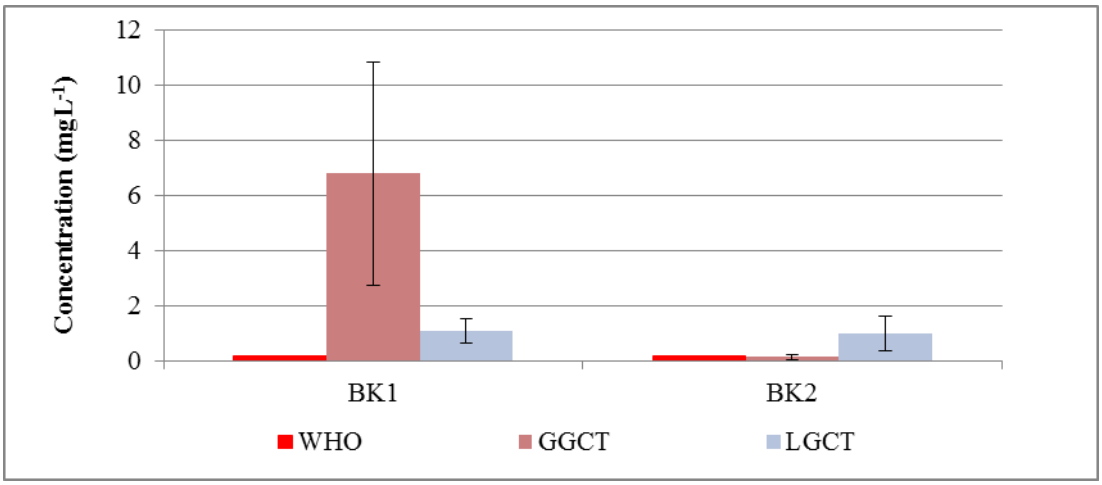
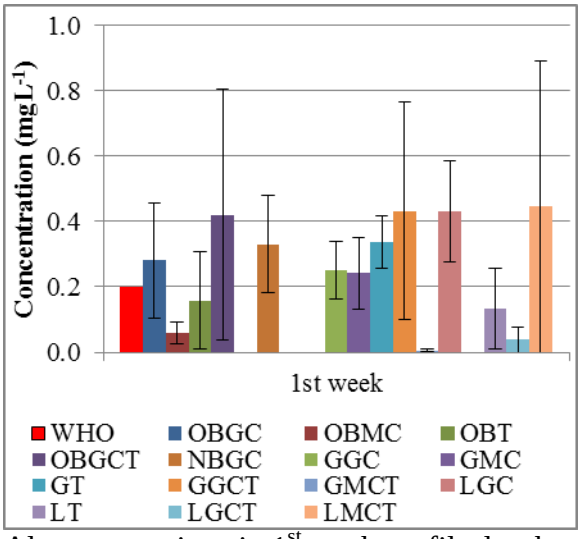
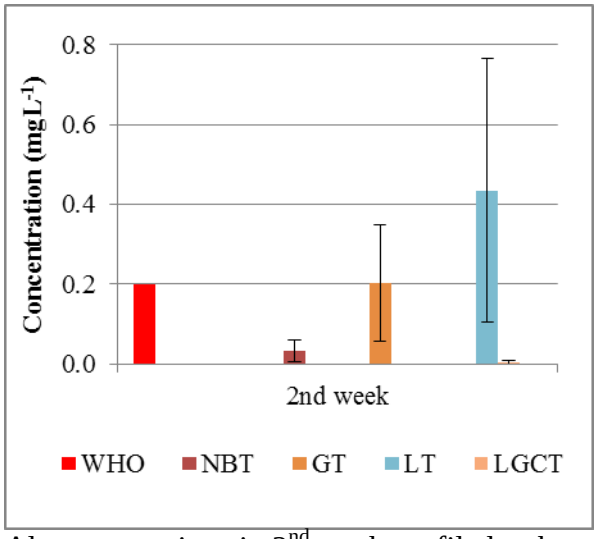


Figure 4.13Ai: Background Aluminium concentrations in leachates obtained from test profiles, compared to the Al WHO (2011) potable water guideline of 0.2mg/L.

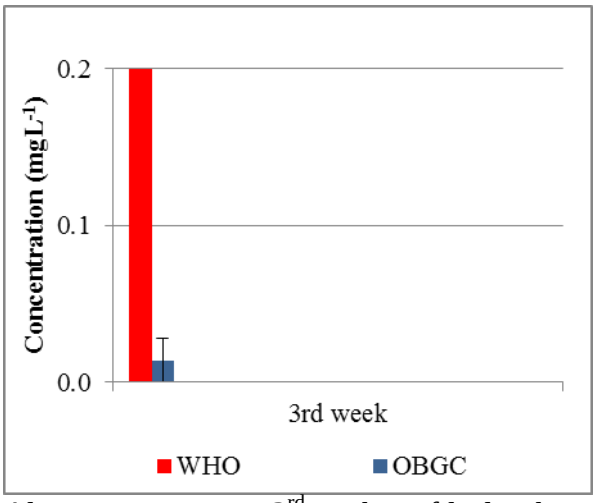
Nine of the twenty profiles produced Al concentrations in leachate that were higher than the Al WHO standard in the first week, most of which were derived from profiles containing GC (figure 4.13Aii). Highest leaching from profiles occurred in the 1st and 6th week (figure 4.13Aii and figure 4.13Avi). Most Al concentrations in the 2nd and 5th weeks were the below limits of detection of 0.002mg/L (Thermo Elemental, 2001) except for LT and OBGC respectively which exceeded the specified WHO (2011) standard (figure 4.13Aiii and figure 4.13Av). Al concentration was below limits of detection in the 3rd week (figure 4.13Aiv) and not detected in 4th and 7th week leachates. By the 8th week, only Al leachates concentrations of OBT and NBT remained above the specified Al WHO standard (figure 4.13Avii).



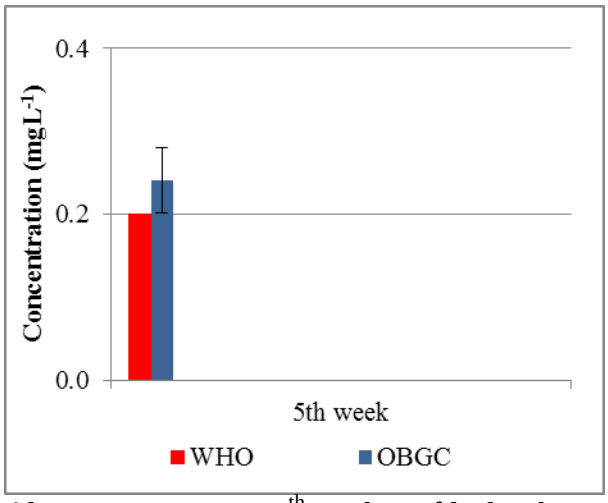
Al concentrations in 1st week profile leachates ii



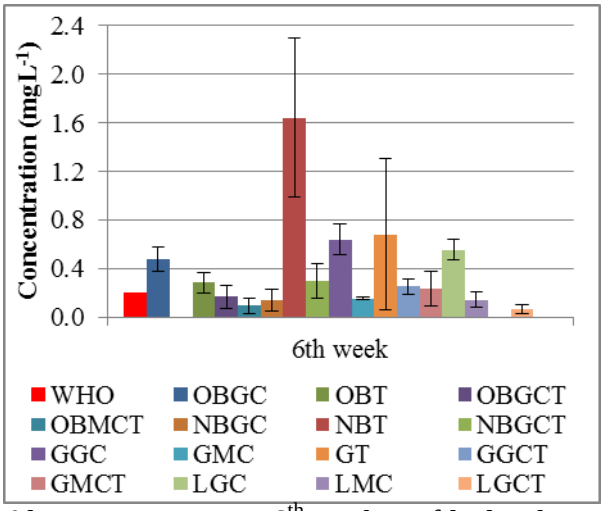
Al concentrations in 2nd week profile leachates iii



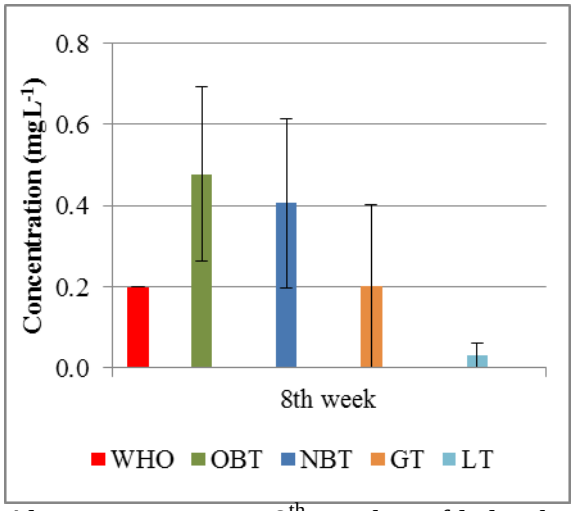
Al concentrations in 3rd week profile leachates iv



Al concentrations in 5th week profile leachates v



Al concentrations in 6th week profile leachates vi



Al concentrations in 8th week profile leachates vii

Figure 4.13Aii-vii: Aluminium concentrations in leachates obtained from test profiles after eight weekly heavy metal additions.

Cadmium

Cadmium was detected in most of the test profile background leachates but concentrations were well below the Cd WHO (2011) potable water guideline of 0.003mg/L (see figure 4.13Bi).

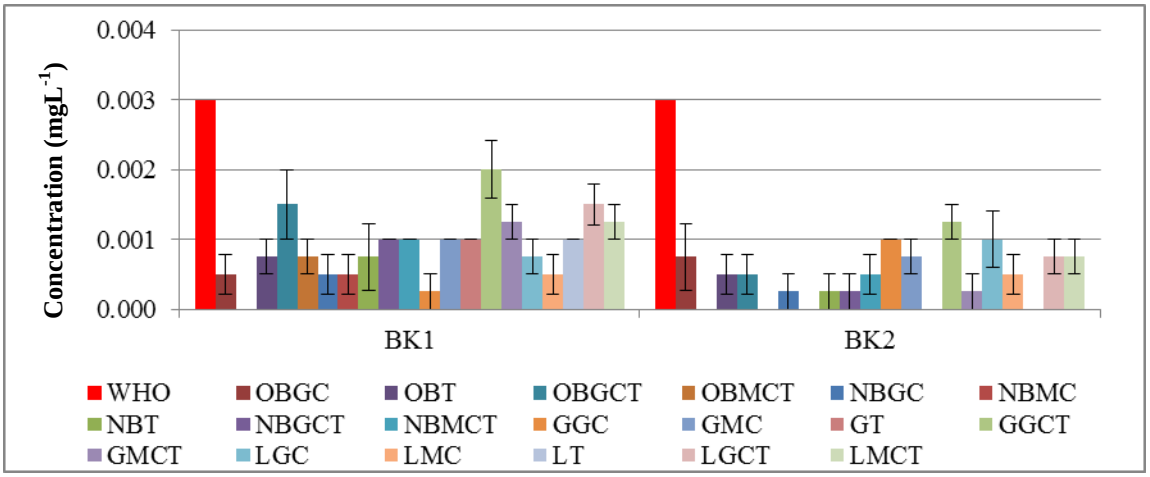
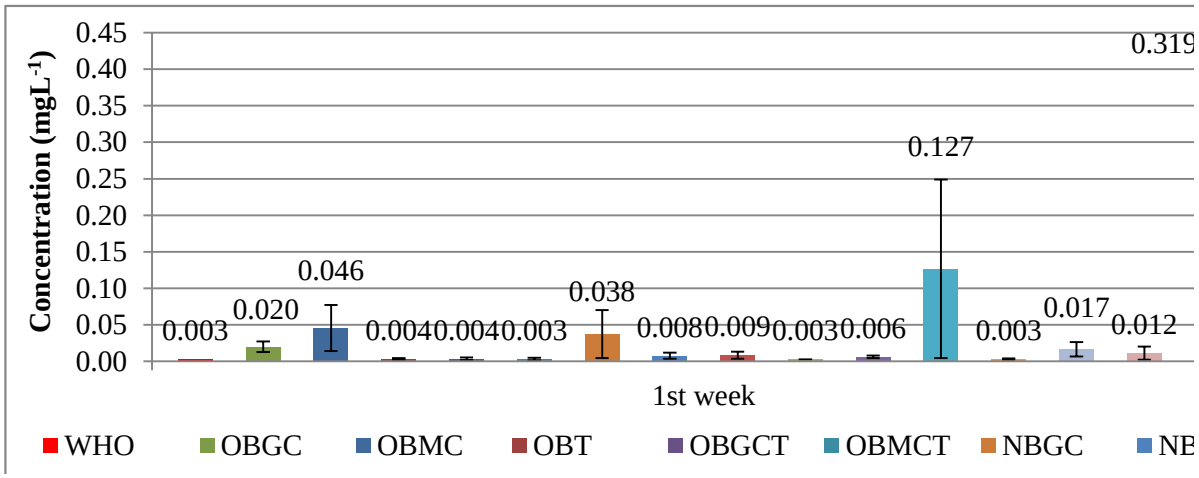


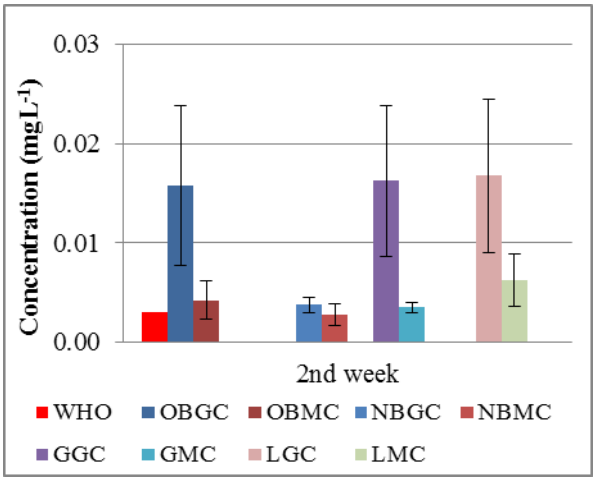
Figure 4.13Bi: Background Cadmium concentrations in leachates obtained from test profiles, compared to the Cd WHO (2011) potable water guideline of 0.003mg/L.

However, after the first heavy metal additions in week 1, Cd concentrations in all the profiles exceeded the Cd WHO (2011) potable water guideline of 0.003mg/L except for NBGCT, GT, LT and LGCT leachates in which Cd was not detected (see figure 4.13Bii). Profiles containing GC had the highest Cd concentrations in leachate. All through the eight weeks of metal additions, OBGC, NBGC, GGC and LGC profiles consistently leached Cd concentrations which were higher than that of the Cd WHO (2011) standard (figure 4.13Biii-ix).



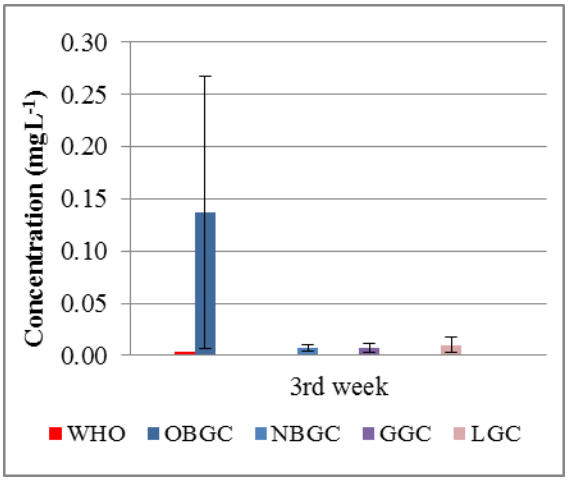
Cd concentrations in 1st week profile leachates

ii



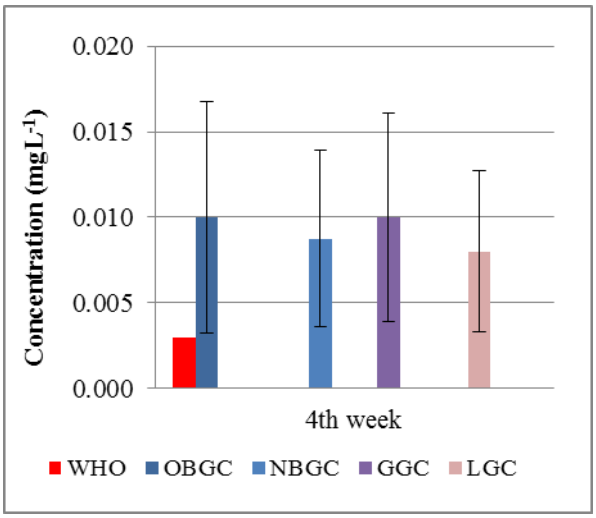
Cd concentrations in 2nd week profile leachates

iii



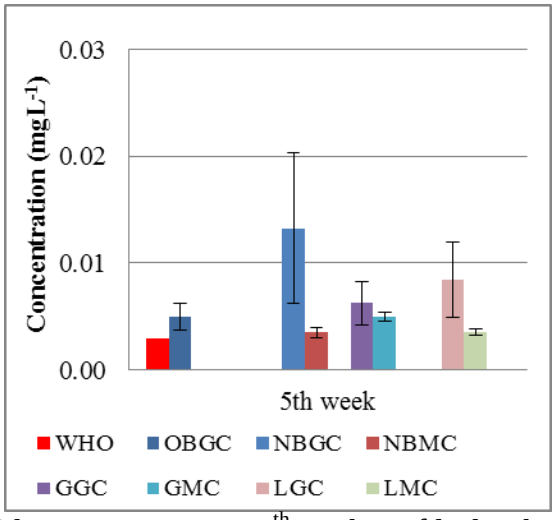
Cd concentrations in 3rd week profile leachates

iv



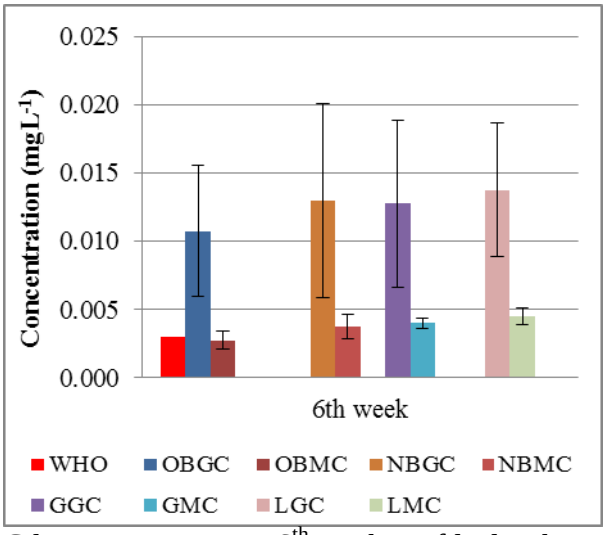
Cd concentrations in 4th week profile leachates

v



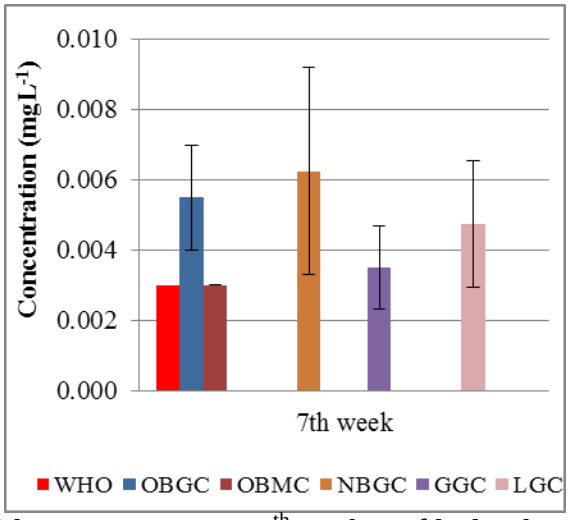
Cd concentrations in 5th week profile leachates

vi



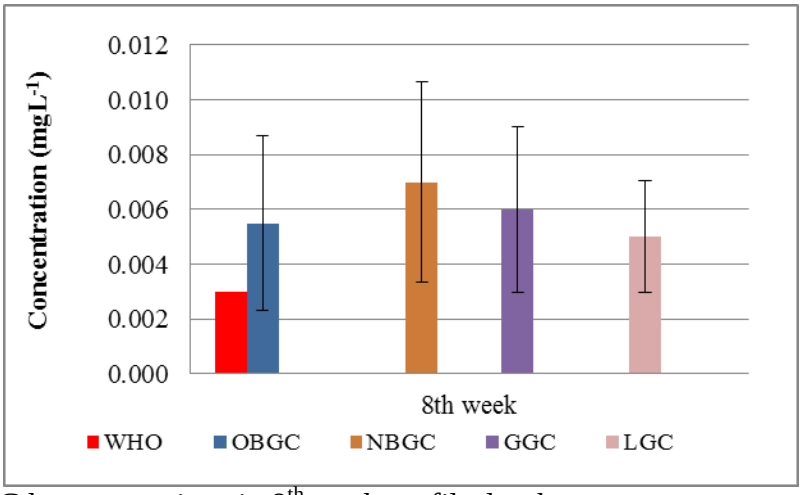
Cd concentrations in 6th week profile leachates

vii



Cd concentrations in 7th week profile leachates

viii



Cd concentrations in 8th week profile leachates ix

Figure 4.13Bii-ix: Cadmium concentrations in leachates obtained from test profiles after eight weekly heavy metal additions.

Chromium

Cr background concentrations were detected in all the test profile leachates but they were well below the specified Cr WHO (2011) potable water guideline of 0.05mg/L (figure 4.13Ci).

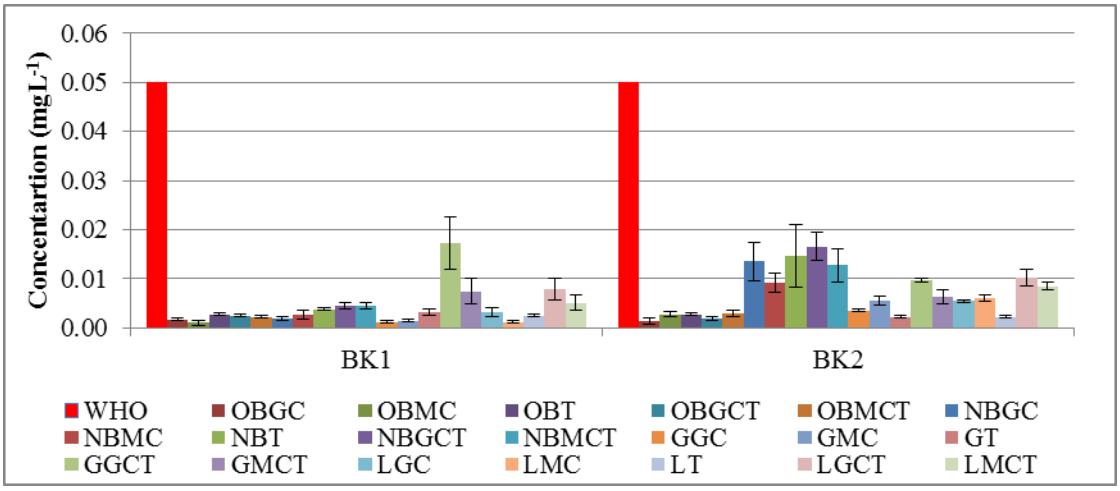
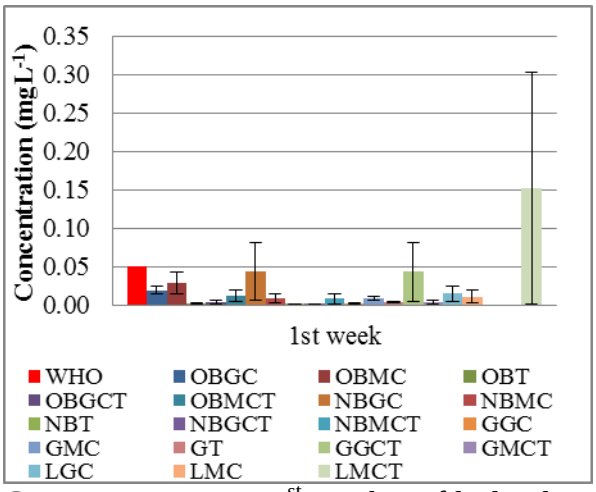


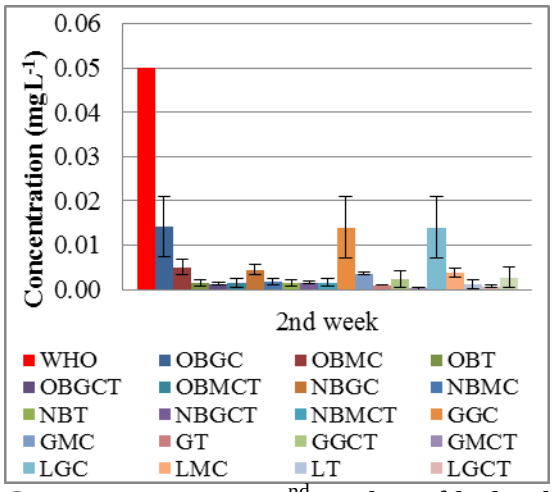
Figure 4.13Ci: Background Chromium concentrations in leachates obtained from test profiles, compared to the Cr WHO (2011) potable water guideline of 0.05mg/L.

During heavy metal additions for the eight-week period, Cr concentrations in all the test profiles fell below the specified standard by the 4th week and remained so till the 8th week (figure 4.13Cii-ix below).



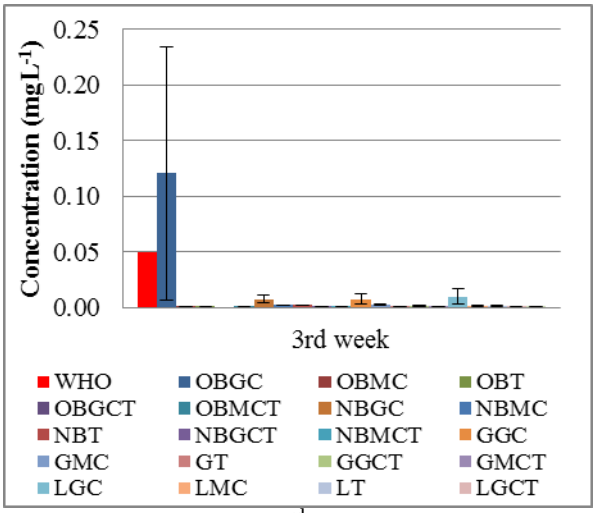
Cr concentrations in 1st week profile leachates

ii



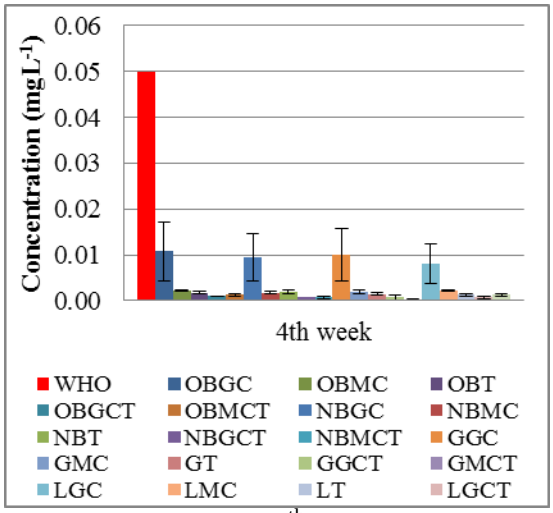
Cr concentrations in 2nd week profile leachates

iii



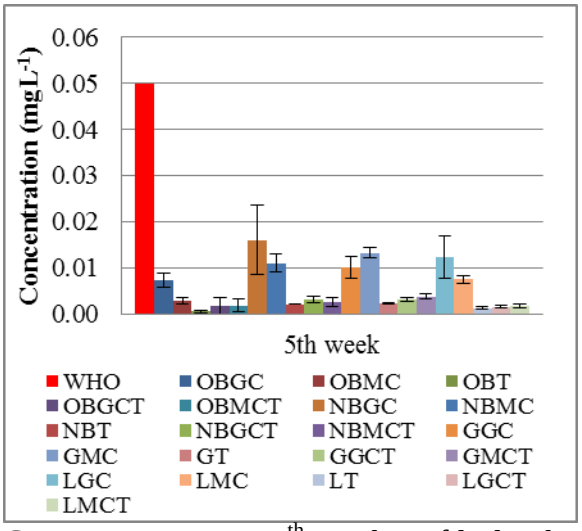
Cr concentrations in 3rd week profile leachates

iv



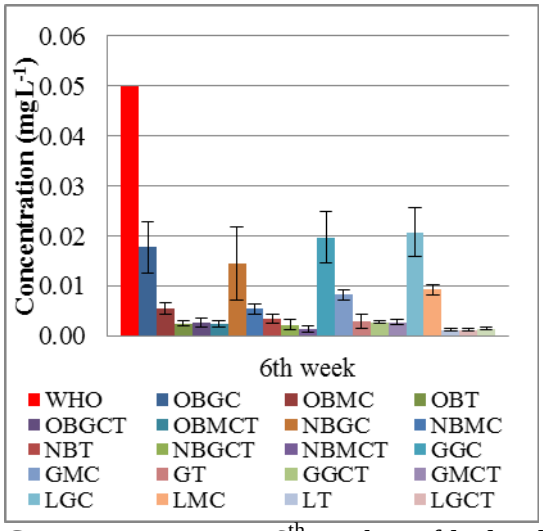
Cr concentrations in 4th week profile leachates

v



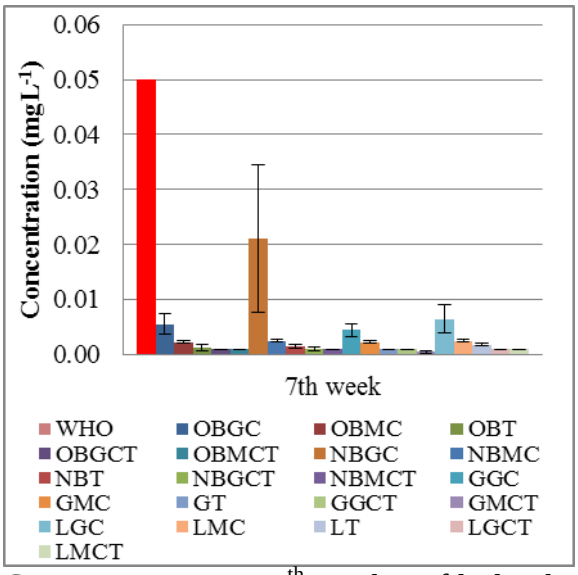
Cr concentrations in 5th week profile leachates

vi



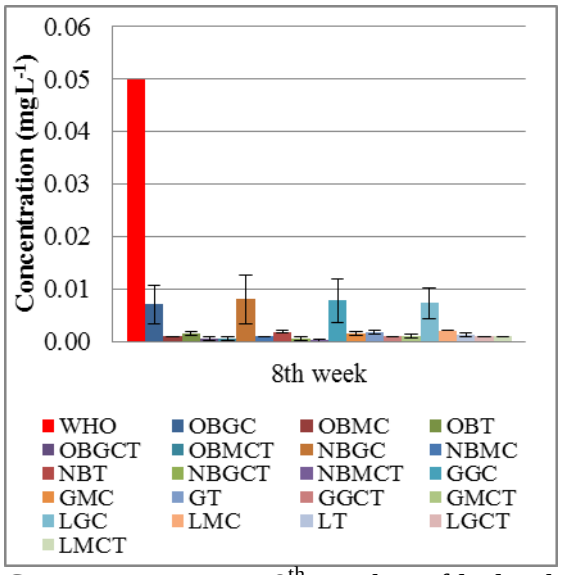
Cr concentrations in 6th week profile leachates

vii



Cr concentrations in 7th week profile leachates

viii



Cr concentrations in 8th week profile leachates

ix

Figure 4.13Cii-ix: Chromium concentrations in leachates obtained from test profiles after eight weekly heavy metal additions.

Copper

Cu concentrations in all background leachate and leachates after heavy metal additions remained well below the WHO (2011) potable water guideline of 2.0mg/L despite eight weeks of metal addition, as seen in figure 4.13Di-ix.

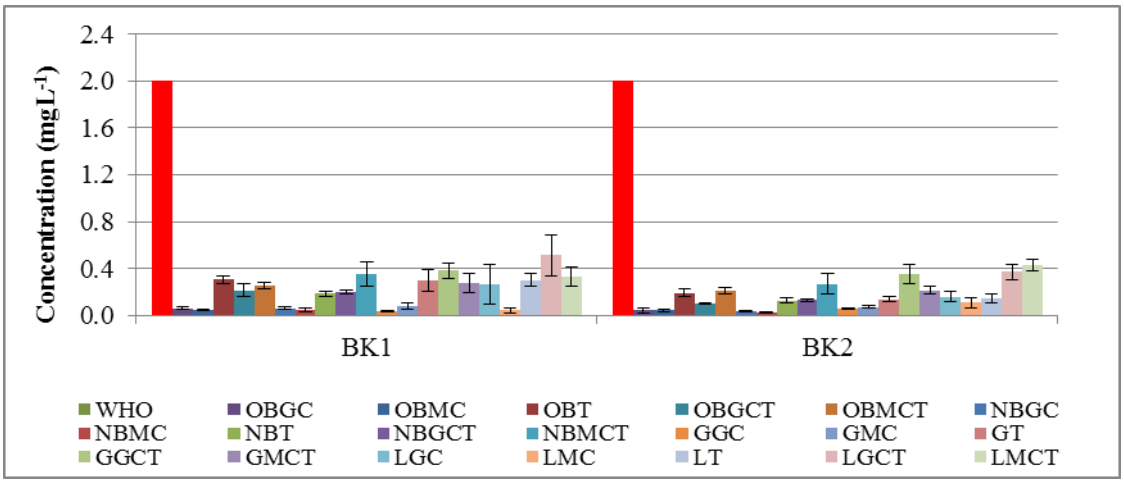
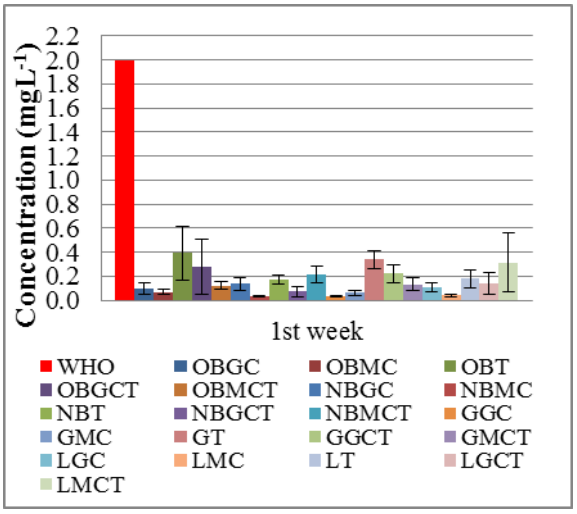
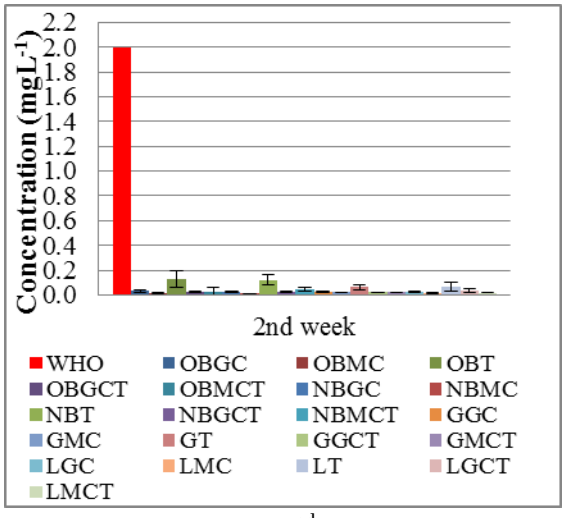


Figure 4.13Di: Background Copper concentrations in leachates obtained from test profiles, compared to the Cu WHO (2011) potable water guideline of 2.0mg/L.



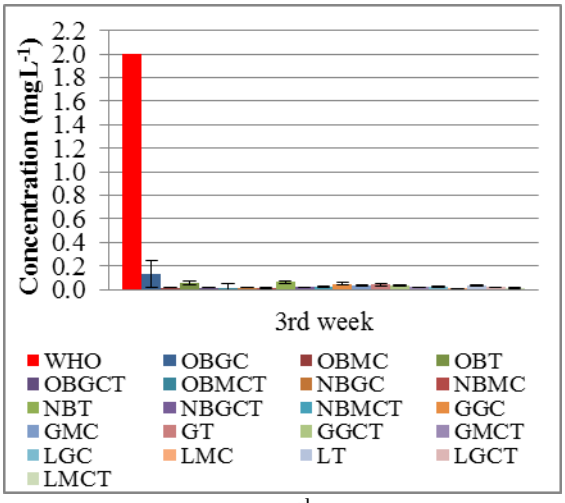
Cu concentrations in 1st week profile leachates

ii

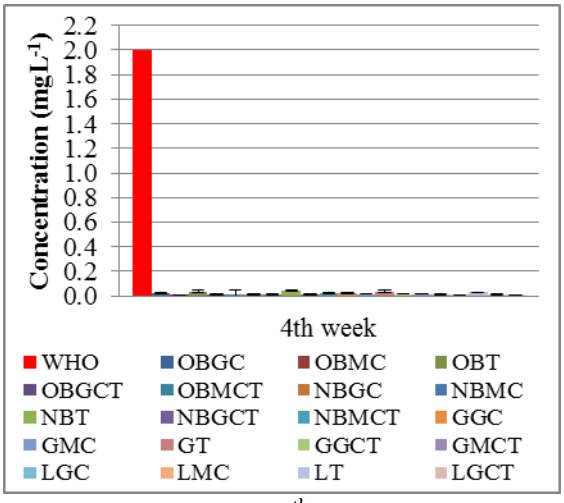


Cu concentrations in 2nd week profile leachates

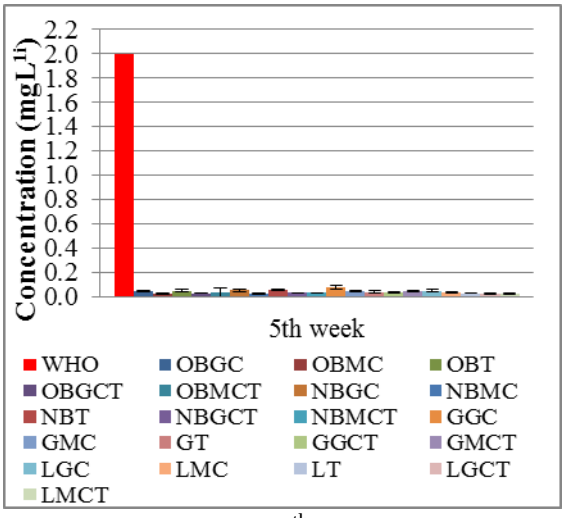
iii



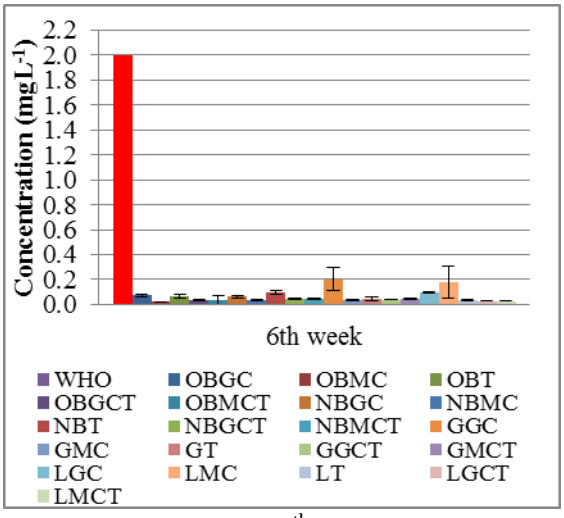
Cu concentrations in 3rd week profile leachates iv



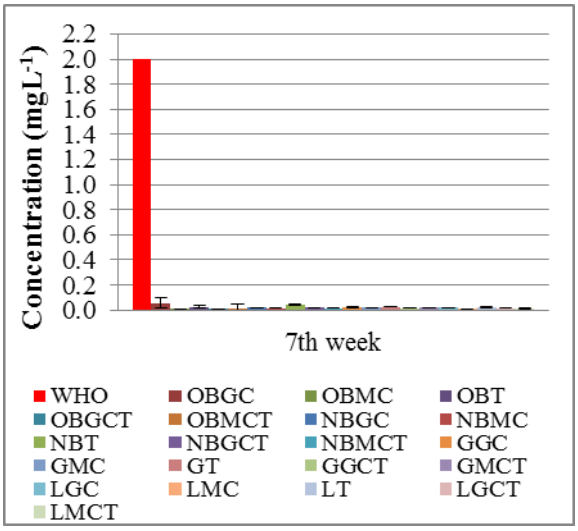
Cu concentrations in 4th week profile leachates v



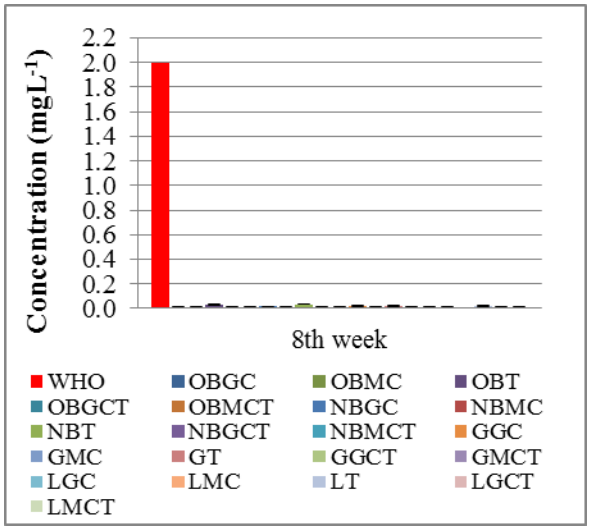
Cu concentrations in 5th week profile leachates vi



Cu concentrations in 6th week profile leachates vii



Cu concentrations in 7th week profile leachates viii



Cu concentrations in 8th week profile leachates ix

Figure 4.13Dii-ix: Copper concentrations in leachates obtained from test profiles after eight weekly heavy metal additions.

Iron

Iron concentrations in all background leachates were detectable but were well below the Fe WHO (2011) potable water guideline of 2.0mg/L, except for GGCT and LGCT (figure 4.13Ei).

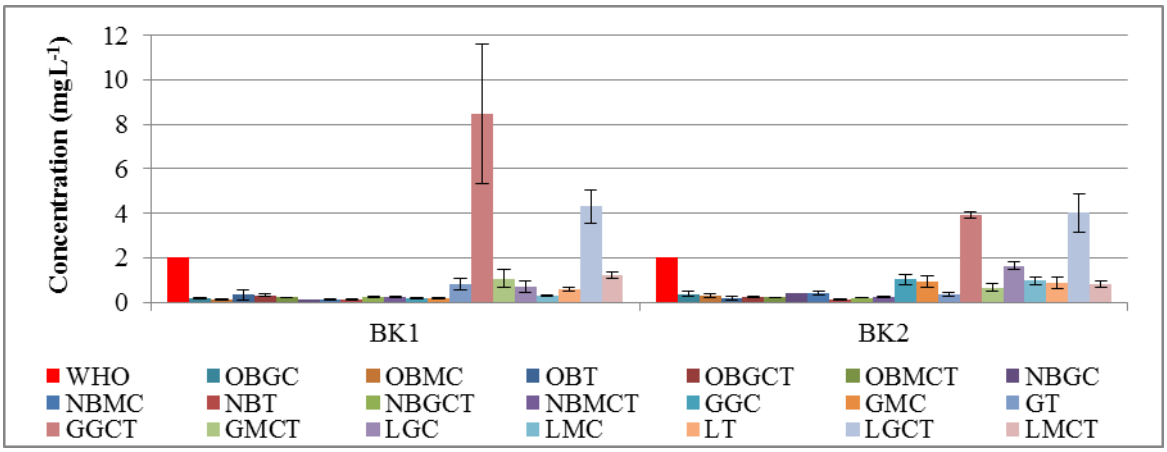
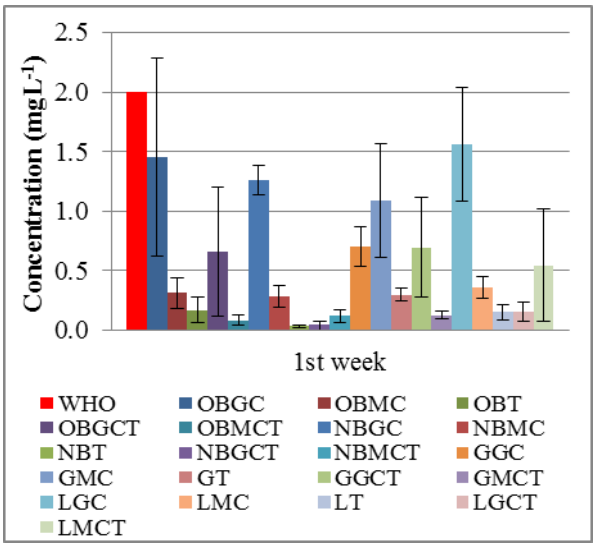


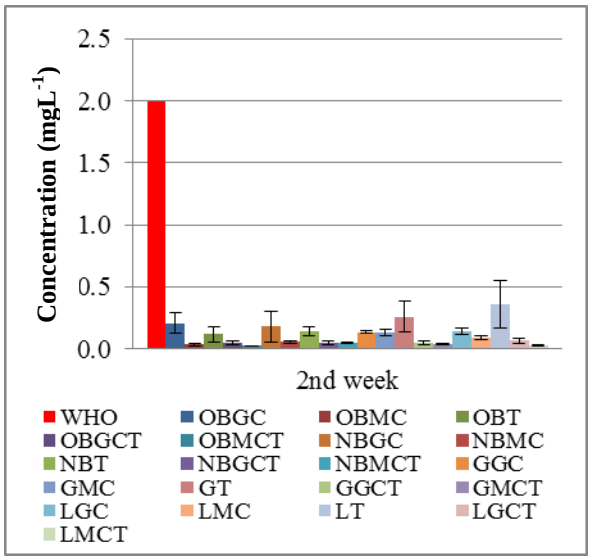
Figure 4.13Ei: Background Iron concentrations in leachates obtained from test profiles, compared to the Fe WHO (2011) potable water guideline of 2.0mg/L.

However by the first week of metal addition, Fe concentrations in GGCT and LGCT had fallen below the specified standard and remained so till the eighth week, as seen in figure 4.13Eii-ix below.



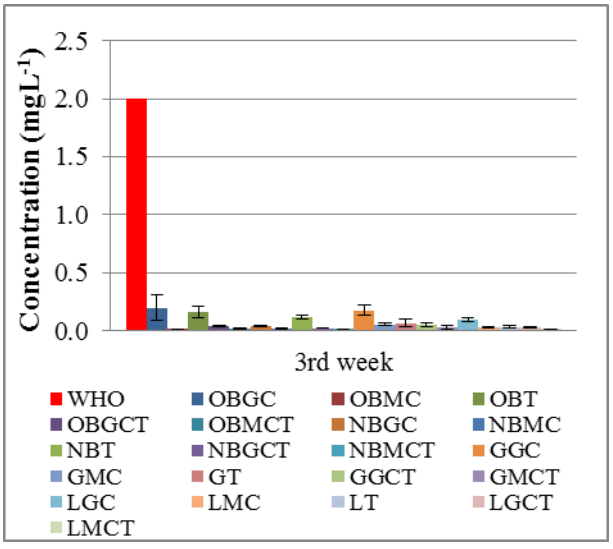
Fe concentrations in 1st week profile leachates

ii



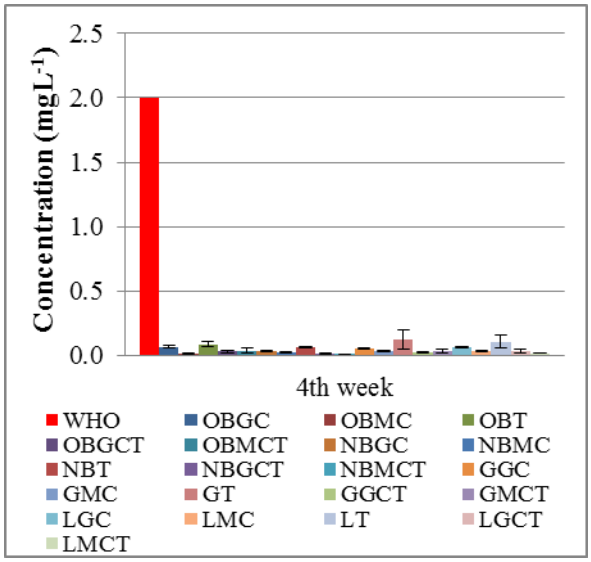
Fe concentrations in 2nd week profile leachates

iii



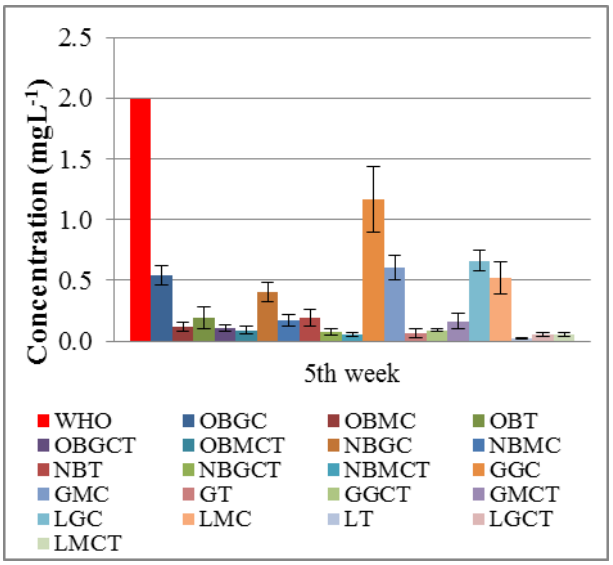
Fe concentrations in 3rd week profile leachates

iv



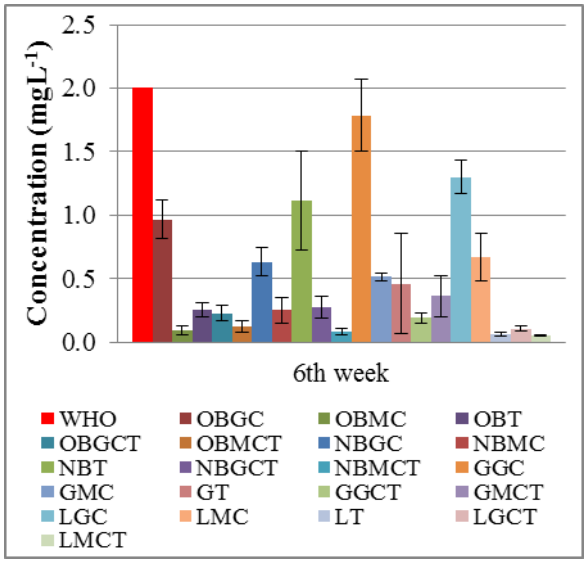
Fe concentrations in 4th week profile leachates

v



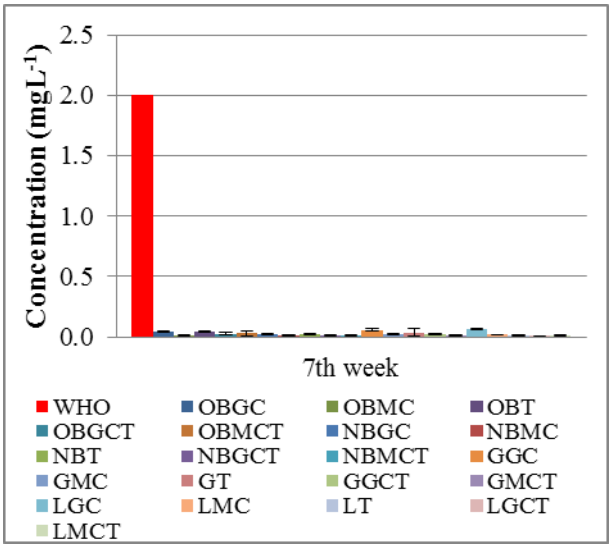
Fe concentrations in 5th week profile leachates

vi



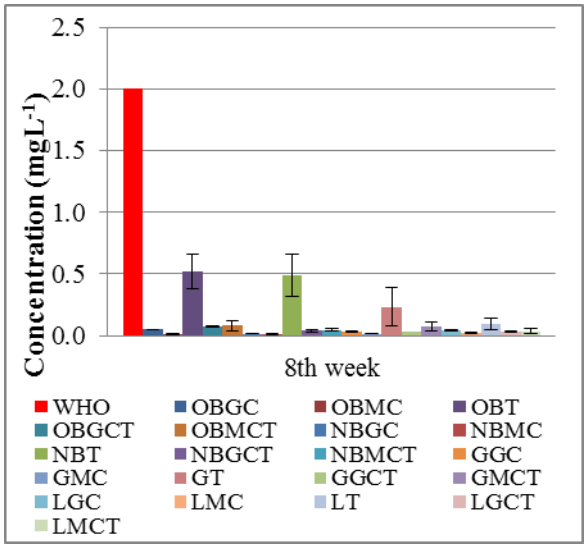
Fe concentrations in 6th week profile leachates

vii



Fe concentrations in 7th week profile leachates

viii



Fe concentrations in 8th week profile leachates

ix

Figure 4.13Eii-ix: Iron concentrations in leachates obtained from test profiles after eight weekly heavy metal additions.

Manganese

Manganese concentrations in background leachate were detectable but below the WHO (2011) potable water guideline of 0.4mg/L as seen in figure 4.13Fi.

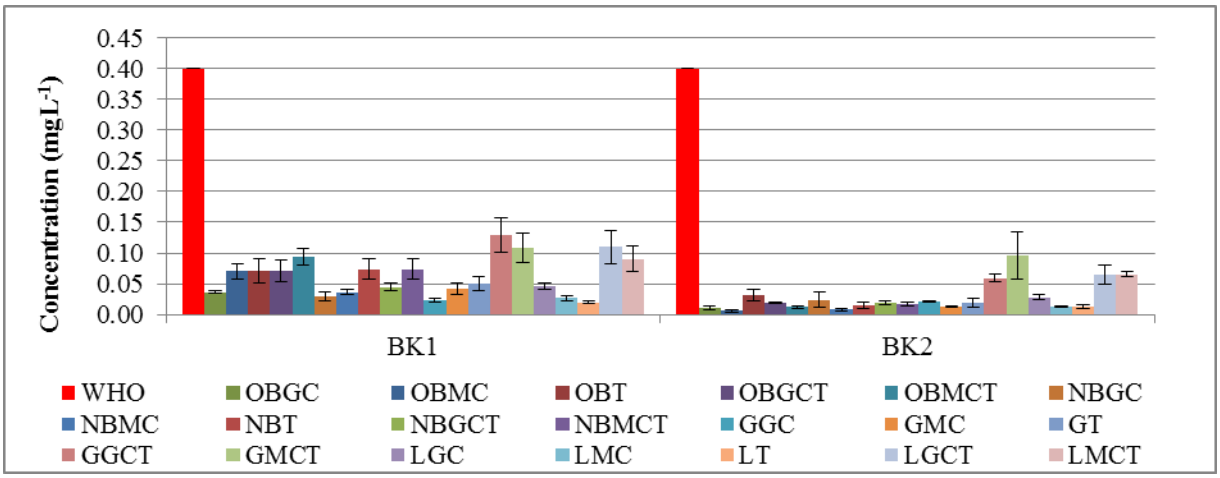
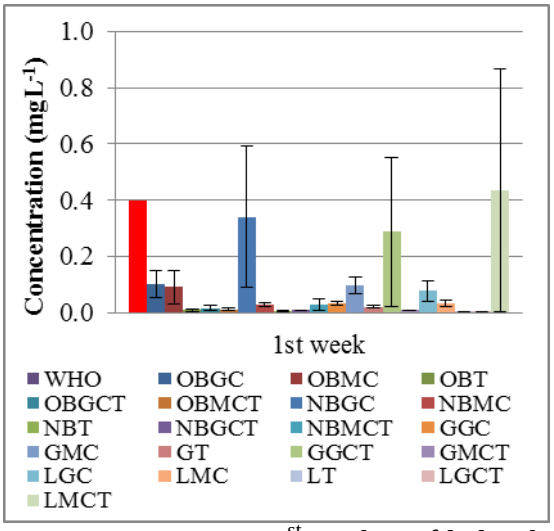


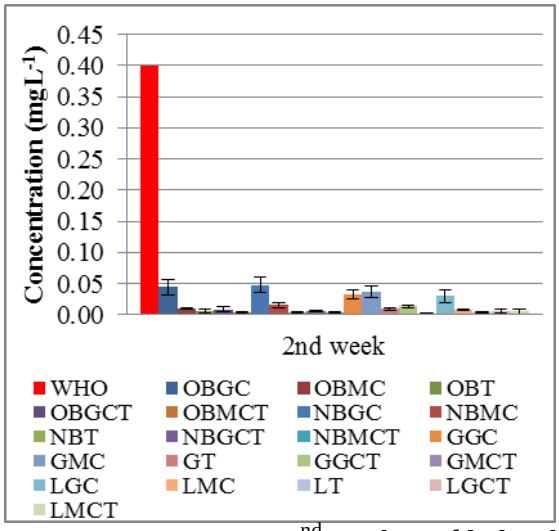
Figure 4.13Fi: Background Manganese concentrations in leachates obtained from test profiles, compared to the Mn WHO (2011) potable water guideline of 0.4mg/L.

All through the experiment, Mn concentrations remained below the specified standard, except for LMCT in week one which went slightly above the standard at 0.43mg/L as shown in figure 4.13Fii-ix below.



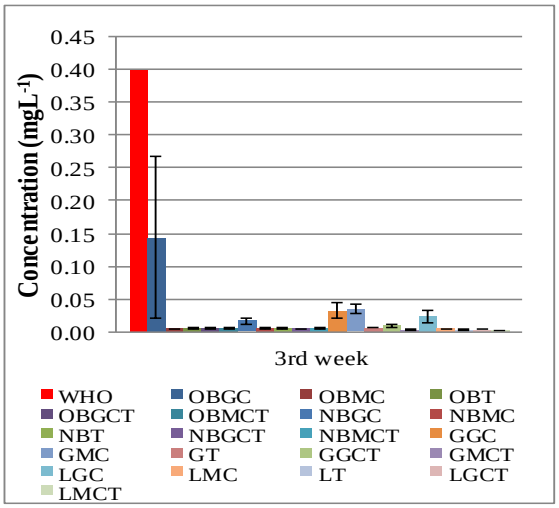
Mn concentration in 1st week profile leachates

ii



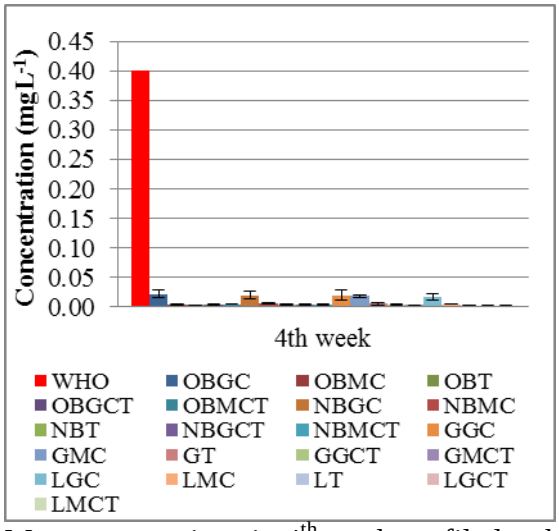
Mn concentrations in 2nd week profile leachates

iii



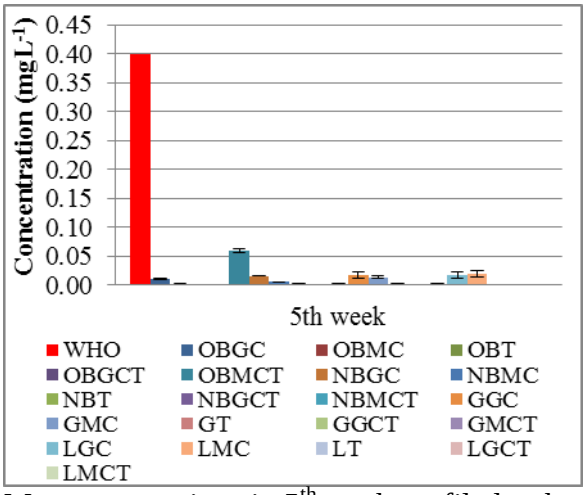
Mn concentrations in 3rd week profile leachates

iv



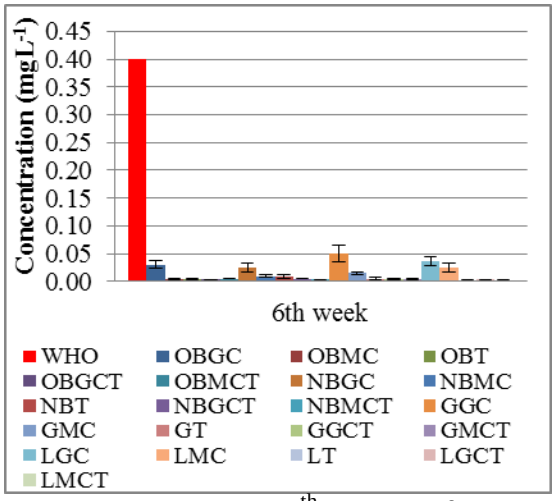
Mn concentrations in 4th week profile leachates

v



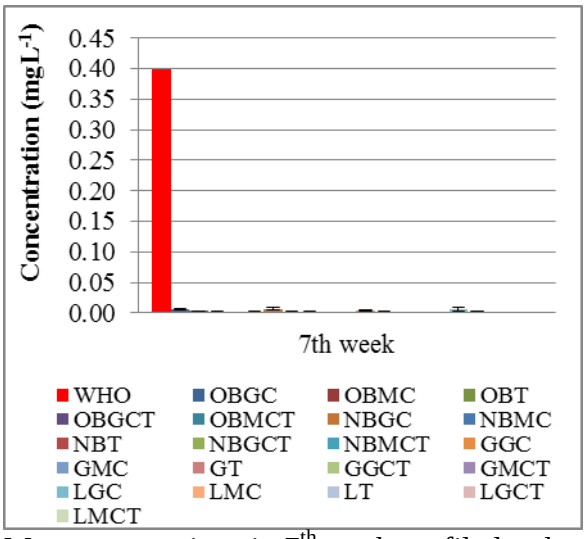
Mn concentrations in 5th week profile leachates

vi



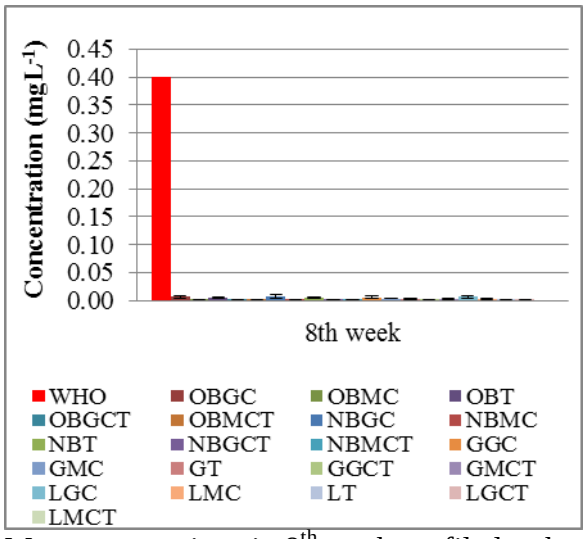
Mn concentrations in 6th week profile leachates

vii



Mn concentrations in 7th week profile leachates

viii



Mn concentrations in 8th week profile leachates

ix

Figure 4.13Fii-ix: Manganese concentrations in leachates obtained from test profiles after eight weekly heavy metal additions.

Nickel

Background leachate concentration of Ni in all the test profiles were below the WHO (2011) potable water guideline of 0.07mg/L (see figure 4.13Gi).

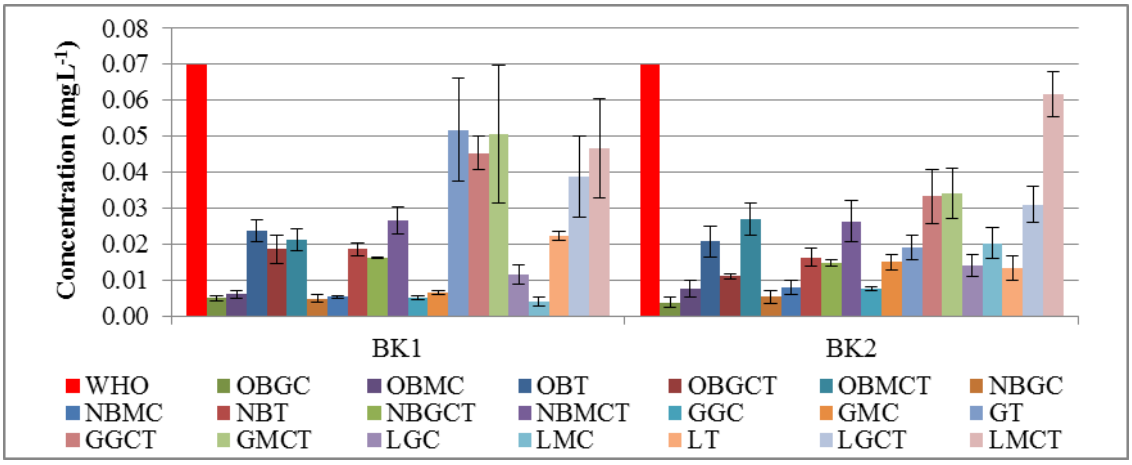
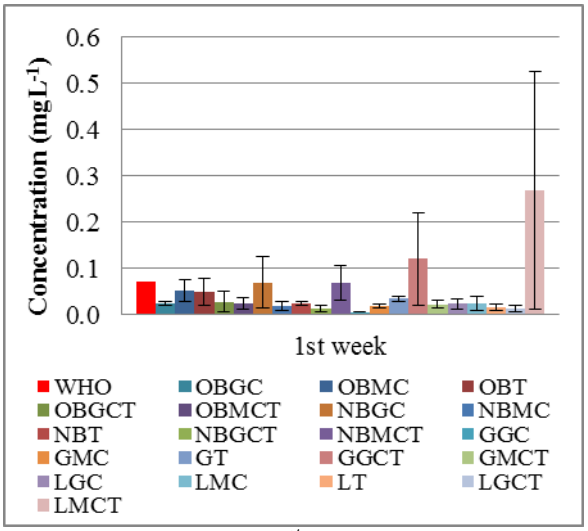


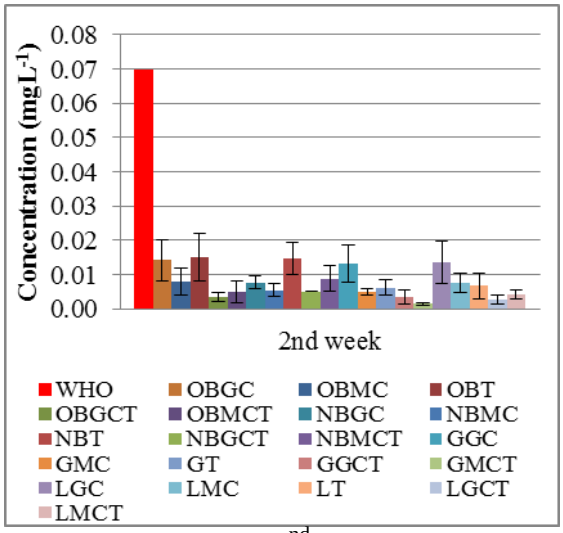
Figure 4.13Gi: Background Nickel concentrations in leachates obtained from test profiles, compared to the Ni WHO (2011) potable water guideline of 0.07mg/L.

Ni concentrations in all profile leachates remained below the Ni WHO (2011) potable water guideline till the eight weeek, as shown in figure 4.13Gii-ix below.



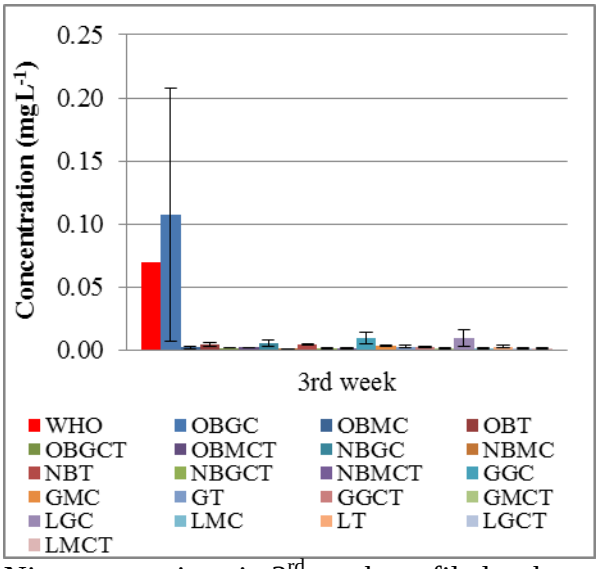
Ni concentrations in 1st week profile leachates

ii



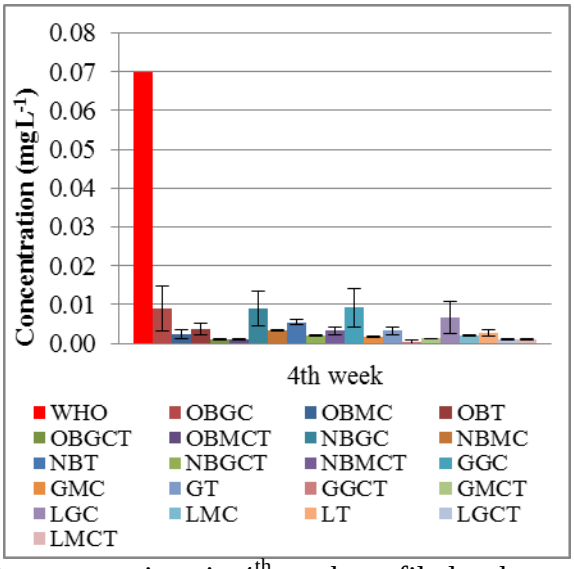
Ni concentrations in 2nd week profile leachates

iii



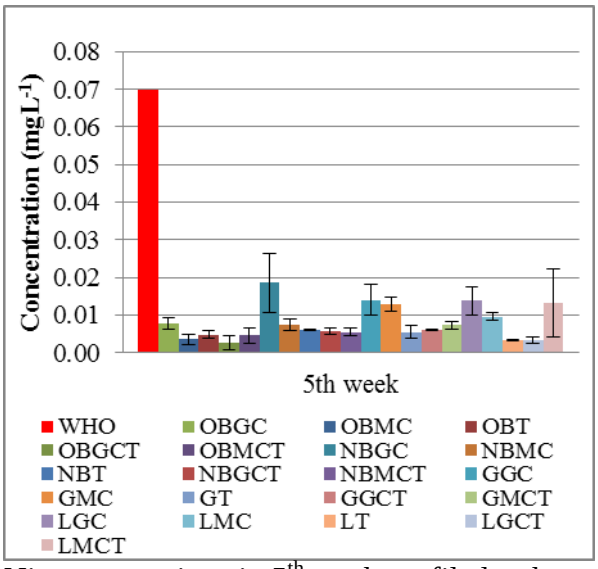
Ni concentrations in 3rd week profile leachates

iv



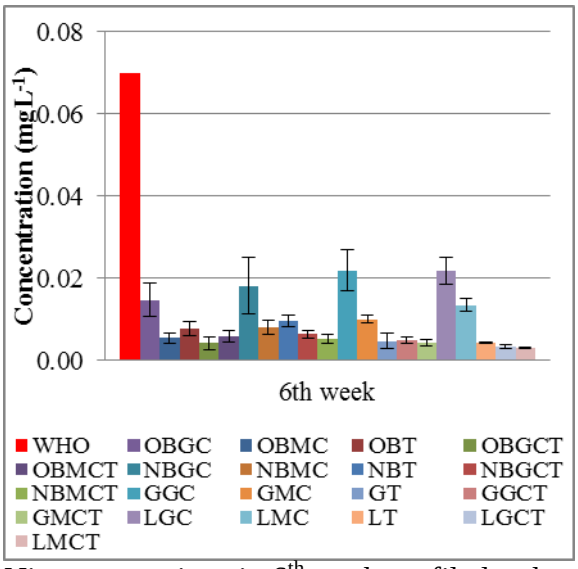
Ni concentrations in 4th week profile leachates

iv



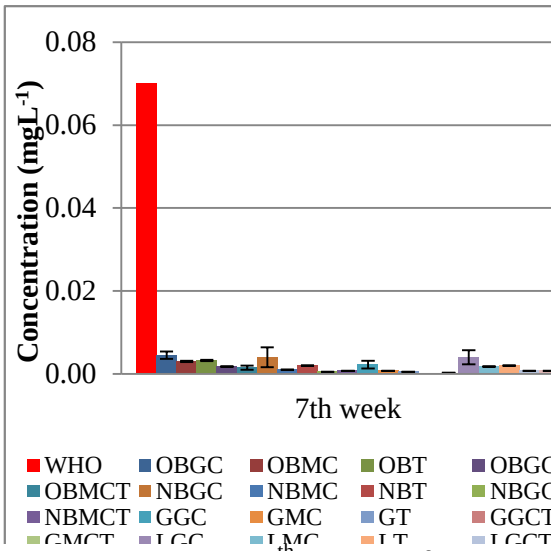
Ni concentrations in 5th week profile leachates

vi

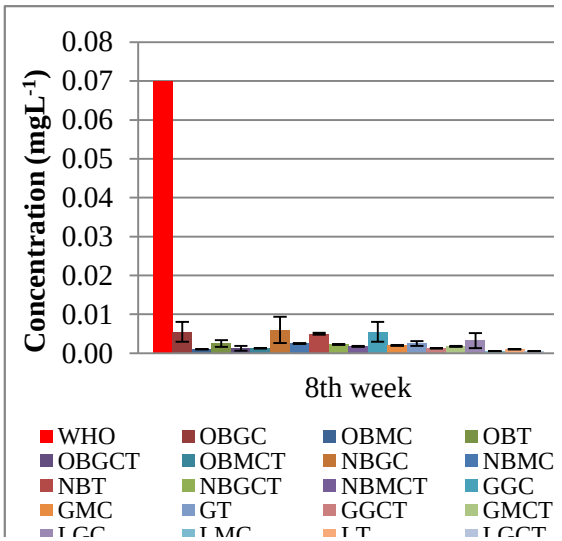


Ni concentrations in 6th week profile leachates

vi



Ni concentrations in 7th week profile leachates viii



Ni concentrations in 8th week profile leachates viii

Figure 4.13Gii-ix: Nickel concentrations in leachates obtained from test profiles after eight weekly heavy metal additions.

Lead

Background Pb concentrations in most of the leachates exceeded the WHO (2011) potable water guideline of 0.01mg/L (figure 4.13Hi).

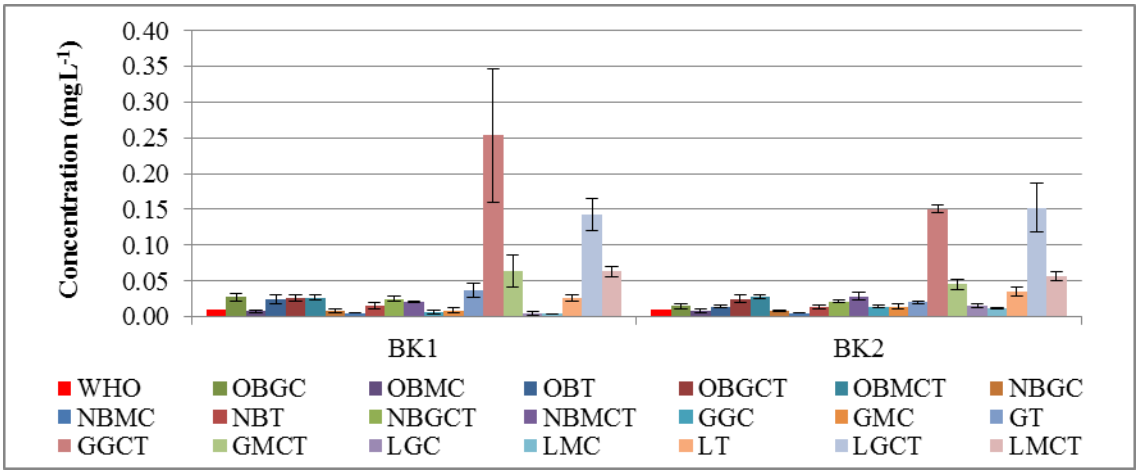
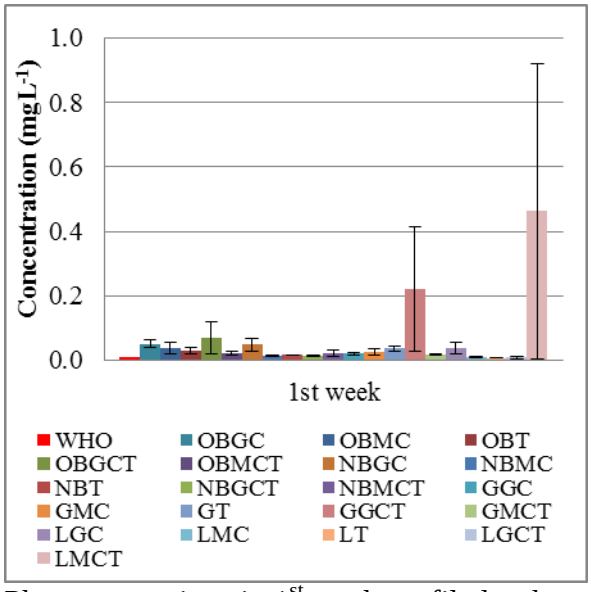


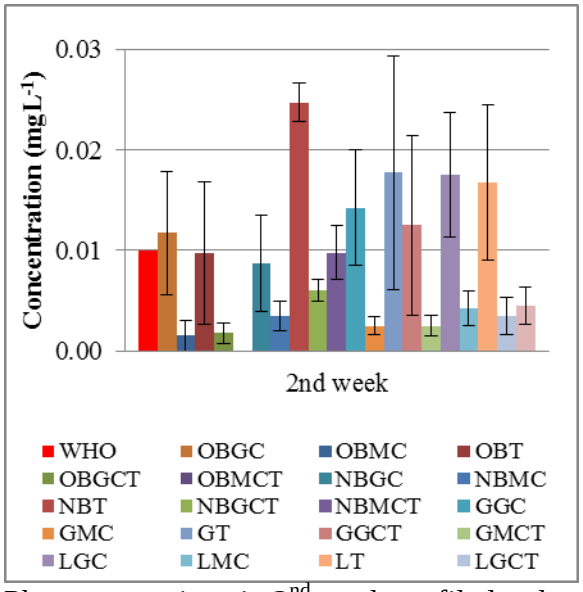
Figure 4.13Hi: Background Lead concentrations in leachates obtained from test profiles, compared to the Pb WHO (2011) potable water guideline of 0.01mg/L.

By the eighth week, Pb concentrations had fallen below the specified standard except in profiles OBT, NBT and GT as shown in figure 4.13Hii-ix below.



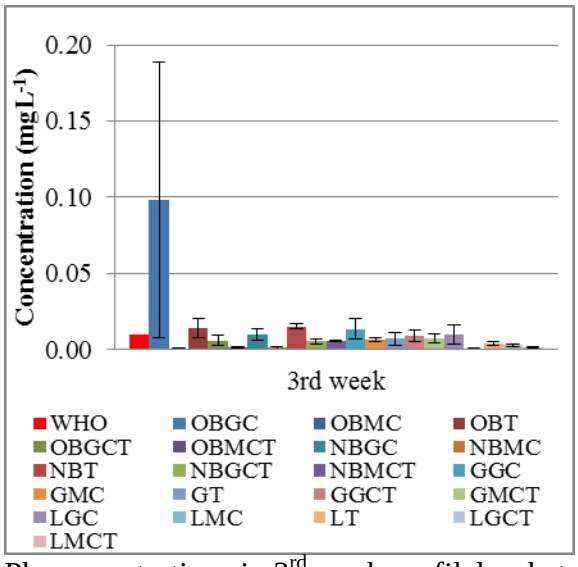
Pb concentrations in 1st week profile leachates

ii



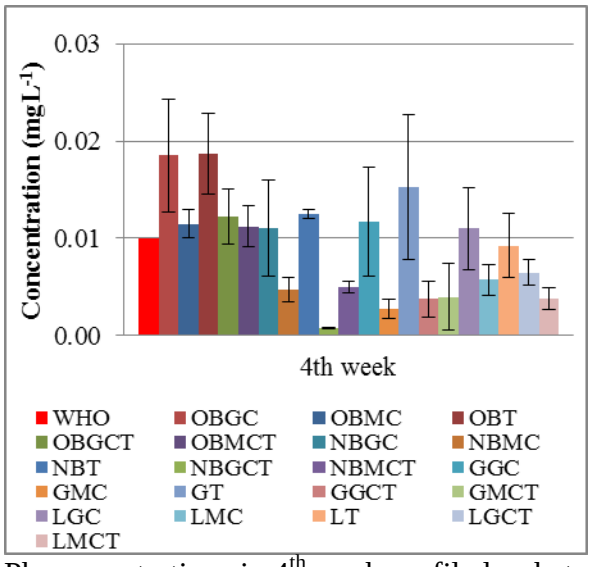
Pb concentrations in 2nd week profile leachates

iii



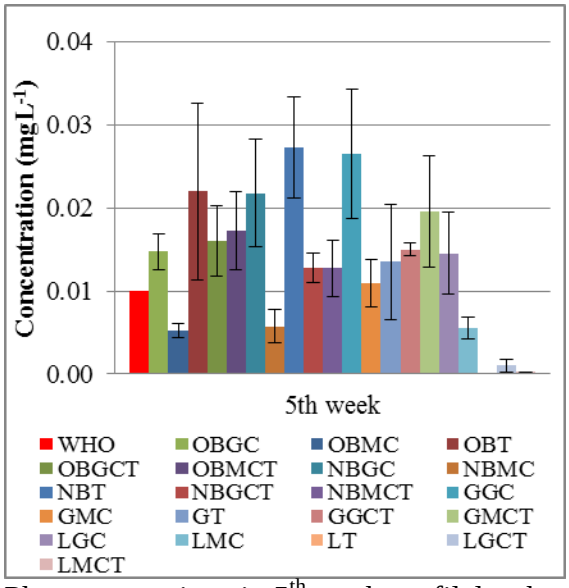
Pb concentrations in 3rd week profileleachates

iv

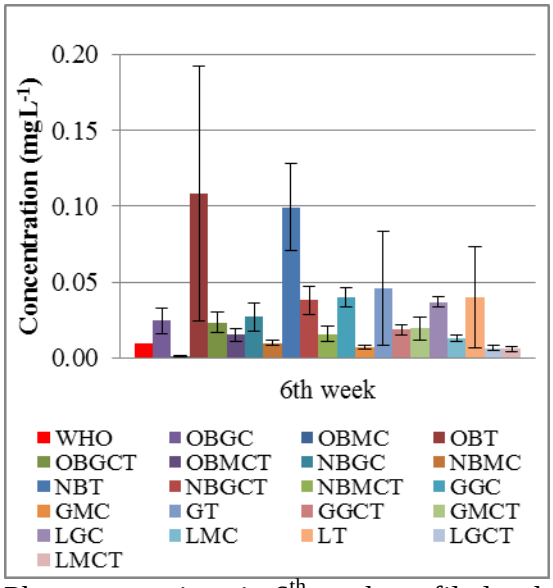


Pb concentrations in 4th week profile leachates

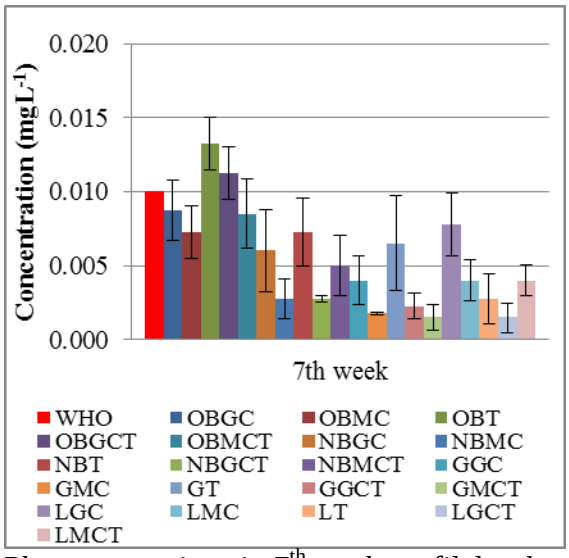
v



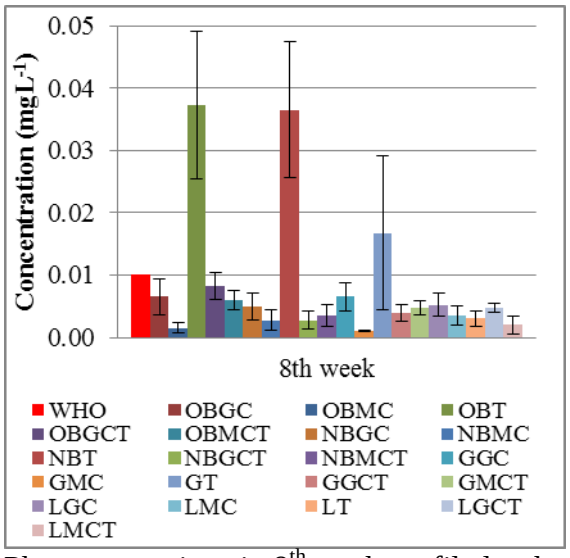
Pb concentrations in 5th week profileleachates vi



Pb concentrations in 6th week profile leachates vi



Pb concentrations in 7th week profileleachates vii



Pb concentrations in 8th week profile leachates viii

Figure 4.13Hii-ix: Lead concentrations in leachates obtained from test profiles after eight weekly heavy metal additions.

Zinc

Zinc concentrations in background leachate as well as leachate after heavy metal addition were well below the WHO (2011) potable water guideline of 3.0mg/L (see figure 4.13Ii-ix).

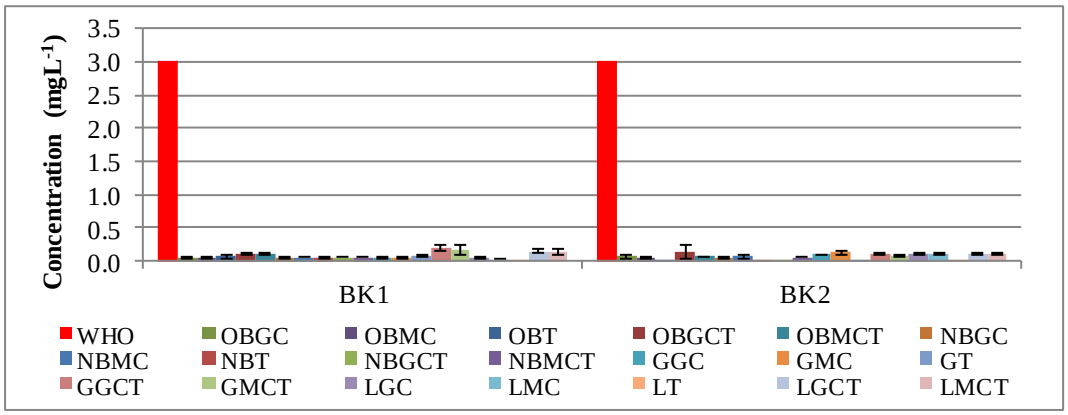
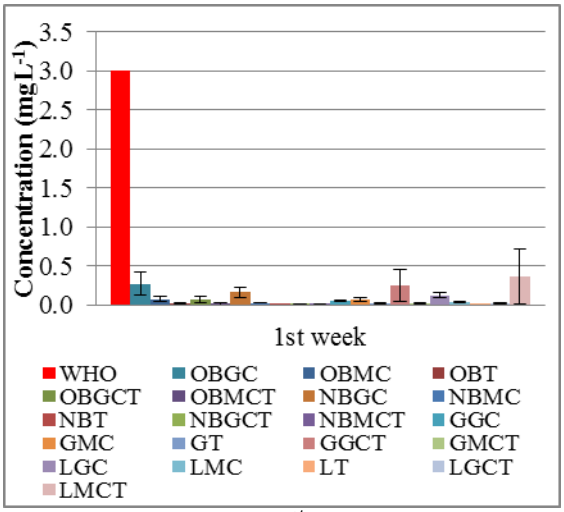
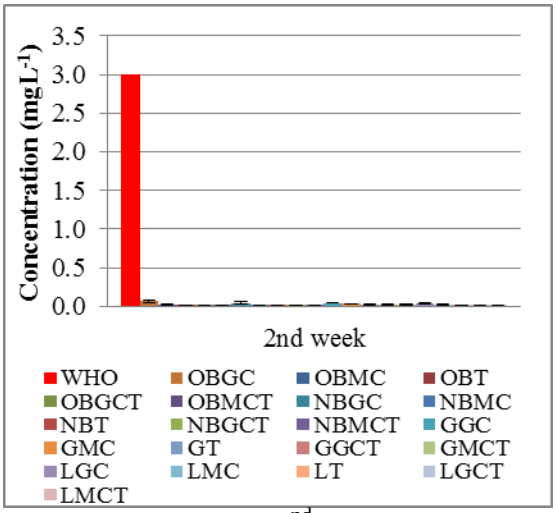


Figure 4.13Ii: Background Zinc concentrations in leachates obtained from test profiles, compared to the Zn WHO (2011) potable water guideline of 3.0mg/L.



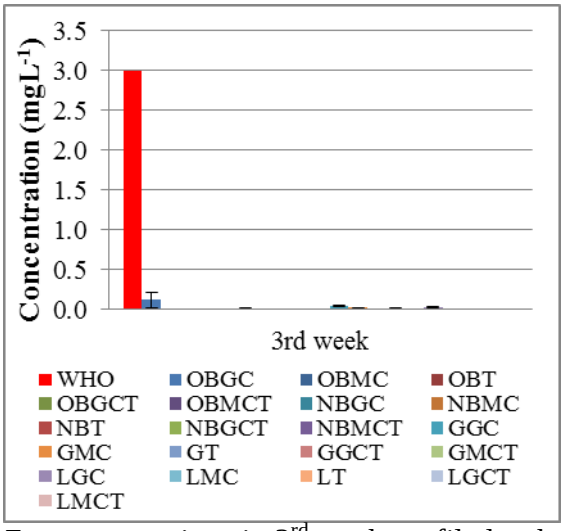
Zn concentrations in 1st week profile leachates

ii

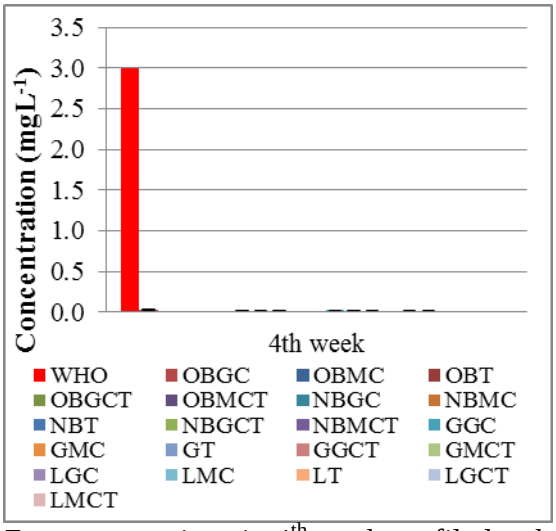


Zn concentrations in 2nd week profile leachates

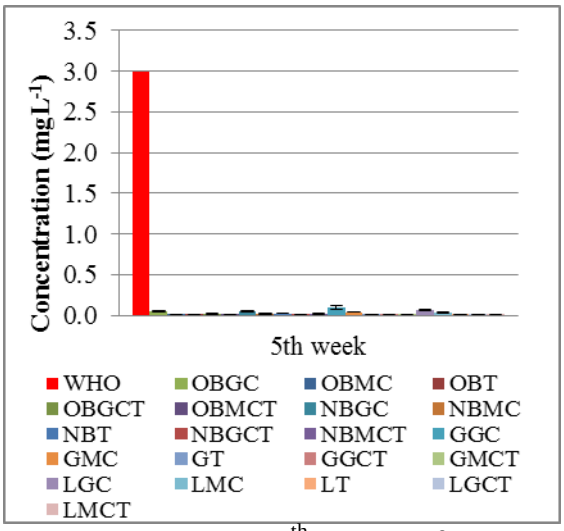
iii



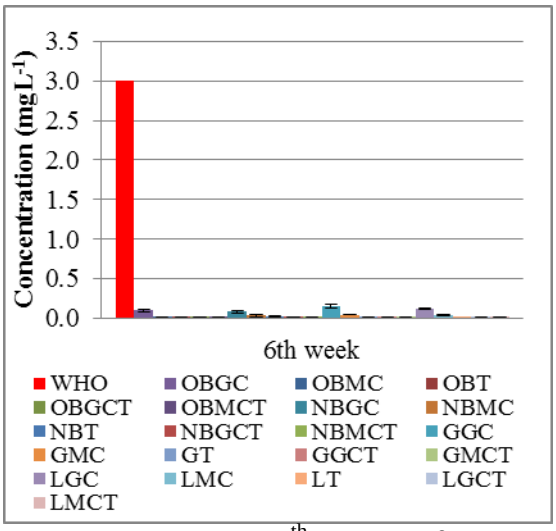
Zn concentrations in 3rd week profile leachates iv



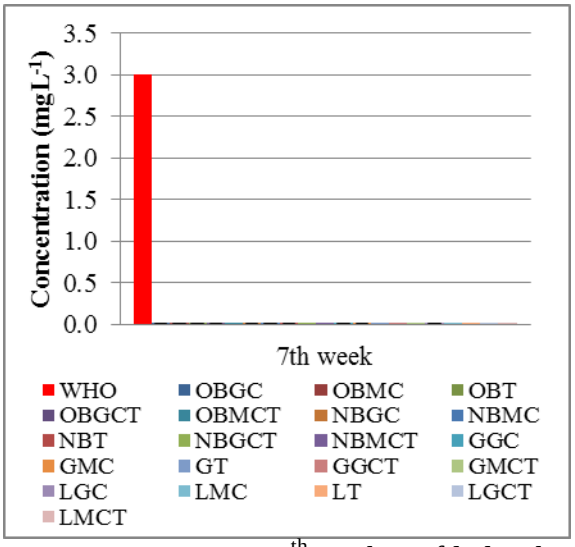
Zn concentrations in 4th week profile leachates v



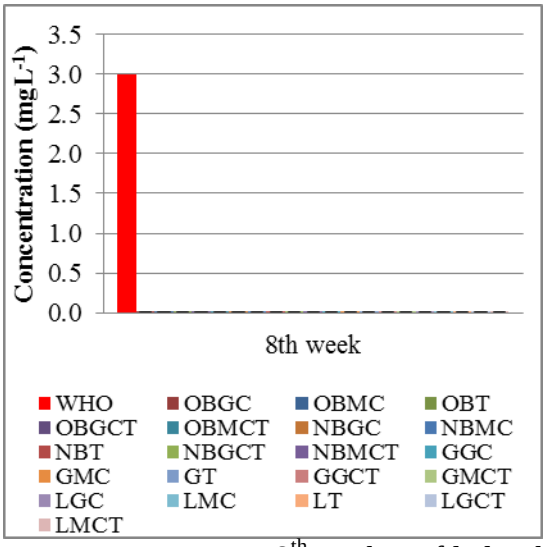
Zn concentrations in 5th week profile leachates vi



Zn concentrations in 6th week profile leachates vii



Zn concentrations in 7th week profile leachates viii



Zn concentrations in 8th week profile leachates ix

Figure 4.13Iii-ix: Zinc concentrations in leachates obtained from test profiles after eight weekly heavy metal additions.

Ultimately, results showed that after adding 200mg/L of each heavy metal (i.e. Cr, Cu, Fe, Mn, Ni, Pb and Zn) concentrations in leachates were below the WHO (2011) potable water guidelines by the eighth week, while Al, Cd and Pb leachate concentrations in some test profiles remained higher than the WHO (2011) standard by the eighth week. However, these values are relatively low compared to Al and Cd toxicity levels for freshwater organisms as shown in table 4.14:

LC_{50} doses (mgL^{-1})	24hr	48hr	72hr	96hr	References
Al	61.66	59.57	57.94	56.92	Anandhan and Hemalatha (2009)
Cd	1.440	0.459	0.392	0.102	Shuhaimi-Othman, Nadzifah and Ahmad (2010)
Pb	2.51	1.88	-	1.65	Offem and Ayotunde (2008)

Table 4.14: LC_{50} doses for Al and Cd in freshwater organisms.

In order to estimate the percentage total concentration of heavy metals leached from each test profile over the eight-week test period, the total concentration of heavy metals recovered in leachate were converted to percentages i.e.:

$$\left(\frac{\text{Total concentration of heavy metal in leachate}}{\text{Total concentration of heavy metals added to profiles}} * 100\right)$$

For example, total Al concentration in OBGC leachate = 1.017mg/L.

total Al concentrations added to profiles = 200mg/L

Applying the equation above,

$$\% \text{ total Al concentration in leachate} = \left(\frac{1.017\text{mg/L}}{200\text{mg/L}} * 100\right) = 0.5\%$$

Test profiles	Total leachate concentration (%)								
	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
OBGC	0.5	0.1	0.1	0.4	1.8	0.2	0.1	0.1	0.3
OBMC	**0.0**	**0.0**	**0.0**	0.2	0.3	0.1	**0.0**	**0.0**	0.1
OBT	0.5	**0.0**	**0.0**	0.8	0.8	**0.0**	**0.0**	0.1	**0.0**
OBGCT	0.3	**0.0**	**0.0**	0.5	0.6	**0.0**	**0.0**	0.1	0.1
OBMCT	0.1	**0.0**	**0.0**	0.3	0.2	**0.0**	**0.0**	**0.0**	**0.0**
NBGC	0.2	**0.0**	0.1	0.3	1.3	0.2	0.1	0.1	0.2
NBMC	**0.0**	**0.0**	**0.0**	0.2	0.4	**0.0**	**0.0**	0.0	0.1
NBT	1.0	**0.0**	**0.0**	0.6	1.1	**0.0**	**0.0**	0.1	**0.0**
NBGCT	0.2	**0.0**	**0.0**	0.3	0.3	**0.0**	**0.0**	**0.0**	**0.0**
NBMCT	**0.0**	**0.0**	**0.0**	0.5	0.2	**0.0**	**0.0**	**0.0**	**0.0**
GGC	0.4	**0.0**	**0.0**	0.4	2.1	0.1	**0.0**	0.1	0.2
GMC	0.2	**0.0**	**0.0**	0.3	1.2	0.1	**0.0**	**0.0**	0.1
GT	0.7	**0.0**	**0.0**	0.6	0.8	**0.0**	**0.0**	0.1	0.1
GGCT	0.3	0.1	**0.0**	0.4	0.6	0.2	0.1	0.1	0.2
GMCT	0.1	**0.0**	**0.0**	0.3	0.4	**0.0**	**0.0**	**0.0**	0.1
LGC	0.5	**0.0**	**0.0**	0.3	2.0	0.1	**0.0**	0.1	0.2
LMC	0.1	**0.0**	**0.0**	0.3	0.9	**0.0**	**0.0**	**0.0**	0.1
LT	0.3	**0.0**	**0.0**	0.4	0.4	**0.0**	**0.0**	**0.0**	**0.0**
LGCT	0.1	**0.0**	**0.0**	0.3	0.2	**0.0**	**0.0**	**0.0**	**0.0**
LMCT	0.2	0.2	0.1	0.5	0.4	0.2	0.1	0.2	0.2

Table 4.15: Percentage total concentrations of nine heavy metals leached from the twenty test profiles over the eight week test period.

Table 4.15 shows that of all the heavy metals added to each profile (200mg/L per heavy metal), every profile yielded at least two heavy metals into leachate with NBMCT leaching the least number of metals (i.e. Cu and Fe) and OBGC and LMCT leaching all nine heavy metals into leachate. Overall, the table shows that ≥98% of all the metals were retained within components of the test profiles i.e. either within the grasses, grass roots or growth media, further supporting the low concentrations of heavy metals in leachate.

Statistical analysis of leaching experiments

Statistical analysis on the results of the leaching experiments was carried out in order to assess the influence of compost and aggregates on heavy metal leaching. In order to achieve this, a two-way ANOVA analysis was carried out on the cumulative heavy metal concentrations in leachate, for the eight weekly additions of heavy metals for each test profile (see appendix 15-22).

p-values	**Al**	**Cd**	**Cr**	**Cu**	**Fe**	**Mn**	**Ni**	**Pb**	**Zn**
Growth media	**0.000**	0.412	0.022	**0.000**	**0.000**	0.058	0.517	0.552	**0.000**
Aggregates	0.725	0.741	0.837	0.441	0.107	0.940	0.927	0.944	0.477
Interactions	0.458	0.447	0.625	**0.018**	0.210	0.298	0.561	0.446	0.237

Where $p < 0.05$ = significant; $p < 0.01$ = very significant; $p < 0.001$ = highly significant

Table 4.16: Results of Two-way ANOVA analysis carried out on total heavy metal concentrations in leachate.

Table 4.16 shows that interactions between growth media and aggregates for the leaching of heavy metals from test profiles were non-significant ($p > 0.05$), except for Cu, and so comments were made based on the p-values for all metals except Cu. The influence of growth media on leaching of heavy metals from the test profiles was significant for Cr and highly significant for Al, Fe and Zn, suggesting that growth media encouraged the leaching of these metals into solution. However, there were no significant differences in the influence of growth media on the leaching Cd, Mn, Ni and Pb which implied that growth media did not influence the leaching of these metals. The influence of aggregates on the leaching of heavy metals,

except Cu, were not significant implying that aggregates had no influence on the leaching of metals. There were significant interactions between growth media and aggregates for Cu and so a post-hoc test was carried out to investigate this relationship. The post-hoc test was also required to determine which of the media specifically influenced the leaching of Al, Cr, Fe and Zn and maybe Cu.

Post-hoc test	
Heavy metal	Growth media
Al	**GC**,**T**
Cr	**GC**
Fe	**GC**, MC, **T**
Zn	**GC**
Cu	MCT, **T**

Table 4.17: Results of Post-hoc test for leaching experiment indicating which growth media influenced leaching of heavy metals.

Post-hoc testing confirmed the initial statistical result obtained for Cu in table 4.16, that aggregates had no significant influence on the leaching of Cu while growth media significantly influenced its leaching. Overall, table 4.17 shows that GC and/or T mostly encouraged the leaching of heavy metals i.e. Al, Cr, Fe, Zn and Cu.

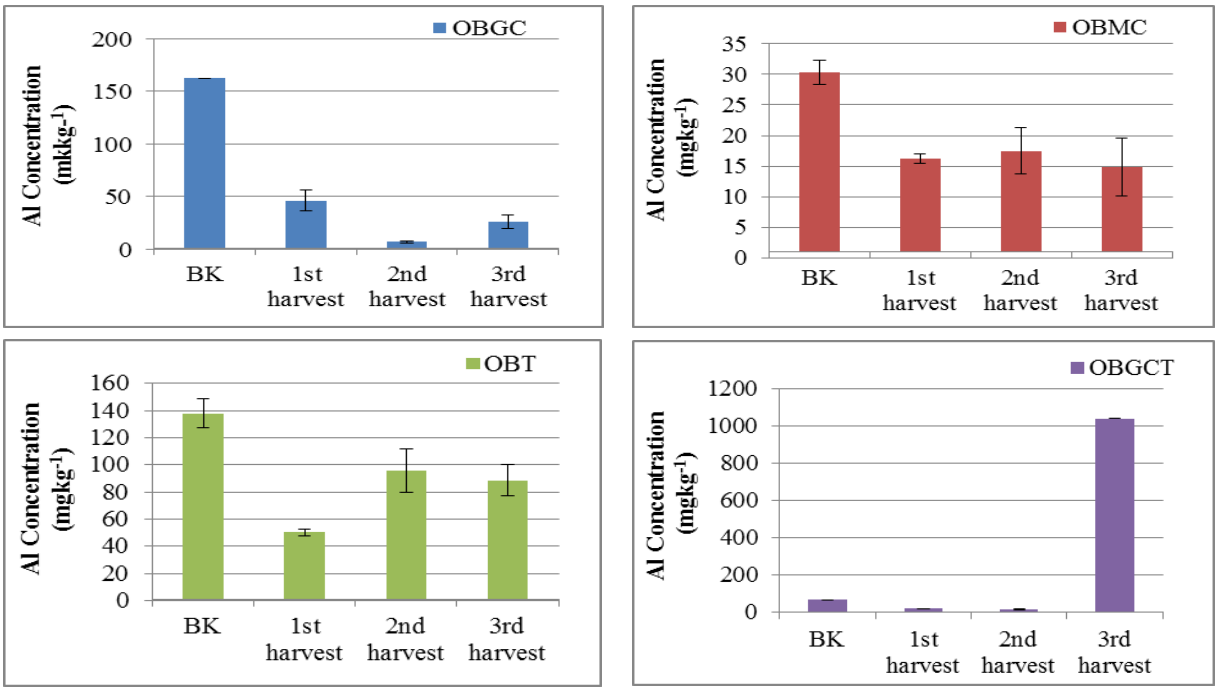
4.3.3.2 Monitoring of heavy metal concentrations in harvested grasses, grass roots and growth media

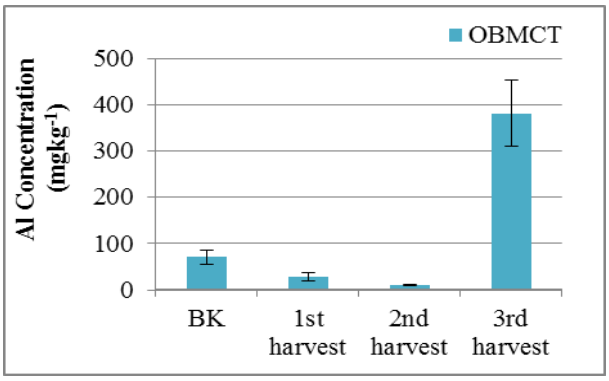
Results of leaching experiments described in the previous section showed that ≥98% of added heavy metals were retained within the test profiles and their components (i.e. grasses, grass roots and/or growth media), and so each of these test profile components were analysed for heavy metals because plant shoots and roots play an important role in heavy metal uptake by aiding the movement of heavy metals (stormwater pollutants) to and from soil (Kalis *et al*., 2007). For grass heavy metal analyses, dried grass cuttings derived from grass biomass determination described in section 3.4.2, were used. Results from background grass heavy

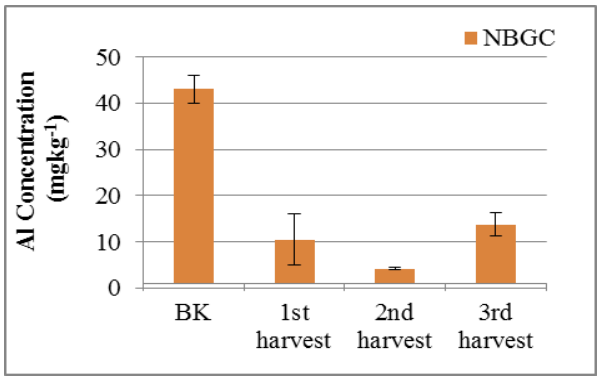
concentrations were compared with that of three grass harvests i.e. 1st harvest after four weeks of heavy metal dosing, 2nd harvest after another four weeks of dosing and 3rd harvest with no dosing at all. Also, cumulative heavy metal concentrations of the background and three harvests were compared to total heavy metal concentrations in both grass roots and growth media so as to identify where each heavy metal was predominantly retained within the profiles by the end of the testing period.

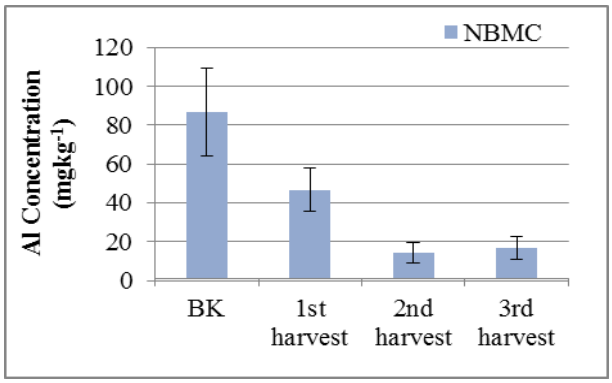
Aluminium

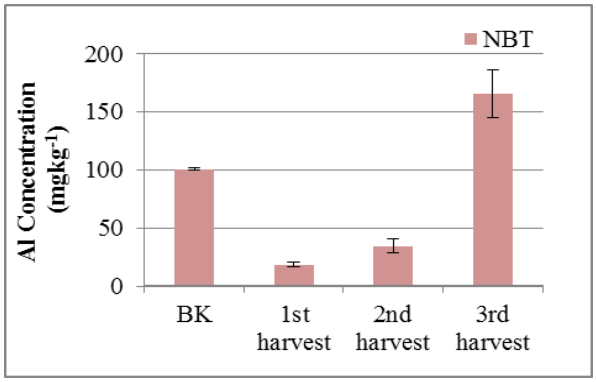
Figure 4.14a shows that 1st and 2nd harvest Al concentrations were predominantly below background concentrations while the 3rd harvest Al concentration exceeded background concentrations in almost half of the profiles. This result shows that the uptake of Al by the profiles in the 1st and 2nd harvest was less than that for background harvests indicating that the addition of Al to the profiles within the two-month spiking period did not necessarily lead to an increase in Al concentrations in the 1st and 2nd grass harvest, however, the effect of Al spiking was prominent by the 3rd harvest when no Al was added. This is a reflection of the movement of residual heavy metals in growth media long after contamination has occurred (Sánchez-Moreno *et al*., 2006; Ideriah *et al*., 2013).

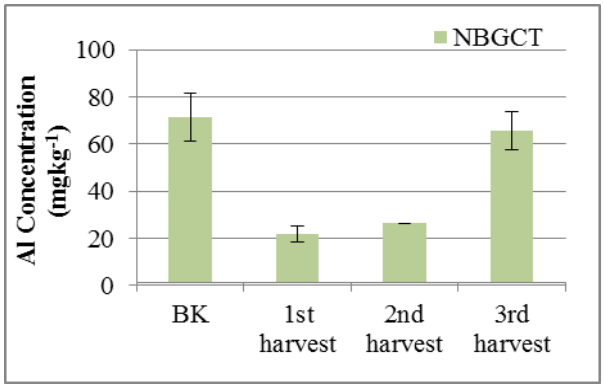


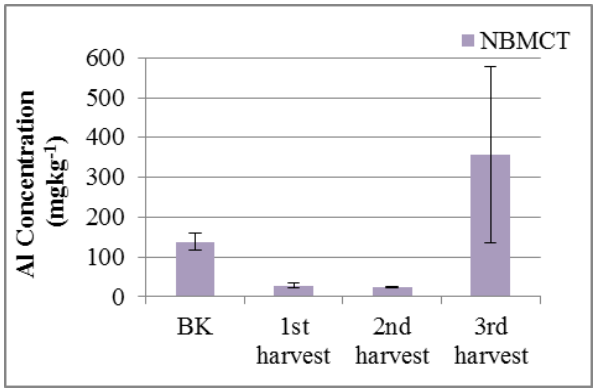

OBMCT
Al Concentration (mgkg-1)
500
400
300
200
100
0
BK
1st harvest
2nd harvest
3rd harvest

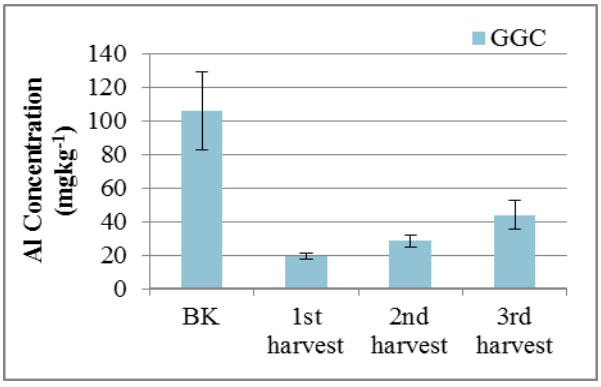

NBGC
Al Concentration (mgkg-1)
50
40
30
20
10
0
BK
1st harvest
2nd harvest
3rd harvest

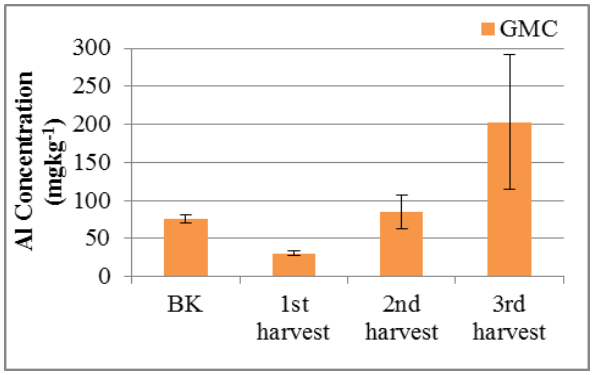

NBMC
Al Concentration (mgkg-1)
120
100
80
60
40
20
0
BK
1st harvest
2nd harvest
3rd harvest


NBT
Al Concentration (mgkg-1)
200
150
100
50
0
BK
1st harvest
2nd harvest
3rd harvest


NBGCT
Al Concentration (mgkg-1)
100
80
60
40
20
0
BK
1st harvest
2nd harvest
3rd harvest


NBMCT
Al Concentration (mgkg-1)
600
500
400
300
200
100
0
BK
1st harvest
2nd harvest
3rd harvest


GGC
Al Concentration (mgkg-1)
140
120
100
80
60
40
20
0
BK
1st harvest
2nd harvest
3rd harvest


GMC
Al Concentration (mgkg-1)
300
250
200
150
100
50
0
BK
1st harvest
2nd harvest
3rd harvest

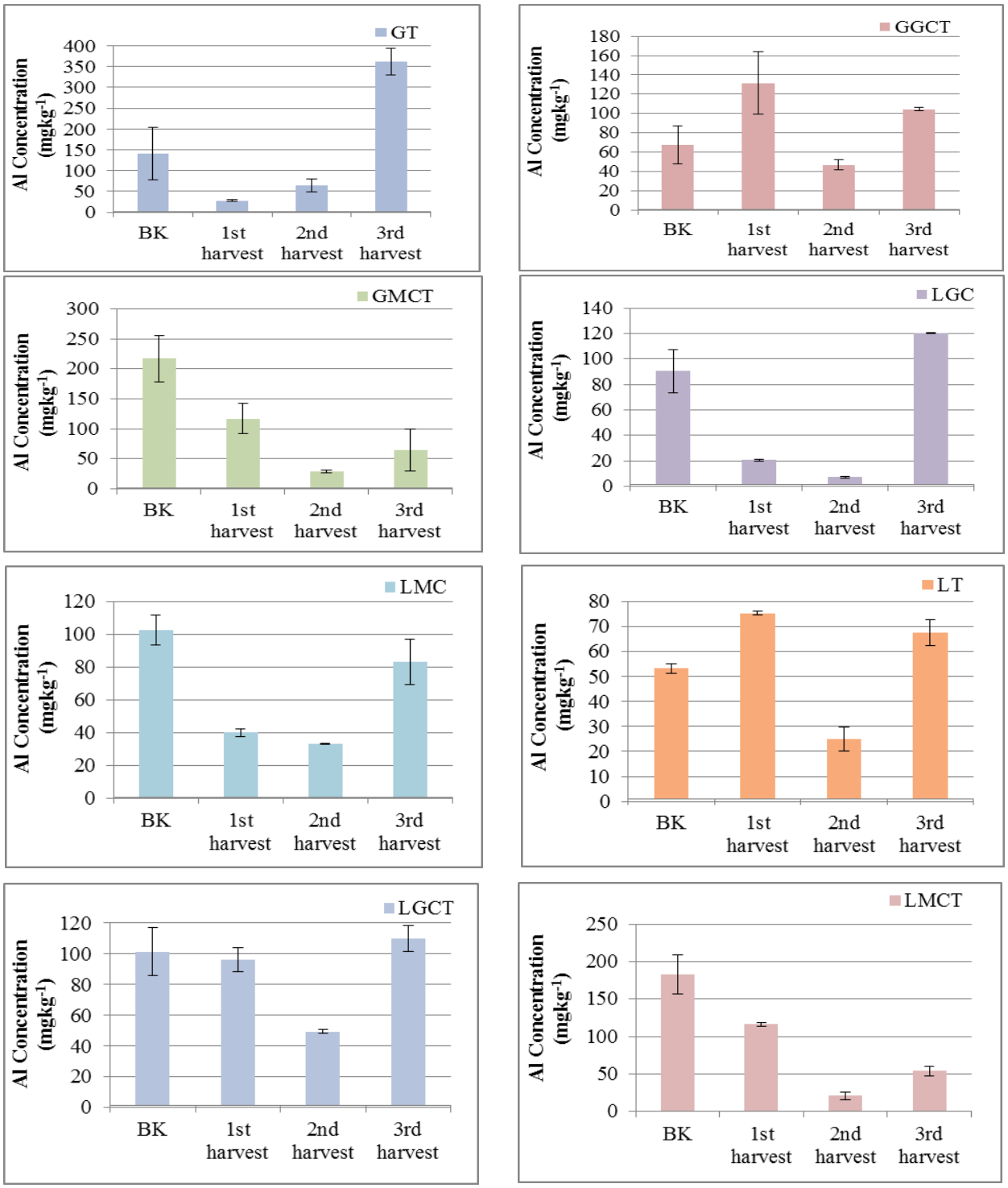
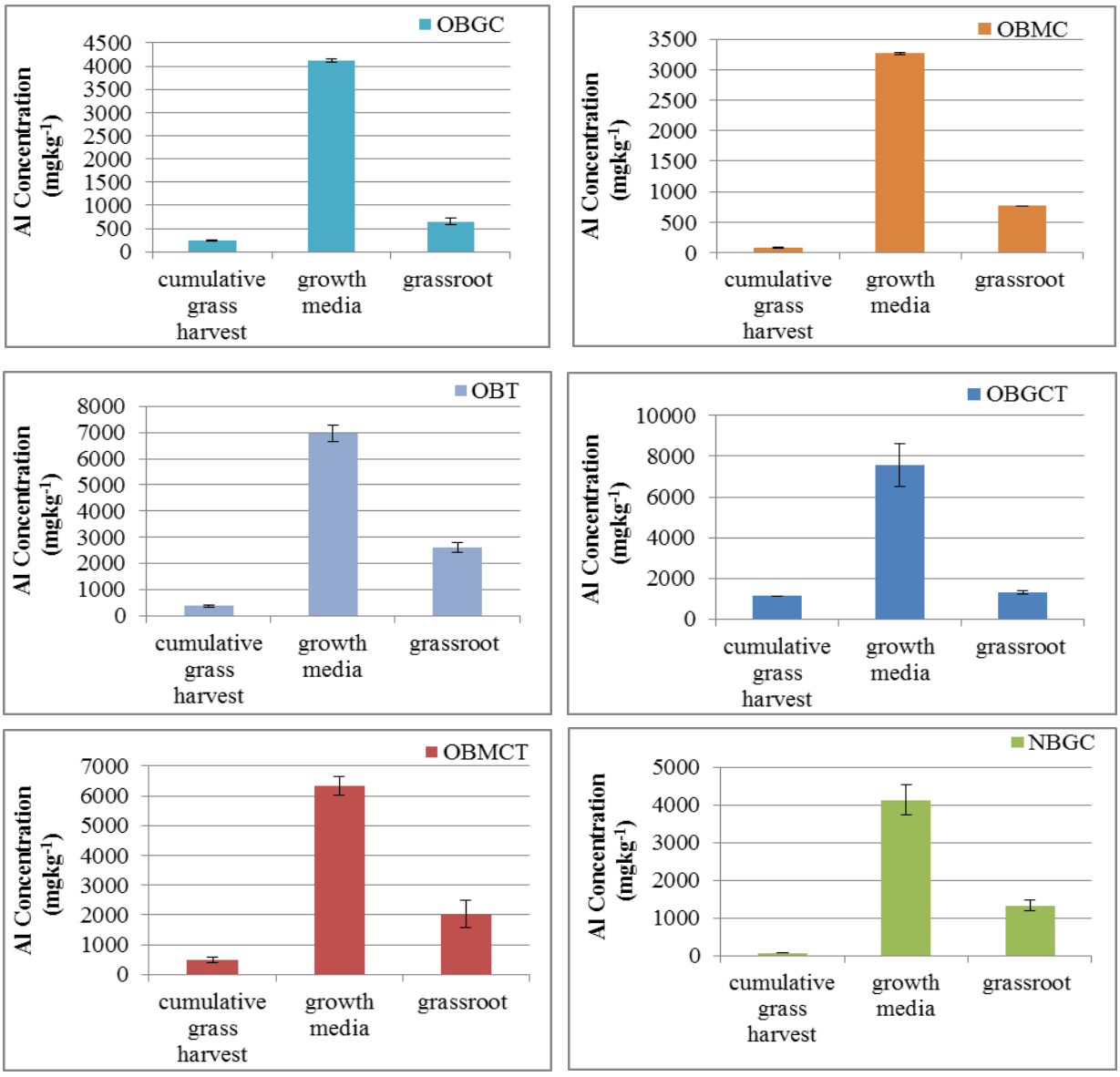
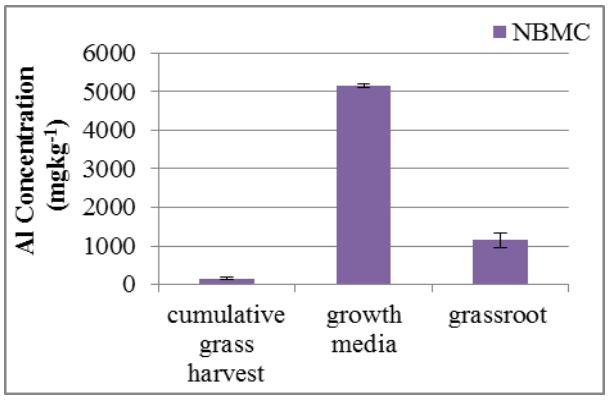
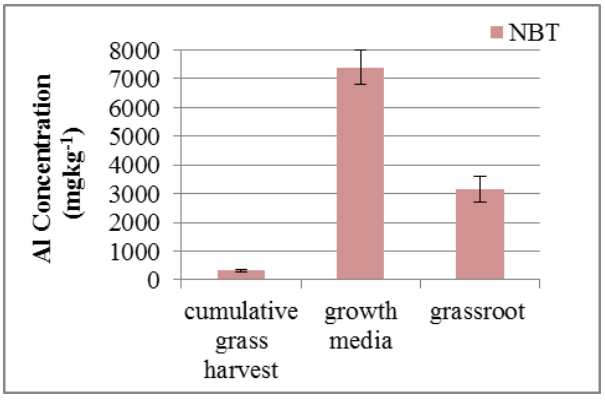


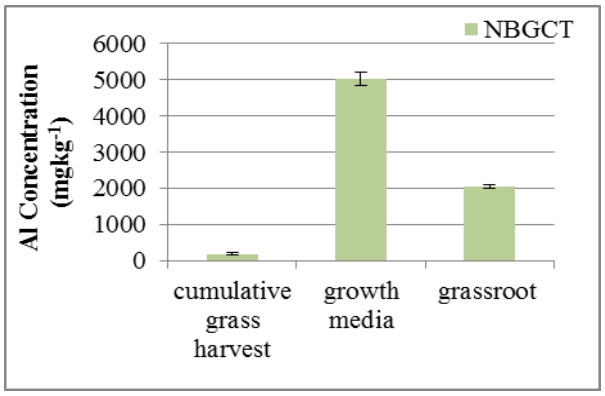
Figure 4.14a: Trends of aluminium concentrations in background and three monthly grass harvests derived from test profiles.

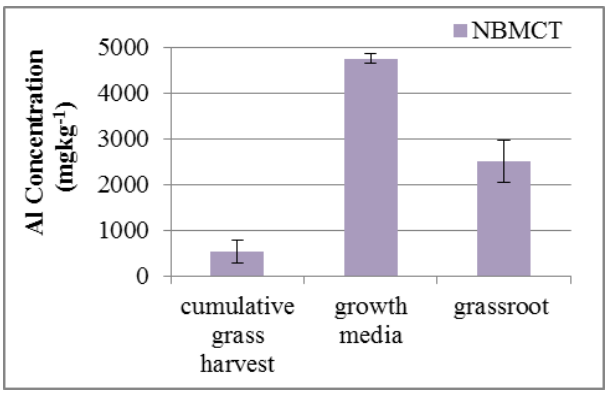
Figure 4.14b shows that at the end of the test period, growth media retained most of the added Al concentrations followed by grass roots, as Al concentrations in these two components far exceeded the Al concentrations taken up by the grasses.

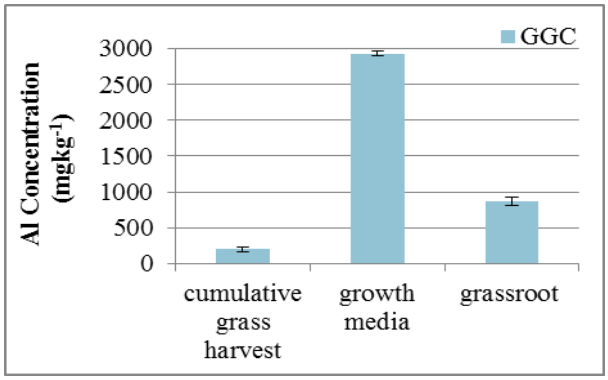


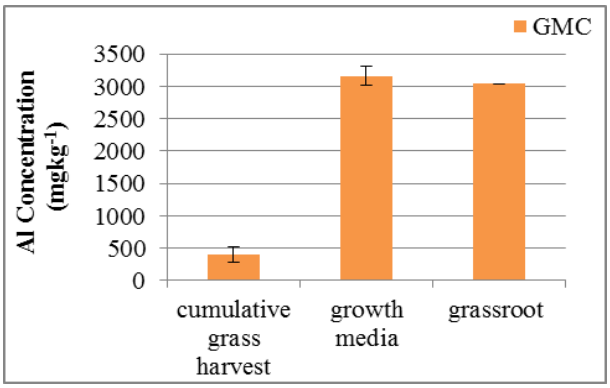

NBMC
Al Concentration (mgkg-1)
6000
5000
4000
3000
2000
1000
0
cumulative grass harvest
growth media
grassroot

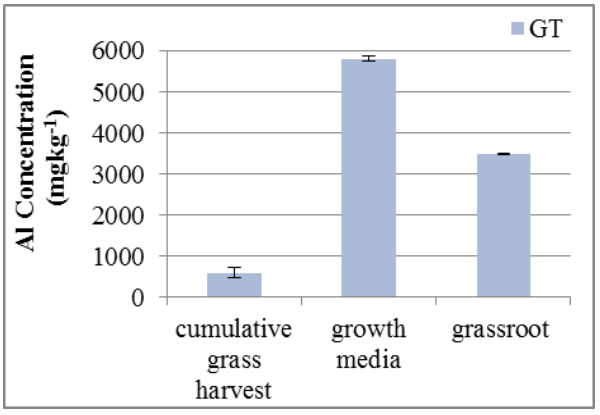

NBT
Al Concentration (mgkg-1)
8000
7000
6000
5000
4000
3000
2000
1000
0
cumulative grass harvest
growth media
grassroot

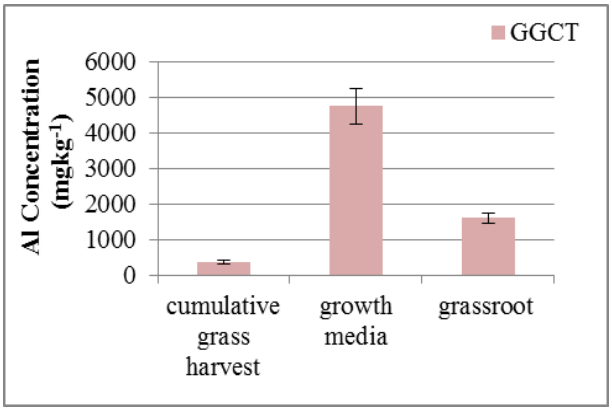

NBGCT
Al Concentration (mgkg-1)
6000
5000
4000
3000
2000
1000
0
cumulative grass harvest
growth media
grassroot


NBMCT
Al Concentration (mgkg-1)
5000
4000
3000
2000
1000
0
cumulative grass harvest
growth media
grassroot


GGC
Al Concentration (mgkg-1)
3000
2500
2000
1500
1000
500
0
cumulative grass harvest
growth media
grassroot


GMC
Al Concentration (mgkg-1)
3500
3000
2500
2000
1500
1000
500
0
cumulative grass harvest
growth media
grassroot


GT
Al Concentration (mgkg-1)
6000
5000
4000
3000
2000
1000
0
cumulative grass harvest
growth media
grassroot


GGCT
Al Concentration (mgkg-1)
6000
5000
4000
3000
2000
1000
0
cumulative grass harvest
growth media
grassroot

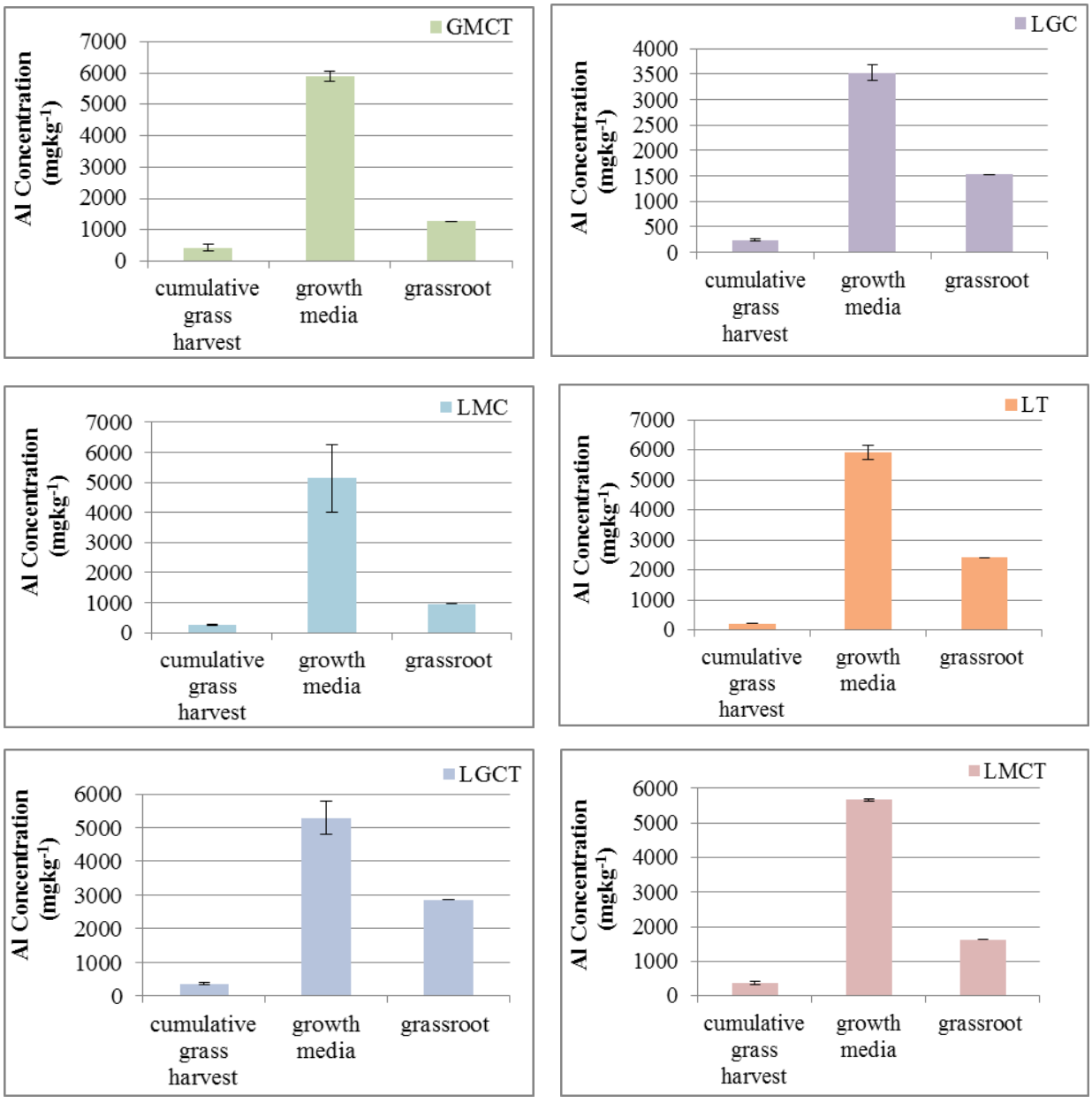
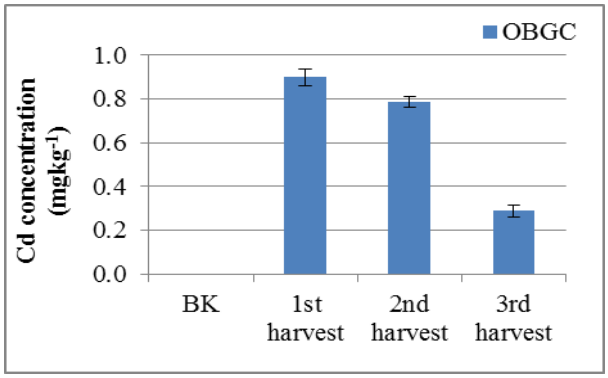
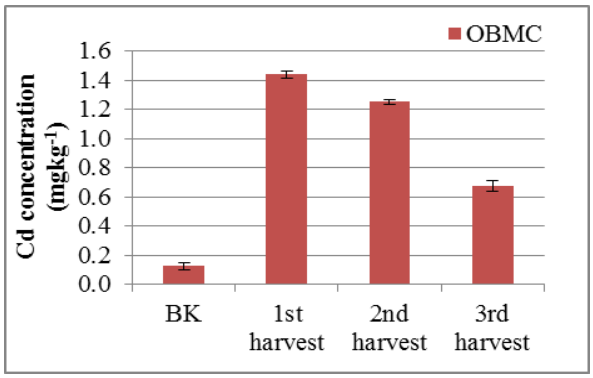


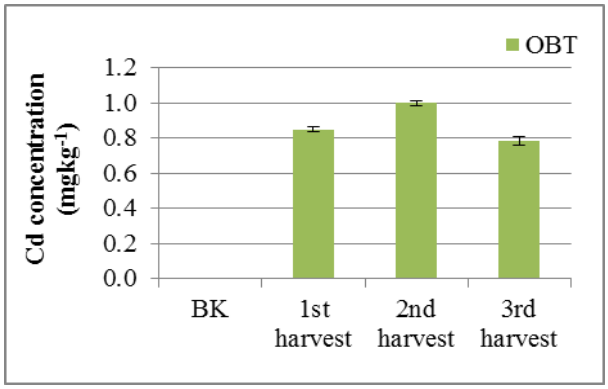
Figure 4.14b: Trends of aluminium concentrations in cumulative grass harvests, growth media and grass roots

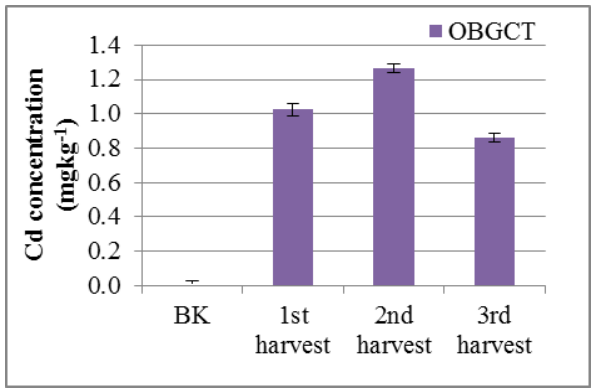
Cadmium

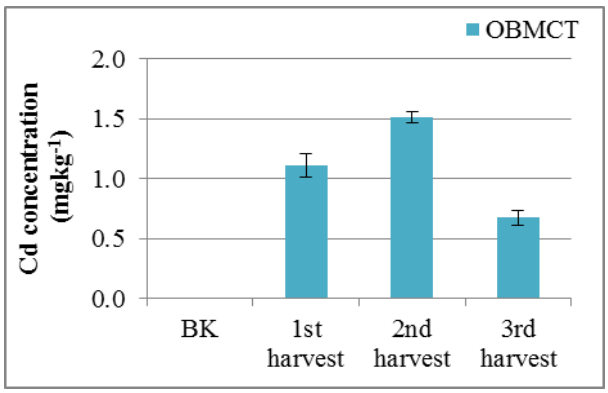
Background analysis of the grasses showed that Cd concentrations were mostly not detected and where detected, they were much lower than in the 1st, 2nd and 3rd harvests (see figure 4.14c). All three spiked grass harvests exceeded background concentrations with the 2nd harvest having the highest uptake of Cd in almost all the profiles. The 3rd harvest showed the least uptake of Cd in all the profiles.

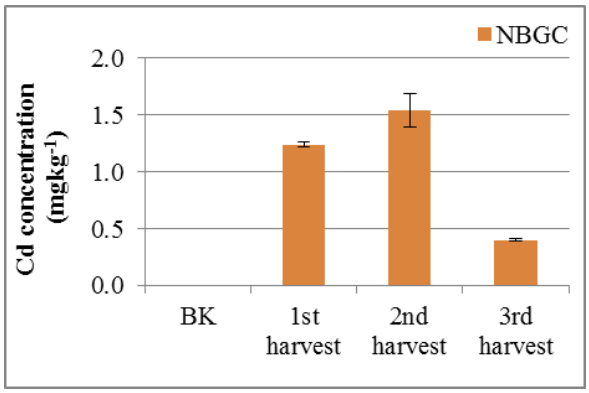


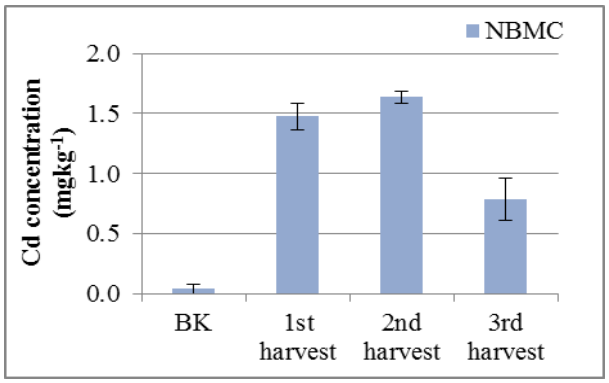


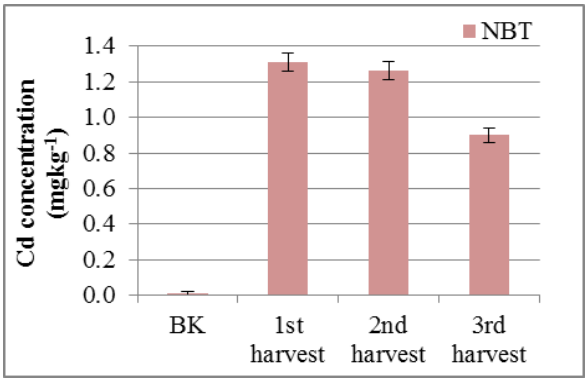


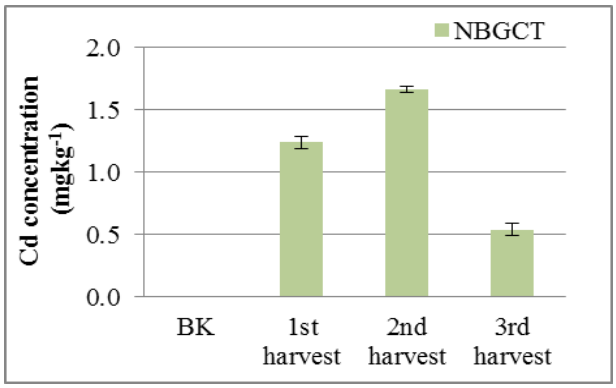


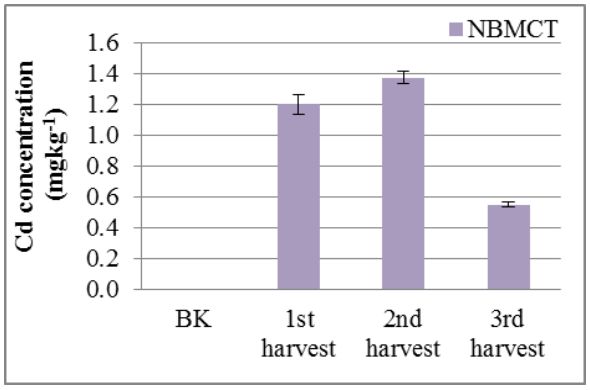


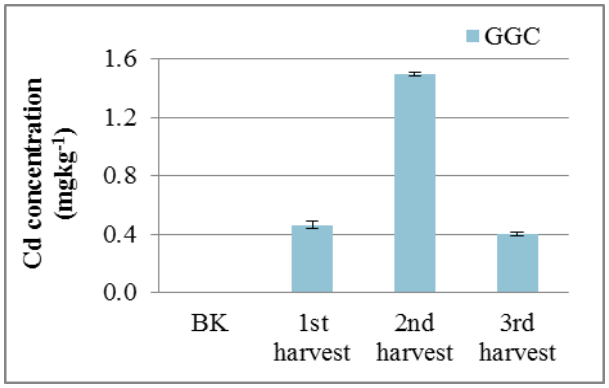


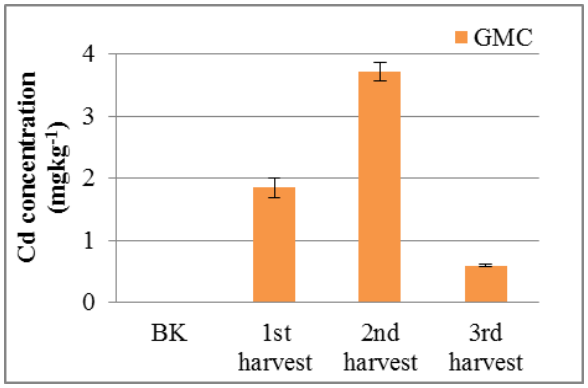

NBMC
Cd concentration (mgkg-1)
2.0
1.5
1.0
0.5
0.0
BK
1st harvest
2nd harvest
3rd harvest

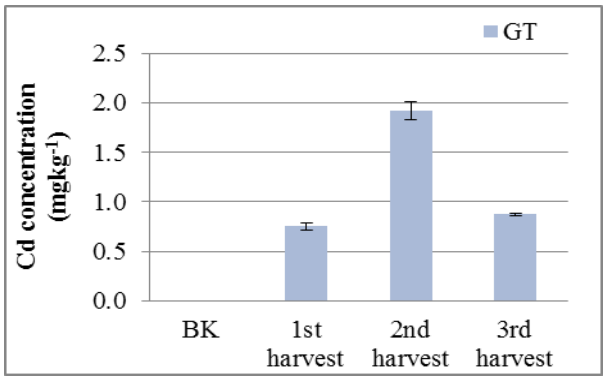

NBT
Cd concentration (mgkg-1)
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
BK
1st harvest
2nd harvest
3rd harvest

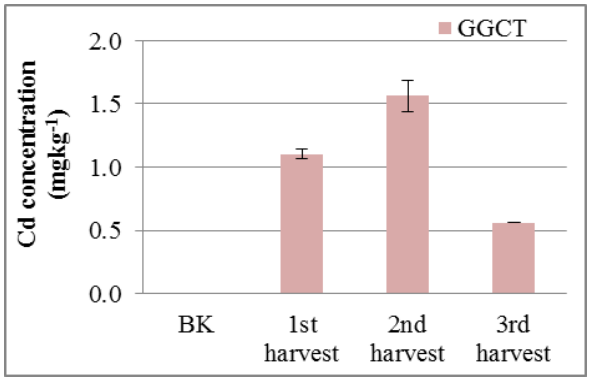

NBGCT
Cd concentration (mgkg-1)
2.0
1.5
1.0
0.5
0.0
BK
1st harvest
2nd harvest
3rd harvest


NBMCT
Cd concentration (mgkg-1)
1.6
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
BK
1st harvest
2nd harvest
3rd harvest


GGC
Cd concentration (mgkg-1)
1.6
1.2
0.8
0.4
0.0
BK
1st harvest
2nd harvest
3rd harvest


GMC
Cd concentration (mgkg-1)
4
3
2
1
0
BK
1st harvest
2nd harvest
3rd harvest


GT
Cd concentration (mgkg-1)
2.5
2.0
1.5
1.0
0.5
0.0
BK
1st harvest
2nd harvest
3rd harvest


GGCT
Cd concentration (mgkg-1)
2.0
1.5
1.0
0.5
0.0
BK
1st harvest
2nd harvest
3rd harvest

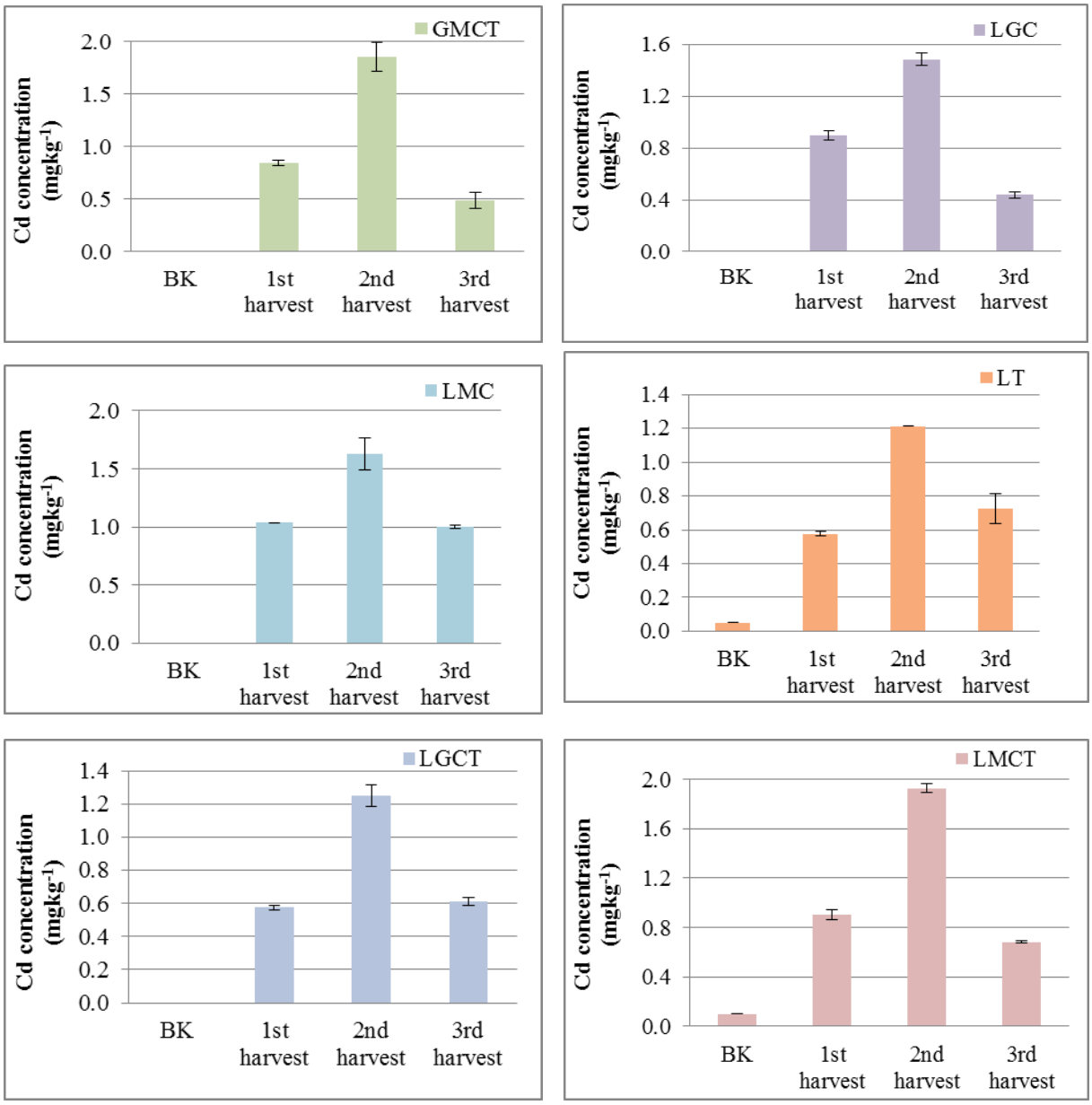
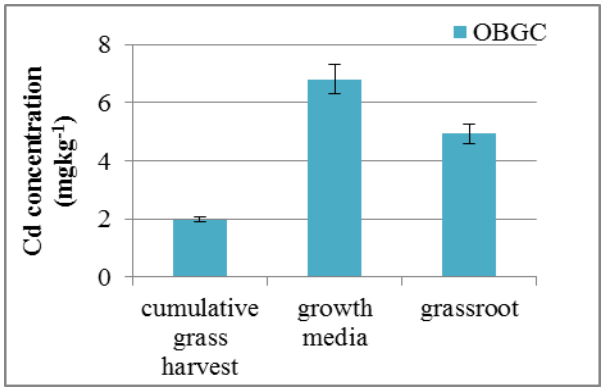
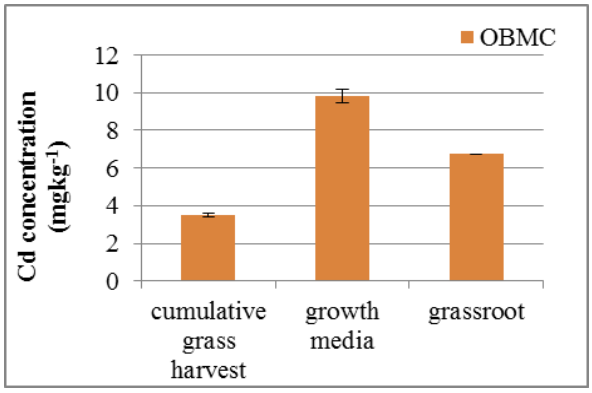
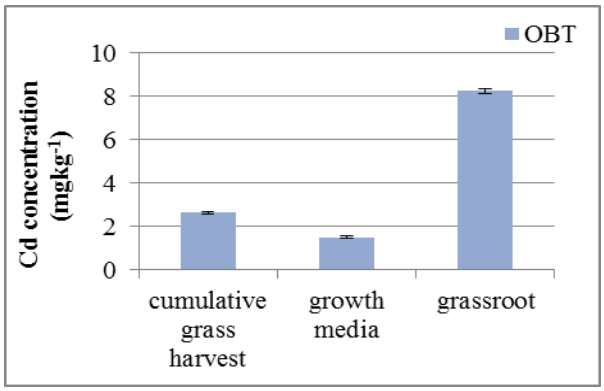


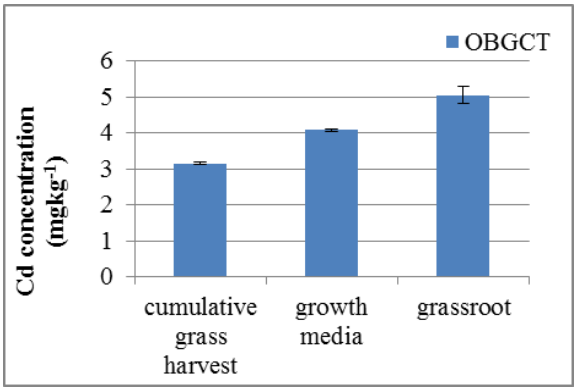
Figure 4.14c: Trends of cadmium concentrations in background and three monthly grass harvests derived from test profiles

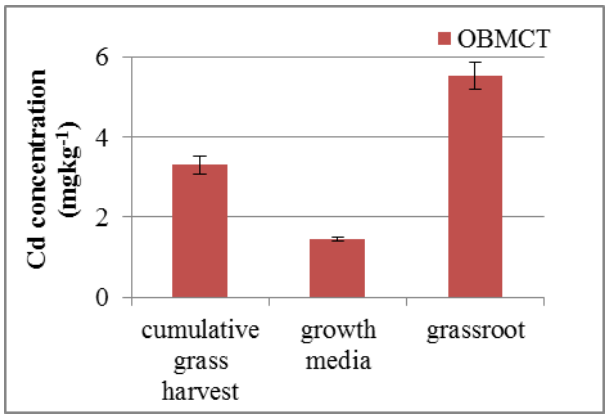
Overall, grass roots retained the highest Cd concentrations compared to the grasses and growth media in almost all the profiles (see figure 4.14d). Cd concentrations in growth media were mostly higher than grass concentrations except in a few profiles where Cd concentrations fell below grass concentrations such as in OBT, OBMCT, NBT, NBGCT, GT, GGCT, LT and LMCT.

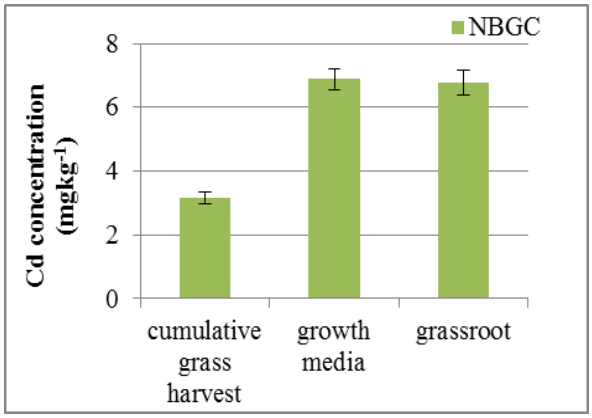


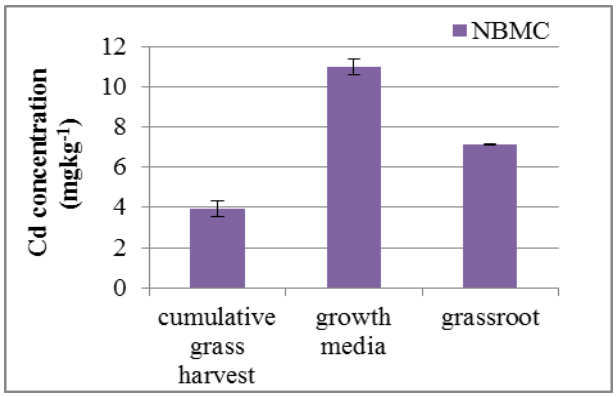


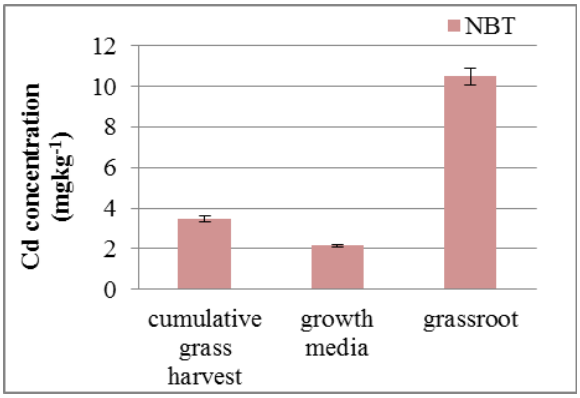


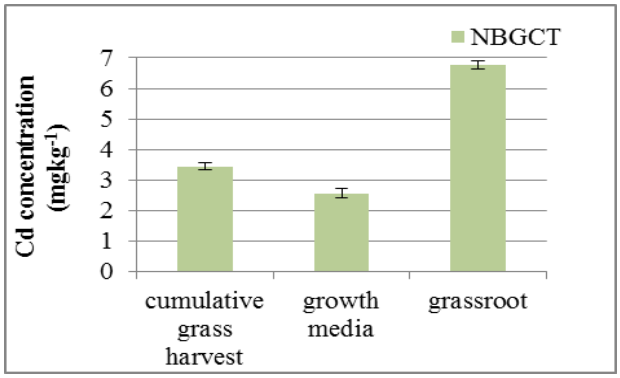


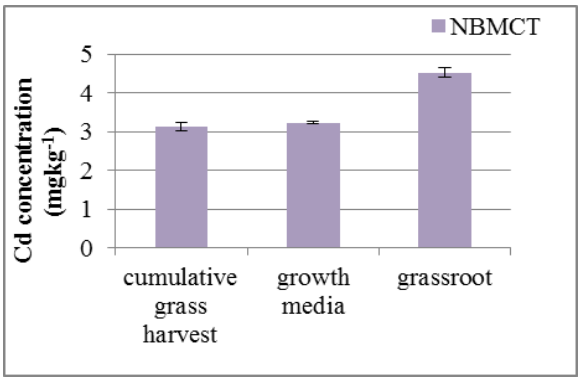


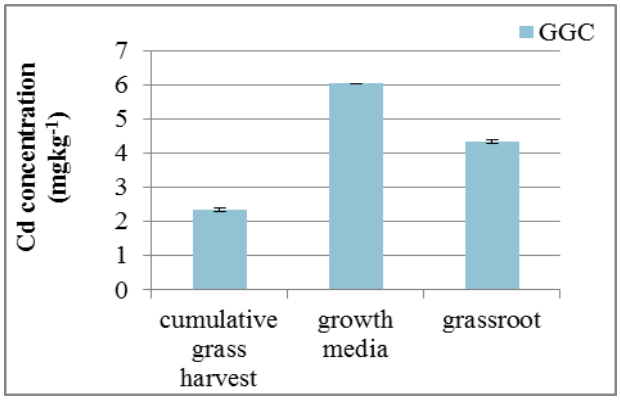


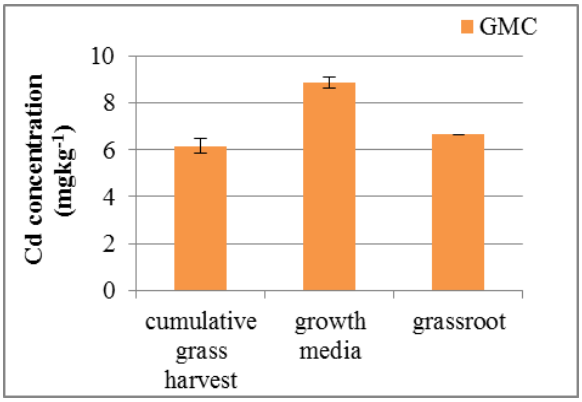

NBMC
Cd concentration (mgkg-1)
0
2
4
6
8
10
12
cumulative grass harvest
growth media
grassroot

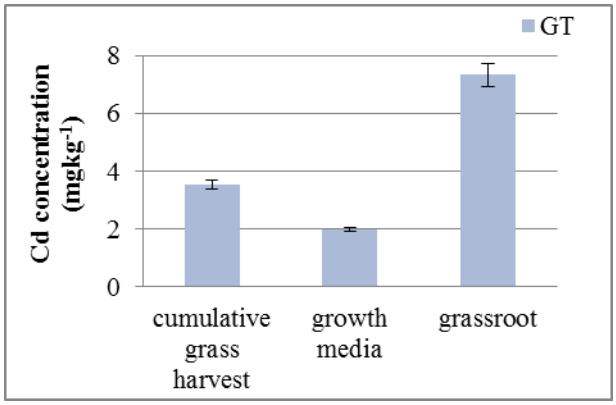

NBT
Cd concentration (mgkg-1)
0
2
4
6
8
10
12
cumulative grass harvest
growth media
grassroot

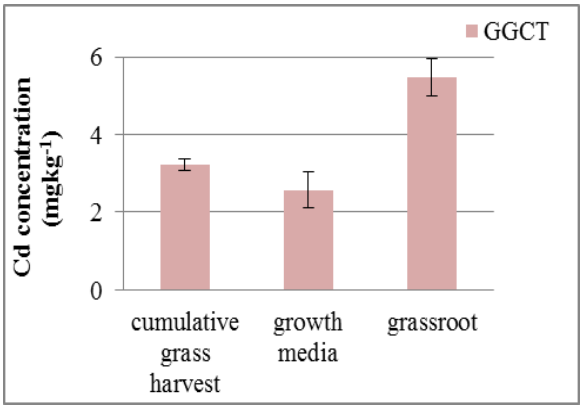

NBGCT
Cd concentration (mgkg-1)
0
1
2
3
4
5
6
7
cumulative grass harvest
growth media
grassroot


NBMCT
Cd concentration (mgkg-1)
0
1
2
3
4
5
cumulative grass harvest
growth media
grassroot


GGC
Cd concentration (mgkg-1)
0
1
2
3
4
5
6
7
cumulative grass harvest
growth media
grassroot


GMC
Cd concentration (mgkg-1)
0
2
4
6
8
10
cumulative grass harvest
growth media
grassroot


GT
Cd concentration (mgkg-1)
0
2
4
6
8
cumulative grass harvest
growth media
grassroot


GGCT
Cd concentration (mgkg-1)
0
2
4
6
cumulative grass harvest
growth media
grassroot

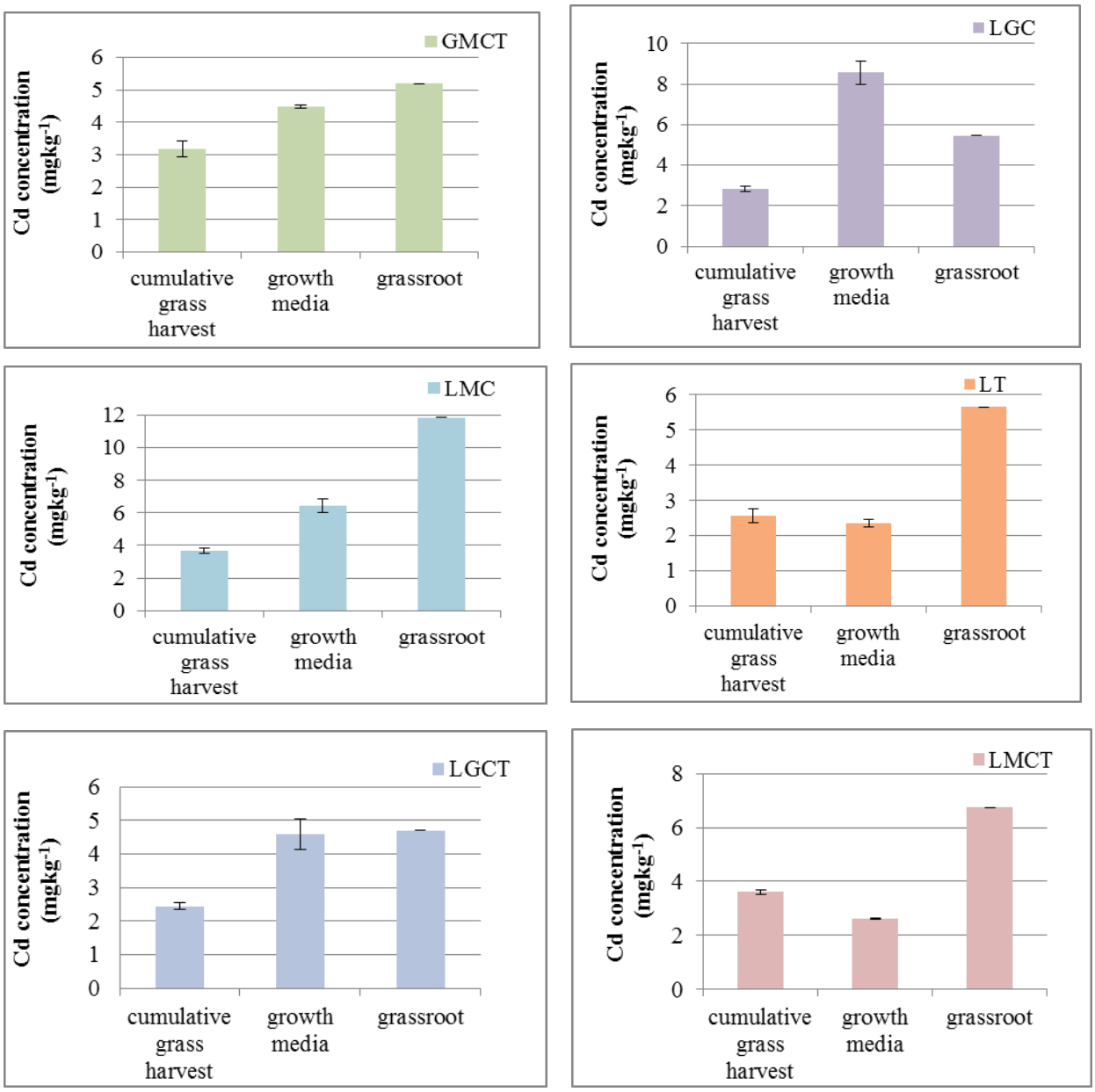
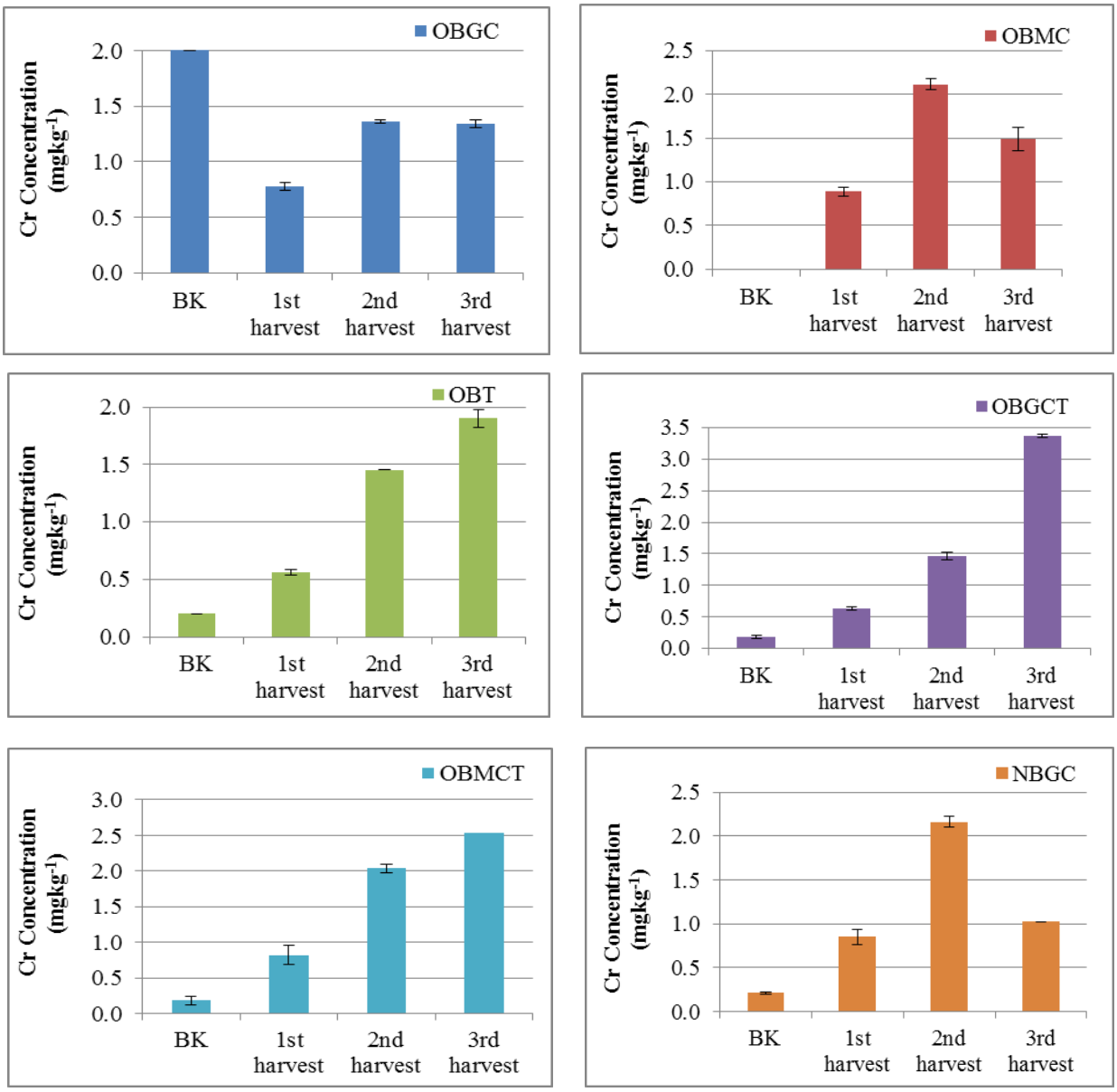
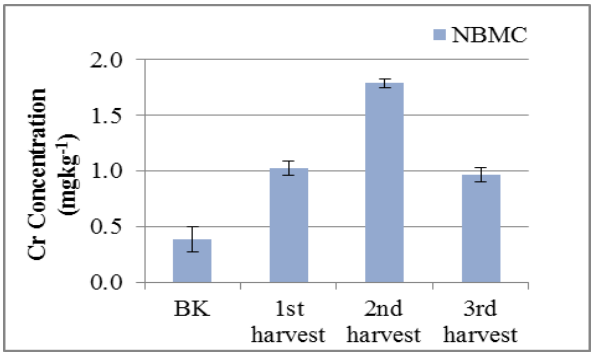


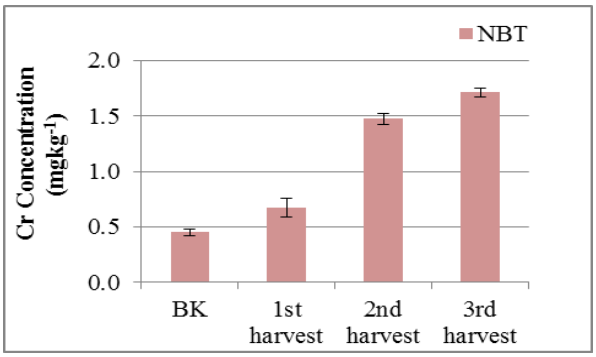
Figure 4.14d: Trends of cadmium concentrations in cumulative grass harvests, growth media and grass roots

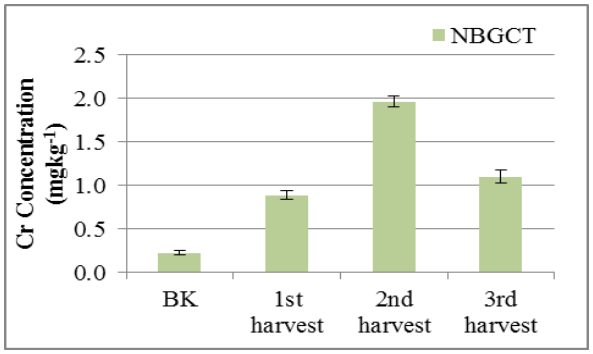
Chromium

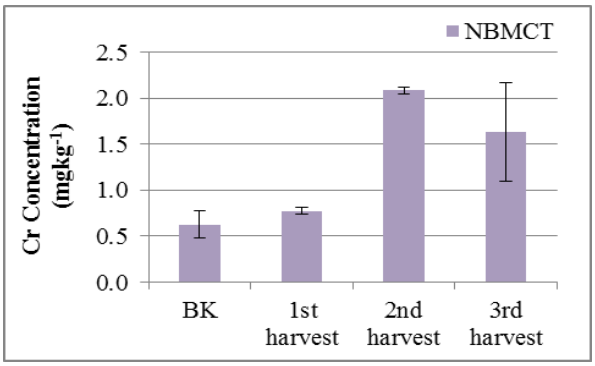
Figure 4.14e shows that 1st, 2nd and 3rd grass Cr concentrations were mostly higher than background concentrations, with the 1st harvest having the lowest Cr uptake and the 2nd harvest having the highest Cr uptake in most profiles.

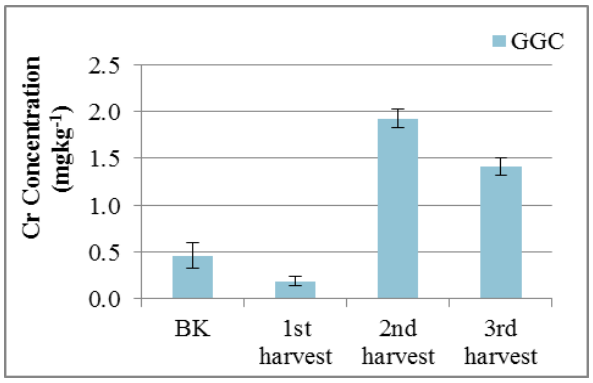


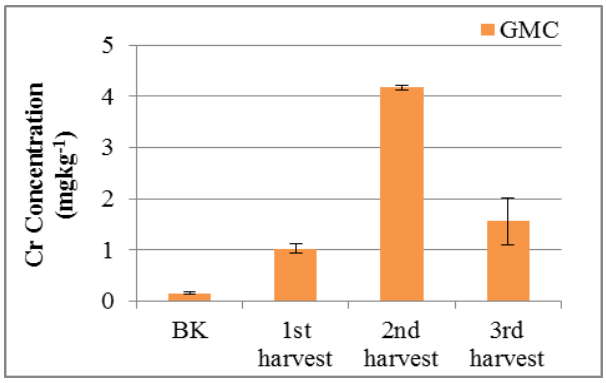

NBMC
Cr Concentration (mgkg-1)
2.0
1.5
1.0
0.5
0.0
BK
1st harvest
2nd harvest
3rd harvest

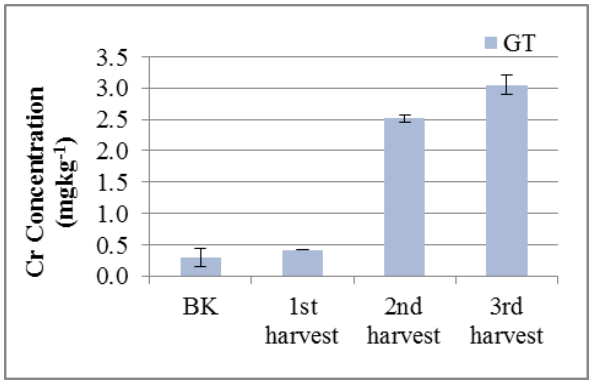

NBT
Cr Concentration (mgkg-1)
2.0
1.5
1.0
0.5
0.0
BK
1st harvest
2nd harvest
3rd harvest

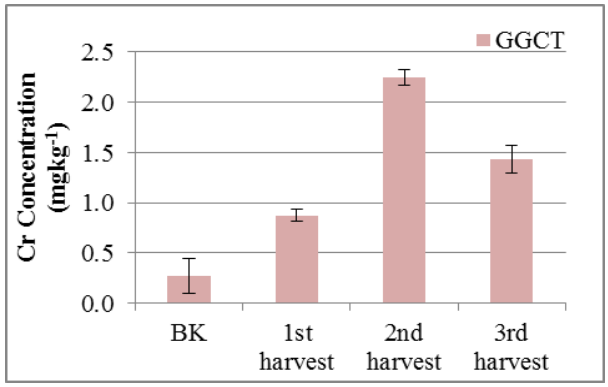

NBGCT
Cr Concentration (mgkg-1)
2.5
2.0
1.5
1.0
0.5
0.0
BK
1st harvest
2nd harvest
3rd harvest


NBMCT
Cr Concentration (mgkg-1)
2.5
2.0
1.5
1.0
0.5
0.0
BK
1st harvest
2nd harvest
3rd harvest


GGC
Cr Concentration (mgkg-1)
2.5
2.0
1.5
1.0
0.5
0.0
BK
1st harvest
2nd harvest
3rd harvest


GMC
Cr Concentration (mgkg-1)
5
4
3
2
1
0
BK
1st harvest
2nd harvest
3rd harvest


GT
Cr Concentration (mgkg-1)
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
BK
1st harvest
2nd harvest
3rd harvest


GGCT
Cr Concentration (mgkg-1)
2.5
2.0
1.5
1.0
0.5
0.0
BK
1st harvest
2nd harvest
3rd harvest

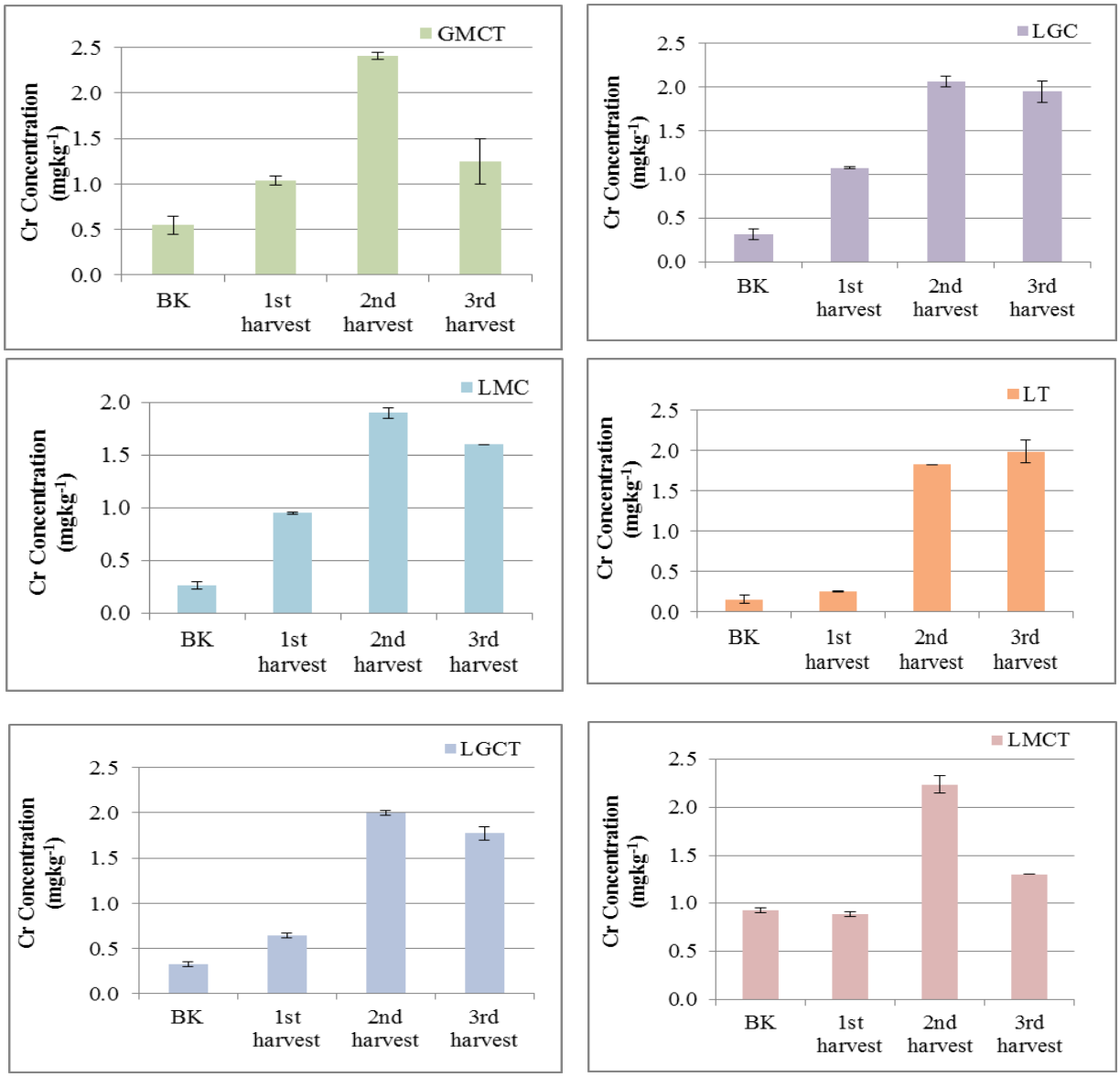
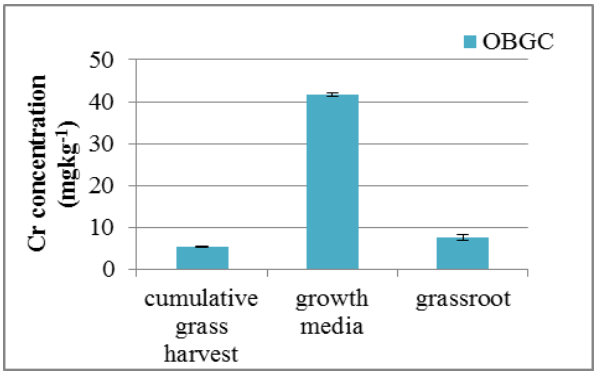
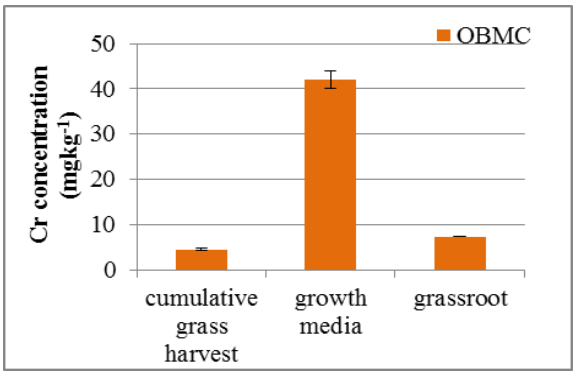
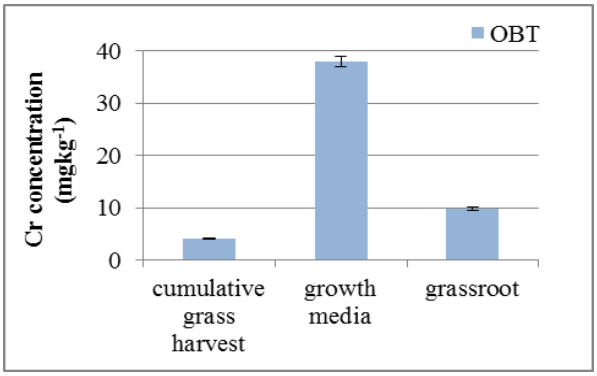


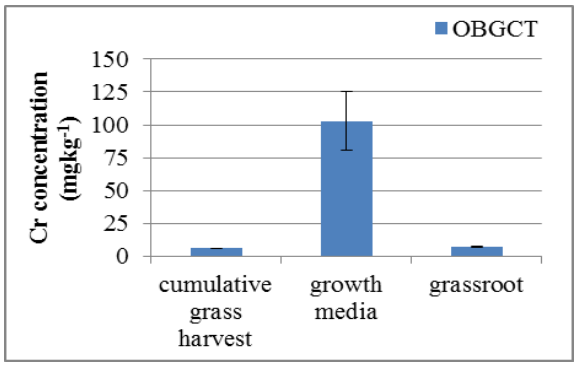
Figure 4.14e: Trends of chromium concentrations in background and three monthly grass harvests derived from test profiles.

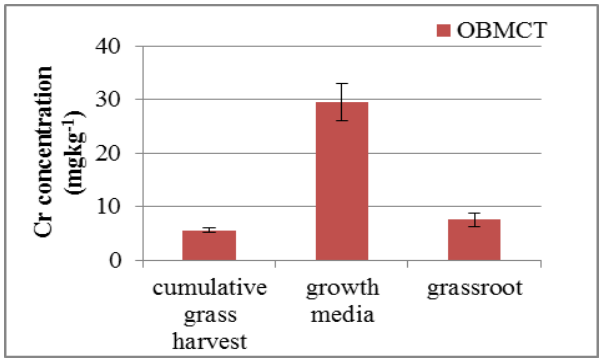
Figure 4.14f shows that growth media retained the highest Cr concentrations in all profiles compared to grasses and grass roots whose concentrations were quite low. GGC retained the highest Cr in growth media.

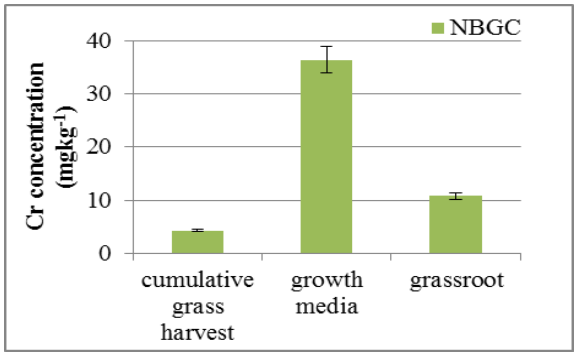


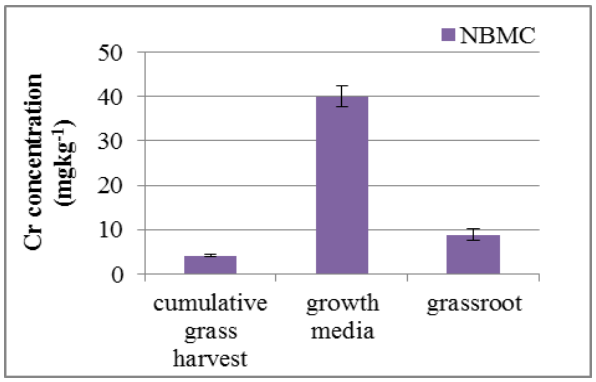


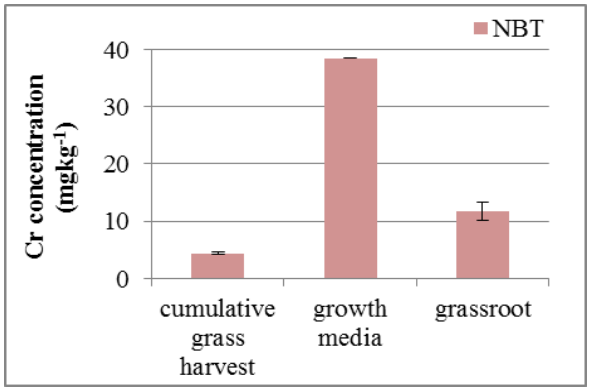


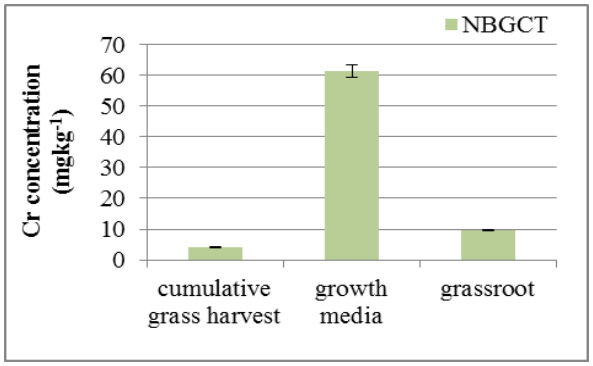


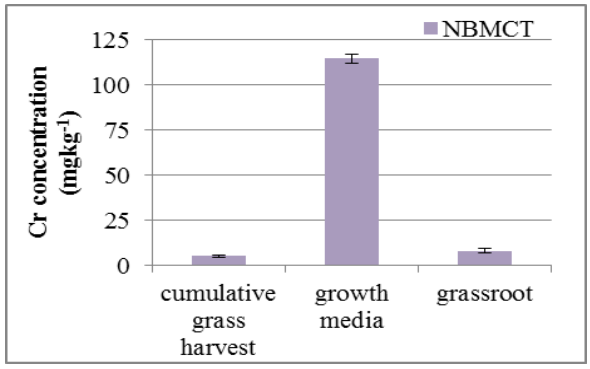


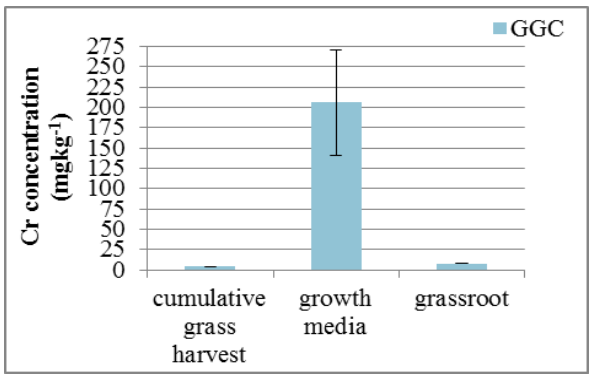


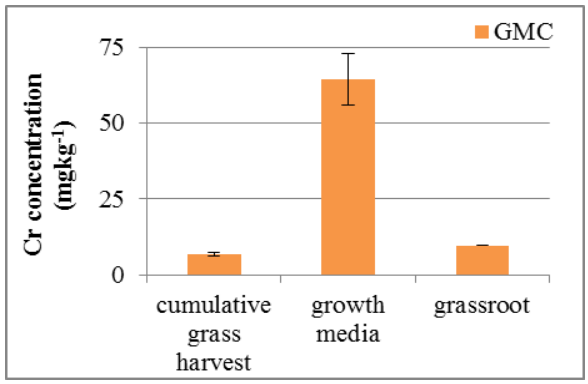

NBMC
Cr concentration (mgkg-1)
50
40
30
20
10
0
cumulative grass harvest
growth media
grassroot

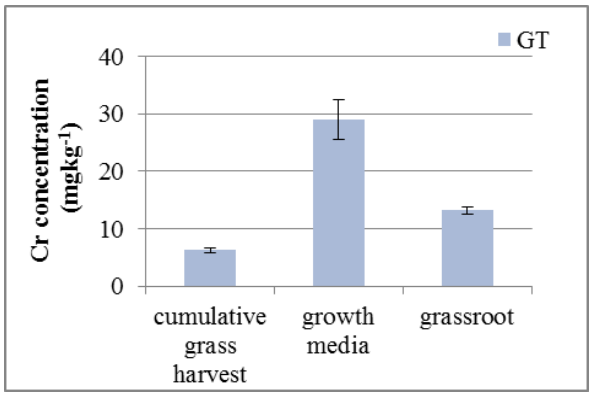

NBT
Cr concentration (mgkg-1)
40
30
20
10
0
cumulative grass harvest
growth media
grassroot

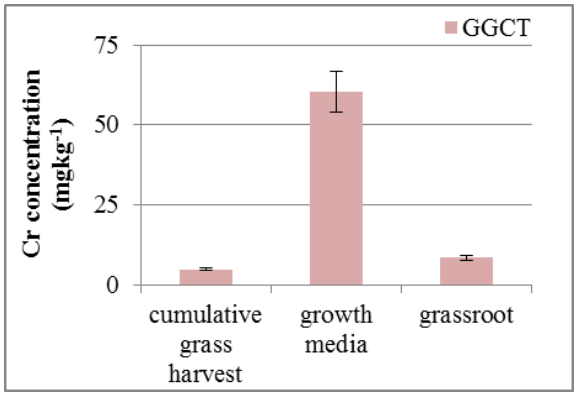

NBGCT
Cr concentration (mgkg-1)
70
60
50
40
30
20
10
0
cumulative grass harvest
growth media
grassroot


NBMCT
Cr concentration (mgkg-1)
125
100
75
50
25
0
cumulative grass harvest
growth media
grassroot


GGC
Cr concentration (mgkg-1)
275
250
225
200
175
150
125
100
75
50
25
0
cumulative grass harvest
growth media
grassroot


GMC
Cr concentration (mgkg-1)
75
50
25
0
cumulative grass harvest
growth media
grassroot


GT
Cr concentration (mgkg-1)
40
30
20
10
0
cumulative grass harvest
growth media
grassroot


GGCT
Cr concentration (mgkg-1)
75
50
25
0
cumulative grass harvest
growth media
grassroot

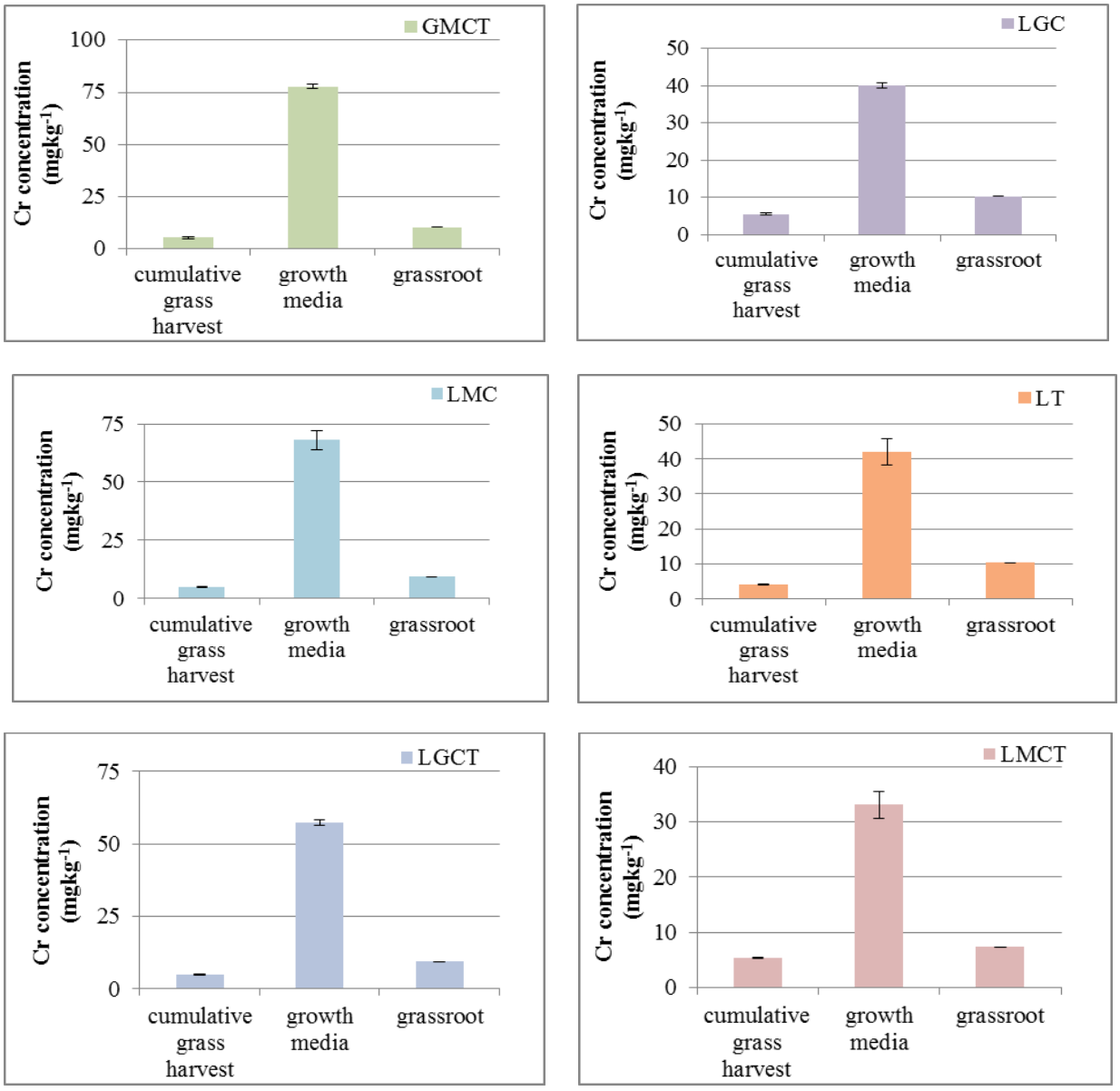
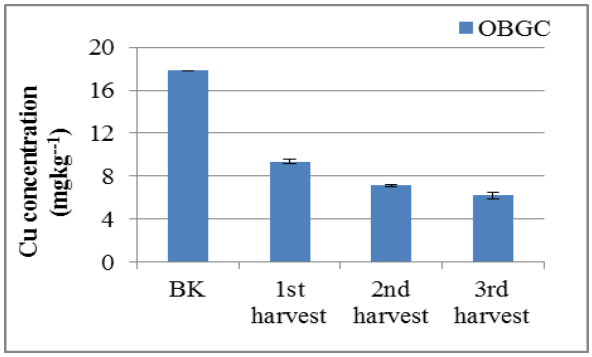
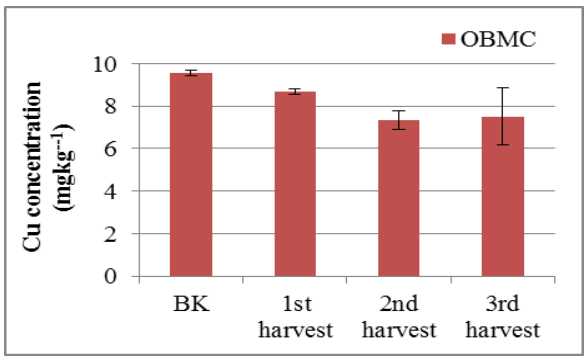


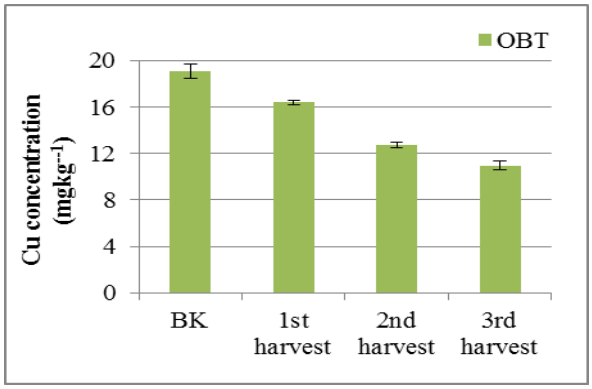
Figure 4.14f: Trends of chromium concentrations in cumulative grass harvests, growth media and grass roots

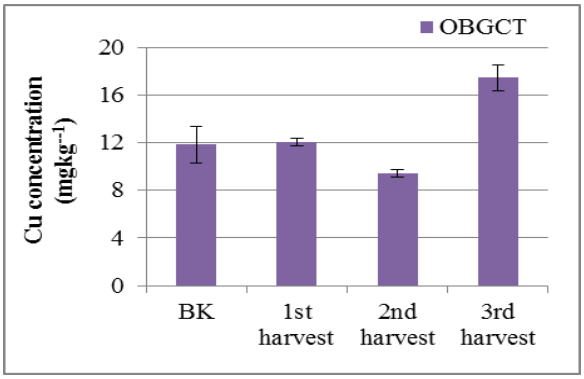
Copper

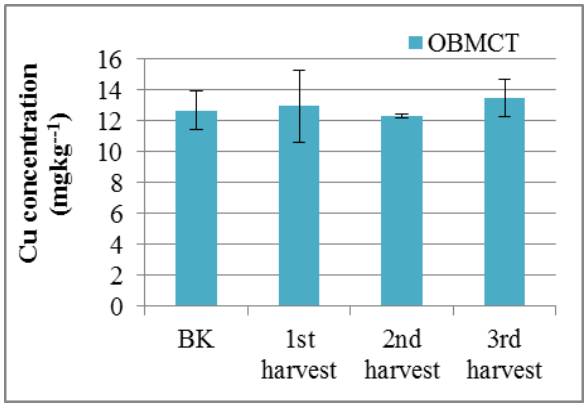
For most profiles, Cu concentrations in the 1st, 2nd and 3rd harvest were mostly below or around background concentrations (see figure 4.14g).

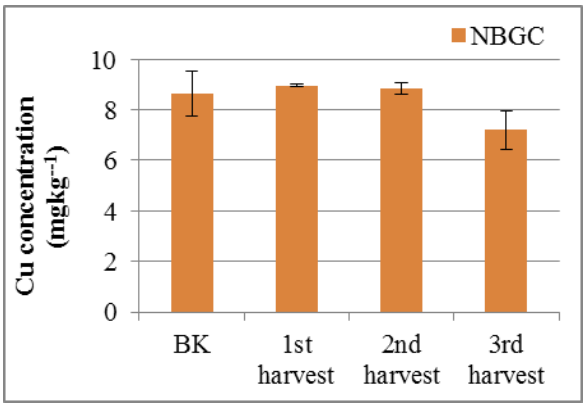


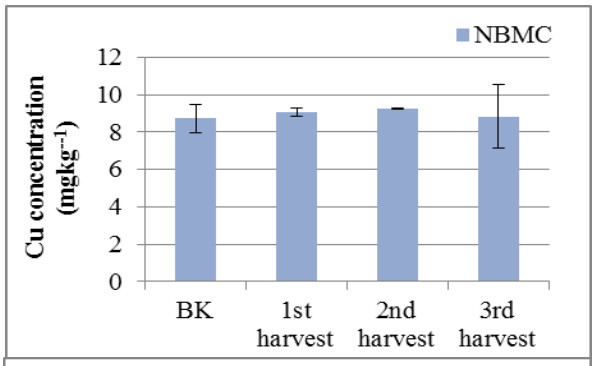


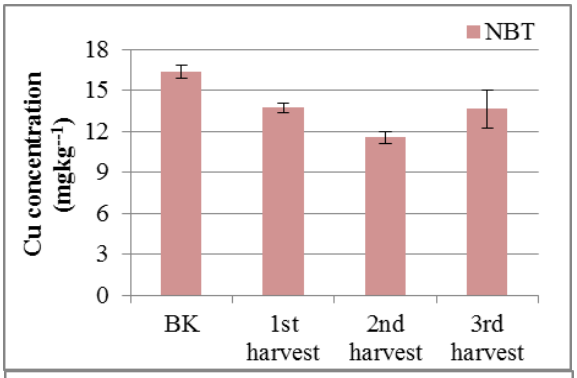


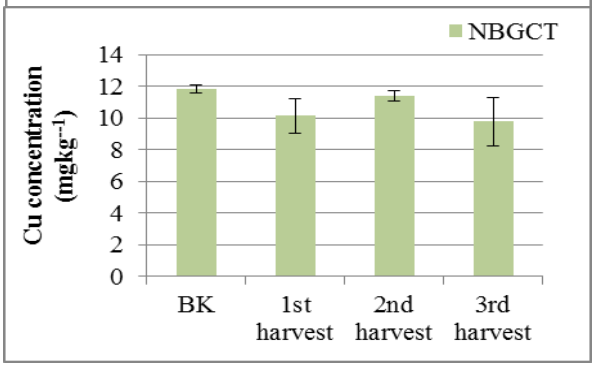


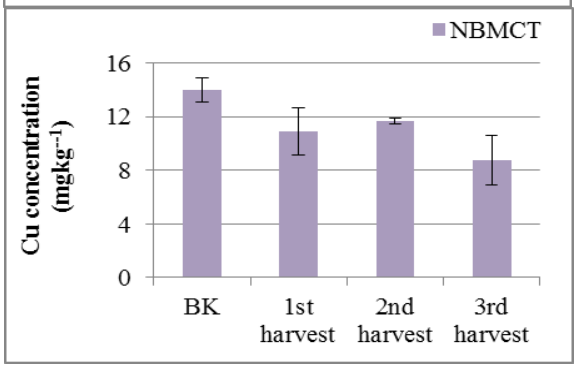


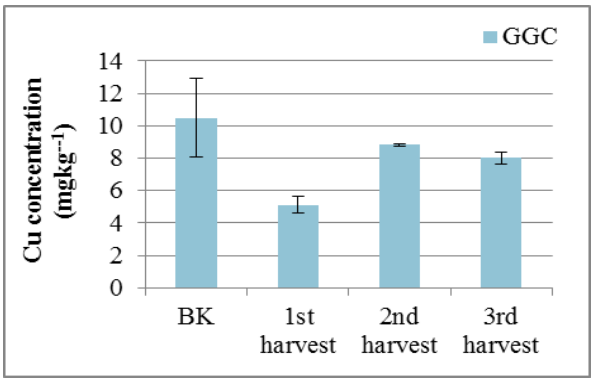


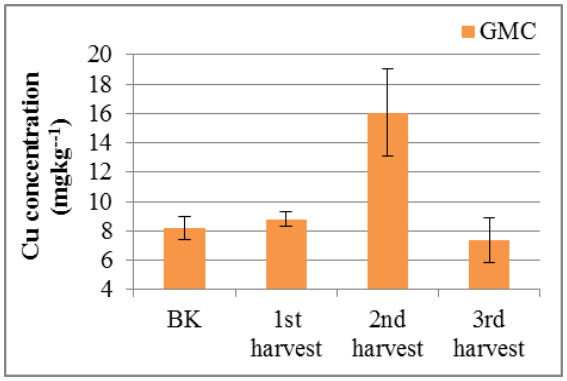

NBMC
Cu concentration (mgkg⁻¹)
12
10
8
6
4
2
0
BK
1st harvest
2nd harvest
3rd harvest

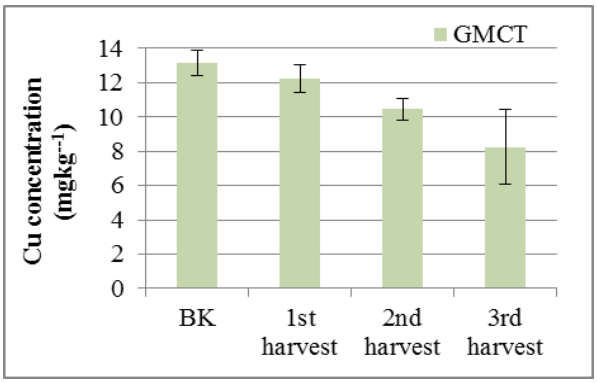

NBT
Cu concentration (mgkg⁻¹)
18
15
12
9
6
3
0
BK
1st harvest
2nd harvest
3rd harvest

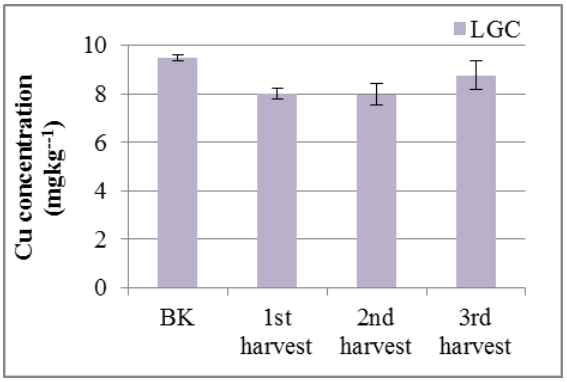

NBGCT
Cu concentration (mgkg⁻¹)
14
12
10
8
6
4
2
0
BK
1st harvest
2nd harvest
3rd harvest


NBMCT
Cu concentration (mgkg⁻¹)
16
12
8
4
0
BK
1st harvest
2nd harvest
3rd harvest


GGC
Cu concentration (mgkg⁻¹)
14
12
10
8
6
4
2
0
BK
1st harvest
2nd harvest
3rd harvest


GMC
Cu concentration (mgkg⁻¹)
20
18
16
14
12
10
8
6
4
BK
1st harvest
2nd harvest
3rd harvest


GMCT
Cu concentration (mgkg⁻¹)
14
12
10
8
6
4
2
0
BK
1st harvest
2nd harvest
3rd harvest


LGC
Cu concentration (mgkg⁻¹)
10
8
6
4
2
0
BK
1st harvest
2nd harvest
3rd harvest

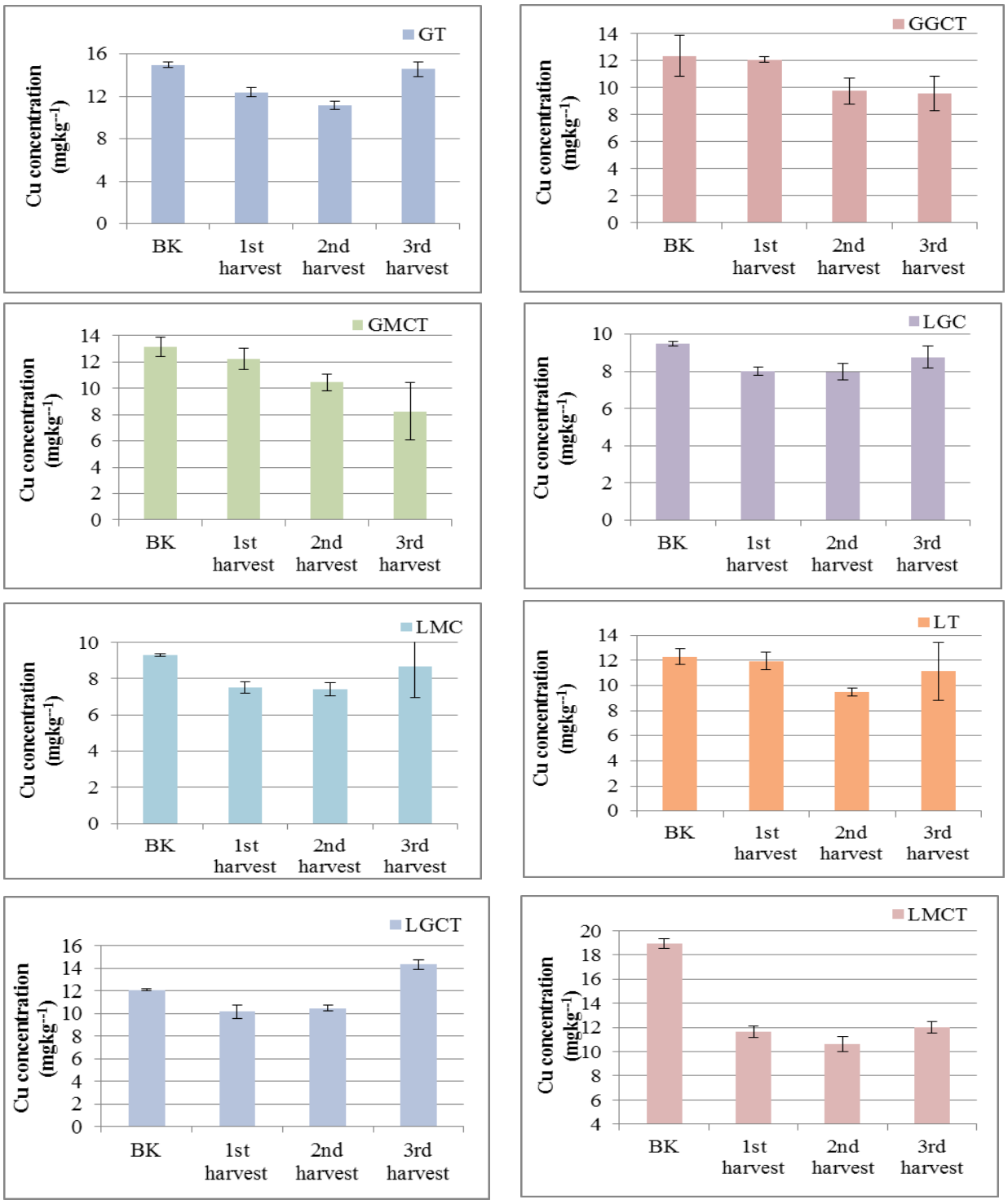
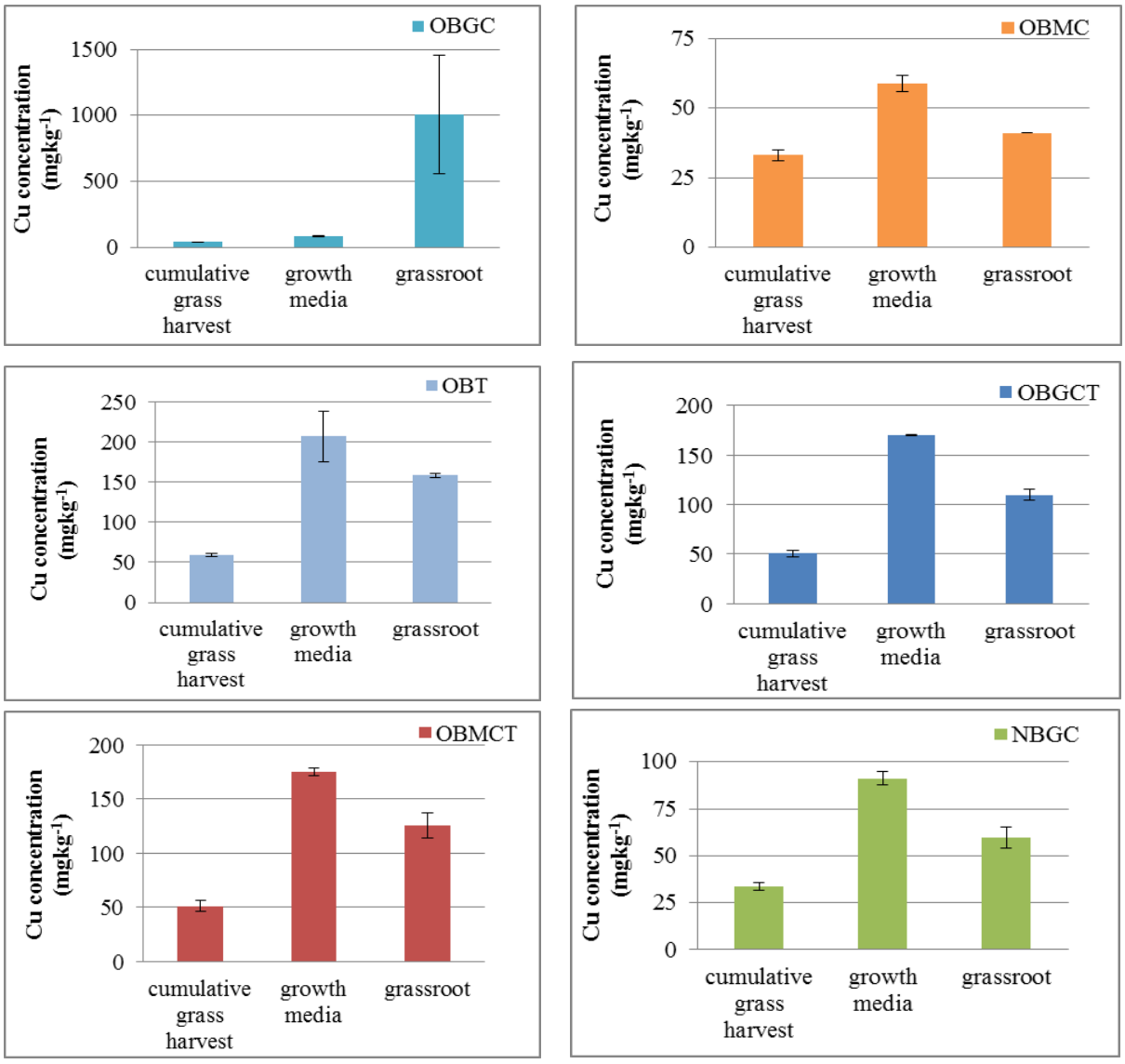
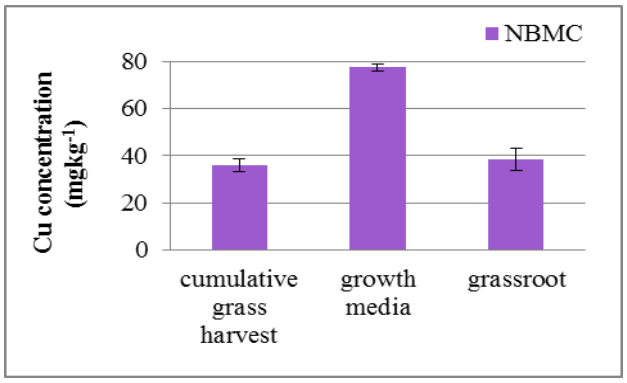
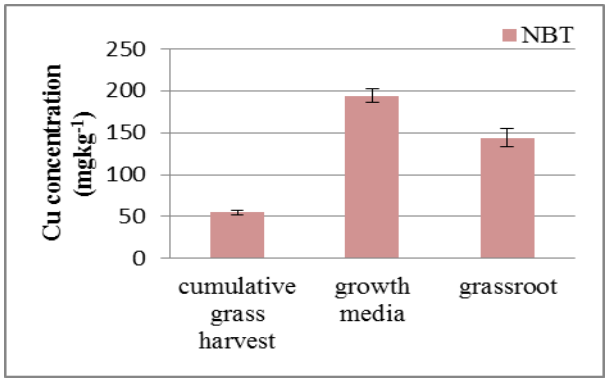


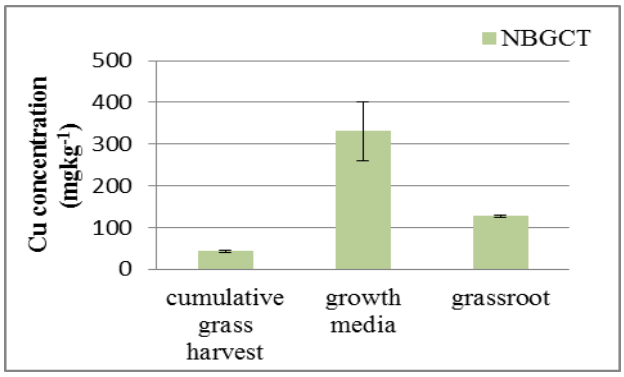
Figure 4.14g: Trends of copper concentrations in background and three monthly grass harvests derived from test profiles.

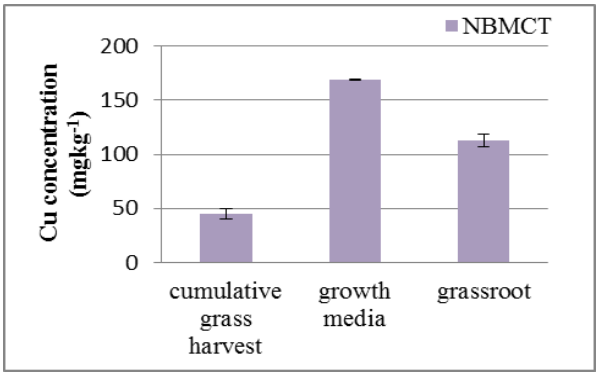
Generally, growth media retained the highest Cu concentrations closely followed by the grass root in all profiles, except for OBGC which had the highest concentrations in grassroots. GMCT retained the highest Cu concentrations in growth media, (see figure 4.14h).

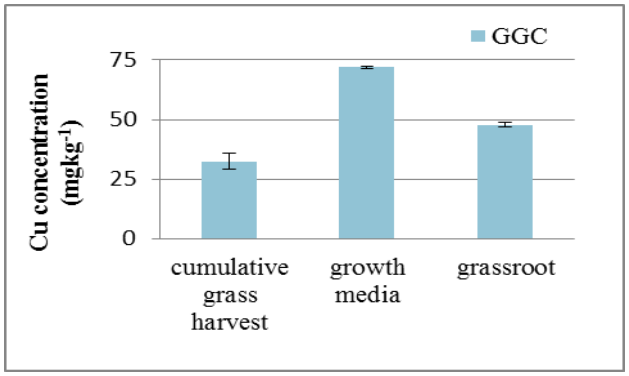


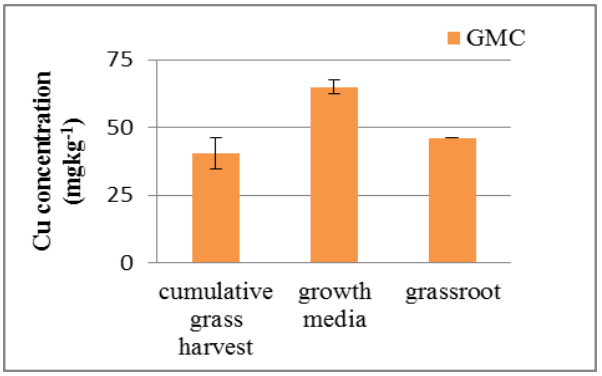

NBMC
Cu concentration (mgkg-1)
80
60
40
20
0
cumulative grass harvest
growth media
grassroot

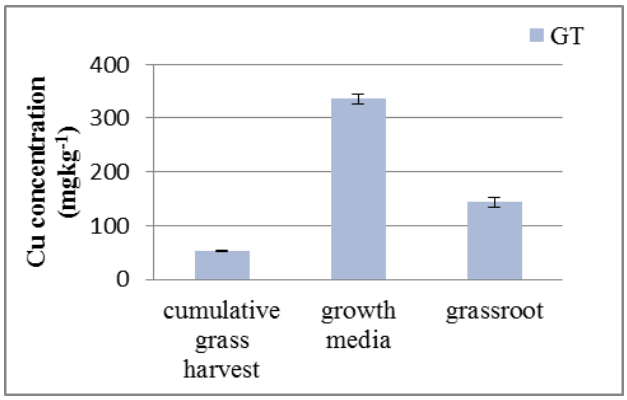

NBT
Cu concentration (mgkg-1)
250
200
150
100
50
0
cumulative grass harvest
growth media
grassroot

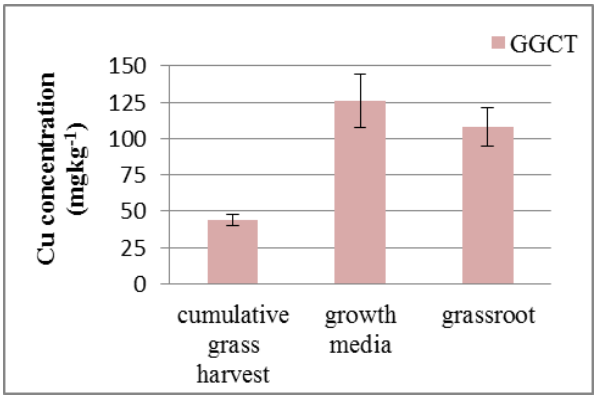

NBGCT
Cu concentration (mgkg-1)
500
400
300
200
100
0
cumulative grass harvest
growth media
grassroot


NBMCT
Cu concentration (mgkg-1)
200
150
100
50
0
cumulative grass harvest
growth media
grassroot


GGC
Cu concentration (mgkg-1)
75
50
25
0
cumulative grass harvest
growth media
grassroot


GMC
Cu concentration (mgkg-1)
75
50
25
0
cumulative grass harvest
growth media
grassroot


GT
Cu concentration (mgkg-1)
400
300
200
100
0
cumulative grass harvest
growth media
grassroot


GGCT
Cu concentration (mgkg-1)
150
125
100
75
50
25
0
cumulative grass harvest
growth media
grassroot

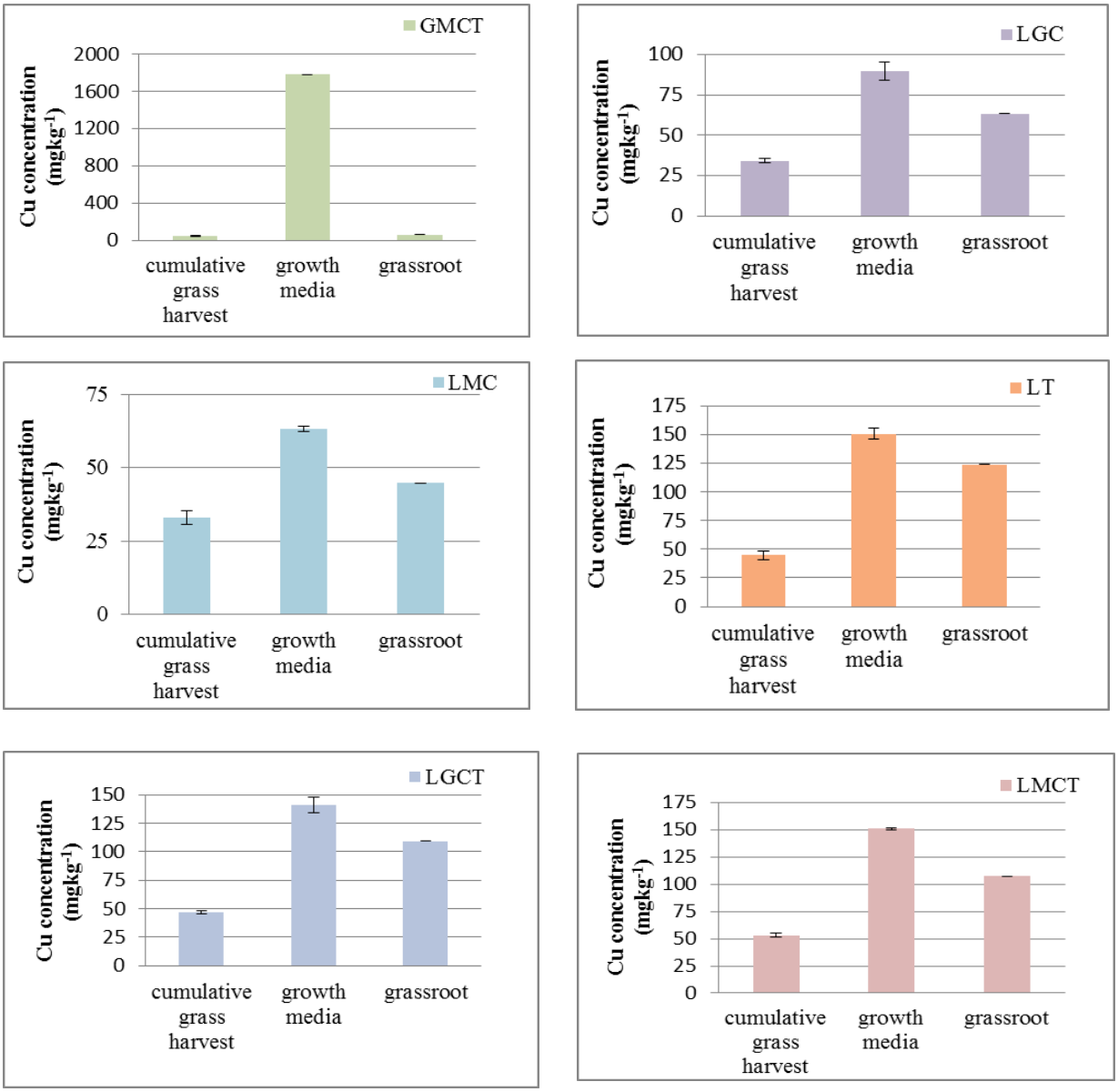
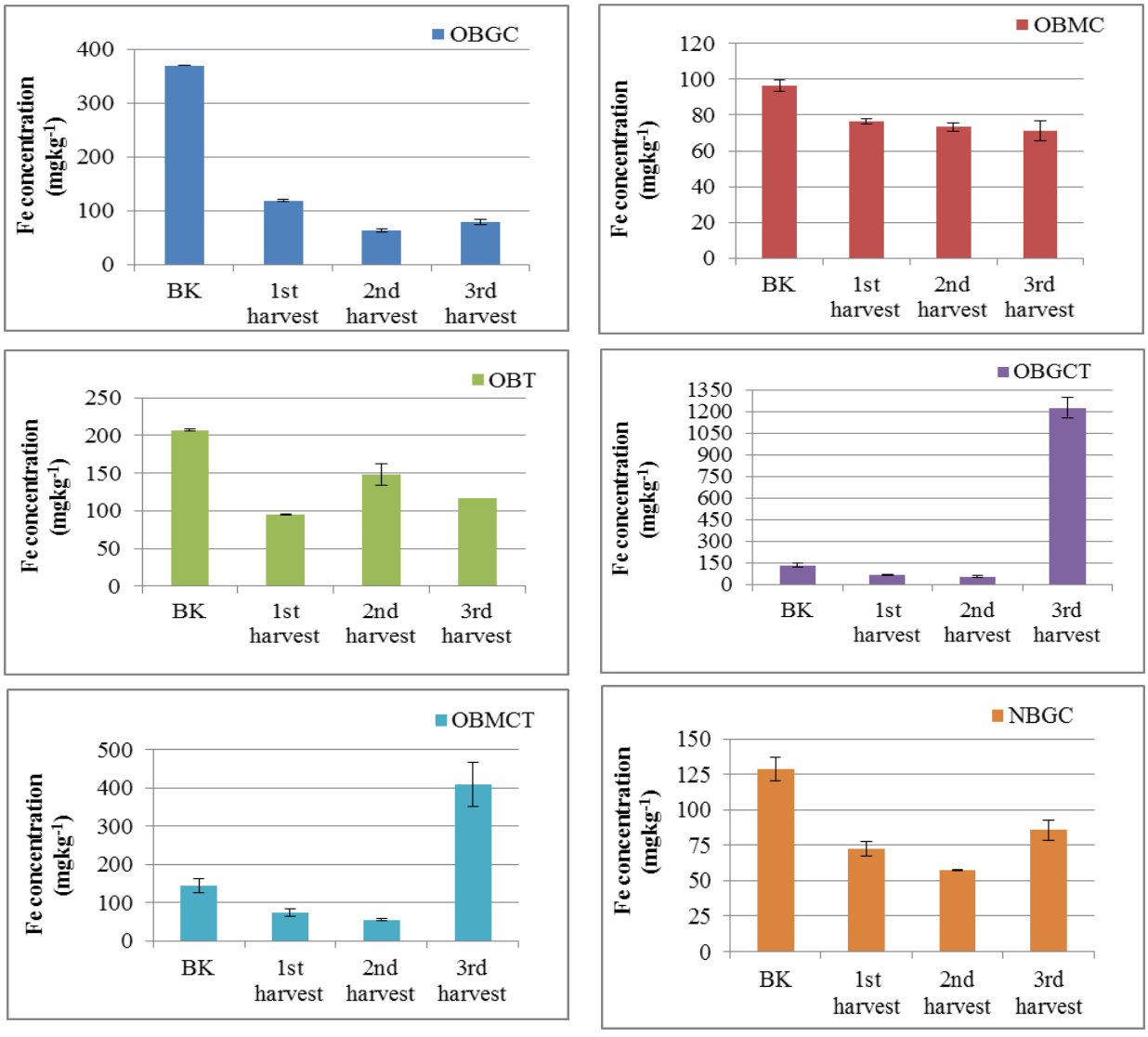
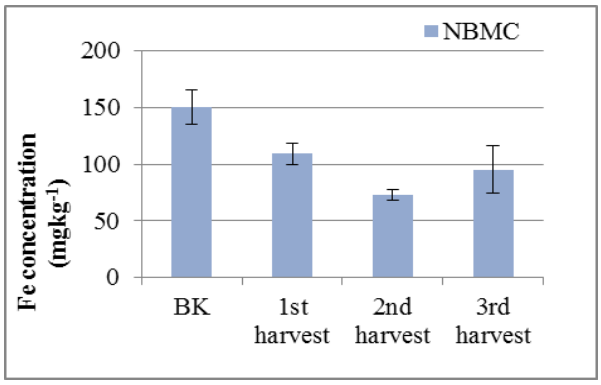


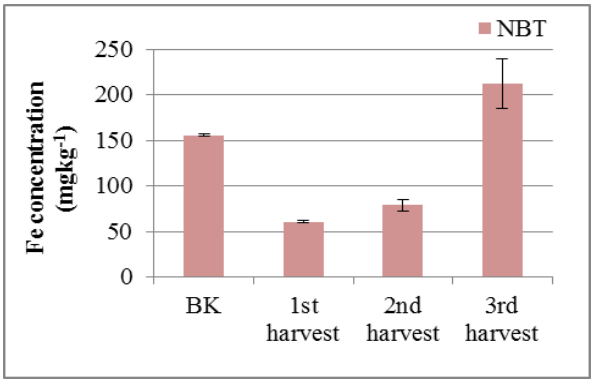
Figure 4.14h: Trends of copper concentrations in cumulative grass harvests, growth media and grass roots

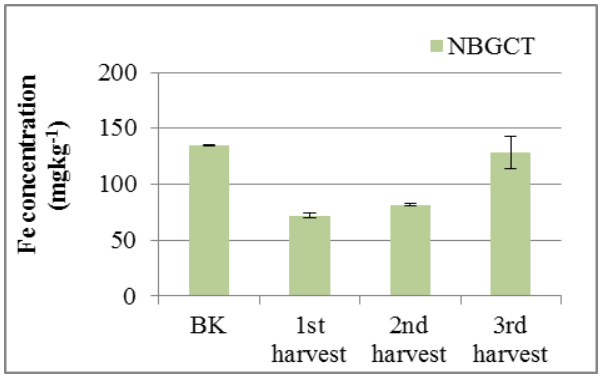
Iron

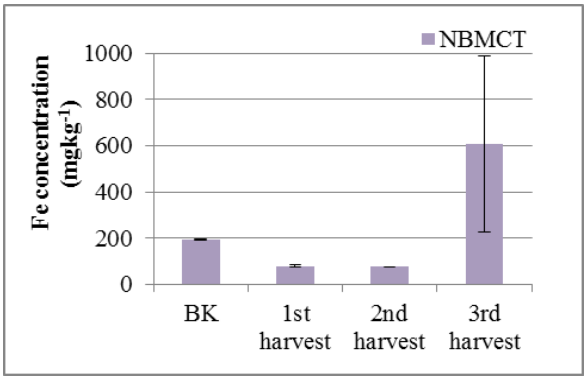
As shown in figure 4.14i, 1st and 2nd harvests were predominantly below background concentrations while the 3rd harvest exceeded background concentrations in about half of the profiles. OBGCT followed by NBMCT grasses retained the highest Fe concentrations.

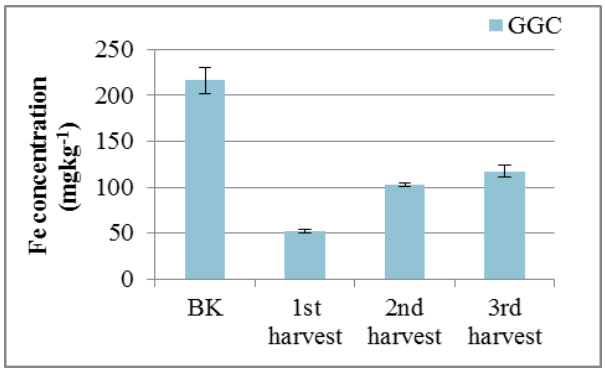


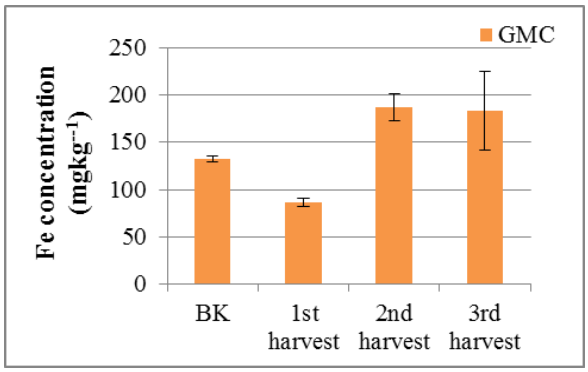

NBMC
Fe concentration (mgkg-1)
200
150
100
50
0
BK
1st harvest
2nd harvest
3rd harvest

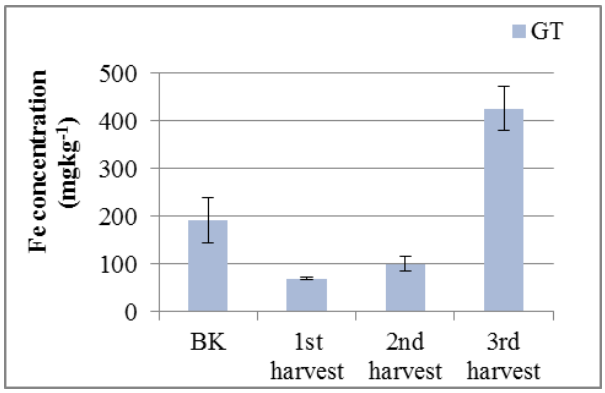

NBT
Fe concentration (mgkg-1)
250
200
150
100
50
0
BK
1st harvest
2nd harvest
3rd harvest

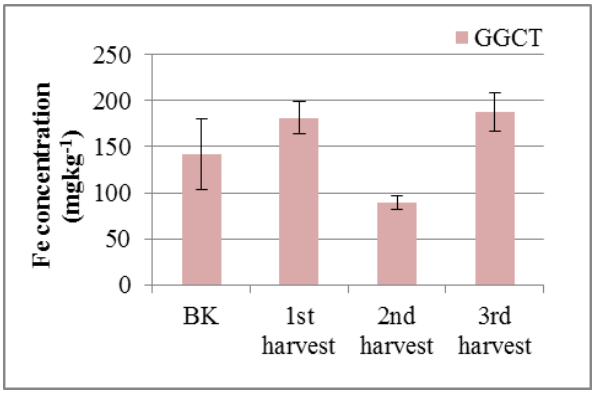

NBGCT
Fe concentration (mgkg-1)
200
150
100
50
0
BK
1st harvest
2nd harvest
3rd harvest


NBMCT
Fe concentration (mgkg-1)
1000
800
600
400
200
0
BK
1st harvest
2nd harvest
3rd harvest


GGC
Fe concentration (mgkg-1)
250
200
150
100
50
0
BK
1st harvest
2nd harvest
3rd harvest


GMC
Fe concentration (mgkg-1)
250
200
150
100
50
0
BK
1st harvest
2nd harvest
3rd harvest


GT
Fe concentration (mgkg-1)
500
400
300
200
100
0
BK
1st harvest
2nd harvest
3rd harvest


GGCT
Fe concentration (mgkg-1)
250
200
150
100
50
0
BK
1st harvest
2nd harvest
3rd harvest

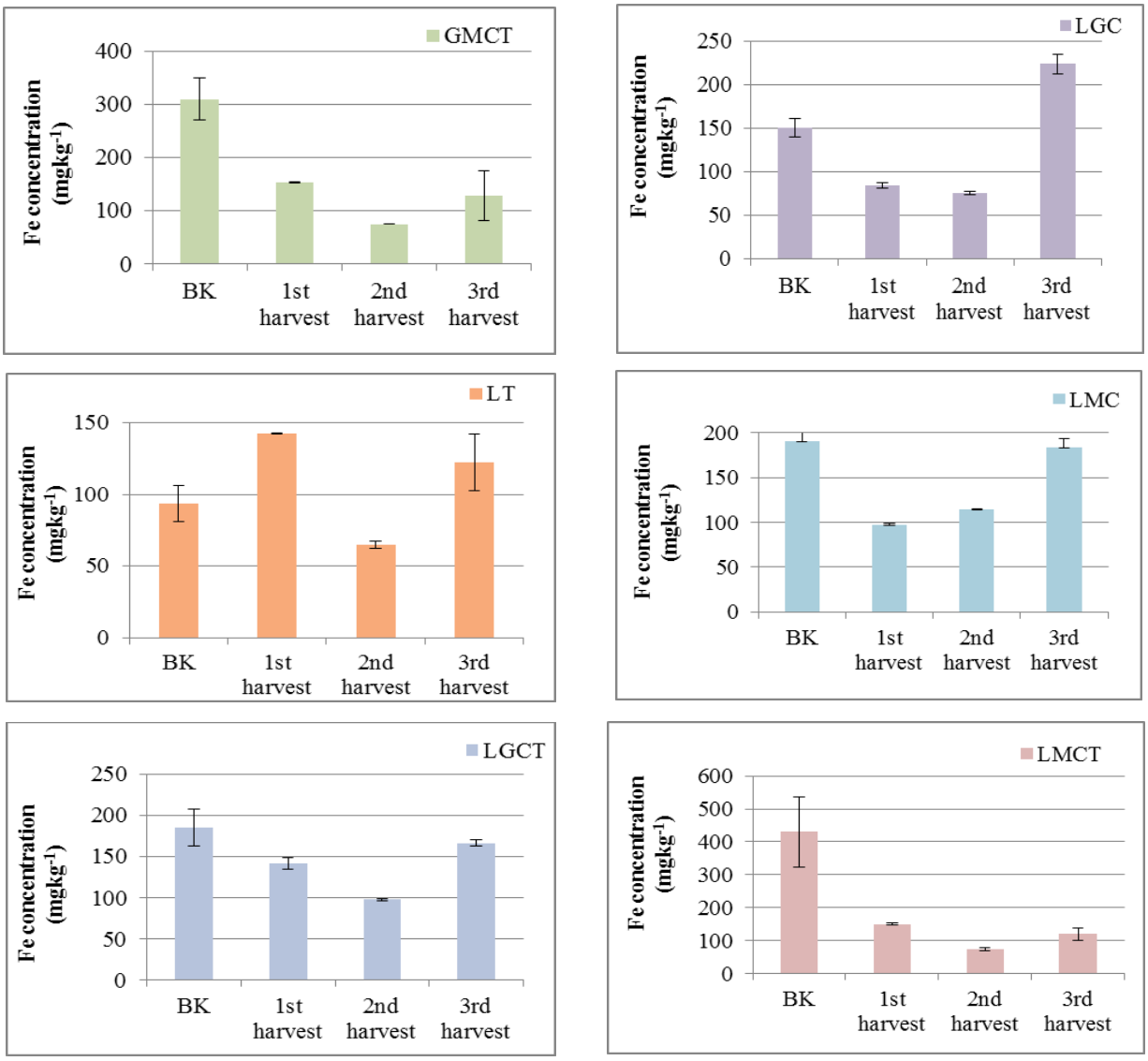
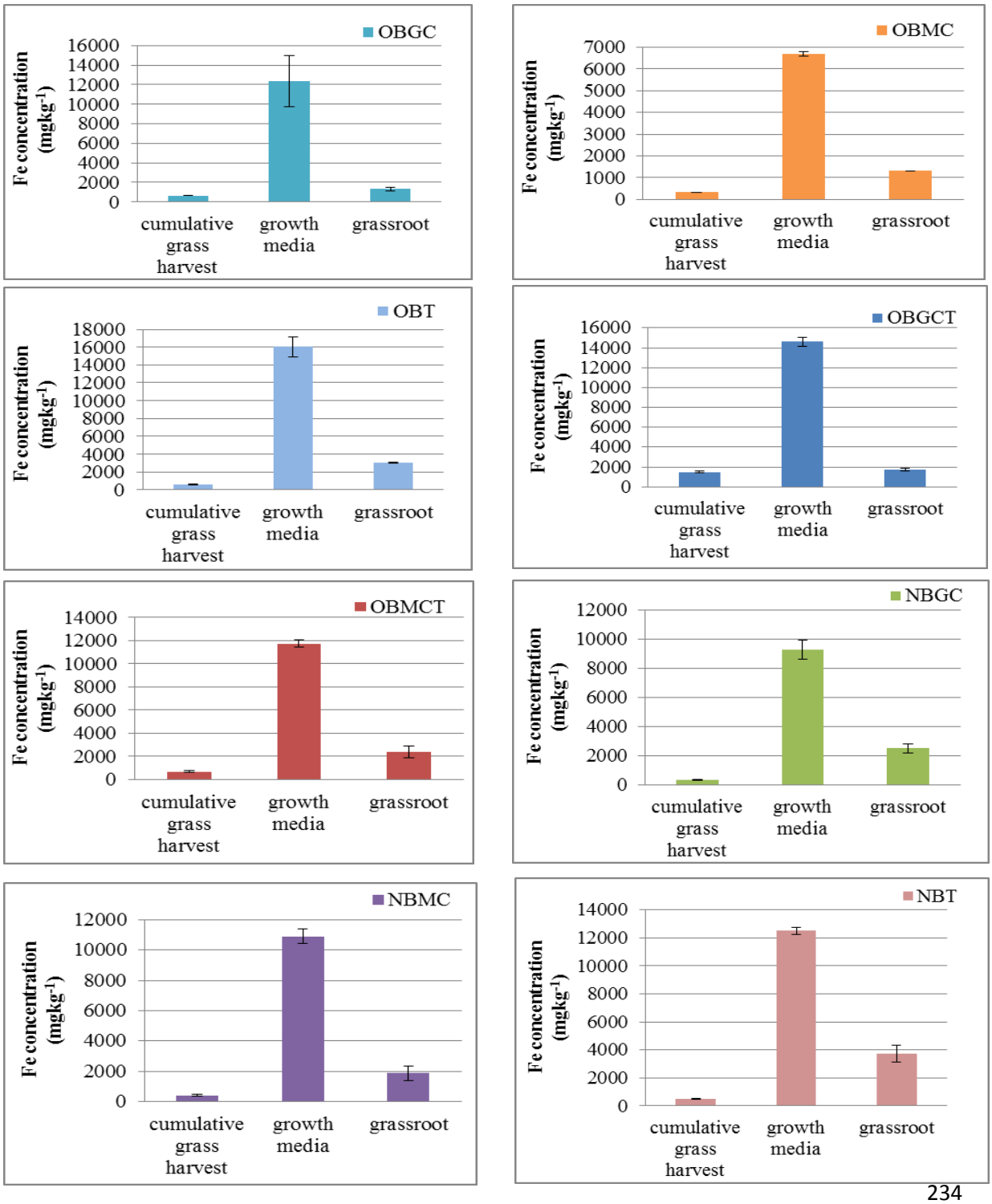
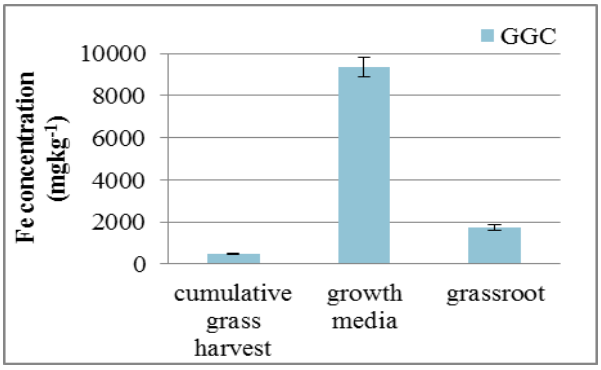
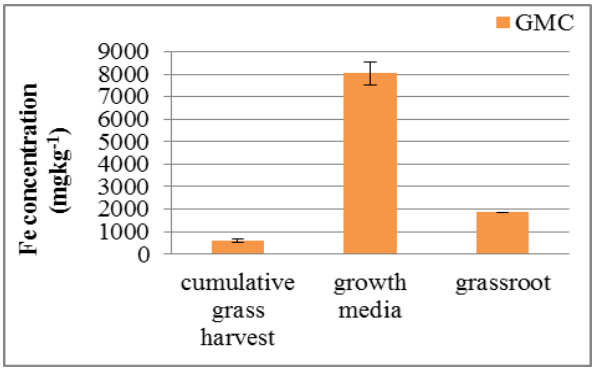


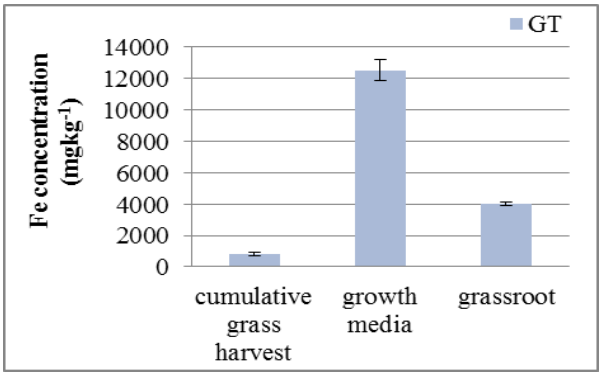
Figure 4.14i: Trends of iron concentrations in background and three monthly grass harvests derived from test profiles

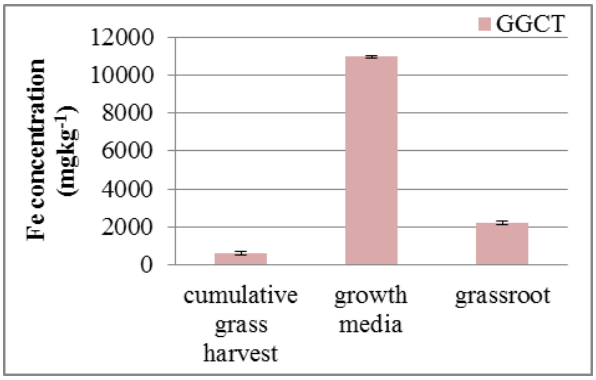
Figure 4.14j shows that the highest Fe concentrations were retained within the growth media followed by the grass roots with the least concentrations in grasses.

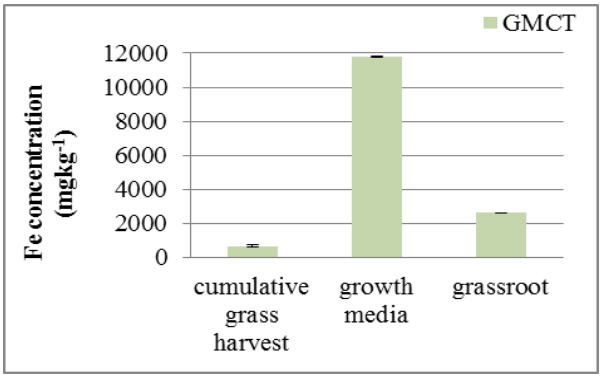


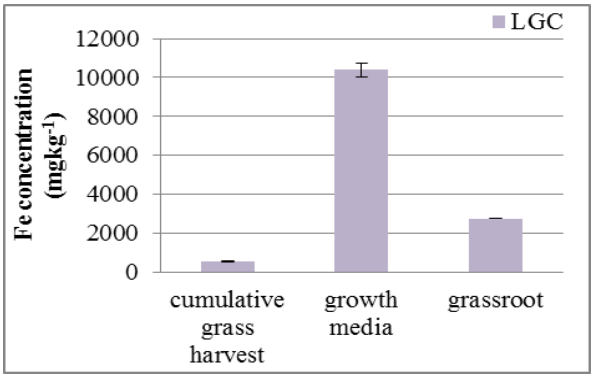

GGC
Fe concentration (mgkg-1)
10000
8000
6000
4000
2000
0
cumulative grass harvest
growth media
grassroot

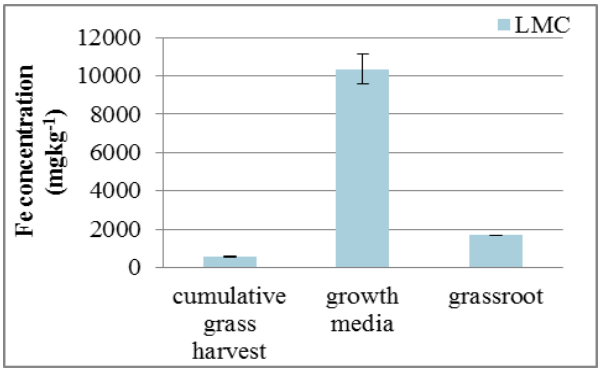

GMC
Fe concentration (mgkg-1)
9000
8000
7000
6000
5000
4000
3000
2000
1000
0
cumulative grass harvest
growth media
grassroot

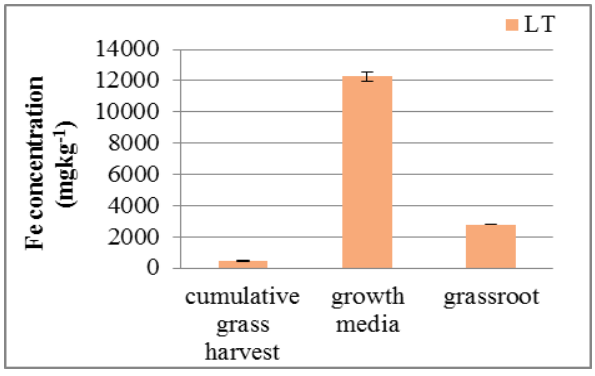

GT
Fe concentration (mgkg-1)
14000
12000
10000
8000
6000
4000
2000
0
cumulative grass harvest
growth media
grassroot


GGCT
Fe concentration (mgkg-1)
12000
10000
8000
6000
4000
2000
0
cumulative grass harvest
growth media
grassroot


GMCT
Fe concentration (mgkg-1)
12000
10000
8000
6000
4000
2000
0
cumulative grass harvest
growth media
grassroot


LGC
Fe concentration (mgkg-1)
12000
10000
8000
6000
4000
2000
0
cumulative grass harvest
growth media
grassroot


LMC
Fe concentration (mgkg-1)
12000
10000
8000
6000
4000
2000
0
cumulative grass harvest
growth media
grassroot


LT
Fe concentration (mgkg-1)
14000
12000
10000
8000
6000
4000
2000
0
cumulative grass harvest
growth media
grassroot

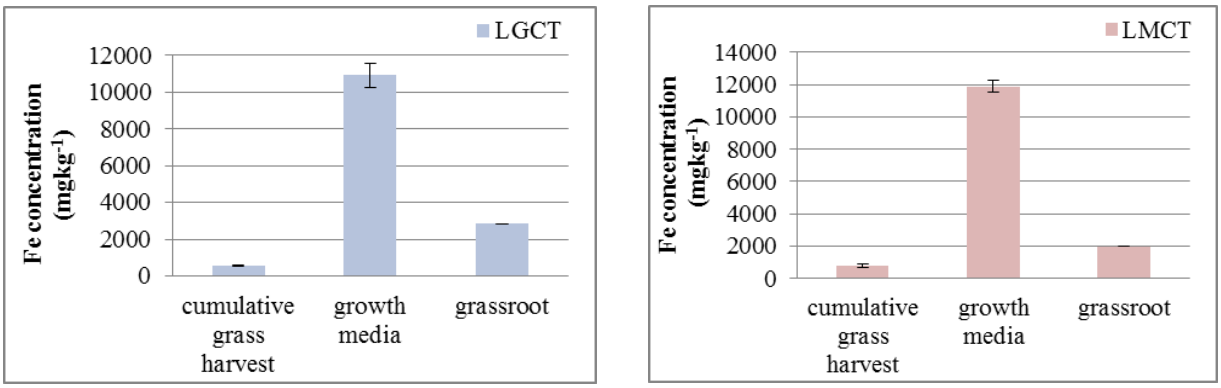
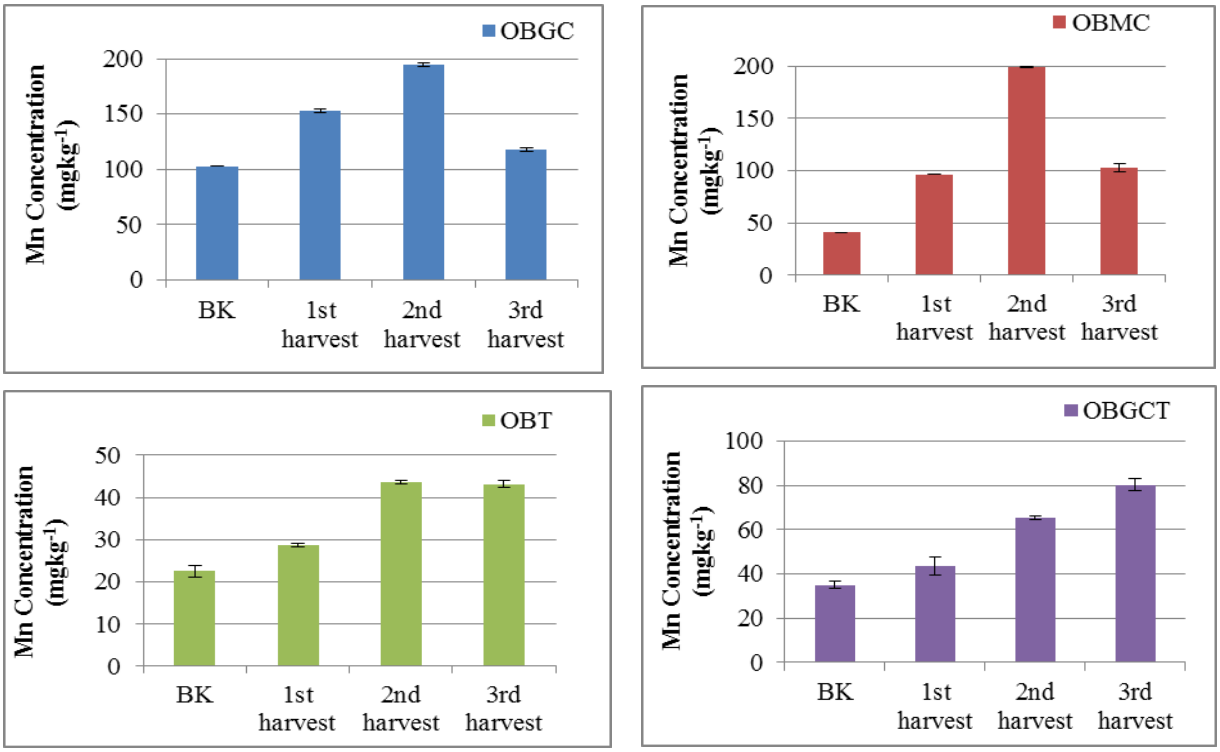
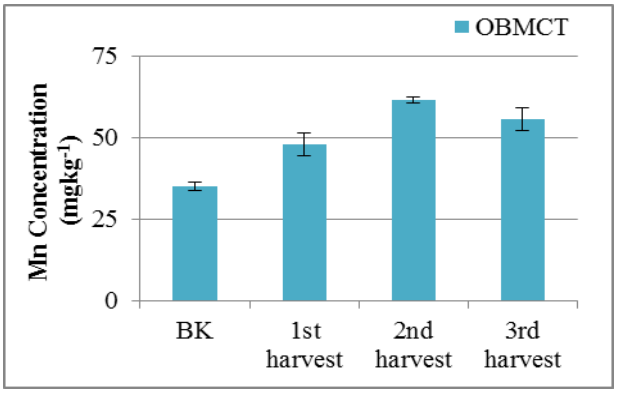


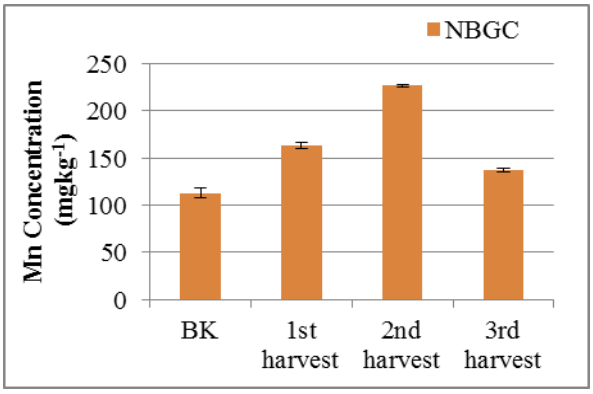
Figure 4.14j: Trends of iron concentrations in cumulative grass harvests, growth media and grass roots

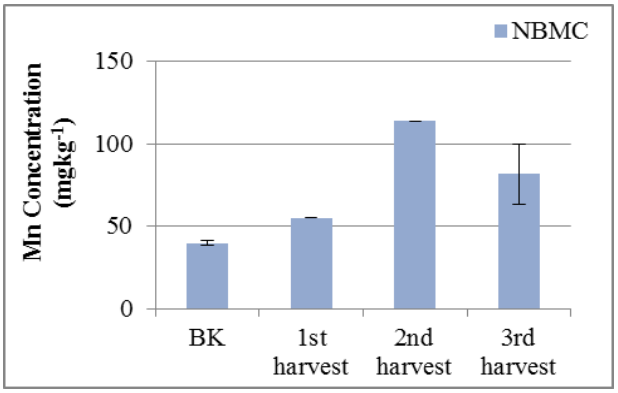
Manganese

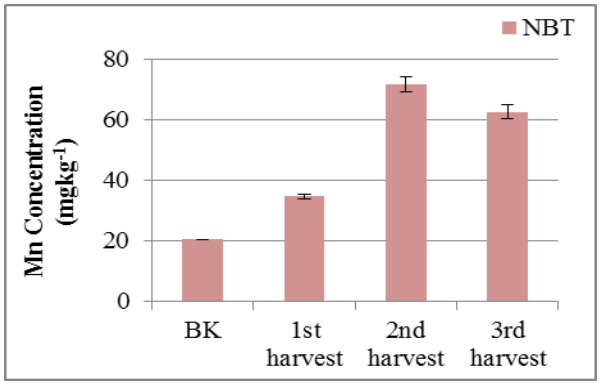
As shown in figure 4.14k, Mn concentrations in the three harvests were predominantly above background concentrations with the 2nd harvest having the highest concentrations in almost all the profiles.

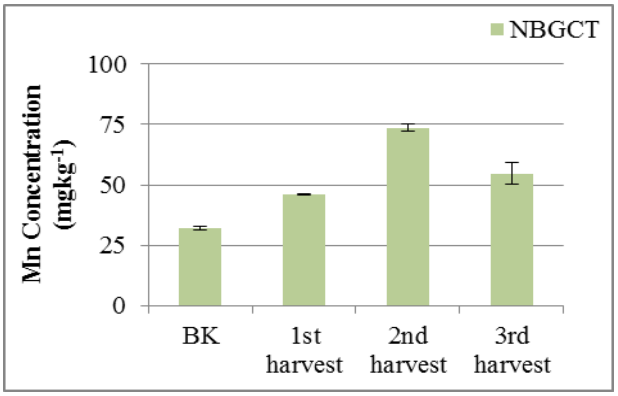


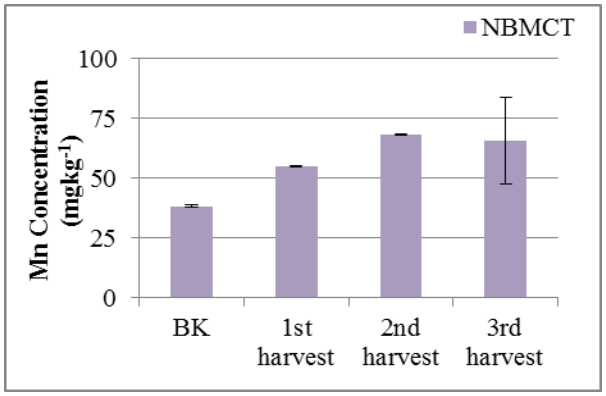

OBMCT
Mn Concentration (mgkg-1)
75
50
25
0
BK
1st harvest
2nd harvest
3rd harvest

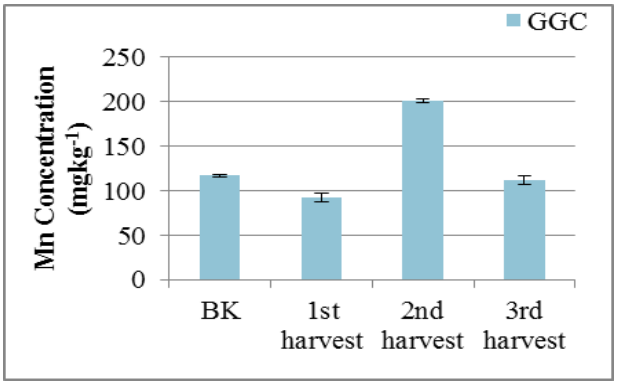

NBGC
Mn Concentration (mgkg-1)
250
200
150
100
50
0
BK
1st harvest
2nd harvest
3rd harvest

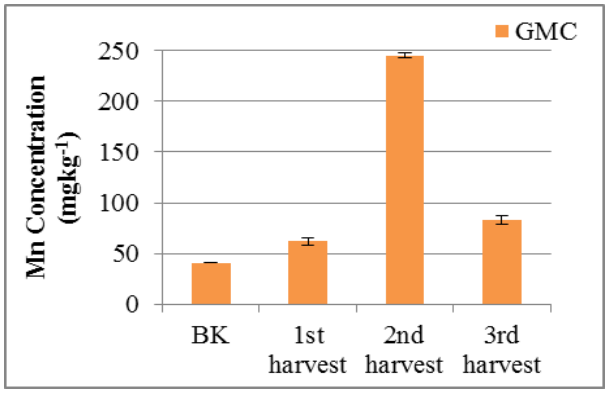

NBMC
Mn Concentration (mgkg-1)
150
100
50
0
BK
1st harvest
2nd harvest
3rd harvest


NBT
Mn Concentration (mgkg-1)
80
60
40
20
0
BK
1st harvest
2nd harvest
3rd harvest


NBGCT
Mn Concentration (mgkg-1)
100
75
50
25
0
BK
1st harvest
2nd harvest
3rd harvest


NBMCT
Mn Concentration (mgkg-1)
100
75
50
25
0
BK
1st harvest
2nd harvest
3rd harvest


GGC
Mn Concentration (mgkg-1)
250
200
150
100
50
0
BK
1st harvest
2nd harvest
3rd harvest


GMC
Mn Concentration (mgkg-1)
250
200
150
100
50
0
BK
1st harvest
2nd harvest
3rd harvest

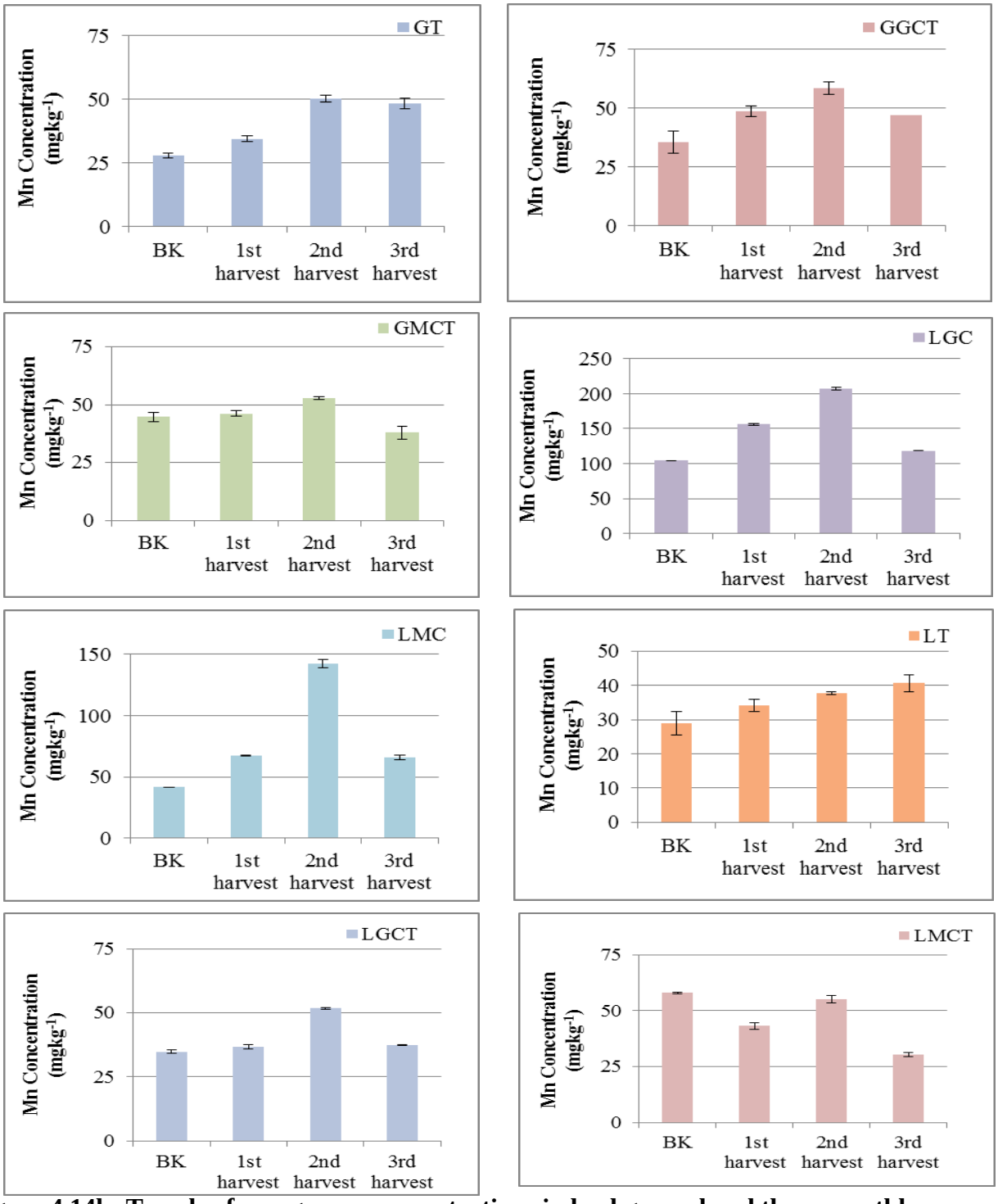
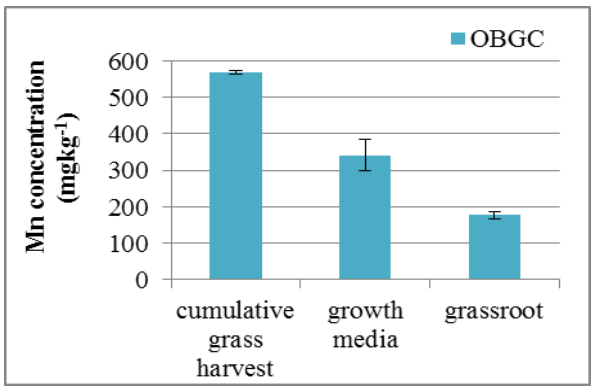
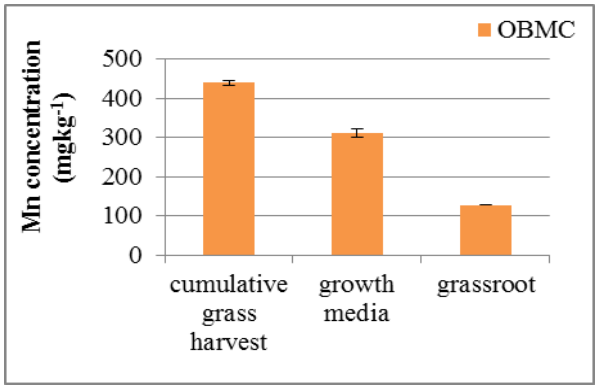
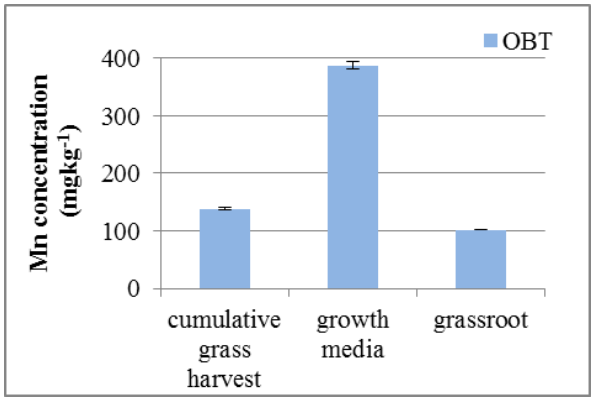


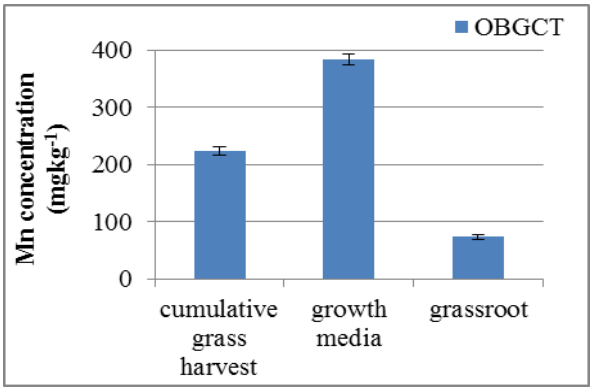
Figure 4.14k: Trends of manganese concentrations in background and three monthly grass harvests derived from test profiles

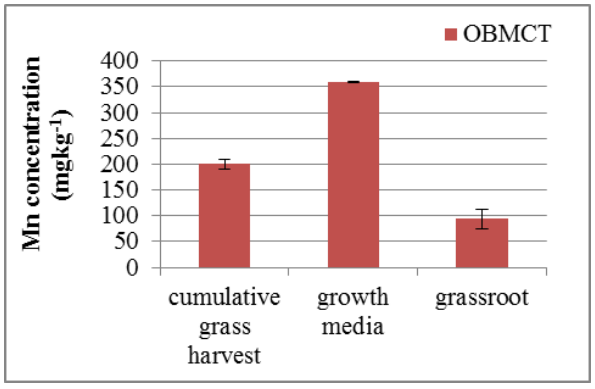
According to figure 4.14l, grass roots had the lowest Mn concentrations while growth media had the highest Mn concentrations in most of the profiles except for OBGC, OBMC, NBCG, GGC, GMC and LGC.

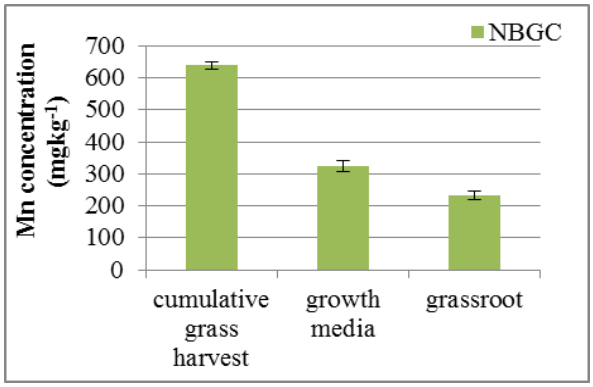


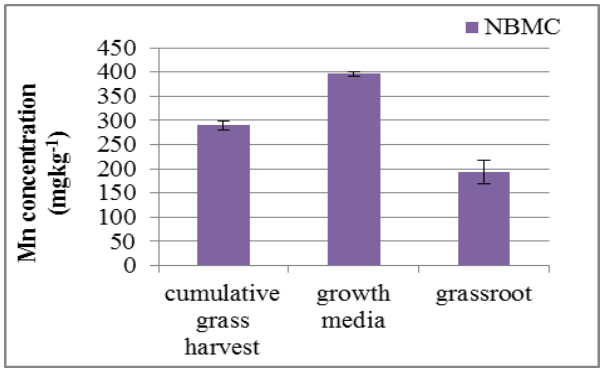


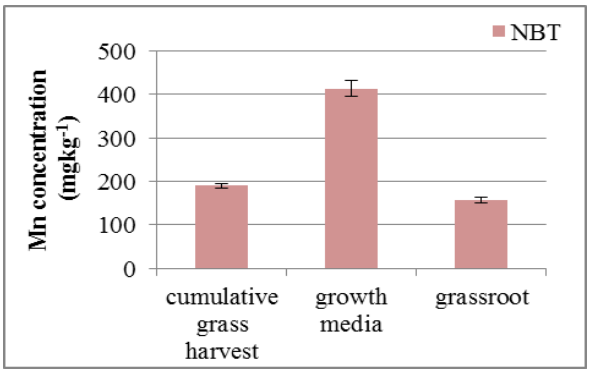


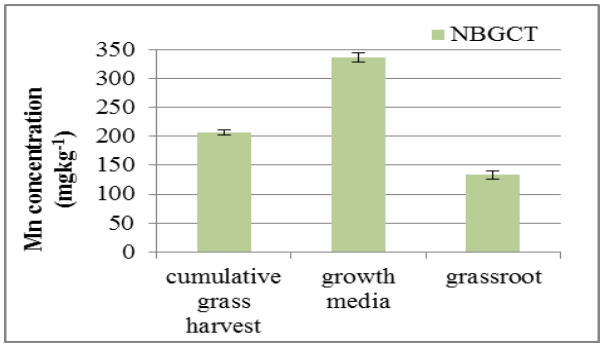


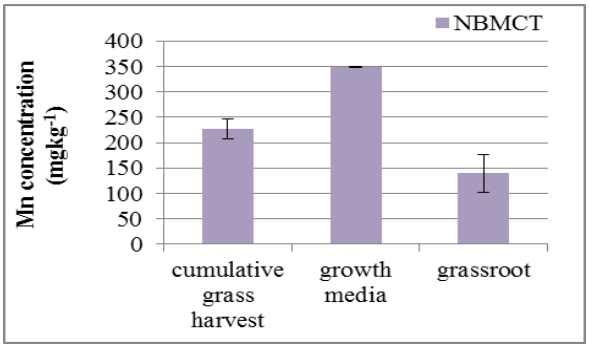


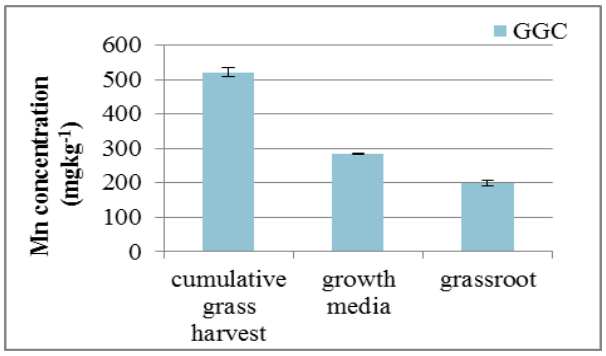


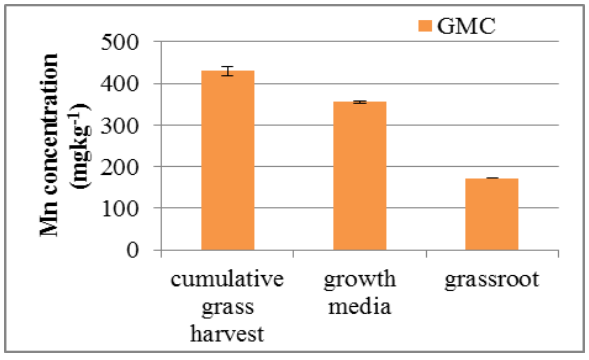

NBMC
Mn concentration (mgkg-1)
450
400
350
300
250
200
150
100
50
0
cumulative grass harvest
growth media
grassroot

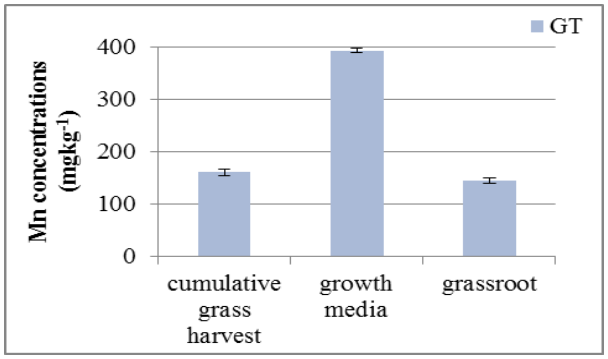

NBT
Mn concentration (mgkg-1)
500
400
300
200
100
0
cumulative grass harvest
growth media
grassroot

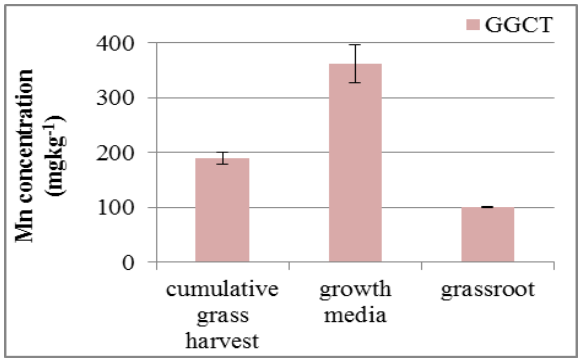

NBGCT
Mn concentration (mgkg-1)
350
300
250
200
150
100
50
0
cumulative grass harvest
growth media
grassroot


NBMCT
Mn concentration (mgkg-1)
400
350
300
250
200
150
100
50
0
cumulative grass harvest
growth media
grassroot


GGC
Mn concentration (mgkg-1)
600
500
400
300
200
100
0
cumulative grass harvest
growth media
grassroot


GMC
Mn concentration (mgkg-1)
500
400
300
200
100
0
cumulative grass harvest
growth media
grassroot


GT
Mn concentrations (mgkg-1)
400
300
200
100
0
cumulative grass harvest
growth media
grassroot


GGCT
Mn concentration (mgkg-1)
400
300
200
100
0
cumulative grass harvest
growth media
grassroot

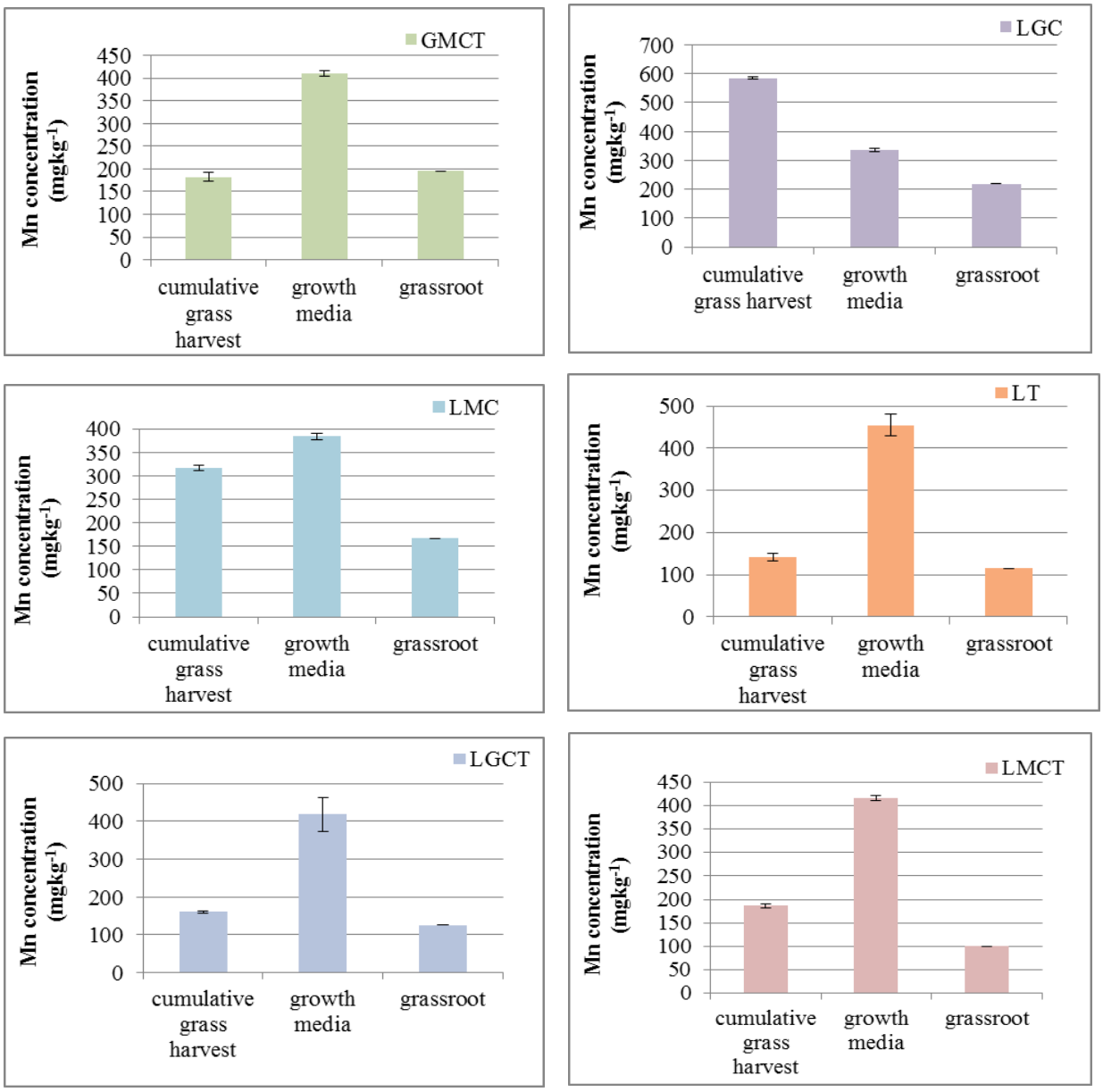
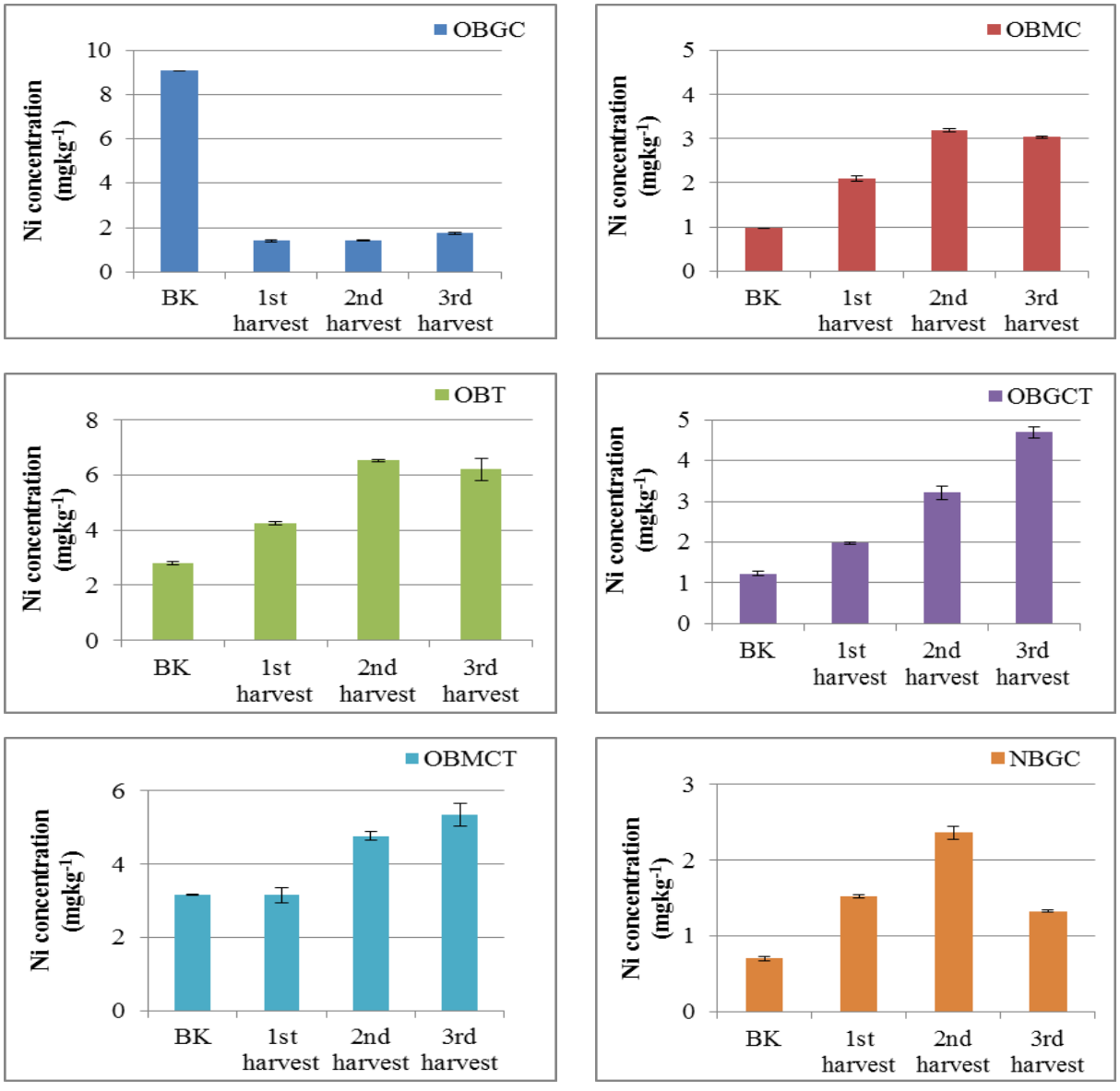
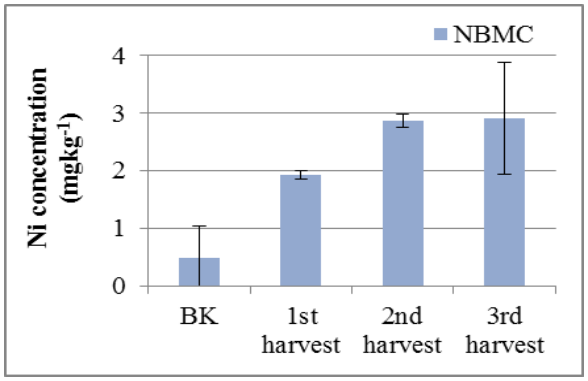


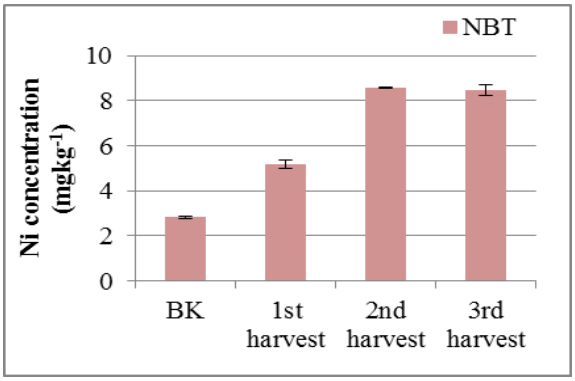
Figure 4.14l: Trends of manganese concentrations in cumulative grass harvests, growth media and grass roots

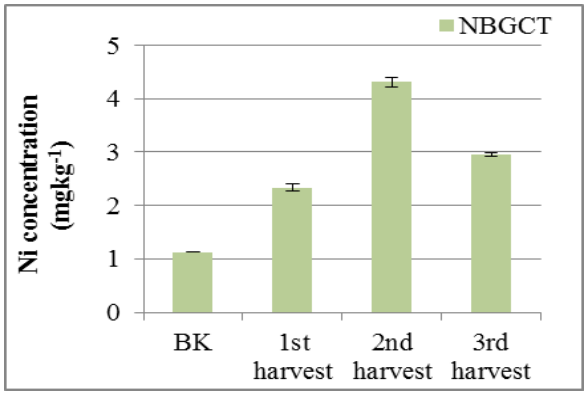
Nickel

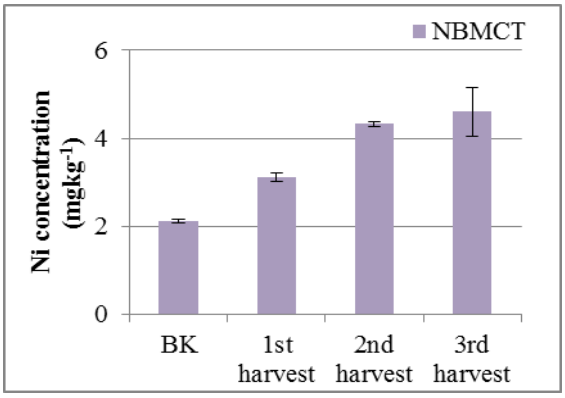
Figure 4.14m shows that Ni concentrations were lowest in background harvests followed by 1st harvest, except for OBGC where Ni background concentrations were the highest of the four harvests. The highest Ni concentrations were found in both 2nd and 3rd harvests in almost all the profiles, with GT possessing the highest concentrations in the 2nd harvest.

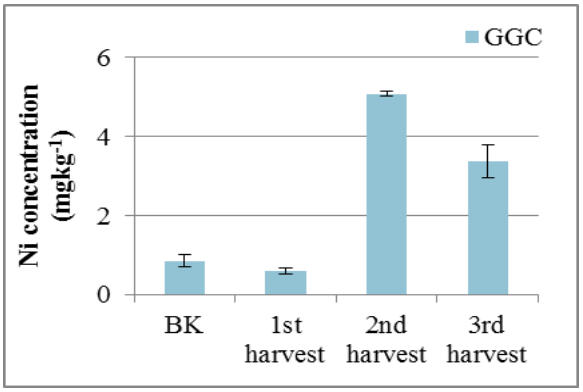


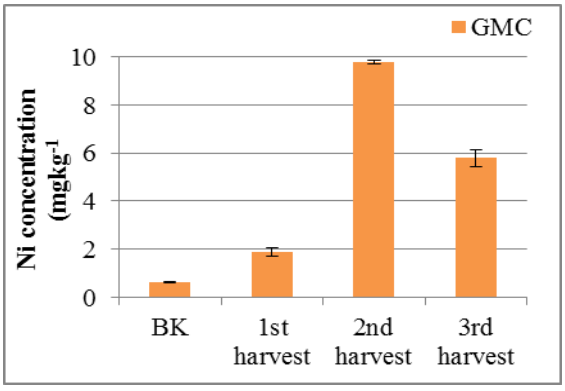

NBMC
Ni concentration (mgkg-1)
0
1
2
3
4
BK
1st harvest
2nd harvest
3rd harvest

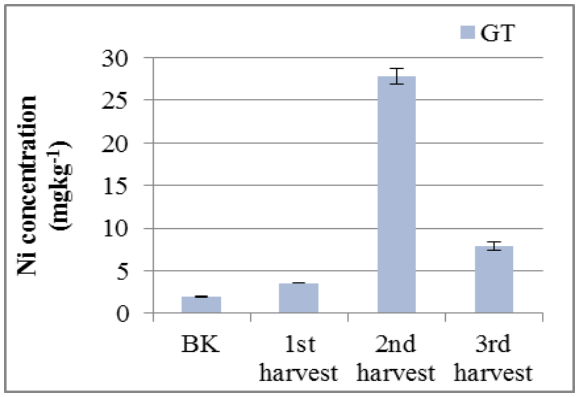

NBT
Ni concentration (mgkg-1)
0
2
4
6
8
10
BK
1st harvest
2nd harvest
3rd harvest

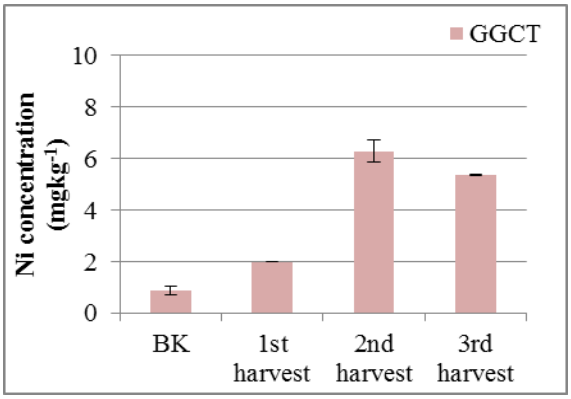

NBGCT
Ni concentration (mgkg-1)
0
1
2
3
4
5
BK
1st harvest
2nd harvest
3rd harvest


NBMCT
Ni concentration (mgkg-1)
0
2
4
6
BK
1st harvest
2nd harvest
3rd harvest


GGC
Ni concentration (mgkg-1)
0
2
4
6
BK
1st harvest
2nd harvest
3rd harvest


GMC
Ni concentration (mgkg-1
0
2
4
6
8
10
BK
1st harvest
2nd harvest
3rd harvest


GT
Ni concentration (mgkg-1)
0
5
10
15
20
25
30
BK
1st harvest
2nd harvest
3rd harvest


GGCT
Ni concentration (mgkg-1)
0
2
4
6
8
10
BK
1st harvest
2nd harvest
3rd harvest

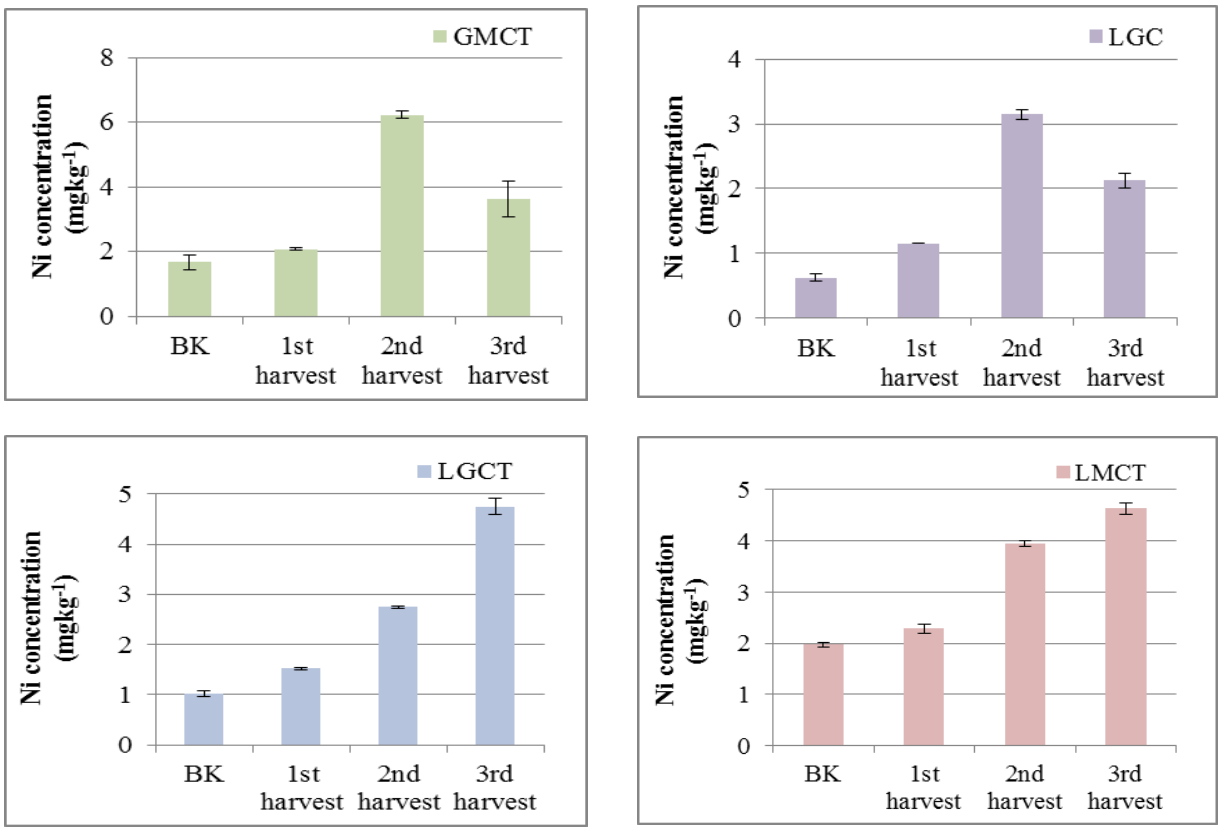
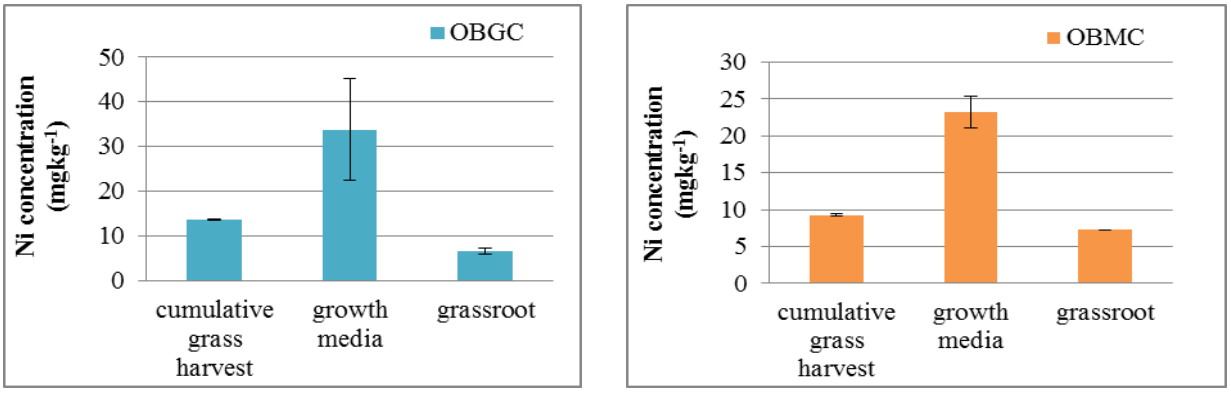
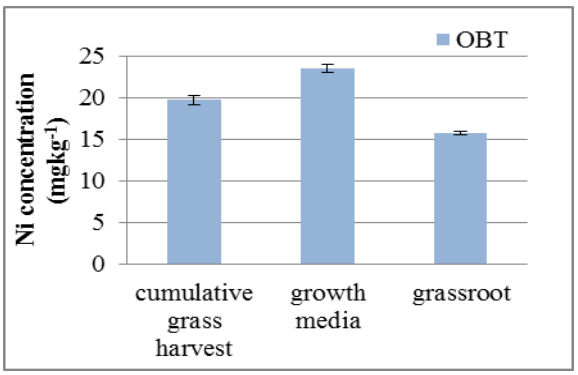
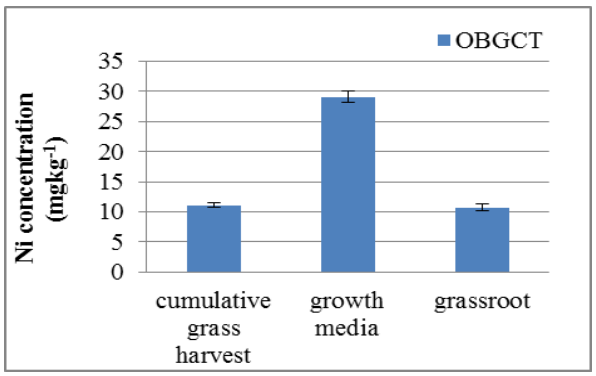


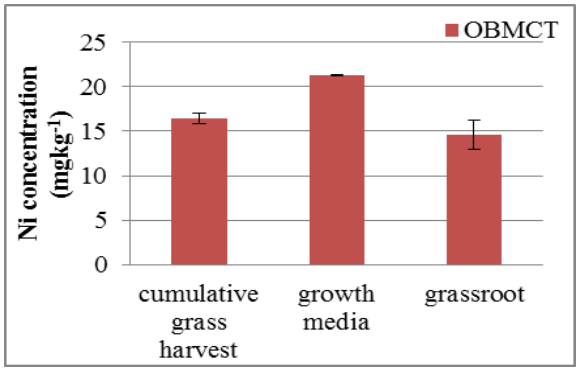
Figure 4.14m: Trends of nickel concentrations in background and three monthly grass harvests derived from test profiles

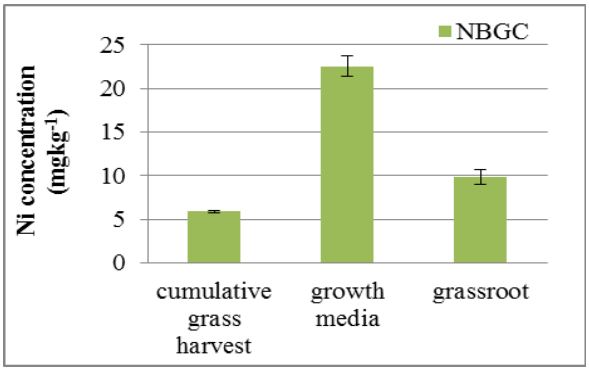
Overall, growth media retained the highest Ni concentrations in almost all the profiles, especially the GMCT profile. Ni concentrations in grasses and grass roots were both lower than growth media concentrations (see figure 4.14n).

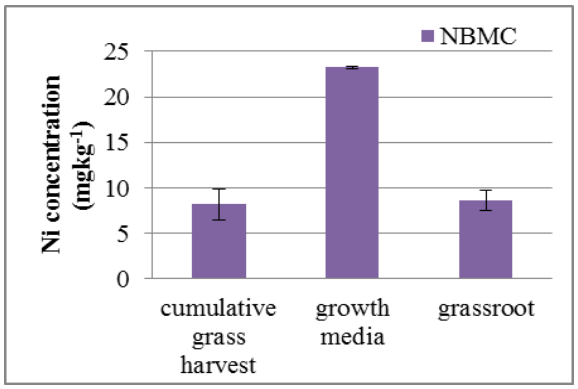


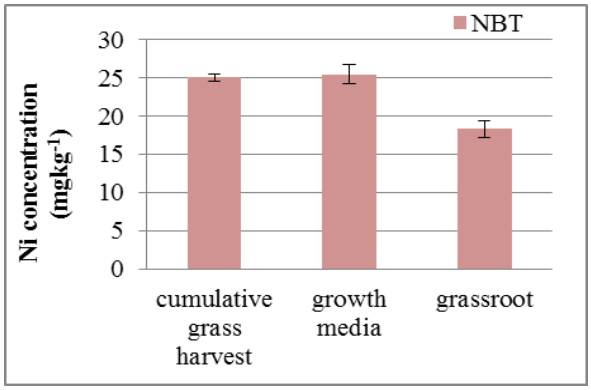

OBT
Ni concentration (mgkg-1)
25
20
15
10
5
0
cumulative grass harvest
growth media
grassroot

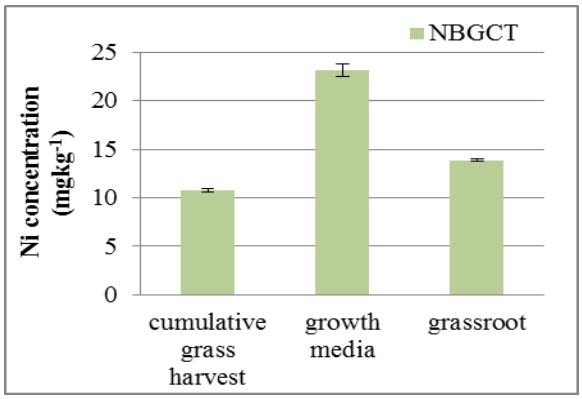

OBGCT
Ni concentration (mgkg-1)
35
30
25
20
15
10
5
0
cumulative grass harvest
growth media
grassroot

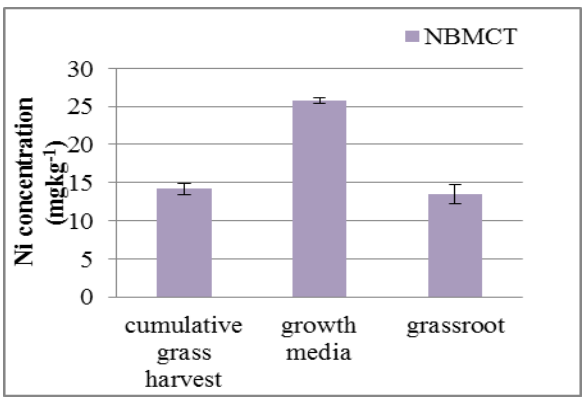

OBMCT
Ni concentration (mgkg-1)
25
20
15
10
5
0
cumulative grass harvest
growth media
grassroot

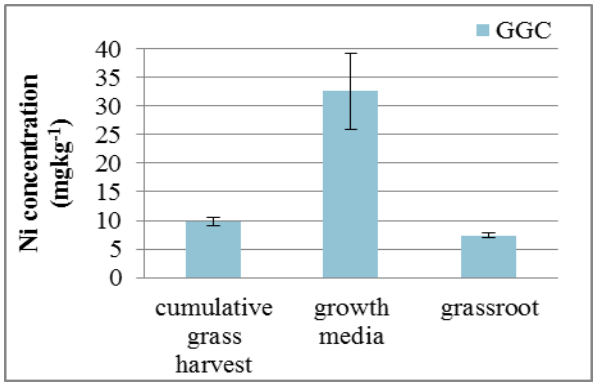

NBGC
Ni concentration (mgkg-1)
25
20
15
10
5
0
cumulative grass harvest
growth media
grassroot

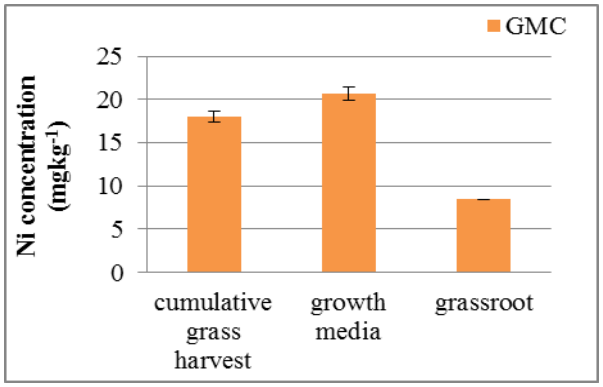

NBMC
Ni concentration (mgkg-1)
25
20
15
10
5
0
cumulative grass harvest
growth media
grassroot

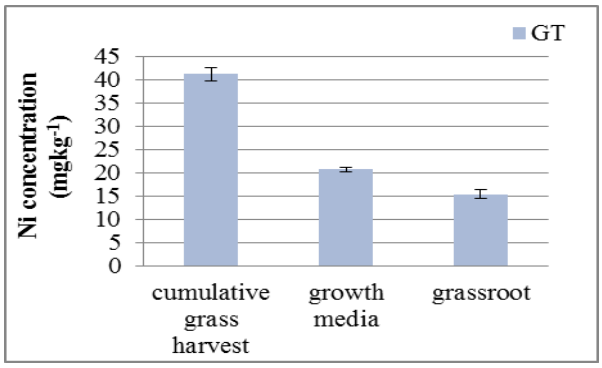

NBT
Ni concentration (mgkg-1)
30
25
20
15
10
5
0
cumulative grass harvest
growth media
grassroot

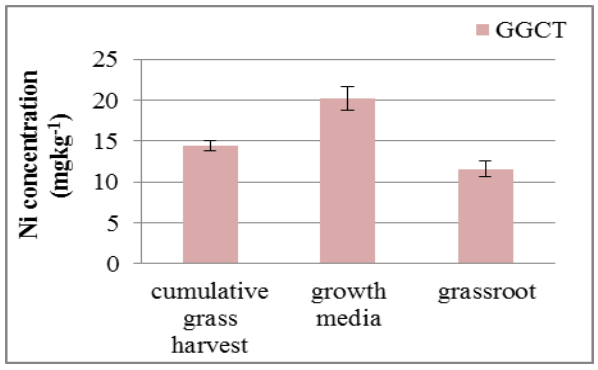

NBGCT
Ni concentration (mgkg-1)
25
20
15
10
5
0
cumulative grass harvest
growth media
grassroot

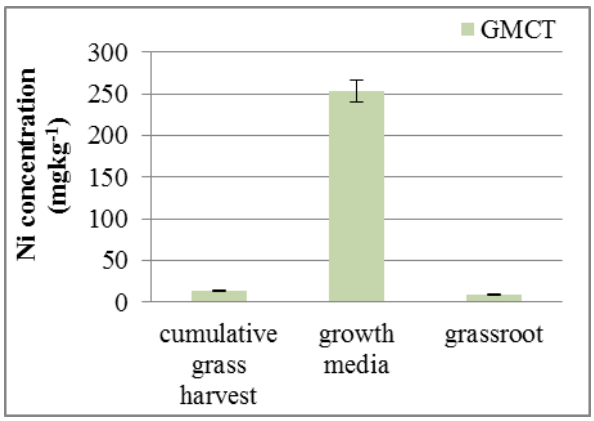

NBMCT
Ni concentration (mgkg-1)
30
25
20
15
10
5
0
cumulative grass harvest
growth media
grassroot

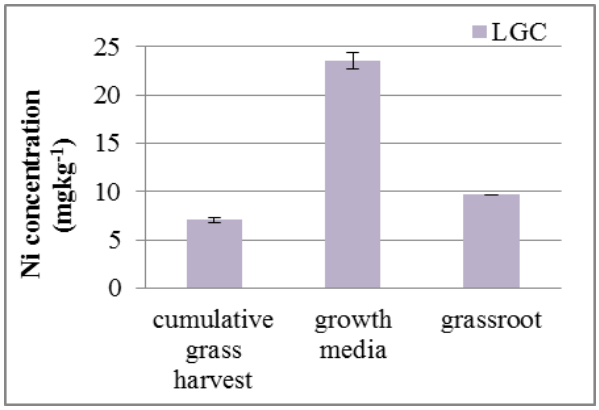

GGC
Ni concentration (mgkg-1)
40
35
30
25
20
15
10
5
0
cumulative grass harvest
growth media
grassroot

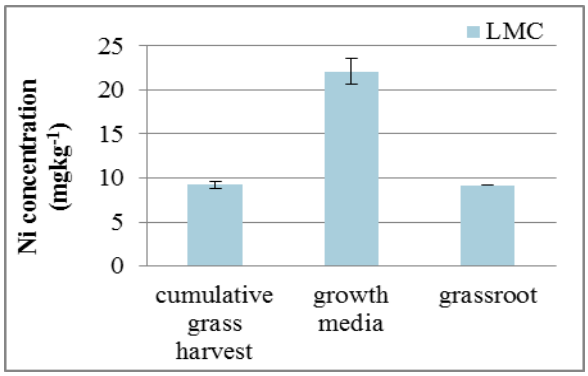

GMC
Ni concentration (mgkg-1)
25
20
15
10
5
0
cumulative grass harvest
growth media
grassroot

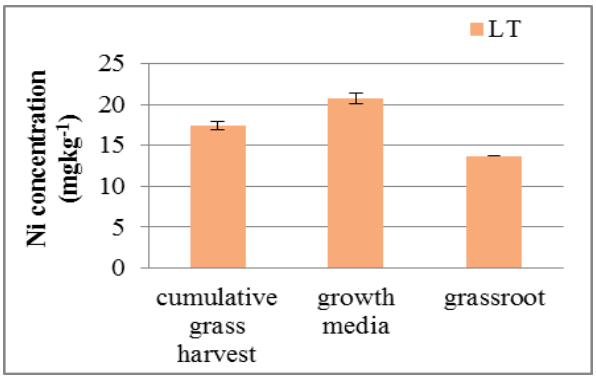

GT
Ni concentration (mgkg-1)
45
40
35
30
25
20
15
10
5
0
cumulative grass harvest
growth media
grassroot


GGCT
Ni concentration (mgkg-1)
25
20
15
10
5
0
cumulative grass harvest
growth media
grassroot


GMCT
Ni concentration (mgkg-1)
300
250
200
150
100
50
0
cumulative grass harvest
growth media
grassroot


LGC
Ni concentration (mgkg-1)
25
20
15
10
5
0
cumulative grass harvest
growth media
grassroot


LMC
Ni concentration (mgkg-1)
25
20
15
10
5
0
cumulative grass harvest
growth media
grassroot


LT
Ni concentration (mgkg-1)
25
20
15
10
5
0
cumulative grass harvest
growth media
grassroot

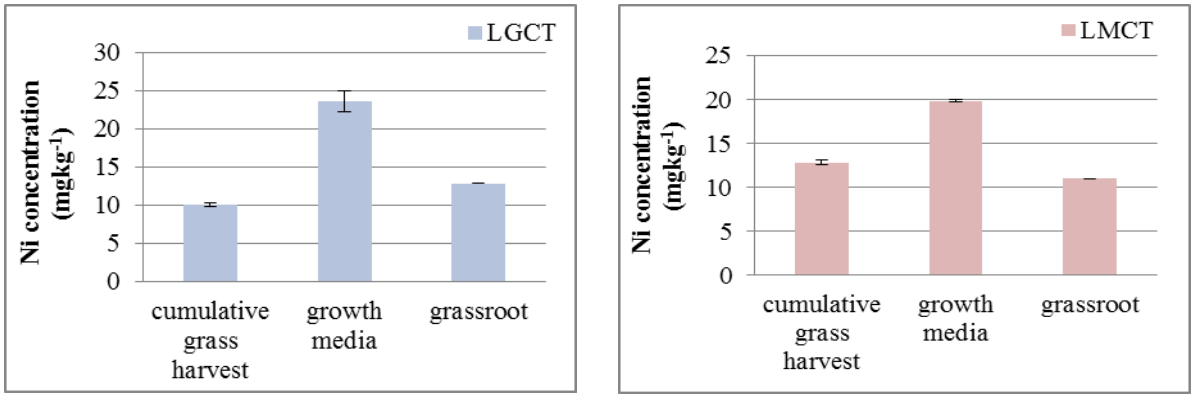
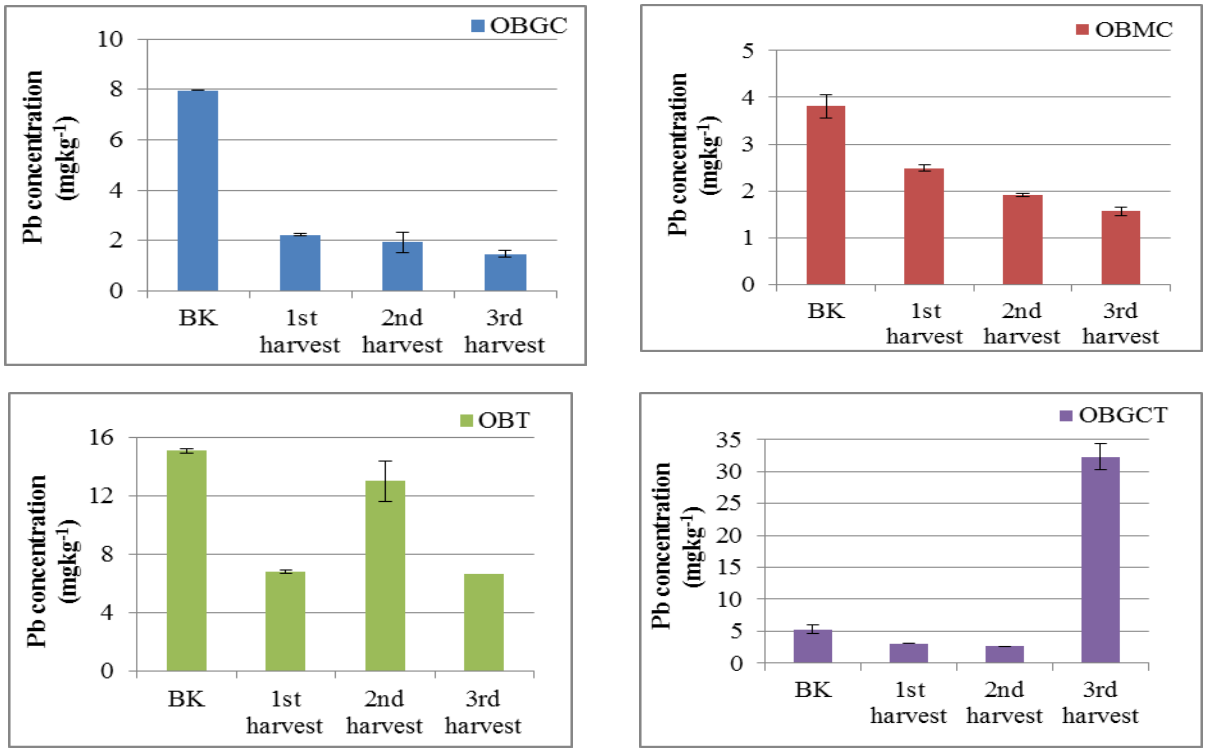
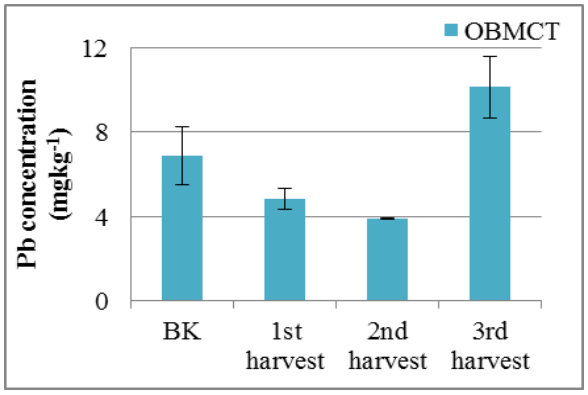


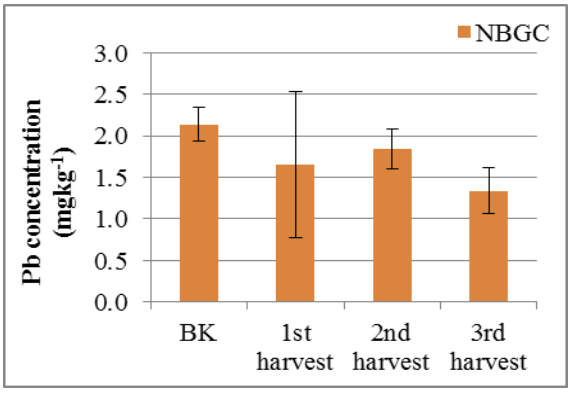
Figure 4.14n: Trends of nickel concentrations in cumulative grass harvests, growth media and grass roots

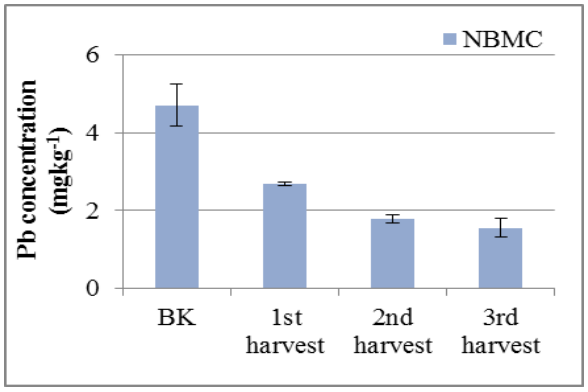
Lead

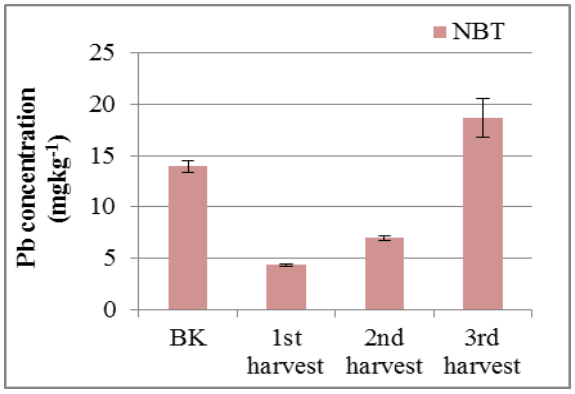
Pb concentrations in the 1st and 2nd harvest grasses were below background concentrations in half of the profiles, however, the 3rd harvest concentrations were above background concentrations in OBGCT, OBMCT, NBT, NBMCT GT, LGC and LMC (see figure 4.14o).

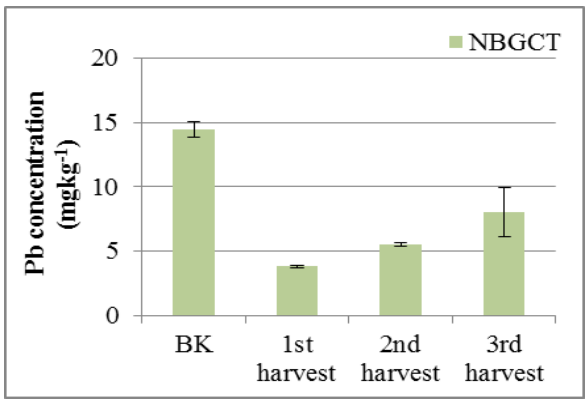


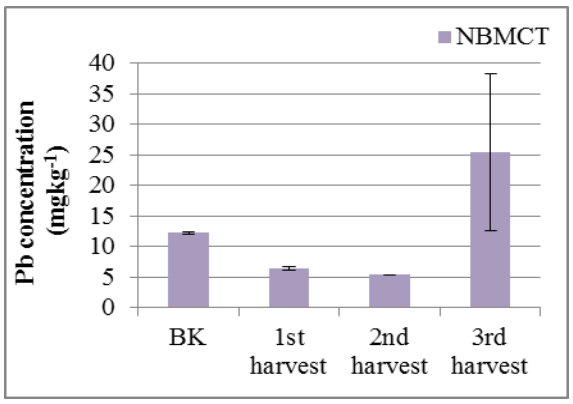

OBMCT
Pb concentration (mgkg-1)
12
8
4
0
BK
1st harvest
2nd harvest
3rd harvest

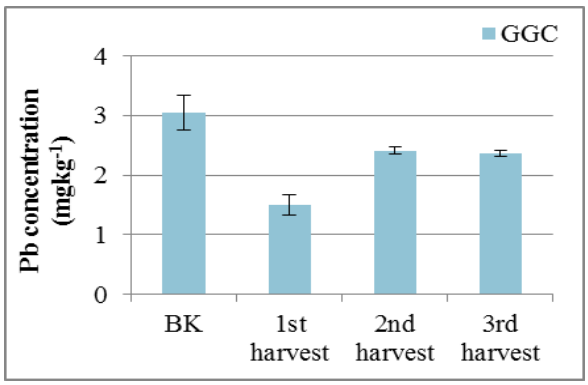

NBGC
Pb concentration (mgkg-1)
3.0
2.5
2.0
1.5
1.0
0.5
0.0
BK
1st harvest
2nd harvest
3rd harvest

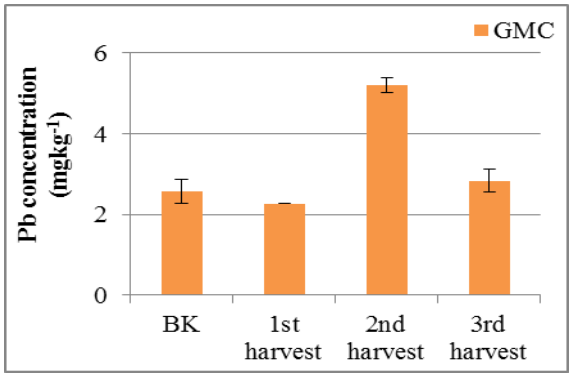

NBMC
Pb concentration (mgkg-1)
6
4
2
0
BK
1st harvest
2nd harvest
3rd harvest


NBT
Pb concentration (mgkg-1)
25
20
15
10
5
0
BK
1st harvest
2nd harvest
3rd harvest


NBGCT
Pb concentration (mgkg-1)
20
15
10
5
0
BK
1st harvest
2nd harvest
3rd harvest


NBMCT
Pb concentration (mgkg-1)
40
35
30
25
20
15
10
5
0
BK
1st harvest
2nd harvest
3rd harvest


GGC
Pb concentration (mgkg-1)
4
3
2
1
0
BK
1st harvest
2nd harvest
3rd harvest


GMC
Pb concentration (mgkg-1)
6
4
2
0
BK
1st harvest
2nd harvest
3rd harvest

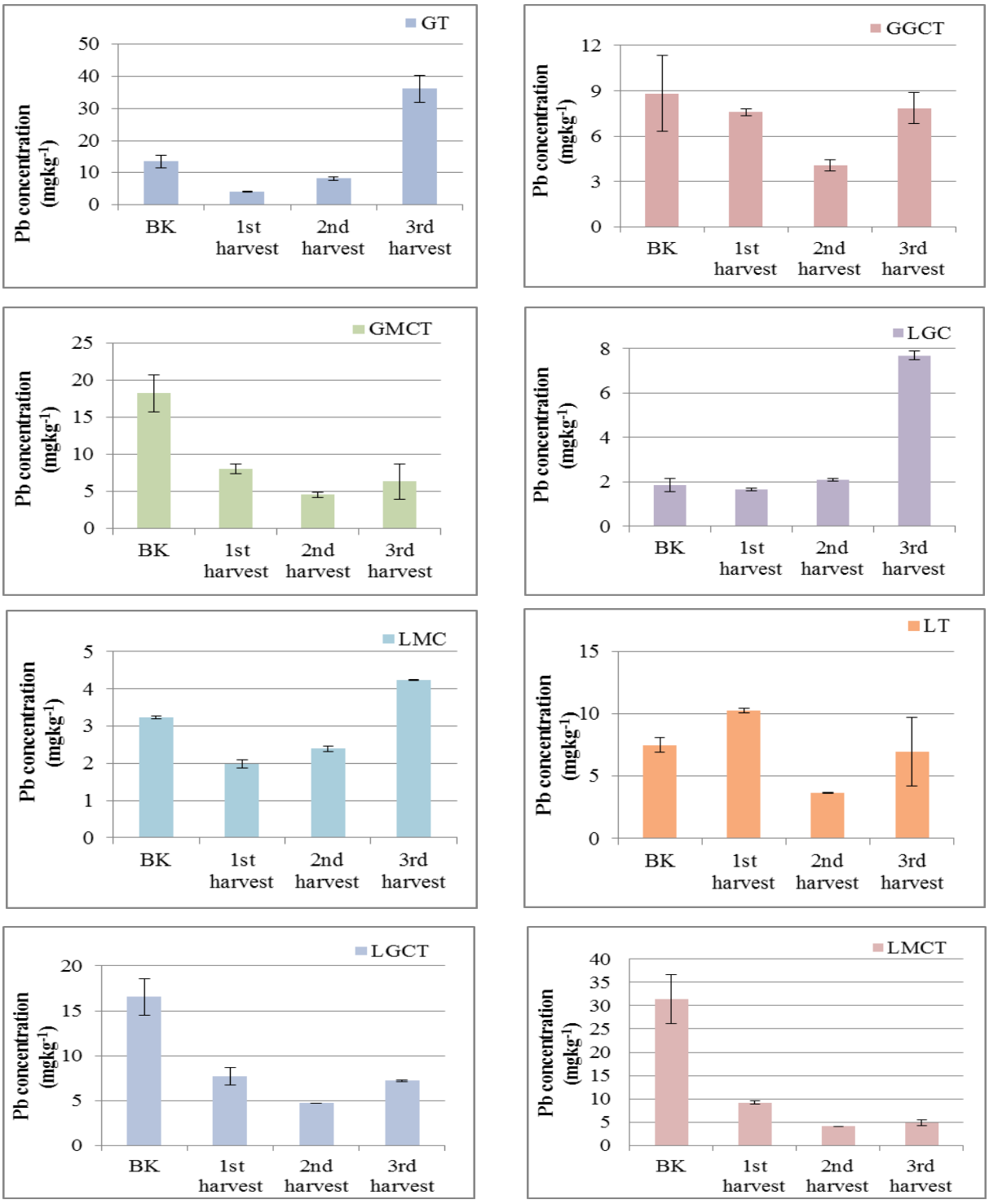
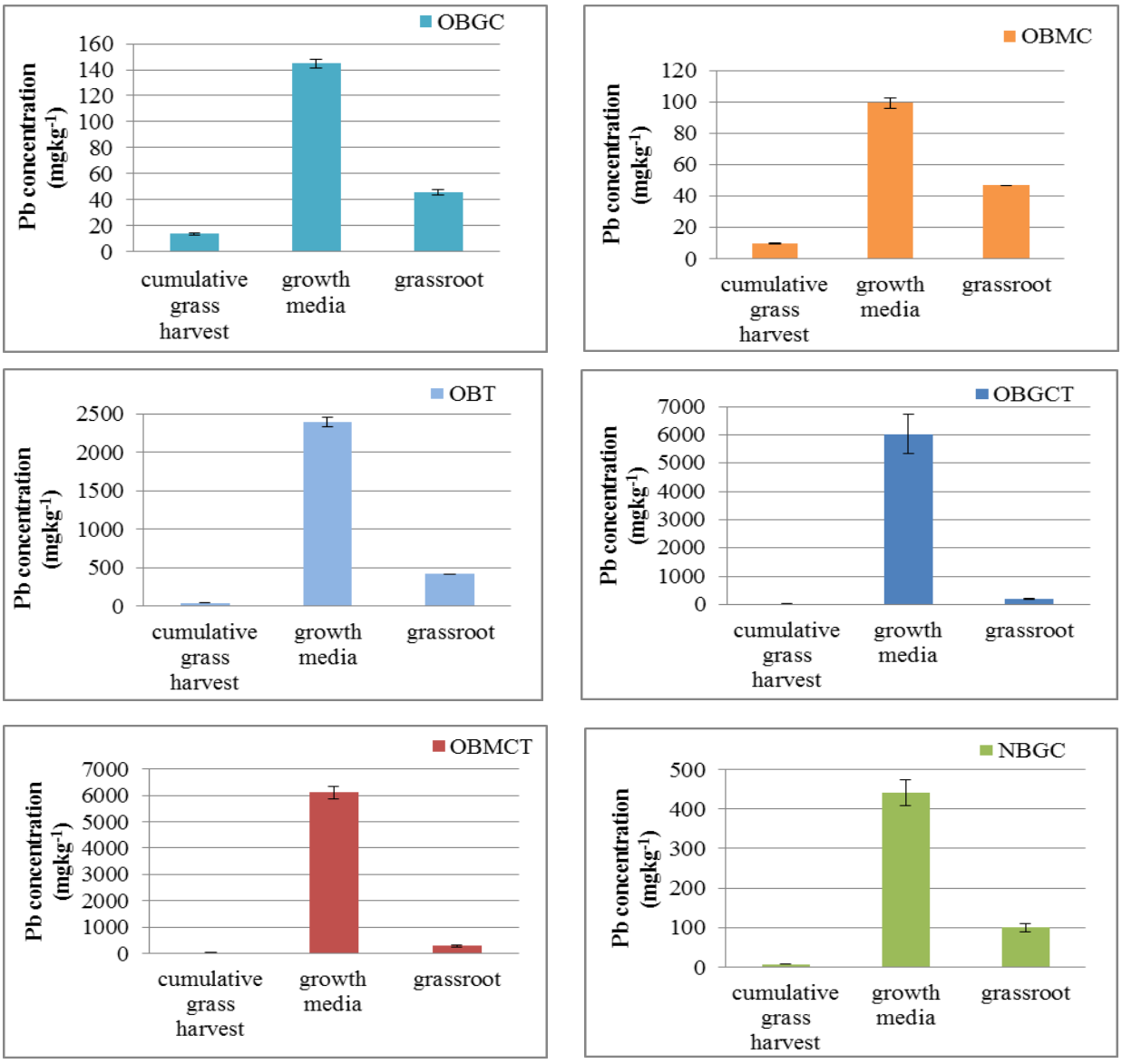
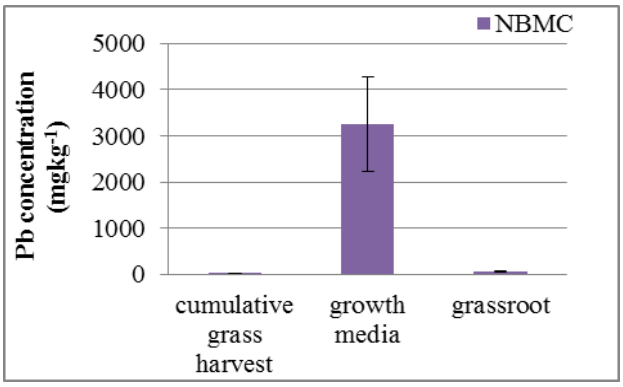
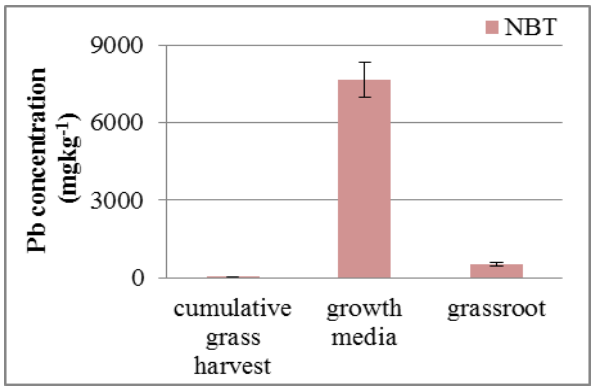


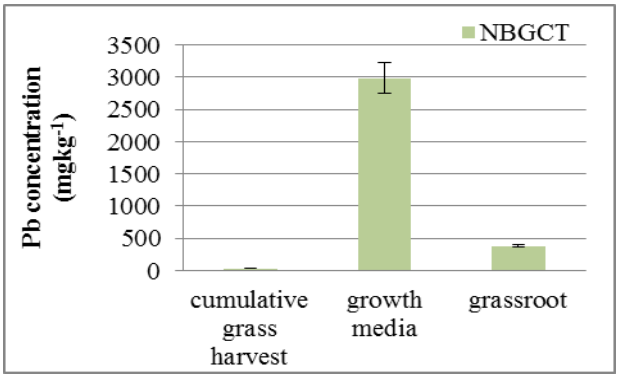
Figure 4.14o: Trends of lead concentrations in background and three monthly grass harvests derived from test profiles

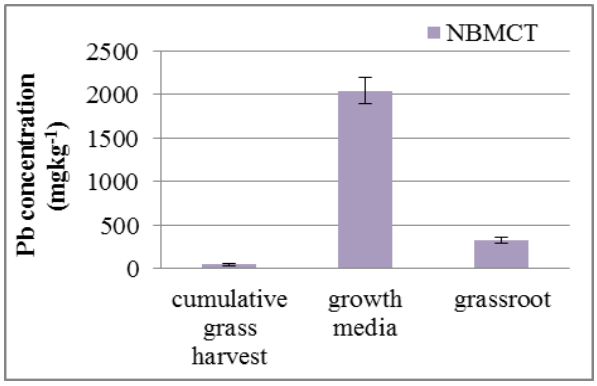
Overall, growth media in all the profiles retained the highest Pb concentrations followed by grass roots and then the grasses. Grasses and grass roots Pb concentrations were very low compared to growth media concentrations as shown in figure 4.14p.

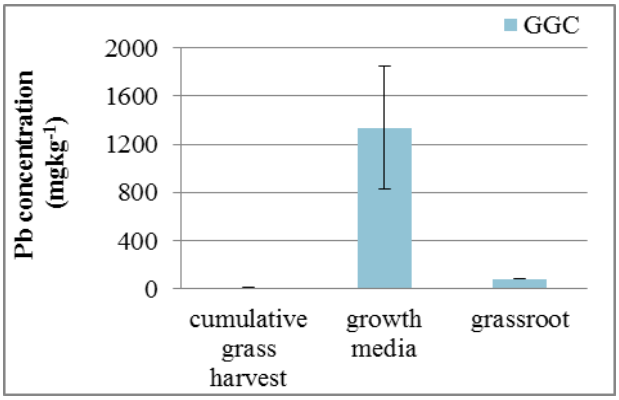


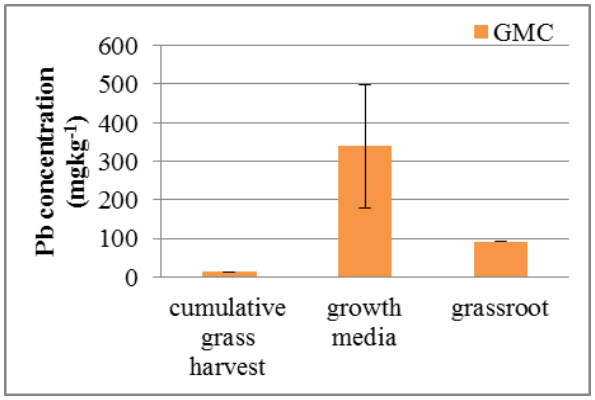

NBMC
Pb concentration (mgkg-1)
5000
4000
3000
2000
1000
0
cumulative grass harvest
growth media
grassroot

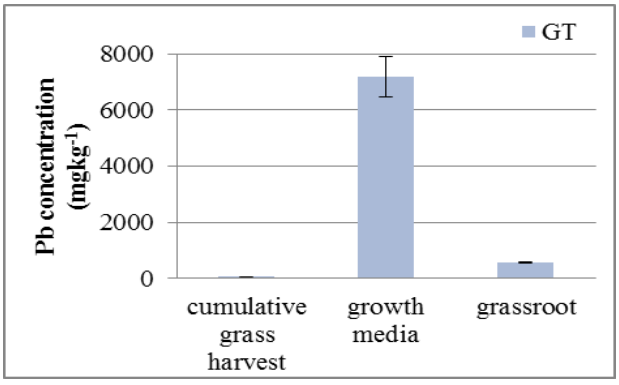

NBT
Pb concentration (mgkg-1)
9000
6000
3000
0
cumulative grass harvest
growth media
grassroot

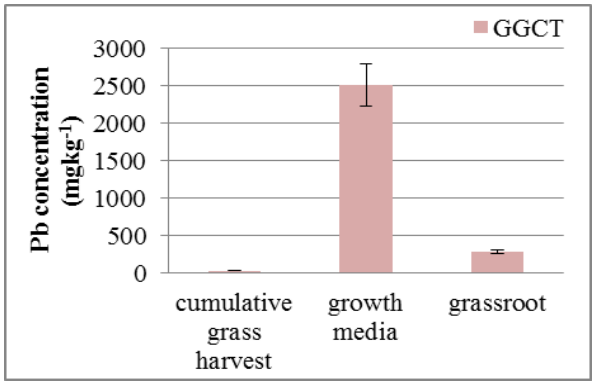

NBGCT
Pb concentration (mgkg-1)
3500
3000
2500
2000
1500
1000
500
0
cumulative grass harvest
growth media
grassroot


NBMCT
Pb concentration (mgkg-1)
2500
2000
1500
1000
500
0
cumulative grass harvest
growth media
grassroot


GGC
Pb concentration (mgkg-1)
2000
1600
1200
800
400
0
cumulative grass harvest
growth media
grassroot


GMC
Pb concentration (mgkg-1)
600
500
400
300
200
100
0
cumulative grass harvest
growth media
grassroot


GT
Pb concentration (mgkg-1)
8000
6000
4000
2000
0
cumulative grass harvest
growth media
grassroot


GGCT
Pb concentration (mgkg-1)
3000
2500
2000
1500
1000
500
0
cumulative grass harvest
growth media
grassroot

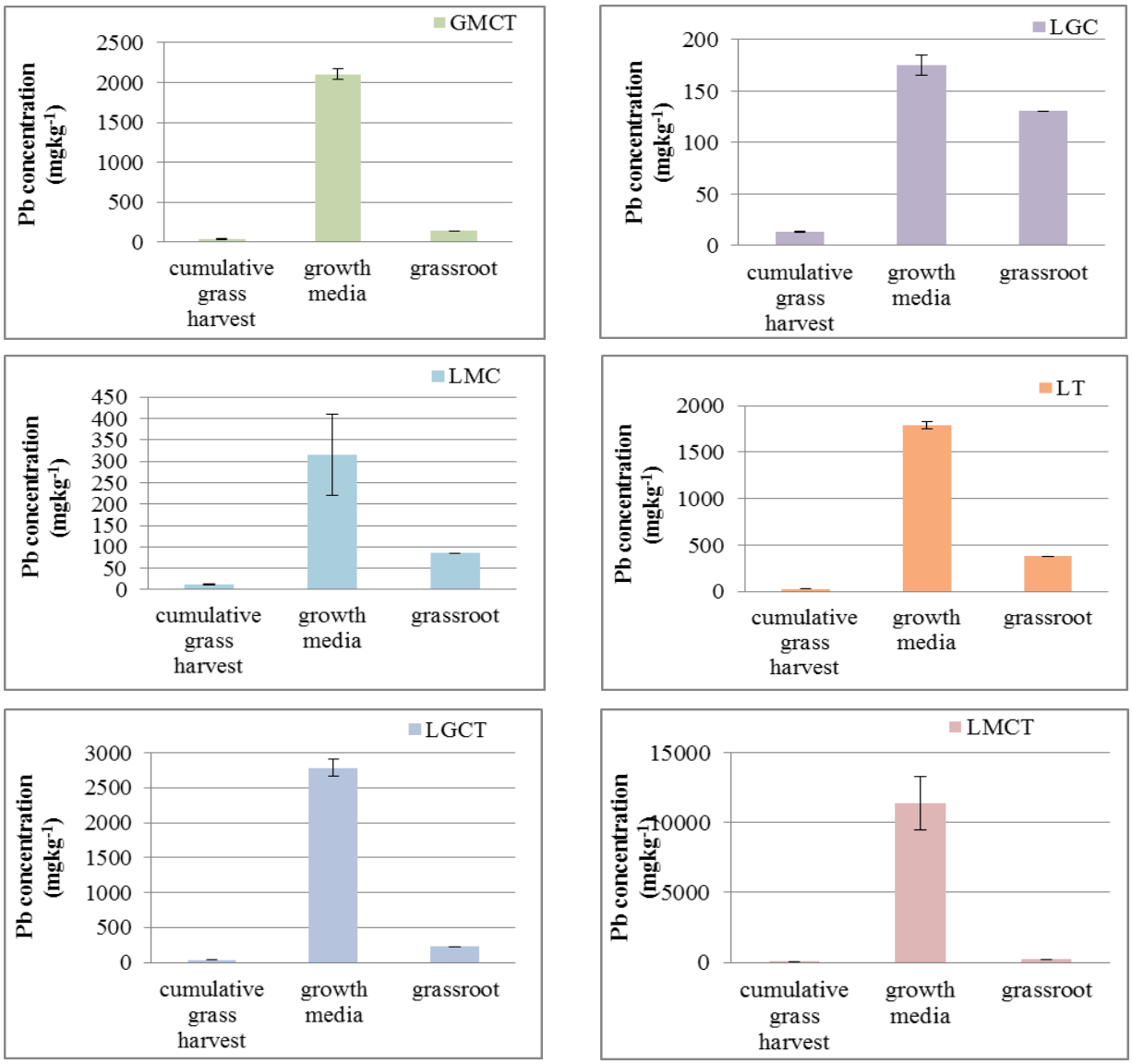
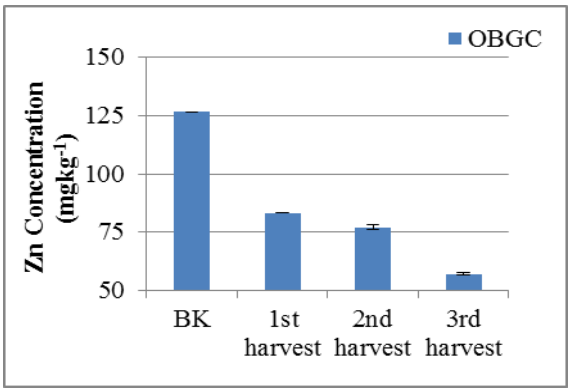
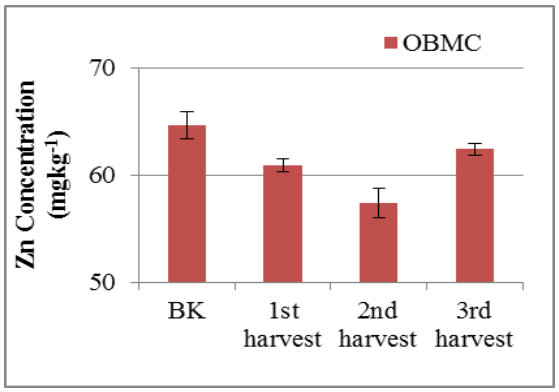


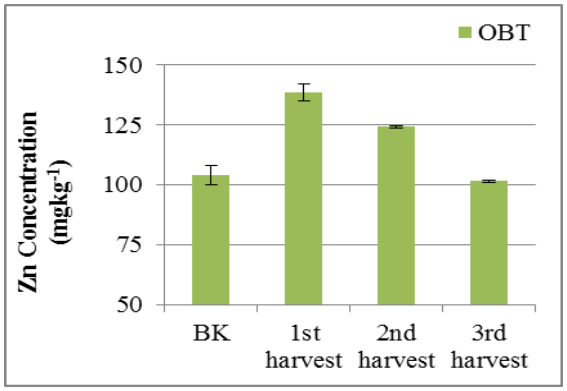
Figure 4.14p: Trends of lead concentrations in cumulative grass harvests, growth media and grass roots

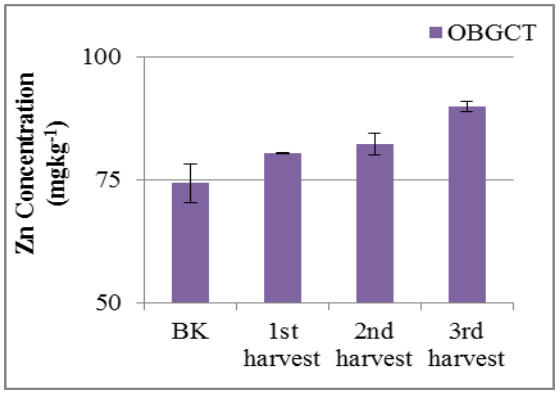
Zinc

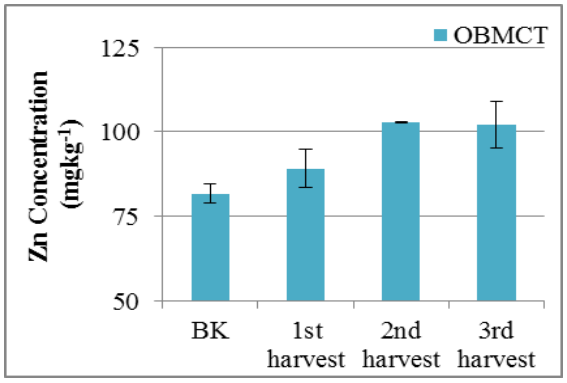
According to figure 4.14q, Zn concentrations were mostly above background concentrations in 1st and 2nd harvests and mostly below background concentrations in the 3rd harvests.

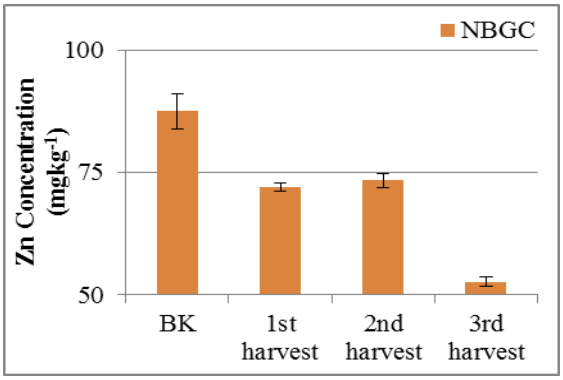


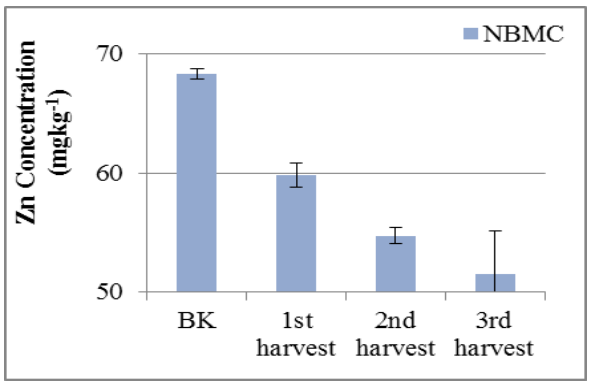


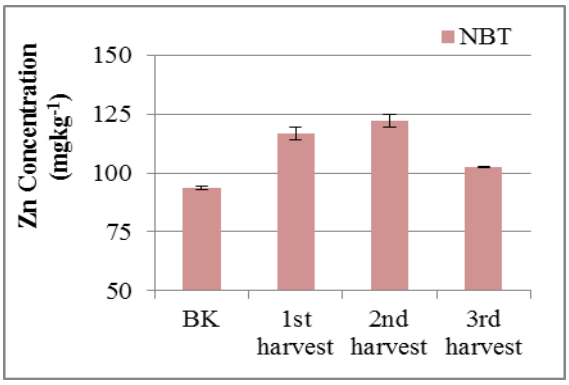


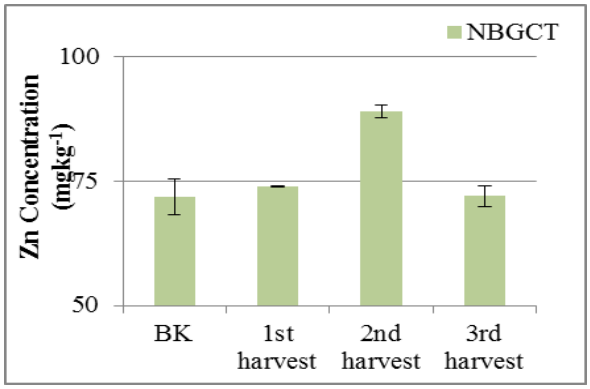


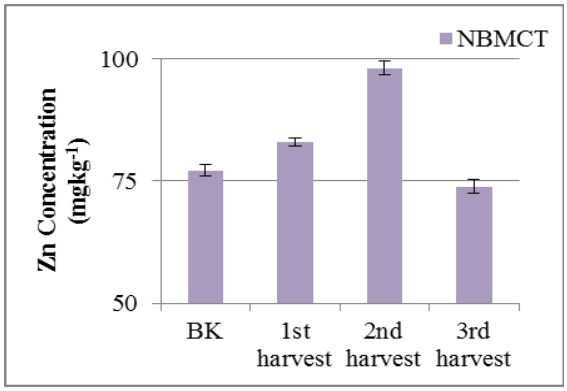


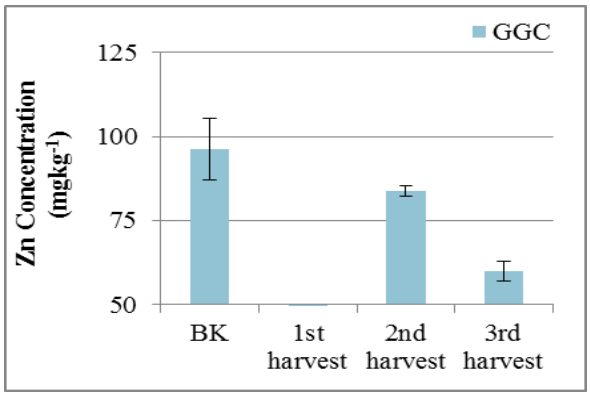


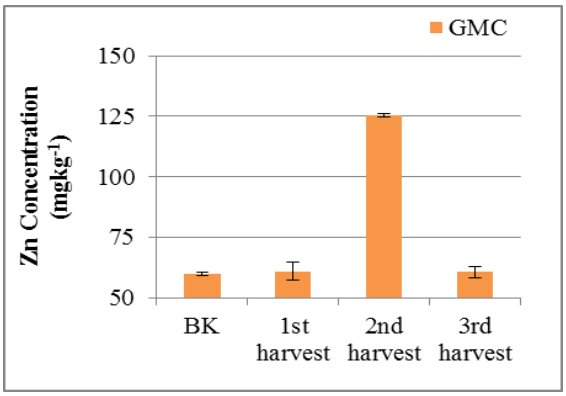

NBMC
Zn Concentration (mgkg-1)
70
60
50
BK
1st harvest
2nd harvest
3rd harvest

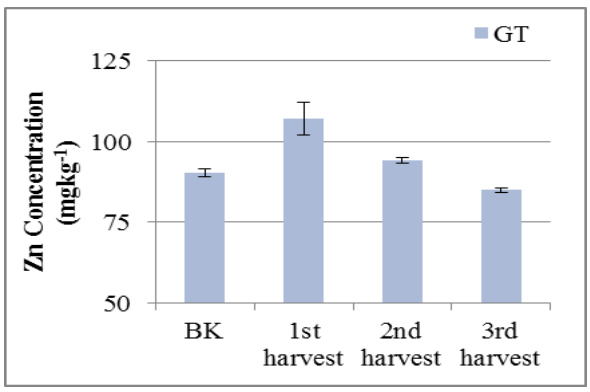

NBT
Zn Concentration (mgkg-1)
150
125
100
75
50
BK
1st harvest
2nd harvest
3rd harvest

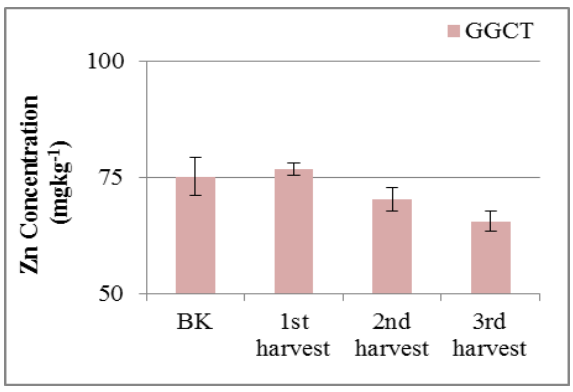

NBGCT
Zn Concentration (mgkg-1)
100
75
50
BK
1st harvest
2nd harvest
3rd harvest


NBMCT
Zn Concentration (mgkg-1)
100
75
50
BK
1st harvest
2nd harvest
3rd harvest


GGC
Zn Concentration (mgkg-1)
125
100
75
50
BK
1st harvest
2nd harvest
3rd harvest


GMC
Zn Concentration (mgkg-1)
150
125
100
75
50
BK
1st harvest
2nd harvest
3rd harvest


GT
Zn Concentration (mgkg-1)
125
100
75
50
BK
1st harvest
2nd harvest
3rd harvest


GGCT
Zn Concentration (mgkg-1)
100
75
50
BK
1st harvest
2nd harvest
3rd harvest

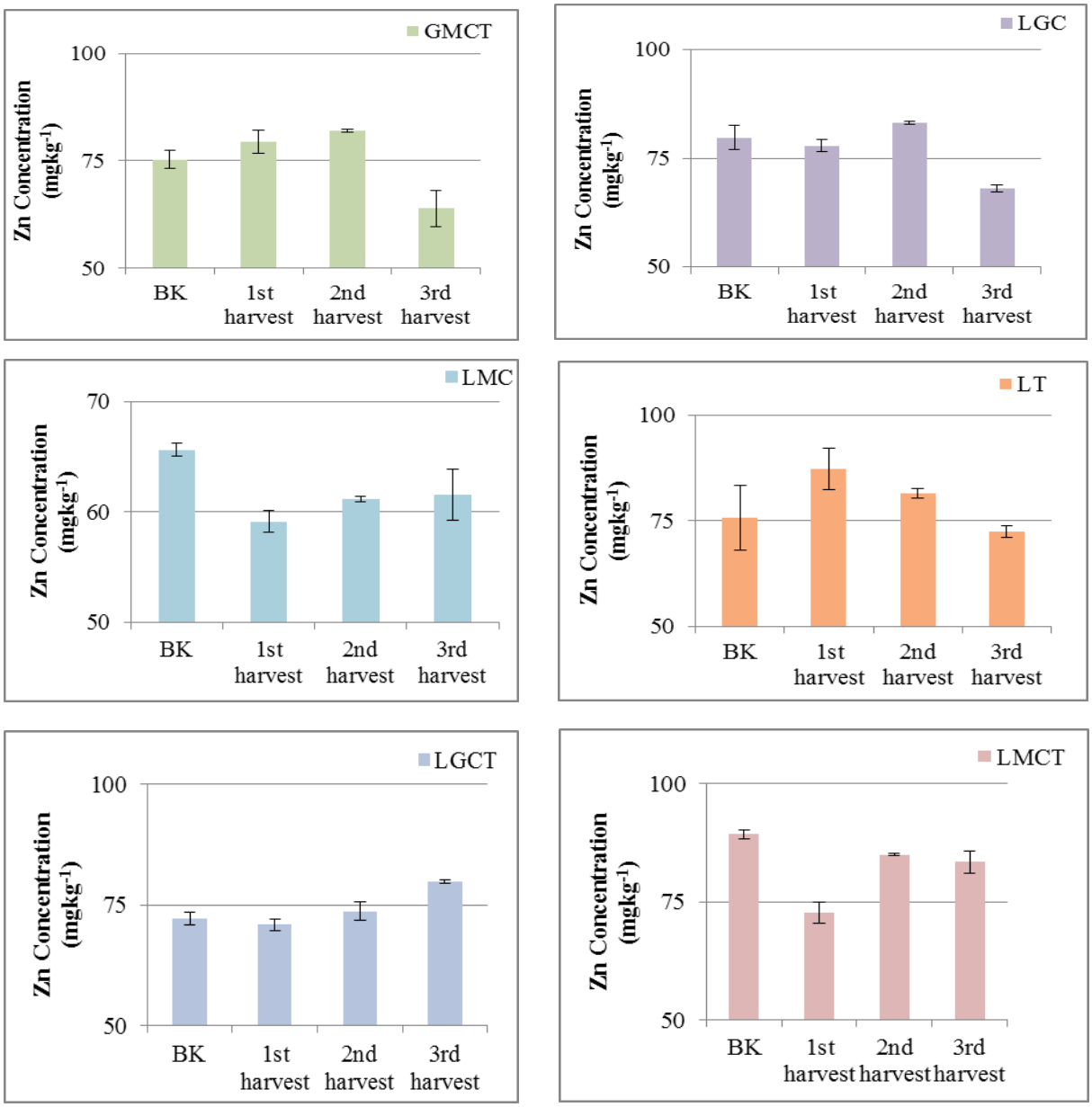
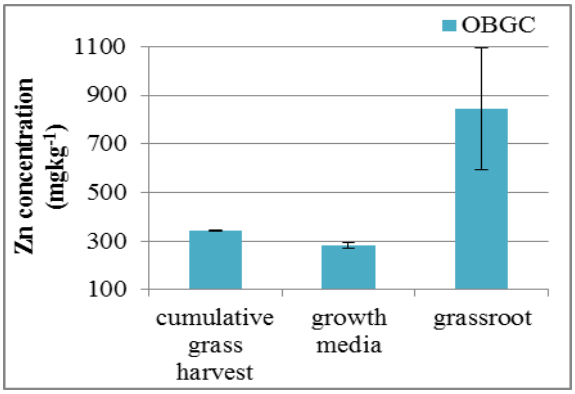
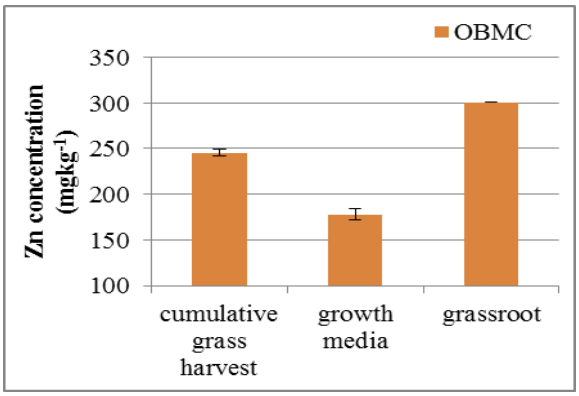
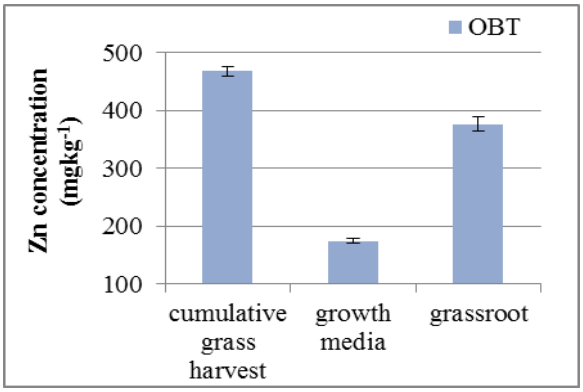


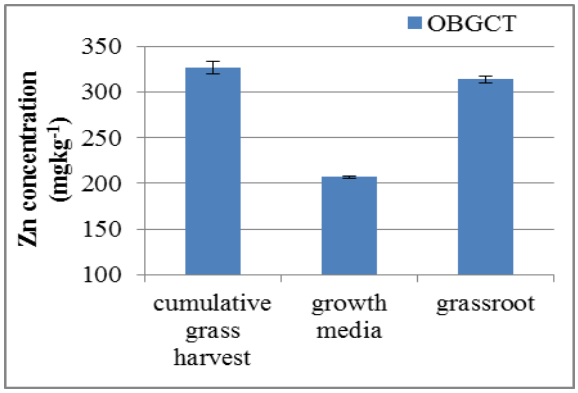
Figure 4.14q: Trends of zinc concentrations in background and three monthly grass harvests derived from test profiles.

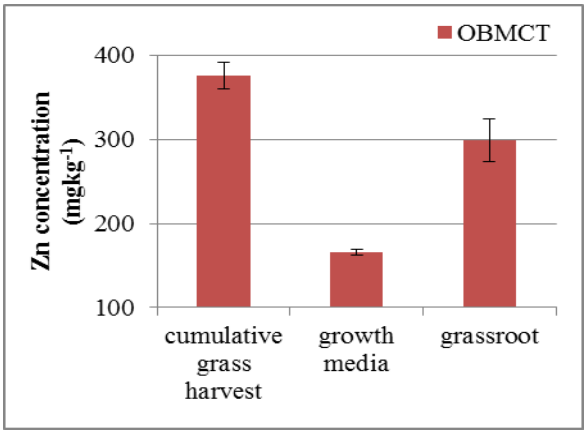
Overall, highest Zn concentrations were retained in both grasses and grass roots with the lowest concentrations found in growth media for almost all the profilesas seen in figure 4.14r.

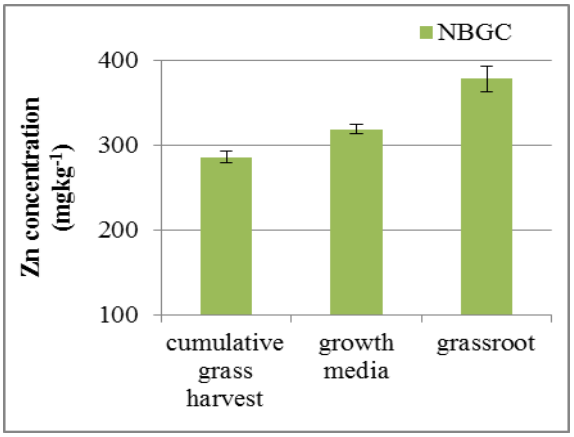


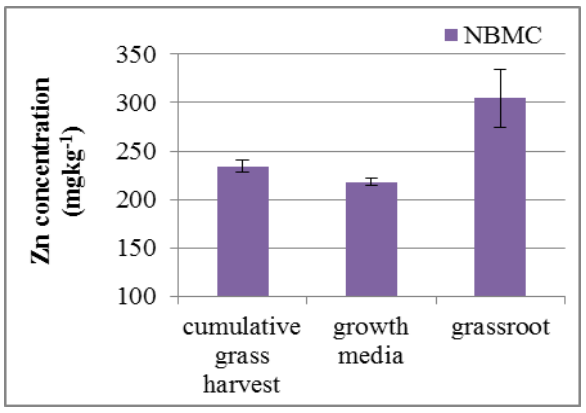


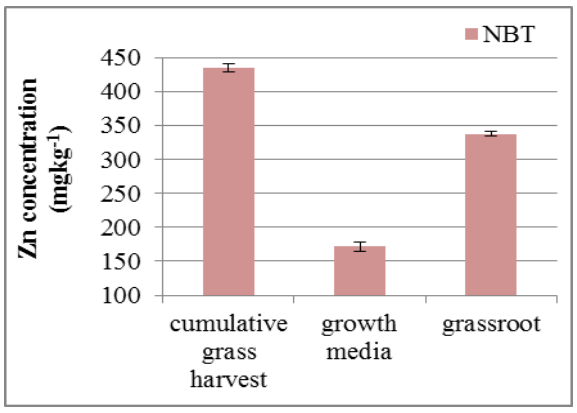


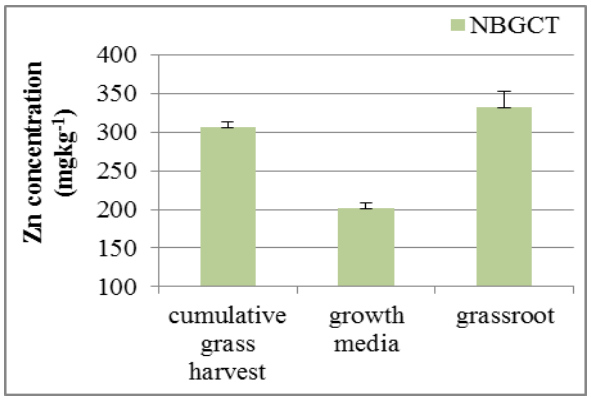


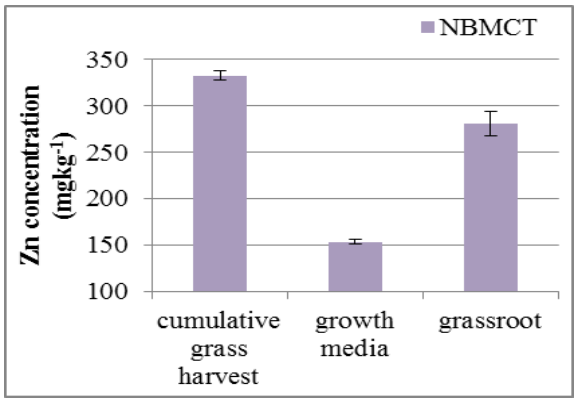


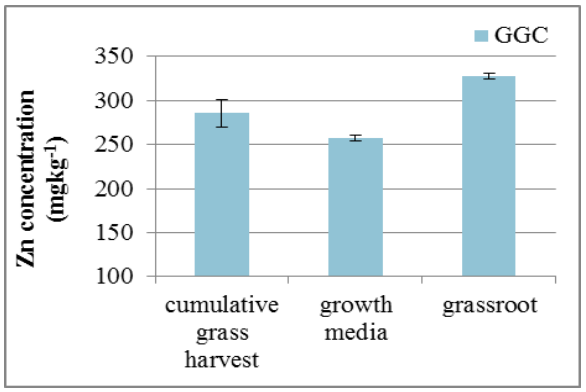


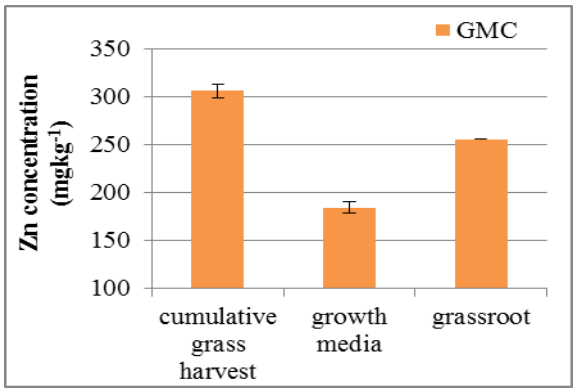

NBMC
Zn concentration (mgkg-1)
350
300
250
200
150
100
cumulative grass harvest
growth media
grassroot

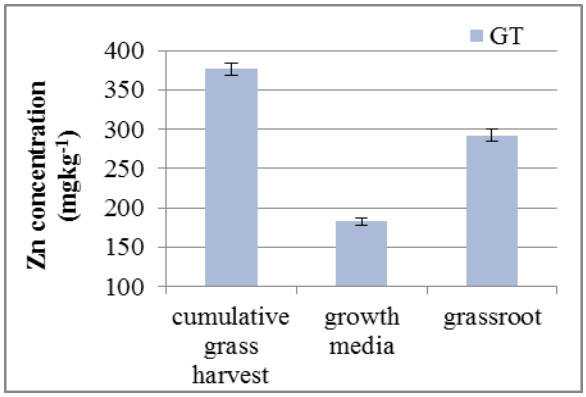

NBT
Zn concentration (mgkg-1)
450
400
350
300
250
200
150
100
cumulative grass harvest
growth media
grassroot

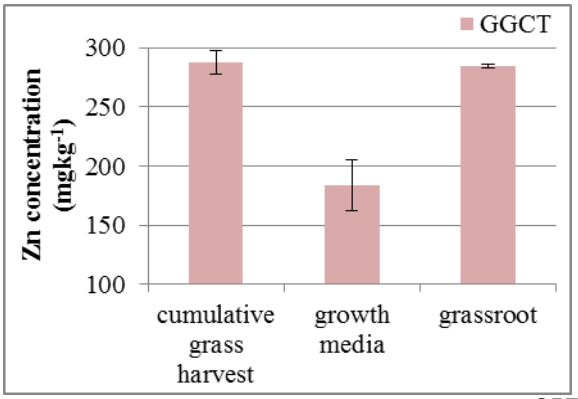

NBGCT
Zn concentration (mgkg-1)
400
350
300
250
200
150
100
cumulative grass harvest
growth media
grassroot


NBMCT
Zn concentration (mgkg-1)
350
300
250
200
150
100
cumulative grass harvest
growth media
grassroot


GGC
Zn concentration (mgkg-1)
350
300
250
200
150
100
cumulative grass harvest
growth media
grassroot


GMC
Zn concentration (mgkg-1)
350
300
250
200
150
100
cumulative grass harvest
growth media
grassroot


GT
Zn concentration (mgkg-1)
400
350
300
250
200
150
100
cumulative grass harvest
growth media
grassroot


GGCT
Zn concentration (mgkg-1)
300
250
200
150
100
cumulative grass harvest
growth media
grassroot

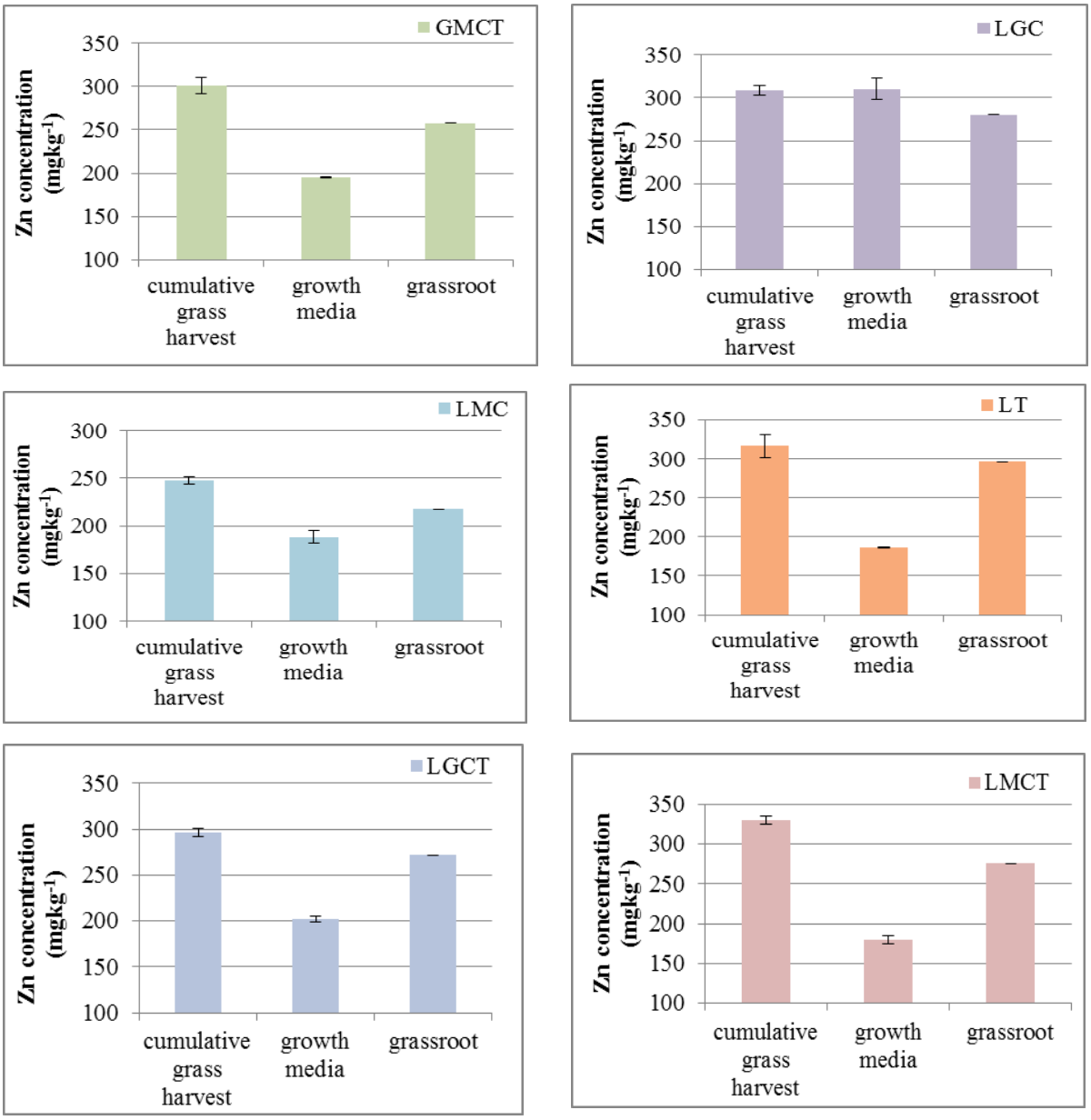


Figure 4.14r: Trends of zinc concentrations in cumulative grass harvests, growth media and grass roots.

In summarising the results of heavy metal concentrations in the cumulative grass harvest, growth media and grass roots across the test profiles, growth media retained the highest concentrations of added heavy metals followed by grass roots; grass shoots retained the least heavy metals as shown by the scales of the graphs in figures 4.15a-c.

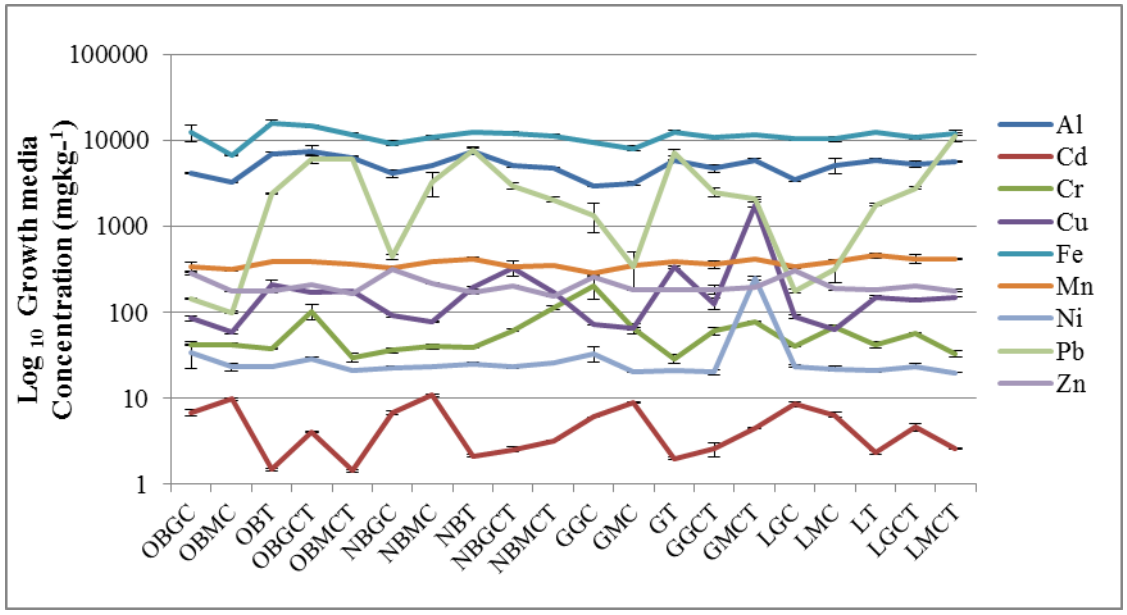


Figure 4.15a: Heavy metal concentrations in growth media derived from test profiles.

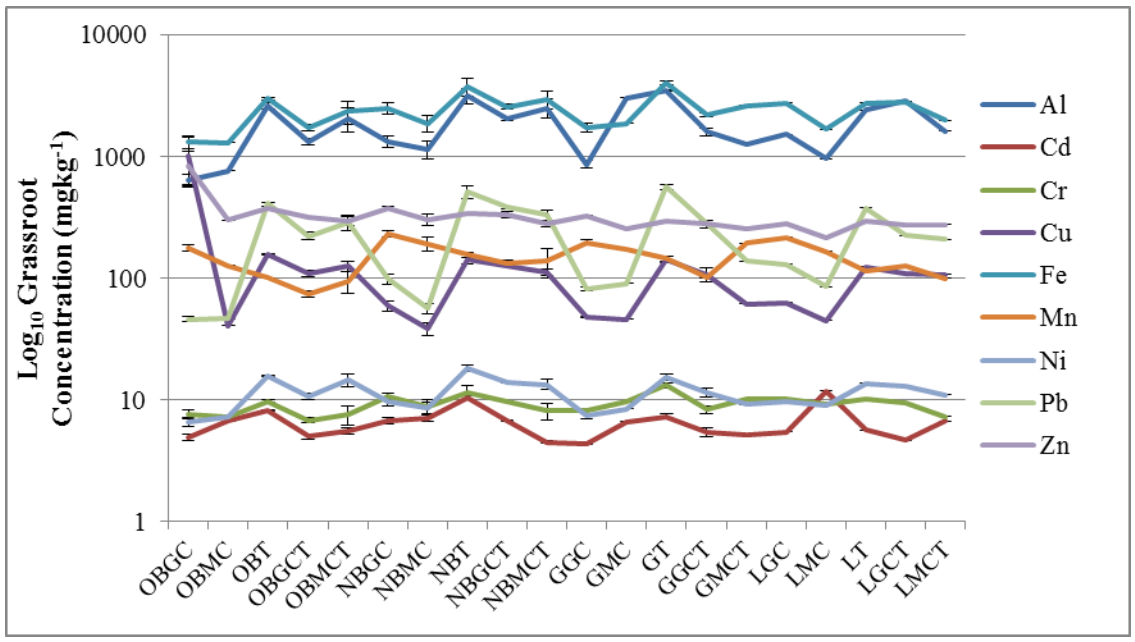


Figure 4.15b: Heavy metal concentrations in grass roots derived from test profiles

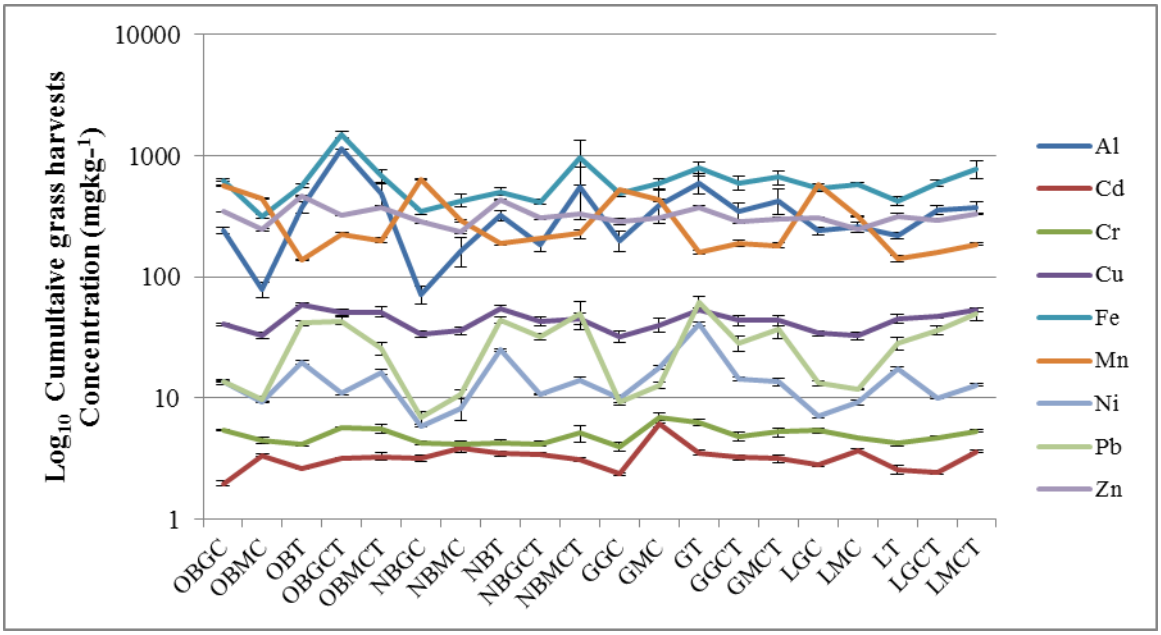


Figure 4.15c: Heavy metal concentrations in grasses derived from test profiles

Fe and Al predominantly had the highest concentrations in growth media, grass roots and grasses while Cd had the lowest concentrations in the three components as shown in figures 4.15a-c. The high concentrations of Al and Fe are explained by the initial high concentrations of Al and Fe in the test samples (see tables 4.1 and 4.2). However these high values are still within typical soil values for Al and Fe as described in section 4.2.7.

Statistical analysis of heavy metal concentrations in harvested grasses

Results of analyses on the grasses, growth media and grass roots in section 4.3.3.2 showed that most profiles retained highest concentrations of seven of the nine test heavy metals within their growth media. However, to determine which growth media and/or aggregates were directly responsible for the trends of heavy metal retention, statistical analyses were carried out on the results derived from the heavy metal analysis of the harvested grasses, growth media and grass roots (appendix 24-32).

Two-way ANOVA analysis was carried out on the total heavy metal concentrations present in three monthly grass harvests derived from the test profiles. The values used represented the

cumulative heavy metal concentrations present in the grass shoots over three months, after eight weeks of heavy metal spiking. Table 4.18 shows the results of the statistical analysis:

p-values	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
Growth media	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**
Aggregates	**0.000**	**0.000**	**0.000**	0.079	**0.019**	**0.000**	**0.000**	0.144	**0.000**
Interactions	**0.000**	**0.000**	**0.000**	**0.001**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**

Where p <0.05 = significant; p<0.01 = very significant; p<0.001 = highly significant

Table 4.18: Results of Two-way ANOVA analysis carried out on heavy metal concentrations present in harvested grasses

Table 4.18 shows that there were highly significant interactions between growth media and aggregates for the nine grass metal concentrations and as such the relationship between growth media and aggregates could not be commented upon separately. A post hoc test was therefore carried out to identify where significant differences were as shown in Table 4.19.

Post hoc test	Growth media	Aggregates
Al	T, **GCT, MCT**	OB, **G**
Cd	MC, T, **GCT, MCT**	NB, L, **G**
Cr	GC,MC, **GCT, MCT**	NB, L, **G**
Cu	MC, T, **GCT, MCT**	OB
Fe	**GCT, MCT**	OB
Mn	GC, MC, **GCT, MCT**	OB, NB, **G**
Ni	MC, T, **GCT, MCT**	NB, **G**
Pb	T, **GCT, MCT**	**G**
Zn	GC, T, **GCT, MCT**	OB ,NB, **G**

Table 4.19: Results of post hoc tests showing growth media and aggregates whose interaction significantly influenced the retention of heavy metals in grasses

The results of the post hoc testing showed that interaction between the highlighted (in bold) growth media and aggregates shown in Table 4.19 significantly influenced the retention of heavy metals. Generally, the post hoc test showed that interactions between growth media GCT and MCT and aggregate G consistently influenced most heavy metal retention by grasses.

Statistical analysis of heavy metal concentrations in growth media

In addition to grasses, a two-way ANOVA test was also carried out on growth media heavy metal concentrations derived from the test profiles. The values used represented the cumulative heavy metal concentrations present in the growth media after three months (i.e. two months of heavy metal spiking and one month after spiking stopped).

p-values	**Al**	**Cd**	**Cr**	**Cu**	**Fe**	**Mn**	**Ni**	**Pb**	**Zn**
Growth media	**0.000**	**0.000**	**0.009**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**
Aggregates	**0.004**	0.128	**0.003**	**0.000**	**0.015**	**0.003**	**0.000**	0.329	**0.020**
Interactions	**0.002**	**0.000**	**0.000**	**0.000**	**0.003**	**0.041**	**0.000**	**0.000**	**0.000**

Where $p < 0.05$ = significant; $p < 0.01$ = very significant; $p < 0.001$ = highly significant

Table 4.20: Results of Two-way ANOVA analysis carried out on heavy metal concentrations present in growth media.

Table 4.20 shows that interactions between growth media and aggregates were highly significant for heavy metals, and so the relationship between growth media and aggregates could not be individually commented upon. Therefore a post hoc test was carried out to identify where significant differences were.

Post hoc test	Growth media	Aggregates
Al	T, **GCT, MCT**	OB, **NB, L**
Cd	GC, MC, **GCT, MCT**	**NB**
Cr	GC, **GCT, MCT**	**G**
Cu	T,**GCT, MCT**	**NB**, **G**
Fe	GC, T, **GCT, MCT**	OB
Mn	MC, T, **GCT, MCT**	**L**
Ni	**MCT**	**G**
Pb	T, **GCT, MCT**	-
Zn	GC, MC, **GCT**	**NB**, **L**

Table 4.21: Results of post hoc tests showing growth media and aggregates whose interaction significantly influenced the retention of heavy metals within growth media

Table 4.21 shows that significant interactions between the highlighted growth media and aggregates influenced the retention of test heavy metals in growth media, except for Pb where significant differences were found only in the stated growth media. Generally, interactions between growth media GCT and MCT and aggregates NB, G and L consistently influenced retention of heavy metals within growth media.

Statistical analysis of heavy metal concentrations in grass roots

Statistical analysis was carried out on heavy metal concentrations present in the grass roots in order to determine which growth media and/or aggregates influenced the retention of heavy metals by grass roots. This was achieved by carrying out a two-way ANOVA on the cumulative heavy metal concentrations present in the grass roots over three months after eight weeks of heavy metal spiking.

p-values	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
Growth media	**0.000**	**0.000**	**0.010**	0.258	**0.000**	**0.000**	**0.000**	**0.000**	0.051
Aggregates	**0.019**	**0.000**	**0.018**	0.125	**0.018**	**0.001**	**0.014**	**0.006**	0.052
Interactions	**0.007**	**0.000**	0.401	0.071	0.360	0.421	0.129	**0.011**	0.136

Where $p < 0.05$ = significant; $p < 0.01$ = very significant; $p < 0.001$ = highly significant

Table 4.22: Results of Two-way ANOVA analysis carried out on heavy metal concentrations present within grassroots.

Table 4.22 shows that there were highly significant interactions between growth media and aggregates for Al, Cd and Pb and so the relationship between growth media and aggregates could not be separately commented upon, therefore a post hoc test were carried out. There were very significant differences in growth media and the aggregates for Cr, Fe, Mn and Ni which implies that both growth media and aggregates highly influenced the retention of these heavy metals in grass roots. However, there was no significant difference for Cu and Zn suggesting that growth media, aggregates or their interaction did not influence heavy metal retention by grass roots. The post hoc test was also carried out to identify where significance was in both growth media and aggregates.

Post hoc test	Growth media	Aggregates
Al	GCT, MCT, **T**	**NB**, G
Cd	MC, **T**	**NB**, L
Pb	GCT ,MCT, **T**	**NB**, G
Cr	**T**	**NB, G**
Fe	MCT, **T**	**NB**, G
Mn	GC, MC, **T**	L, **NB**, G
Ni	GCT, MCT, **T**	**NB**
Cu	GC	OB
Zn	GC	OB

Table 4.23: Results of post hoc tests showing growth media and aggregates and their interaction in the retention of heavy metals within grass roots

Table 4.23 shows that significant interactions between the highlighted growth media and aggregates for Al, Cd and Pb influenced the retention of heavy metals within grass roots. Although there were no significant interactions between growth media and aggregates for Cr, Fe, Mn and Ni, table 4.23 highlights the components that significantly influenced heavy metal retention by grass roots. There were no significant differences and interactions between growth media and aggregates for Cu and Zn and this was influenced by GC and OB. Overall, interactions between T and NB influenced the retention of heavy metals by grass roots except for Cu and Zn. GC and OB did not influence heavy metal retention by grass roots.

4.3.3.3 Monitoring of motor oil concentrations in leachates from test profiles

Motor oil analysis showed that after adding 4.8ml of clean and used motor oils (described in section 3.4.3) which was equivalent to a year's worth of oil in a typical urban environment (Wilson *et al*., 2003), all test profiles retained most of their oil contents over the three months test period, as oil concentrations in leachates were below limits of detection of 1mg/L.

4.3.4 Fifth objective: Monitoring motor oil retention in aggregates

Motor oil retention experiments on six varying weights of each of the four test aggregates were carried out with clean motor oil (C) and used motor oil (U), as described in appendix 33, to observe their oil retention capacities. The experiment was carried out until aggregate weights became fairly constant (i.e. eight weeks for OB and NB, and four weeks for G and L) and the results are represented below as percentage increase in weight:

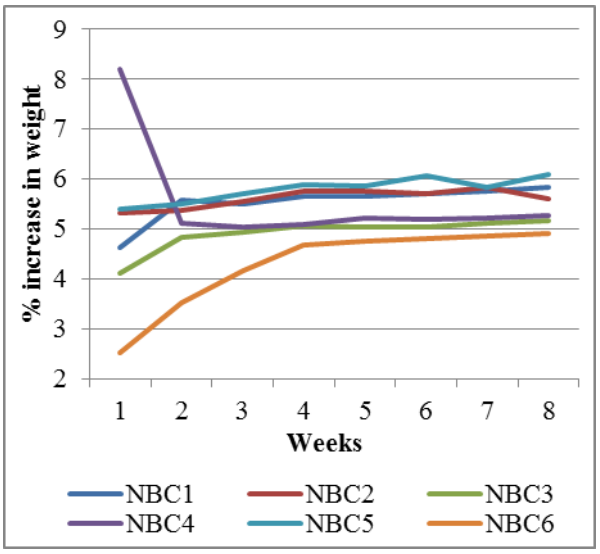


Figure 4.16a: Percentage increase in weight of new bricks in clean oil

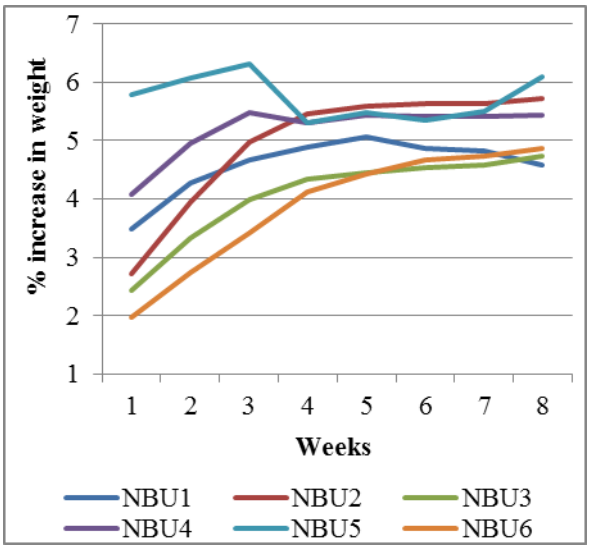


Figure 4.16b: Percentage increase in weight of new bricks in used oil

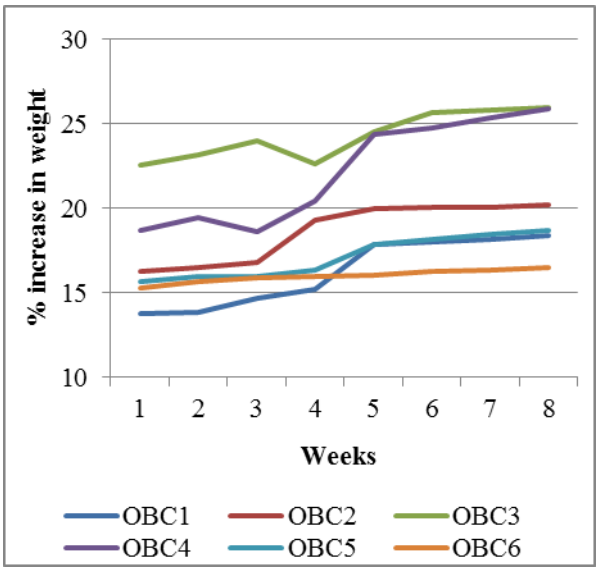


Figure 4.16c: Percentage increase in weight of old bricks in clean oil

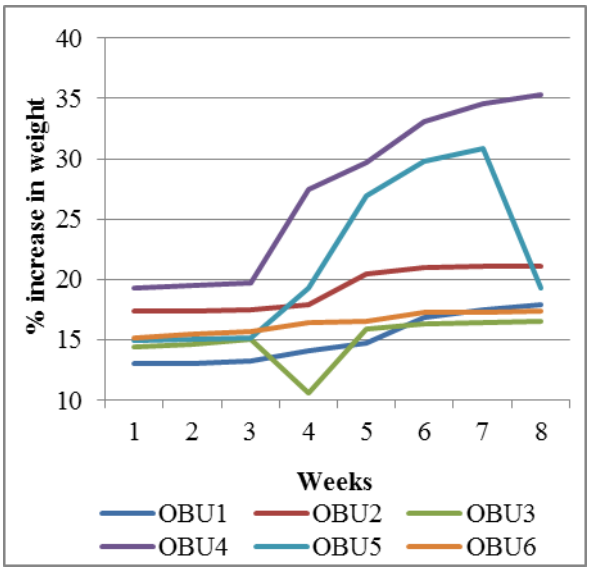


Figure 4.16d: Percentage increase in weight of old bricks in used oil

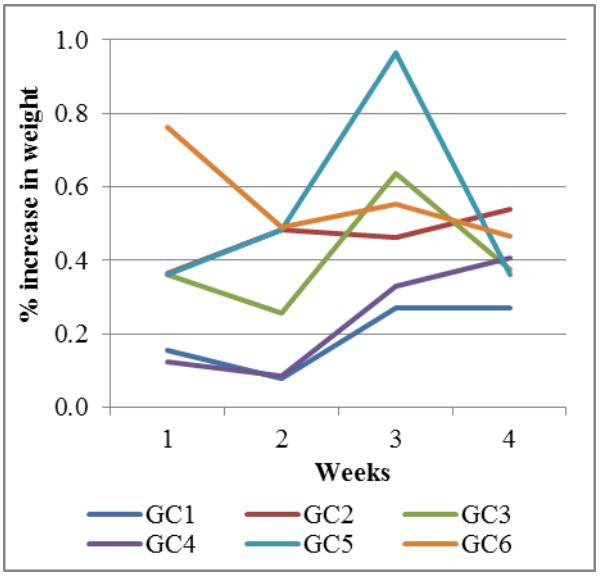


Figure 4.16e: Percentage increase in weight of gravel in clean oil

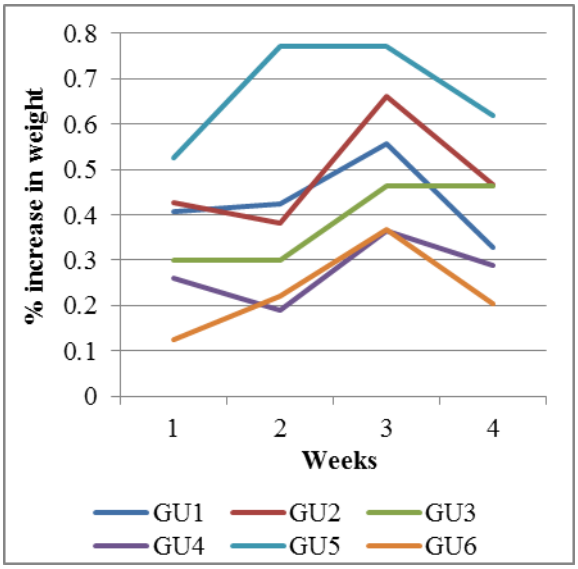


Figure 4.16f: Percentage increase in weight of gravel in used oil

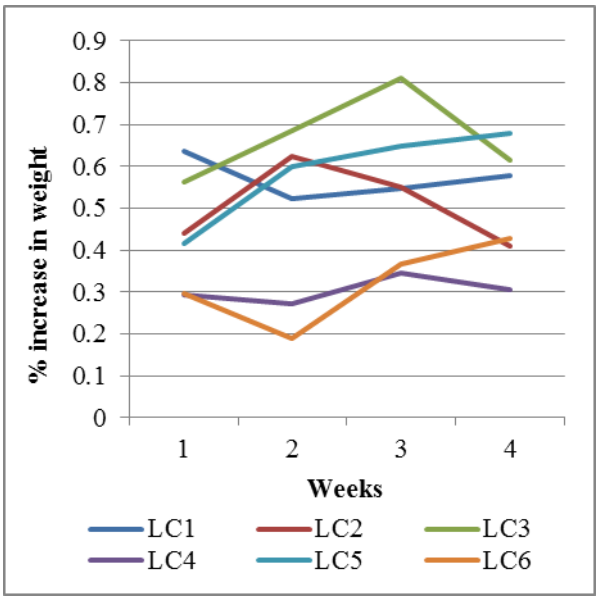


Figure 4.16g: Percentage increase in weight of limestone in clean oil

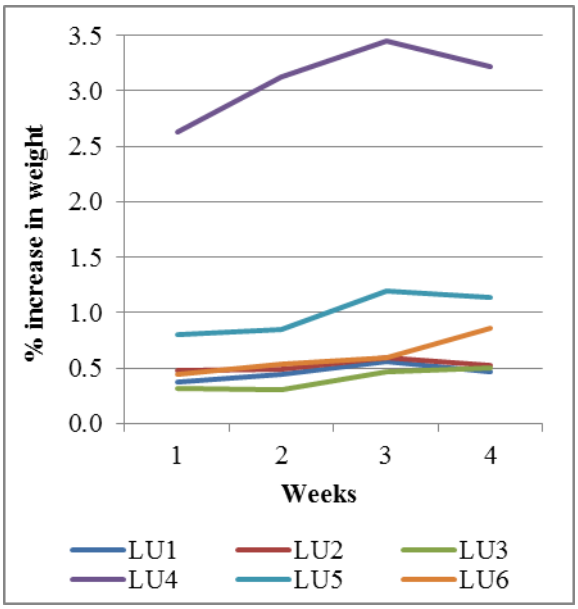


Figure 4.16h: Percentage increase in weight of limestone in used oil

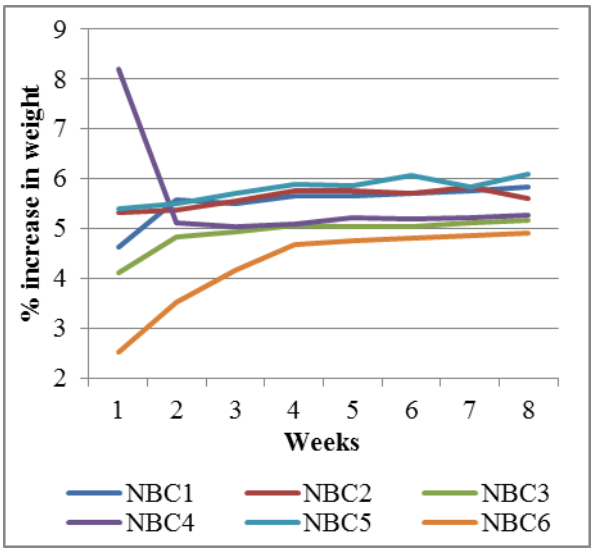


Figure 4.16a: Percentage increase in weight of new bricks in clean oil

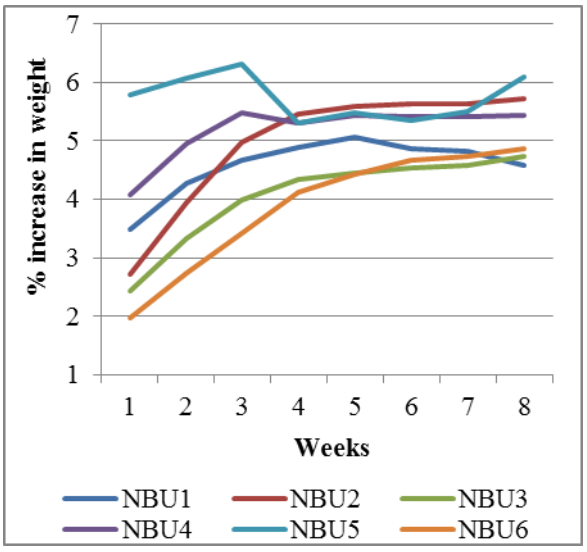


Figure 4.16b: Percentage increase in weight of new bricks in used oil

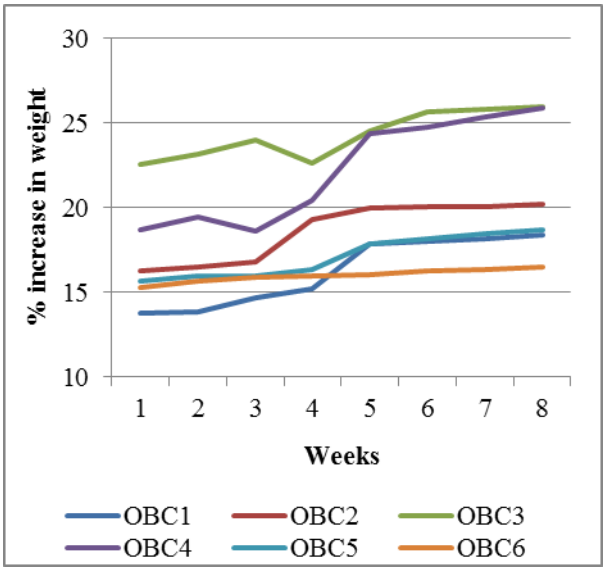


Figure 4.16c: Percentage increase in weight of old bricks in clean oil

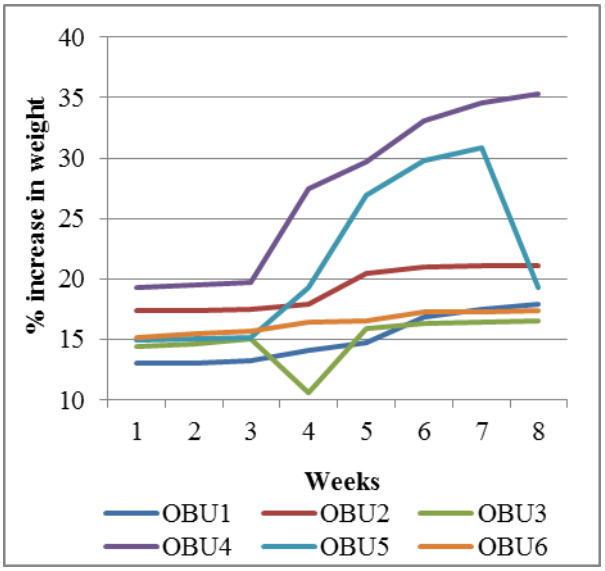


Figure 4.16d: Percentage increase in weight of old bricks in used oil

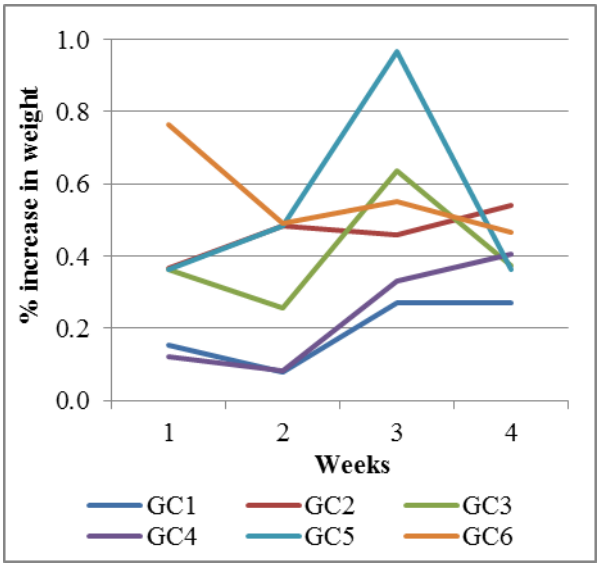


Figure 4.16e: Percentage increase in weight of gravel in clean oil

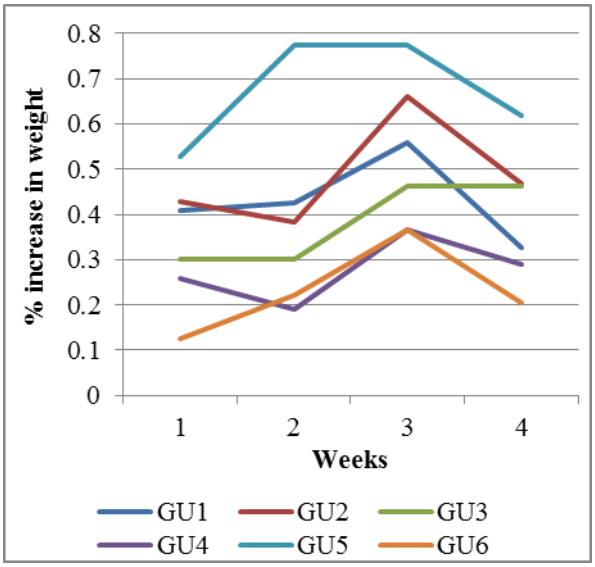


Figure 4.16f: Percentage increase in weight of gravel in used oil

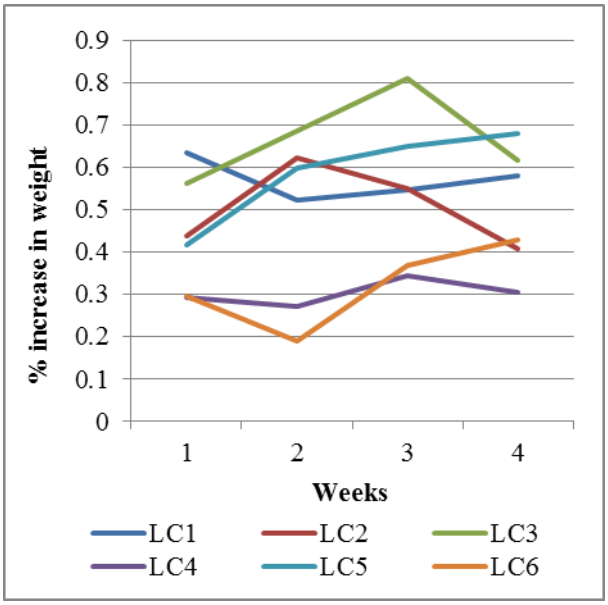


Figure 4.16g: Percentage increase in weight of limestone in clean oil

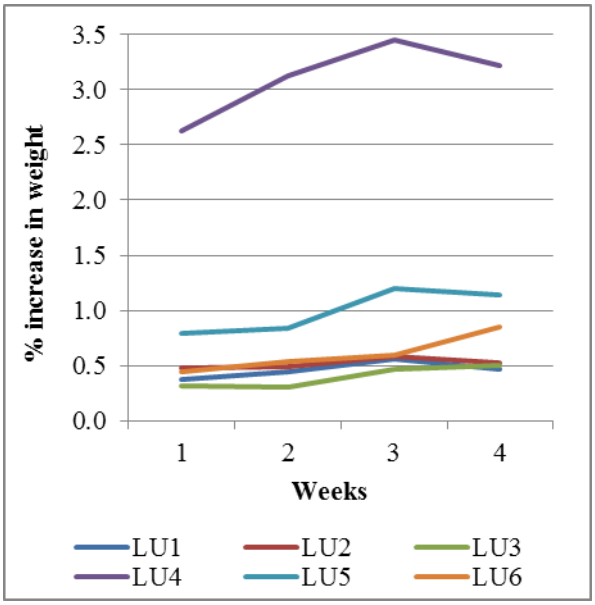


Figure 4.16h: Percentage increase in weight of limestone in used oil

Figures 4.16a-h show that over time, aggregates were able to absorb motor oil, with OB being the highest absorbers of clean and used oil, as indicated by their increase in weight. The least oil absorbers were G and L. NB and G absorbed clean and used oil similarly however OB and

L absorbed more used oil than clean oil. Also, as seen in the appendix 33, larger aggregates did not necessarily absorb larger quantities of oil but on the contrary, smaller aggregates absorbed the highest weights of oil across the four aggregates due to their larger surface area (Khalaf and DeVenny, 2005).

Leaching studies carried out on aggregates dosed with used and clean motor oil with initial concentrations of 2767mg/L (an equivalent of eight months' worth of oil loading in a typical urban environment (Wilson *et al.*, 2003) was carried out over three weeks to further determine oil retention capacity of the test aggregates. Concentrations of oil in aggregate leachates were measured and compared to G (control) as described in appendix 33. Figure 4.17 shows that used oil leached from all the aggregates into water in the first week, with G producing the highest leachate used oil concentration at 1.62mg/L. However, the overlapping error bars of the four aggregates in the first week showed that leaching of oil was not statistically significant, indicating that leaching of used oil from the recycled aggregates were similar to that of G regardless of their varying oil leachate concentrations. In the second week, used oil in leachate was below limits of detection for all the aggregates except for G whose leachate oil concentration had reduced to 1mg/L. By the third week, used oil in leachate was below limits of detection in leachates derived from all the aggregates.

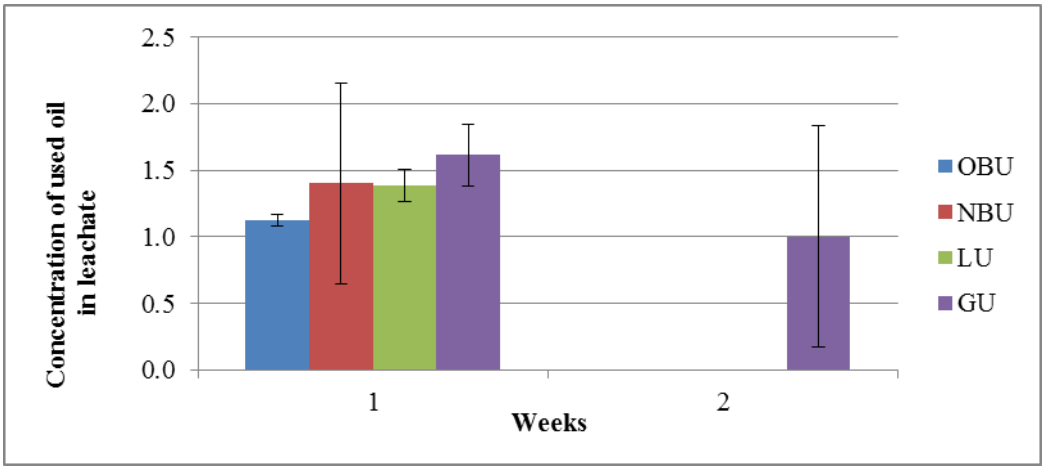


Figure 4.17: Oil concentrations in leachates derived from aggregates dosed with used oil

For clean oil, oil concentrations were all below limits of detection in leachates for all the aggregates except for G in the first and second week with clean oil concentrations of 1.2 mg/L and 1.7mg/L respectively. Overall, these results show that compared to the initial oil concentrations of 2767mg/L, recycled aggregates retained at least 99.9% of added motor oil with OB, NB and L retaining more clean and used oil compared to G.

4.4 Conclusion

To further improve the sustainability of vegetative SuDS, alternative materials to topsoil and gravel, which are natural resources, were analysed. Alternative materials employed consisted of compost as growth media, bioreactor and remediator of pollutants, and RA as sub-base aggregates and remediator of pollutants, and their performances were compared with the controls i.e. T and G. In order to assess the performance of MC and GC compared to T, and OB, NB and L compared to G in vegetative SuDS, baseline analyses followed by experimental designs were established to determine the ability of these materials to deal with pollutants that would otherwise compromise water quality in vegetative SuDS. The analyses were carried out in line with the aims and objectives of the study.

Assessment of test materials commenced with baseline analyses which included determination of moisture content, organic matter, carbonate content, water holding capacity, bulk density, pH, total heavy metal content, heavy metal content, bacterial and fungal enumeration and identification, and wtare quality assessments. Data derived from baseline analyses were compared to PAS 100:2011 (BSI, 2011), CSUGE (WRAP, 2003), ICRCL 59/83 values and Kelly indices guideline values (ContaminatedLand, 2000). This first stage of assessment was in fulfilment of the first objective which was characterisation of test materials by baseline analyses. Baseline analyses suggested that the high moisture content, moderate bulk density and slightly alkaline pH of GC coupled with its background total heavy metal concentration falling within the PAS100:2011 (BSI, 2011) standard for composted materials and its background bioavailable heavy metals in effluent falling well within the WHO (2011) potable water guidelines, would make GC a suitable growth media and bioreactor to replace or

complement the use of T in vegetative SuDS. However, MC possessed some beneficial qualities over GC such as a higher organic matter and carbonate content, higher microbial populations and diversity (oil degraders) and higher alkalinity, which are desired qualities required for pollutant removal and improvement of stormwater quality in vegetative SuDs as well as sustenance of vegetative growth (Rosell *et al*., 2001; Kaschl *et al*., 2002; Lafuente *et al*., 2008). RAs were within the Kelly values for uncontaminated materials (ContaminatedLand, 2000). Baseline results formed the bases for further tests which were carried out on the test materials to assess pollutant retention and degradation and vegetative growth, in order to assess the abilities of the test materials in improving water quality in vegetative SuDS.

The second objective of investigating biofilms development in compost in simulated swale conditions was achieved by carbon dioxide monitoring, for assessing microbial activity which is indicative of biodegradation of pollutants in test compost samples under conditions similar to that which occur in swales at various times. Analyses of microbial activity showed that under conditions of restricted oxygen at low levels of moisture, there was a decline in microbial activity over time in GC, MC and T with the lowest activity occurring significantly in T; however, there was no significant difference in microbial activity between GC and MC. In aerobic, wet conditions there was increased microbial activity over time in the compost samples, with T still maintaining the lowest microbial activities and no significant difference in microbial activities between GC and MC. However, microbial activities were more prolific in the light than in the absence of light for all the samples. These results suggested that GC and MC would be suitable as replacements for topsoil, in terms of pollutants biodegradation in vegetative SuDS devices in varying swale conditions.

The third objective of grass development in simulated swale conditions was achieved by measuring grass biomass derived from pot trials. At this stage, apart from being assessed individually, the recycled materials were also combined with control materials giving rise to twenty test profiles, and these were also employed in the remaining analyses. These combinations were carried out to ascertain the qualities of the recycled materials both as a

substitute and supplement to the controls. In assessing grass biomass yield over a five-month period, profiles containing GC followed by MCT produced the highest biomass yield while MC produced the least over time. Also, all profiles containing L consistently produced the highest grass biomass compared to G, especially from the third to the fifth month, while profiles containing OB produced the least biomass by the fifth month. Biomass yield from test profiles was statistically compared to biomass yield from control profiles i.e. G and T and it was found that GC, GCT and MCT interacted with aggregates OB and NB to produce significant grass biomass, with GC and MCT producing significant biomass increase compared to T; and L performing as well as G. The implication of this result was that the interaction of aggregates with growth media could significantly influence biomass yield. This is because RA may contain nutrients, useful structural properties or harmful compounds which may enhance or be detrimental to biomass yield (Khalaf and DeVenny, 2004; Dhir and Paine, 2007).

The fourth objective was to investigate the efficacy of compost and RA in remediating pollutants in simulated swale conditions and this was achieved by carrying out leaching analyses on effluents derived from test profiles dosed with heavy metals and motor oil in swale simulations, in order to assess their ability to remove pollutants. Also, total heavy metal and motor oil concentration analyses of test profiles were carried out. Results showed that after the addition of high concentrations of heavy metals to the profiles, almost all heavy metal concentrations analysed in leachate remained below the WHO (2011) potable water guideline by the end of the experimental period except for Al and Cd, which remained above the WHO (2011) guideline in some profiles containing GC and T but below toxicity levels for freshwater organisms. Comparing concentrations of leached heavy metals with concentrations added to test profiles; results showed that ≥98% of all added heavy metals were retained within the test profiles. Statistical analyses showed that unlike grass biomass yield, aggregates and their interactions with growth media had no significant effect on leaching of heavy metals and so leaching was significantly influenced by growth media alone, specifically GC and T. Oil analyses of leachates derived from profiles dosed with an equivalent of a year's worth of

oil in a typical urban environment (Wilson *et al*., 2003) showed that oil concentrations in leachates were below limits of detection. The implication of these results to vegetative SuDS is that in worst case scenarios such as runoff from land heavily contaminated with heavy metals (ContaminatedLand 2000), compost can perform as well as T in vegetative SuDS in retaining ≥98% of common heavy metal pollutants in runoff; with ≤2% of heavy metals being leached into groundwater (especially influenced by GC and T). Also in cases of heavy spillages of oil into the environment, test profiles will perform as well as profiles comprising G and T in the remediation of motor oils.

Due to the high retention of heavy metals (≥98%) within the test profiles, profile components (i.e. grass shoots, grass roots and growth media) were analysed for total heavy metal concentrations. Analyses showed that growth media retained the highest concentrations of added heavy metals followed by grass roots, while grass shoots retained the least heavy metal concentrations. Seven of the nine test heavy metals were mostly retained within the growth media of most profiles i.e. Al, Cr, Cu, Fe, Mn, Ni, and Pb. Cd was predominantly retained within the grassroots while Zn was retained both in the growth media and grass roots. Statistically, interactions between GCT and MCT, and NB, G and L consistently influenced heavy metal retention by grasses and growth media, while T and NB influenced all heavy metal retention by grass roots except for Cu and Zn. This analysis further confirmed that for remediation of heavy metals within vegetative SuDS, growth media would predominantly act as a sink for most heavy metal pollutants (specifically for Al, Cr, Cu, Fe, Mn, Ni, Pb and Zn), while grass roots would predominantly act as a sink for Cd and Zn. Varying heavy metal concentrations in the three grass harvests suggested that heavy metals within vegetative SuDS devices are capable of migrating to and from growth media and roots into grasses thereby encouraging phytoremediation, but proper maintenance has to be in place to prevent heavy metals in grasses from returning into groundwater especially when the plants die (Wilson *et al.*, 2004;Woods-Ballard *et al.*, 2007). Analysis of motor oil concentrations in test samples were not carried out in this study, but studies by Napier *et al*. (2008) have shown that 81% of motor oil added to topsoil in SuDS conditions were degraded, and therefore it was expected in

this study that most of the motor oil retained within the test profiles would have been degraded (Bernal, Alburquerque, and Moral, 2009).

The fifth objective of this study was to investigate the effect of RA on water quality and oil pollution attenuation in simulated swale conditions. To fulfill this aim, oil absorption studies and oil leachate analyses was carried out on test RA. Oil absorption studies showed that OB was the highest absorber of clean and used oil, while G and L were the least absorbers. Also, smaller aggregates absorbed the highest weights of oil across the four aggregates compared to larger aggregates. Leaching studies showed that G yielded the highest concentrations of used and clean motor oil in leachate compared to the recycled aggregates.

Apart from achieving the aims and objectives of this research, these results have shown that compost and recycled aggregates would fare better either as supplements or as substitutes for T. Growth media GC and MCT can substitute for T in grass development, biodegradation of organic pollutants and in the uptake of heavy metals in vegetative SuDS profiles. OB can replace G in vegetative SuDS for oil absorption thereby improving water quality, and L can be used as aggregate supplements for enhanced biomass yield.

Chapter five: Discussion

5.1 Introduction

To successfully establish vegetative SuDS devices on a particular site, there are various factors which need to be considered including (but not limited to): soil type, soil pH, infiltration and drainage of surface runoff, quality of surface runoff, types and treatment of runoff pollutants, vegetation type and biomass development, climatic conditions such as wet and dry weather, microbial activities and type of aggregate bases (Centre for Alternative Land Use, 2006). However, the over-riding factor that needs to be addressed is the issue of sustainability. Construction of vegetative SuDS involves the use of natural resources, such as gravel and topsoil, which have significant environmental impacts such as resource depletion, harmful emissions and waste generation (Shaffer *et al*., 2009; WRAP, 2010). For vegetative SuDS to be effective:

- there must be dense vegetative cover for attenuation and primary treatment of runoff (Highways Agency, 2006; Woods-Ballard *et al*., 2007),
- there must be adequate soil layer for infiltration and further treatment of runoff and its attendant pollutants, as described in table 2.5 and 2.6 (Highways Agency, 2006),
- they should incorporate gravel beds for water retention and stability of the structure (American Rivers, 2004),
- they must contribute to the landscape/amenity value of a particular area (Revitt and Ellis, 2001; Highways Agency, 2006) and ultimately,
- they help to reduce urban heat island effects (Wilby and Perry, 2006) and enhance carbon sequestration thereby mitigating the effects of climate change (Tratalos *et al.*, 2007; Charlesworth, 2010).

The purpose of this research was to improve the sustainability of conventional vegetative SuDS components (i.e. gravel and topsoil) by replacing or supplementing them with recycled materials, which are more sustainable. The recycled materials employed included compost and recycled aggregates. Previous chapters have described how these recycled materials were sourced and how background characterisation helped to identify the properties they possessed

which then formed the basis for further experimental analyses. Experiments were carried out on composts to determine their suitability as growth media, bioreactors and pollutant mitigators; and on recycled aggregates to determine their suitability as sub-base aggregates and pollutant mitigators, in vegetative SuDS. Compost samples consisted of green compost and mixed compost (GC and MC) with topsoil (T) as control, while recycled aggregates comprised of crushed old and new bricks, and limestone aggregates (OB, NB and L), with gravel (G) as control. Findings from this study showed that compost and RA were able to fulfill the roles described above as well as T and G, and in some cases performed better than the conventional materials. It was also discovered that the benefits of recycled materials can be maximised if they are combined with each other or with the conventional materials. This chapter discusses the beneficial qualities of compost and RA in line with the stated aims and objectives of this study as well as their applicability in improving water quality and sustainability in vegetative SuDS devices. Limitations encountered during the course of the research were also highlighted as well as recommendations for further research on this work.

5.2 Characterisation of compost and recycled aggregates

The first objective of this study, which was the characterisation of test materials, was carried out and achieved by comparing derived baseline data with requirements for PAS 100:2011 (BSI, 2011), CSUGE (WRAP, 2003), ICRCL 59/83 values and Kelly indices guideline values (ContaminatedLand, 2000), as well as data obtained in literature.

Moisture content in growth media

Moisture content determination on green and mixed compost as well as topsoil showed that initial moisture contents of GC followed by MC were significantly higher than T. Soils rich in organic matter, such as soil amended with GC or MC, are usually characterised by increased microbial activity and decomposition of organic matter, with one of the limiting factors being availability of moisture (Stark and Firestone, 1995; Rey *et al*., 2002). Studies by Keith *et al*., (1997) and Rey *et al*., (2002) have shown that moisture is a good predictor of soil microbial activity with high microbial respiration rates being observed in winter, spring and autumn, coinciding with periods of lower evaporation due to lower temperatures, and after rainfall

events in the summer. Moisture is also directly related to growth of vegetation and plant development (Hewitt, 2004) in a variety of processes such as transpiration and photosynthesis which are required for plant growth and development (Denmead and Shaw 1962); root growth (Katterer *et al*., 1995), plant biomass development (Wellard, 1987; Bowman *et al*., 1995) and plant growth-promoting microorganisms such as rhizobacteria (Kloepper *et al*., 1980). In addition, moisture is vital in the treatment of runoff pollutants (Hewitt 2004) and maintaining microbial activity and diversity in the growth media (Schnürer, 1986; Rey *et al.*, 2002), which in turn enhances the biodegradation of organic compounds such as hydrocarbons in motor oil (Davidson *et al*., 2000) and organic matter (Vallini *et al*., 2002; Liang *et al*., 2003; Pommier and Lefebvre 2009).

When moisture content is low, it can (a) reduce or inhibit microbial activities which is shown by a reduction in carbon dioxide (CO_2) evolution (Nakasaki *et al*., 1994; Ryckeboer *et al*., 2003), (b) act as a limiting factor in nutrient cycles such as the nitrogen and carbon cycles (Malhi and McGill, 1982; Stark and Firestone, 1995; Fierer and Schimel, 2002), (c) decrease photosynthetic rates and biomass production in plants and (d) inhibit the enhancing effect of temperature on plant growth (Kolb *et al.*, 1990; Ambebe and Dang, 2009; Ambebe and Dang, 2010). Low soil moisture content can also reduce infiltration rates of surface runoff because low moisture encourages the compacting of soils (especially clayey and loamy soils) thereby increasing the penetration resistance of the soil by plant roots and reducing the porosity of the soil, which in turn lowers infiltration of runoff (Ayers and Perumpral, 1982; Iijima *et al*., 2003; David and Sousa, 2008).

On the other hand, excessive moisture can depress vegetative growth due to saturation of soil which leads to the depletion of oxygen required for plant respiration and anaerobic conditions which could be toxic to biomass development (Grewal and Williams, 2000; FAO, 2005). Excessive moisture can also limit the availability of soil nutrients (e.g. nitrates) to plants as excessive water can transport nutrients to regions within the soil that might be inaccessible to plant roots for uptake (Blevins *et al*., 1983; Kleinhenz *et al.*, 1997). The effects of excessive moisture are more pronounced when soil is compacted by vehicular or human traffic

(Hatchell *et al*., 1970). As a result, a balance has to be reached whereby moisture content of growth media is sufficient for plant growth and development, and biodegradation without inhibiting these processes. The CSUSE specification for moisture content in growth media described this balance as being between 35-55% (WRAP, 2003). GC's moisture content fell within this range and would therefore be able to retain its initial moisture better than MC and T. This suggests that in periods of low/no rainfall or runoff, vegetative SuDS containing compost, especially GC, would retain moisture thereby producing higher grass biomass, higher microbial activity and hence higher biodegradation of stormwater pollutants, compared T (Mahamadou *et al*., 2001; Merdun *et al*., 2008). It must however be highlighted here that there will be loss of moisture by evaporation as the growth media samples, i.e. GC, MC and T, were transported from their sources to point of analysis, and therefore the initial moisture content values obtained in this study might be lower than the actual moisture content at the source.

Water holding capacity of growth media

As with moisture content, soils high in organic matter such as loamy and compost-amended soils help in the proper drainage of soil, as the organic matter they contain helps to retain water without being water-logged (Markham, 2006; Thakur, 2006). Studies by Hernando *et al*. (1989), Shiralipour *et al*. (1992) and Mamo *et al*. (2000) have shown that one of the effects of compost in soil is to increase water holding capacity (WHC), and this is required in vegetative SuDS as proper water retention encourages sedimentation and biodegradation processes (described in sections 2.16 and 2.17). Studies by Tester (1990), Berman (1994), Edwards *et al*. (2000) and Celik *et al*. (2004) showed that compost-amended soils increased field capacity (the amount of soil-retained moisture) and available water content (moisture available for plant use); and these properties are directly related to soil porosity (Aggelides and Londra, 2000). Analysis on water holding capacity (WHC) of growth media samples in this study showed that T had the highest WHC followed by GC, while MC had the least WHC. This result suggests that when applied to vegetative SuDS devices, T and GC would retain moisture/runoff thereby enabling treatment processes to be carried out. However, the length of time for water retention is also important because prolonged water retention in soil

can lead to water-logging and development of anaerobic conditions which could slow down treatment processes, which require the presence of oxygen, in vegetative SuDS (Woods-Ballard *et al*., 2007).

Organic matter content of growth media

The presence of organic matter in soil usually helps in the absorption of moisture and improves WHC (Bot and Benites, 2005; FAO, 2005), improves soil structure and porosity (Zeytin and Baran, 2003; Celik *et al*., 2004), and increases soil microbial activity in vegetative SuDS (Marinari *et al*., 2000). Also, effective porosity is dependent on levels of organic matter content because according to Vidal-Beaudet and Charpentier (2000), the amendment of soil with 40% by volume of peat had a higher percolation rate compared to just 20% peat by volume. It therefore expected that GC would have the highest organic matter content because it had the highest moisture content. However, contrary to expectations, MC contained the highest organic matter followed by GC while T had the least organic matter content.

Bulk density of growth media

Results from analysis on growth media bulk density showed that T had the highest bulk density followed by GC with MC having the least bulk density. The very low bulk density of MC indicated that the pore spaces between its aggregates were large and loosely packed and the higher bulk density of T corresponded with its closely packed aggregate pore spaces (Celik *et al*., 2004).

At this point it must be noted that it was observed that moisture content, WHC, organic matter content and bulk density are all inter-related. Results from baseline analysis showed that MC contained the highest organic matter content compared to GC and T, and it was therefore expected that MC's initial moisture content and water retention will be the highest of the three samples because soils high in organic matter have increased capacity to store water (FAO, 2005). It was also expected that MC's bulk density would be $<1.0 gcm^{-3}$ as this is the value for soils high in organic matter (WRAP, 2003).

However, findings showed that WHC of MC, rather than being high due to its high organic matter, was the lowest of all the test samples, quickly reaching saturation when water was first added. Also initial moisture content of MC was lower than GC though higher than T. MC's bulk density was indeed $<1.0 gcm^{-3}$ but it possessed the lowest bulk density of all the samples which indicated that MC was the most porous sample (FAO, 2005; VanDerZanden and Cook, 2011). The answer to this trend lies in the large pore spaces between MC particles which makes them too porous to retain much water despite its high organic matter content. Low bulk density also explained why MC's WHC was low such that leaching occurred immediately water was added to it. Therefore, the high porosity of MC could consequently lead to leaching of nutrients, heavy metals and pollutants into groundwater thereby polluting it, as runoff in vegetative SuDS devices would not be retained long enough for the treatments described in 2.16 to occur.

Organic matter in GC was lower than MC and therefore its bulk density was higher than that of MC and thus less porous (VanDerZanden and Cook, 2011). It was expected that GC's initial moisture content and water retention values would be less than that of MC due to lower organic matter content (FAO, 2005), but results shows that GC's WHC and moisture content were higher than MC. This trend in GC is explained by its higher bulk density compared to MC, which indicated that GC's particles were more closely packed than that of MC, thereby encouraging water retention. GC retained 100% of water added to it for 24 hours before leaching occurred, which corresponded with the Highways Agency's (2006) recommended minimum residence time of 24 hours which allows the effective treatment of stormwater and runoff to take place. Therefore, applying GC to vegetative SuDS devices should encourage the retention and infiltration of runoff long enough for quality and quantity control processes (such as runoff attenuation, sedimentation of particles and suspended solids, filtration, adsorption and biodegradation of pollutants) to take place, thereby improving the quality of ground water (Wilson *et al.,* 2004; Woods-Ballard *et al.,* 2007).

Organic matter in T was the least amongst the three samples and it was therefore expected that its bulk density, initial moisture content and WHC would be the highest of the three

samples. This is because low organic matter in soil results in smaller pore spaces and dense soil structure which slows down the movement of water leading to prolonged water retention and poor soil drainage (FAO, 2005). These expectations were confirmed with T having the highest WHC and bulk density, with leaching occurring after three days, which exceeded the prescribed minimum residence time of 24 hours recommended by Highways Agency (2006). The high bulk density and extended WHC in T can be explained by the clay component of the T which has smaller pore sizes between its particles thereby retaining water for prolonged periods (Rycroft and Amer, 1995; Puhalla *et al*., 2010; Osman, 2012). T's moisture content was, however, much lower than expected and this could be explained by evaporation of water from the sample before its moisture content was analysed. Wilson *et al.* (2004) and Woods-Ballard *et al.* (2007) stated that prolonged water retention in vegetative SuDS could retard vegetative growth and create anaerobic conditions which could slow down microbial activity and biodegradation due to lack of oxygen required for microbial metabolism, thereby truncating runoff and storm water treatment. Therefore, based on water retention, treatment of runoff by T in vegetative SuDS may be slower or ineffective compared to GC.

Carbonate content of growth media

Although there was no specified standard for carbonate in the PAS 100 and CSUGE specification, Matos *et al*. (2001) and Lafuente *et al*. (2008) showed that the presence of carbonates in soil provides binding sites for heavy metals thereby removing them from solution and improving stormwater quality. Zobeck and Amante-Orozco (2001) considered carbonate concentrations of <3% to be low while Barth *et al*. (2003) reported concentrations of <5% to be low. On the other hand, Plassard, Winiarski and Petit-Ramel (2000) reported high concentrations of carbonates in soil to be ~23%, while Rodriguez-Rubio *et al*. (2003) reported high concentrations to be 30-46%. In this light, carbonate content of the three samples were considered low ranging from 3% in GC to 6% in MC. However, Barth *et al*. (2003) reported that carbonate levels as low as 5% could significantly influence water quality, when leached into water bodies, due to their high solubility. This is because the dissolution of carbonates into solution takes up the CO_2 released from the decomposition of organic matter

in soil and compost thereby producing hydrogen carbonate ions (HCO_3^-) (Barth *et al*., 2003) - see equation below,

$$CO_3^- + H_2O + CO_2 \longrightarrow 2HCO_3^-$$

The presence of HCO_3^- could increase the pH of water thereby affecting the aquatic ecosystem adversely including the death of aquatic animals (Tucker and D'Abramo, 2008). However, when these carbonates bind with available heavy metals thereby removing them from solution, they can improve water quality (Matos *et al*., 2001; Lafuente *et al*., 2008). In this study, MC had the highest carbonate content (6%) and therefore it is expected that by applying MC to vegetative SuDS, it would provide more carbonate binding sites for heavy metals compared to T or GC, thereby mitigating heavy metal pollutants in run off and improving stormwater quality.

Hydrogen ion level (pH) of growth media

Hydrogen ion level affects the populations of soil microbes and the availability of nutrients to plants as biological and chemical reactions within soil depend on the pH of compost particle surfaces in equilibrium with surrounding soil solution (Wong and Fang, 1999; Markham, 2006). For example, Shiralipour *et al*. (1992) and Stamatiadis *et al*. (1999) reported an increase in the pH of acid soils and a decrease in the pH of basic soils (Mahrous *et al*., 2006) due to increased soil buffering capacity brought about by organic acids (humic acid) present in compost organic matter. Heavy metals are less available to plants at high pH and alkaline soils easily take up heavy metals compared to acidic soils, as the presence of alkaline ions free up binding sites on compost thereby providing sorption for these compounds (Elzahabi, 2000; USDA, 2000; Jiang *et al*., 2011), rendering them unavailable to plants, thereby remediating pollutants (Ayres, Davis and Gietka, 1994; BSI, 2011). Macronutrients are less available to plants at low pH due to the precipitation of these nutrients. Hydrogen ion level also affects the activity of enzymes in plants (Sanders and Adam, 1987; de Matos *et al*., 2001 and Amir *et al*., 2005). Harpstead, Sauer and Bennett (2001) reported that pH range of 6.5 to 7.8 was most ideal for crop and biomass yield, and because the compost and topsoil samples fell within this

range, the application of these materials to vegetative SuDS in terms of pH should not pose a problem to grass growth in vegetative SuDS.

Heavy metal analysis of growth media and aggregates

The presence of heavy metals in the environment poses a threat of toxicity to ecosystems both on land and in water because they are not biodegradable and they tend to accumulate along the food chain to toxic levels (Helfrich *et al*., 1998; Dudka and Miller, 1999). Compost has been found to be effective in the removal of heavy metals from stormwater due to the sorption of dissolved heavy metal ions by compost (Quevauviller *et al*., 1996, Seelsaen *et al*., 2007). However, it was reported by Seelsaen *et al*., (2007) that compost particle size is related to rate of sorption as compost with smaller particle sizes had larger surface areas, and therefore greater sorption than compost with larger particle sizes. The affinity for heavy metal ions by compost is as a result of the organic matter (humus) content of compost occasioned by high microbial diversity and activity present in compost (Barker and Bryson, 2002). The higher the humus content of compost, the higher the heavy metal sorption; due to the presence of large amounts of strong metal binding sites such as the carboxyl, alcoholic hydroxyl and phenolic functional groups (McCarthy *et al*., 1990; Huang *et al*., 2005).

The sorption of dissolved heavy metal ions by compost reduces their mobility and hence bioavailability to soil, vegetation and groundwater, as the metals are converted to organically bound forms known as organo-metallic complexes (Chaney and Ryan, 1993; Ciavatta *et al*., 1993; Quevauviller *et al*., 1996; Huang *et al*., 2005). The consequence of this, as reported by Paré *et al*. (1999), Castaldi *et al*. (2004) and Huang *et al*. (2005), was that the concentration of dissolved (bioavailable) heavy metal ions in solution decreases while the concentration of organo-metallic complexes (non-bioavailable heavy metals) in compost increases, with an increase in the total heavy metal concentration. The organo-metallic complexes have very low solubility as the metals interact very strongly with the organic matter in compost. It was therefore concluded by Castaldi *et al*., (2004) and Amir *et al*., (2005) that determination of the concentration of total heavy metals in compost, though useful in determining the environmental impacts of compost, was not an accurate measure of the concentration of

bioavailable heavy metals because heavy metal mobility and behaviour could not be ascertained; and so bioavailable heavy metals have to be analysed separately (Sims and Sklin, 1991; Hsu and Lo, 2001).

Compost can also be a major source of heavy metal pollution in soil, accumulated over a period of time. The amount of heavy metals present in compost depends on the compost source/feedstock. Common heavy metals found in compost include zinc (Zn), copper (Cu), nickel (Ni), lead (Pb), cadmium (Cd) and chromium (Cr) (Pinamonti *et al*., 1997; Nicholson *et al*., 2003; López *et al*., 2004), and these metals can be available for plant uptake in vegetative SuDS and leached into groundwater depending on the pH level of surrounding soil solution, organic matter content and quality of the compost as well as species of vegetation (CAST, 1980; Baldwin and Shelton, 1999; Amir, 2005). Long term effects of the accumulation of heavy metals in soils are phytotoxicity, interruptions to soil microbial processes, consumption of heavy metals by humans through affected vegetation or through livestock that has grazed on affected plants (Nicholson *et al*., 2003; Lei and Run-Dong, 2010).

Results from Table 4.1 showed that total heavy metal concentrations in the growth media samples were within the PAS 100 values except for Cr, which was slightly over the Cr PAS 100 and Kelly indices limits in GC; and Pb which was extremely high in T. Studies have shown that elevated concentrations of Cr in soils have been associated with anthropogenic activities such as metal finishing, corrosion control of metals (Kimbrough *et al*., 1999; Banks, Schwab and Henderson, 2006), leather tanning and finishing (ATSDR, 2008[b]), wood treatment and preservation (Chen and Hao 1998; Solo-Gabriele *et al*., 1998; Kimbrough *et al*., 1999), combustion of coal, waste incineration, cement works, and fugitive emissions from road dusts (Environment Agency, 2002). Any of these processes might have been responsible for the high concentrations of Cr in GC. According to the Canadian Council of Ministers of the Environment (1999), Environment Agency (2002), Jankiewicz and Ptaszyński (2005), Banks, Schwab and Henderson (2006) and Boni and Sbaffoni (2009), Cr in its trivalent state in soil i.e. Cr (III) strongly adsorbs onto soil particles and organic matter, and are relatively insoluble with limited mobility; and therefore the possibility of soluble toxic hexavalent Cr

ions (Cr (VI)) polluting groundwater by leaching is minimised. However, Cr's availability and mobility in soil can be influenced by factors such as pH, soil type, particle size, organic matter content and soil moisture content. Apart from adsorption onto soil particles, Mangkoedihardjo, Ratnawati and Alfianti (2008) reported that some plants are able to take up Cr ions (<90mgkg^{-1}) without any significant effect on aerial parts while Banks, Schwab and Henderson (2006) reported a reduction in root depth as a result of high Cr concentrations in soil (310 mgkg^{-1}) which was indicative of phytotoxicity. This implied that vegetation such as grasses would be capable of taking up Cr concentrations up to 310 mgkg^{-1}from polluted surface runoff and soils before any adverse effects would be noticed which is useful in the phytoremediation of this heavy metal in vegetative SuDS.

Therefore, the elevated levels of Cr in GC should therefore not pose any adverse pollution to groundwater and water bodies because the metal would either be adsorbed onto compost particles or be taken up by vegetation. However, the vegetated components of SuDS devices need to be monitored as elevated concentrations of heavy metals in the plants could lead to phytotoxicity and diminished vegetative growth (Banks, Schwab and Henderson, 2006) which can in turn compromise the effectiveness and water quality of these SuDS devices. In addition, the anaerobic decomposition of plant matter used for phytoremediation in devices such as ponds and wetlands may increase the mobility of heavy metal ions due to the formation of soluble complexes thereby reducing water quality (Environment Agency, 2002).

Pb concentrations in T used in this study corresponded with the range of concentrations derived by USEPA (1986) from the upper layer of soils obtained from roadsides, which was 30–2,000 mgkg^{-1}higher than natural levels of 10-30 mgkg^{-1}, and it was deduced that the soil may have been contaminated with particulates derived from automobiles burning leaded fuel. Pb is a naturally-occurring element in soils but anthropogenic activities have led to elevated levels. Other sources of Pb include lead-based paint, ash from coal/wood combustion, waste incineration, and lead-containing pesticides (Bell 2003; ATSDR, 2007). Most lead-based products have been strictly regulated or eliminated due to the persistence and toxicity of lead in the environment (ATSDR, 2007). However, Bowen (1975) estimated that Pb in soil in the

UK had a residence time of between 400-3000 years which consequently means that even after the removal of sources of Pb exposure, it will still persist in soil and will take time before the effect of this regulation is felt. Lead persists in soils because it strongly adsorbs to soil, is non-biodegradable and will not quickly be absorbed by plants. Hence it is not easily leached into subsoil and groundwater and should not cause phytotoxocity in plants (Bell 2003; ATSDR, 2007).

Results from table 4.2 showed that total heavy metal concentrations in the aggregates were well below the ICRCL 59/83 and Kelly indices values except for OB which exceeded the Kelly indices value at for Mn. Mn is a naturally-occurring element and one of the uses of its ore is as a brick colourant which explains the higher concentrations of Mn in OB and NB compared to G (ATSDR, 2008[c]; USGS, 2008). Mn in recycled limestone aggregates have been attributed to the precipitation of Mn ions on limestone surfaces by oxidation processes (Chopard, Herrmann and Vicsek, 1991; Rose, Shah and Means, 2003) which are catalysed by microorganisms (Vail and Riley, 2000; Morgan, 2005). Sources of Mn in railway ballasts include emissions from moving engine parts (USEPA, 1995) and brakes of trains (Burkhardt, Rossia and Boller, 2008), and these accounted for the higher concentration of Mn in L compared to G.

Heavy metal analysis of leachates derived from growth media and aggregates

Rand, Wells and McCarty (2003) reported that fresh water was the recipient of most toxic substances generated by industries and urban areas which could be detrimental to aquatic ecosystems including fresh water organisms. To determine the potential toxicity levels of Mn, Fe and Pb derived from MC leachate, which were higher than WHO (2011) guidelines, their concentrations were compared to Mn, Fe and Pb lethal toxicity (LC_{50}) values for fresh water organisms (discussed in section 4.2.8). LC_{50} is the lethal concentration of a chemical that causes death in 50% of tested animal populations (Hill and Finster, 2010). A report by the Environment Agency (2007) showed that LC_{50} 48-hr Fe concentrations in *Daphnia magna* was 9.6mg/L, LC_{50} 96-hr Mn concentrations for *Ceriodaphnia dubia* (a species of water fleas) was 3.9mg/L (Lasier, Winger and Bogenrieder, 2000), and Offem and Ayotunde (2008)

reported that the mean acute 96-hr LC_{50} for Pb in *Daphnia magna* was 1.65mg/L. Mn, Fe and Pb concentrations in MC leachate, compared to these toxicity values, were below LC_{50} and therefore their concentrations should pose no threat to aquatic life.

Heavy metal leaching trends in background leachates of the test samples showed a persistence of higher numbers of heavy metals in MC followed by GC, while T contained the least heavy metals in leachate .These results indicate that MC had the least sorption sites for heavy metals followed by GC, while T has the highest sorption sites, and all this was probably a function of their particle sizes as discussed in above. The higher persistence of heavy metals in MC over time can be explained by the fact that its coarser particle sizes provided less surface areas and hence sorption sites for the metals thereby enhancing their mobility (Castaldi *et al*., 2004; Huang *et al*., 2005; Seelsaen *et al*., 2007). The higher surface area of T followed by GC, provided by their smaller particle sizes, created more sorption sites for the heavy metals thereby reducing their mobility and availability in leachate over time (Seelsaen *et al*., 2007). Therefore the use of compost particularly MC in vegetative SuDS may encourage the leaching of pollution heavy metals into groundwater compared to T.

Leaching trends in RA samples and gravel showed that heavy metals were mostly non-detectable in leachate and where leaching occurred, concentrations were well below the WHO (2011) potable water guidelines. This implied that the use of recycled bricks and limestone would pose no threat of heavy metal contamination to groundwater when applied in vegetative SuDS.

Microbial enumeration in growth media

The presence of organic matter in soil increases microbial populations and activity because carbon in organic matter acts as energy (Alvarez, Gagne and Anton, 1995; Weon *et al*., 1999; Chitravadivu *et al*., 2009) and nutrient source for microbial metabolism which results in increased microbial growth (Sessitsch *et al*., 2001; Bradley and Chapelle, 2010; Ros *et al*., 2011). These observations corresponded with results derived from the determination of organic matter content in growth media test samples, as MC had the highest organic matter

content followed by GC and lastly T. This therefore implies that the high organic matter content of MC provided more carbon and nutrients required for microbial growth, metabolism and diversity compared to GC or T.

Microbial identification in growth media

The presence of indigenous oil-degrading microorganisms in soil influences biodegradation of oil because they are familiar with the contaminant as well as their environment (Coker, 2006). Therefore, in identifying oil degrading species in the test growth media which would facilitate the biodegradation of oil pollutants in vegetative SuDS (Coupe, 2004), MC was found to contain the highest diversity of oil degrading microorganisms followed by T, and lastly GC. The results showed that microbial population does not necessarily give an indication of microbial diversity because though GC had higher microbial populations compared to T, T produced more microbial diversity compared to GC. This observation was explained by Sessitsch *et al*., (2001) who reported that soils with smaller soil particle sizes (in this case T) yielded higher microbial diversity compared to soils with larger particle sizes (in this case MC and GC).

Stemmer, Gerzabek and Kandeler (1999) and Kandeler *et al*., (2000) further observed that bacterial populations were highest in soils with smaller particle sizes and fungal populations were highest in soils with larger particles sizes. The reason given for this trend was that smaller soil particles provide a protective habitat for bacterial populations thereby excluding them from predatory grazing by predators such as protozoans (Heynen *et al*., 1988; Postma and van Veen, 1990; van Gestel, Merckx and Vlassek, 1996), while larger sized particles had low nutrient availability caused by increased leaching of nutrients due to higher porosity in larger soil particles and protozoan grazing (Sessitsch *et al*., 2001). The higher microbial diversity in T compared to GC was therefore not as a result of its organic matter content, which was quite low, but it was as a result of its smaller particle sizes and hence larger surface area which created more room for colonisation and helped to preserve microbial diversity compared to GC. It was therefore deduced that for successful biodegradation of organic pollutants in vegetative SuDS, media combining high microbial populations (influenced by

high organic matter content) with high microbial diversity (influenced by large surface areas) would be most appropriate. MC would not meet these criteria because though it had the highest organic matter content to sustain high microbial populations, its larger particle size may eventually cause the leaching of nutrients thereby depleting microbial populations and diversity, and exposing its microbial populations to predatory grazing. Likewise T may be unsuitable because, though its particle sizes were small enough to minimise predatory grazing compared to the compost samples, its organic matter content was low and may not sustain high microbial numbers. However, GC fitted these criteria well because its organic matter content and particle sizes were in-between the that of MC and T (i.e. though GC's organic matter was not as high as MC's, its surface area was smaller than that of MC; and though GC's surface area was not as small as T's, its organic matter content was higher than that of T), and would therefore produce the necessary microbial populations and diversity required for biodegradation in vegetative SuDS.

Coliform tests on the growth media samples showed that *E. coli* cells were present in GC, MC and T, and they were above WHO (2011) drinking water guidelines, as *E. coli* must not be detected in drinking water. However, as discussed in section 4.2.8, effluents from vegetative SuDS are ideally discharged into other SuDS devices or water bodies including groundwater and not for potable uses. Therefore, results obtained from the differential coliform tests were compared with the faecal coliform standard for effluents that have undergone treatment processes such as chlorination and ultra violet radiation (Lin, 2001; Black and Veatch Corporation, 2010), and were discharged into groundwater and surface water environments such as canals, rivers and lagoons (USEPA, 2011). This standard has a limit of 200 faecal coliforms/100ml (Akinde *et al*., 2011; USEPA, 2011) and faecal coliforms in untreated effluents from the test growth samples were well below this limit, and should therefore not pose a threat to the environment.

It is important to note that the materials used in this study were obtained from the sources described in section 3.2 and are unlikely to be representative of materials derived from other sources. If the materials used in this study are obtained from any source other than the ones

specified in this study, it is recommended that an analytical protocol, such as the ones employed in this study, be carried out to assess their baseline properties.

The outcome of the results discussed above has shown showed that compost and RA can be characterised just as well as T and virgin aggregates, and these recycled materials had even better baseline qualities compared to the virgin materials. In characterising T for use in the construction of vegetative SuDS, the history of soil on site has to be investigated to know if it is clean or contaminated because historical activities on land have significant impact on soil properties which could ultimately have detrimental effects on water quality (Rivett, Sadler and Barnes, 2011). Apart from historical site investigations, further assessments including archeology, topography, levels of contamination if present, and ecology, as well as soil physical and chemical analyses, have to be carried out. These assessments would help to characterise the T on site and if they are found to be clean and fit for use, T on site can be used for vegetative SuDS construction (Woods-Ballard *et al*., 2007; DEFRA, 2009[b]). In the case of this study, geochemical analysis showed that T was high in Pb and Cr concentration and they did not pose a problem in terms of water quality. However, in a worst case scenario, Cr and Pb's mobility and availability in soil can be influenced by factors such as pH, soil type, particle size, organic matter content and soil moisture content, which could eventually lead to phytotoxicity and contamination of ground water (ATSDR, 2007; Guanxing *et al*., 2011).

On the other hand, if T is contaminated or unfit for use (e.g. low organic matter content), it would either have to be treated *in-situ* (which may take a long time to treat and uniformity of treatment may not be guaranteed due to inability to properly homogenise and continuously mix the soil), or *ex-situ* (which may be faster and more effective than in-situ treatment but more expensive due to excavation, equipment and land use costs) (Coker, 2006), or excavated and disposed of while the importation of clean and fit-for-use T, usually from green fields, would be necessary (Jones *et al*., 1999; DEFRA, 2009[b]; European Commission, 2012). These two processes have time and cost implications. In cases where T has to be imported into a site, its standardisation would have to include first-hand assessment of its source in addition to other physical and chemical assessments, similar but not restricted to the ones carried out in

this study**.** This is because T is derived from several sources and though it is believed that T is derived from green field sites, there are now a broad range of materials sold as T which have been necessitated by the depletion of available natural T and changes in legislation to encourage recycling, waste recovery, and reduction in the dependence on landfill disposal. In addition to natural T, which is derived from green field sites and is getting depleted due to the decrease in green field sites and increase in construction, other types of T include Manufactured Topsoil and Skip Waste Soil. Manufactured Topsoil is media made up of two or more types of soil in varying proportions (such as 70% sand and 30% natural topsoil) and are expensive as they are processed using high value soil. Skip Waste Soil usually consists of a mixture of natural T, clay and fragments of building waste materials such as brick, concrete, mortar, glass, wood, metal and plastic. Skip Waste Soil is usually extremely alkaline and infertile, with elevated levels of contaminants such as heavy metals and hydrocarbons, which could lead to contamination of groundwater and water bodies. Also its fine structure could collapse if applied to vegetative SuDS thereby leading to compaction, drainage and vegetation growth problems and even flooding (DEFRA, 2009[b]).

Assessments of T are also necessary because commercial T could contain parasites, pathogens and seeds of parasitic weeds which could be detrimental to the establishment of vegetation and water quality (Ogle and Dale, 1997; Elzein and Kroschel, 2003; van der Putten, Klironomos and Wardle, 2007). Other contaminants that T may contain include dioxins which could occur naturally (Prange *et al*., 2002) or anthropogenically due to waste incineration and industrial emissions (USEPA, 2010), heavy metals derived from industrial processes, vehicular emissions (Zinkutė, Taraškevičius and Jankauskaitė, 2009) and auto repair workshops (Ipeaiyeda and Dawodu, 2008), and hydrocarbons derived from coal combustion and vehicular and industrial emissions, (Li *et al*., 2011). Characterisation of G in vegetative SuDS involves sourcing and transportation to site of use. Appropriate sourcing is necessary because processes employed at the processing plant to prepare G for sale depends on the intended use and involves washing, various separation techniques to remove impurities and crushing/screening to divide them into various grades and sizes (British Geological Survey, 2007; Highbeam Business, 2012). Therefore G of appropriate grades and sizes must be

sourced and purchased. However, unlike T, chemical analysis on G aggregates may not necessarily be carried out because they are usually chemically inert and free of hazardous chemicals such as heavy metals (Highbeam Business, 2012); and this was confirmed in this study as G contained the least concentrations of heavy metals compared to the other aggregates tested.

Therefore, the process of externally sourcing for appropriate T and virgin aggregates (specifically gravel) for the construction of vegetative SuDS could be (a) time consuming due to varieties of commercial T and virgin aggregates available (DEFRA, 2009[b]), (b) costly due to purchase, transportation and storage costs (Klimkowska *et al*., 2010), (c) sources of contaminants and (d) unsustainable due to depletion of natural resources and pollution due to transportation (Shaffer *et al*., 2009; WRAP 2010). The application of T and G, which are natural resources, in vegetative SuDS can be considered unsustainable (see section 2.8) mainly because they are excavated and utilised at rates which are faster than replenishment thereby leading to their depletion. Replenishment of these resources is very slow because they involve natural chemical, physical, biological and biochemical processes which may take years to be completed (Smith and Collen, 2004). Also, energy in the form of fuel (derived from fossil fuels and natural resources), is consumed in transporting these materials from source to destination, resulting in depletion of fossil fuels, noise pollution, increased carbon dioxide (CO_2) and particulate emissions, and air pollutants such as nitrogen oxides (NO_x), sulphur dioxide (SO_2), carbon monoxide (CO) and volatile organic carbon compounds (VOCs) (e.g. methane), all of which further lends credence to their unsustainability (Howard, 2000; Huang and Hsu, 2003; Dimoudi and Tompa, 2008).

To improve sustainability in vegetative SuDS, alternatives to T and G were required which will perform at least as well as the virgin materials but whose environmental impacts will be less significant. This study showed that compost and aggregates, when used as base materials in vegetative SuDS, would perform as well as, and even better than T and G in vegetative SuDS.

In terms of time savings, characterisation of compost should be straightforward and less time consuming because although compost components come from various sources just like T, proper composting ensures that these components undergo the same processes (described in section 2.13) thereby homogenising the matrix (Mahimairaja *et al*., 2008). Also, compost history or background may not need to be assessed if the required compost parameters meet the PAS 100 specification. Characterisation of RA on the other hand may be more time consuming because though these materials have to be sourced for (like G) to obtain the right grade and size, RA would have to be analysed to ensure it is fit-for-use and will not compromise water quality, a process which G may not have to undergo.

In terms of cost savings, the cost of purchasing compost is cheaper than that of T, and though Mahimairaja *et al*.(2008) suggested that the cost of transporting compost is more expensive than that of T due to its bulk, this can be compensated for (subject to thorough cost-benefit analysis) by conserving T on site for re-use on other projects or selling it to other consumers that may need it, such as nurseries, gardens and other construction sites (Klimkowska, 2010). There are also significant cost savings to using RA as these materials can be obtained at almost no cost from construction sites (Planning4Minerals, 2006; British Geological Survey, 2007) and further savings can be made if the source of RA is close to the site of use (Highbeam Business, 2012). Also, the application of compost in vegetative SuDS can also be an added advantage for commercial composters in terms of income generation, and for Local Authorities (LA) in terms of income generation and cost-savings in waste management, especially with the increase in landfill tax (Association for Organics Recycling, 2010), and in fulfilling the requirements for Planning Policy Statements (PPS25) regarding stormwater management. PPS25 requires that flood risks assessments be carried out by Local Planning Authorities on new developments, including flood risks from sewers and groundwater. PPS25 supports the control and reduction of runoff and encourages the use of SuDS (National SUDS Working Group, 2004; Bartens and The Mersey Forest Team, 2009).

For income generation, green wastes from households and public places can be collected and composted by the LA, packaged and sold at cheaper rates compared to commercial composts,

to consumers such as farmers and gardeners, thereby generating income for the local council. Establishment of an LA composting plant would create job opportunities for people and if this is not feasible, the cost of waste disposal to landfill can be channelled to contracting credible commercial composters. WRAP (2007[b]) and Association for Organics Recycling (2010) reported that compost generated from segregated waste streams, though largely used in agriculture as mulch, still ended up in landfill. Therefore, the cost of disposing compost to landfill can be used to offset some or all of the initial construction costs of incorporating compost into vegetative SuDS thereby conserving T and saving costs. All that will be required will be land space to store compost until needed.

In terms of sustainability, the social, economic and environmental impacts of using compost and RA would have to be considered (Morse, 2010). Social impacts involve (but are not limited to) protecting and promoting human health (Bell and Morse, 2012) and this can be achieved by using compost and RA in vegetative SuDS because their application will help in the reduction of waste and improve water quality by removing, motor oil, solids, and heavy metals from stormwater runoff (Mahimairaja *et al*., 2008; Farrell and Jones, 2009). However, studies have shown that compost and RA could be sources of contaminants which could affect water quality in vegetative SuDS. Contaminants, such as heavy metals (Dimambro, Lillywhite and Rahn, 2007; Rao, Jha and Misra, 2007; El-Hammadi and Hanchi, 2011), pathogens (Dimambro, Lillywhite and Rahn, 2007; Guan *et al.*, 2008), hydrocarbons (Hartlieb, Marschner and Klein, 2001), herbicides (BurlingtonFreePress.com, 2012), sulphates and chlorides (Debieb *et al*., 2010; Martín-Morales *et al*., 2011) have been identified in compost. However, most of these contaminants are also found in T and virgin aggregates, as shown in this study, due to anthropogenic activities and so in terms of contaminant content, use of recycled materials in vegetative SuDS should not pose serious risks as long as proper assessments, like that carried out in this study, are carried out to determine levels of contamination and suitability for use (Petersen *et al.*, 2003).

The economic impacts of using compost and RA in vegetative SuDS include financial resources and changing consumption patterns (Bell and Morse, 2012), and their application

would provide some cost savings (as discussed above) and would bring about a change among consumers from utilising natural resources to utilising more sustainable materials for construction. However, Petkovic *et al*., (2004) pointed out that countries with abundant reserves of virgin materials and land space and/or small waste volumes are unwilling to utilise recycled materials despite hefty landfill taxes because they can obtain these virgin materials at relatively low costs with enough land space for landfilling waste. But Highbeam Business (2012) suggested that awareness of the fact that the price customers pay for these natural resources actually covers 50% of the producers' transport costs from source to market in the aggregates industry, coupled with the transportation costs of virgin materials to point of use (Howard, 2000), and setting up of functional guidelines for the use of recycled materials in construction (Petkovic *et al*., 2004), would encourage consumers to source for cheaper alternatives in the form of recycled materials.

The main environmental impacts of utilising compost in vegetative SuDS comprises of conservation of natural resources and biodiversity, and protection of the atmosphere (Bell and Morse, 2012). Application of compost and RA in vegetative SuDS will ensure that less T and G is stripped and excavated respectively for construction, thereby conserving these natural resources and associated ecosystems and biodiversity (Mahimairaja *et al*., 2008). Also, compost and RA can be considered renewable resources, and hence sustainable, because municipal and construction waste will always be generated as long as humans exist (Wilson and Davies, 2012). Table 5.1 shows the types and quantities of organic wastes recycled in 2008/09, with data from 2007/08 also shown for comparison.

Table 5.1 Quantity and type of organic wastes recycled in the UK, 2008/09 and 2007/08.

Source: Association for Organics Recycling, 2010

Table 5.1 shows that there was a 14% increase in total input wastes generated for recycling between 2007/08 to 2008/09, and this figure would have been higher if every waste type and quantity generated was accounted for e.g. construction wastes. This information further buttresses the fact that waste/recycled materials are renewable and would continue to be generated due to anthropogenic activities, and are therefore sustainable (Association for Organics Recycling, 2010). However, the application of compost and RA may not necessarily improve noise and air pollution caused by transportation but as discussed above, this can be offset by T conservation and sourcing for recycled materials locally (Highbeam Business, 2012).

Apart from the sustainability of compost in terms of characterisation, compost is also an effective bioreactor, remediating pollutants by biodegradative processes discussed in section 2.17. Relevant literature has shown that high microbial numbers and microbial diversity are two key factors to effective biodegradation of pollutants (Wackett and Hershberger, 2001; Barker and Bryson, 2002; Coupe *et al*., 2006[a]; Bradley and Chapelle, 2010), and therefore the development of microorganisms in compost is discussed in the next section.

5.3 Biofilm development in compost

The second objective of this study was to monitor biofilm development in compost and this was achieved by monitoring microbial activity in compost via CO_2 monitoring, in conditions similar to that found in vegetative SuDS. Findings showed that compost had higher microbial activity and hence higher biodegradation capabilities (Puehmeier *et al*., 2005; Coupe *et al*., 2006[a]; Jefferies *et al*., 2008) compared to T, in various vegetative SuDS conditions such as low moisture and restricted oxygen conditions, and saturated aerobic conditions in the absence and presence of light, as described in section 3.4.1. This result suggested that compost would improve runoff quality when applied to vegetative SuDS.

In low moisture and restricted oxygen conditions, microbial activity decreased in the three samples, significantly in T, mainly because moisture, being one of the essential elements required for microbial growth and metabolism, was exhausted, leading to the demise of most

microbes while other microbes would go into hibernation or form spores, thereby decreasing activity (Griffiths *et al*., 2003; Kukade *et al*., 2011). This result could be also be explained by the fact that the initial high moisture content of GC and high organic matter content of MC (which helps to store moisture) (FAO, 2005) compared to T, helped to maintain microbial activity until moisture was depleted. It is possible that the group of organisms left after the nine-week testing period were mesophilic anaerobes, which could survive in anaerobic conditions without moisture (Insam and de Bertoldi, 2007; Charlesworth *et al*., 2012).Therefore, in adverse conditions such as periods of no/low rainfall or runoff, microbial activities and hence biodegradation of pollutants (Coupe *et al*., 2006[a]) would still be maintained in swales and other vegetative SuDS devices containing GC and MC, though at reduced rates, compared to T, whose microbial activities were significantly low in these same adverse conditions.

In the presence of higher levels of moisture and oxygen, usually common in the upper layer and sub-bases of swales and other vegetative SuDS or during periods of heavy rainfall and runoff, GC and MC would enhance microbial activities and hence biodegradation of pollutants in vegetative SuDS (Qiu *et al*., 2005). In addition, there would be more activity occurring either in the upper layer of swales (layer more accessible to light) than in the sub-layer (layer less accessible to light) or in the daytime than at night probably due to abundance of photosynthetic microbes (Poretsky *et al*., 2009).

Therefore, in terms of microbial activities, GC and MC would be more versatile in coping with varying conditions in vegetative SuDS compared to the conventional T. These results also showed that though microbial diversity is necessary for biodegradation, high microbial numbers is one of the most important criteria required for effective biodegradation (Jefferies *et al*., 2008). This is because, though GC had the lowest microbial diversity as shown in table 4.5, microbial activity in GC was similar to MC (which had the highest diversity of microorganisms) because of its high microbial numbers.

Studies by Napier *et al*. (2008) have shown that biodegradation of pollutants, such as motor oils, occur in vegetative SuDS soils but biodegradation is less effective in devices where soil is submerged e.g. soils in ponds and detention basins. This trend was attributed to the effect of increased moisture content which had an inhibitory effect on oil degradation (Napier *et al*., 2008) because saturated soils are indicative of lower oxygen content thereby creating anaerobic conditions (Jefferies *et al*., 2008; Kukade *et al*., 2011). The consequence of this is that aerobic microbial degradation of pollutants is limited (Malina and Zawierucha, 2007).

Biodegradation is, however, more effective in vegetative SuDS soils that are intermittently dry, such as between rainfall events e.g. soils in swales and detention basins (Napier *et al*., 2008). These devices have to alternate between being wet and dry because if devices are dry for prolonged periods, biodegradation of pollutants is slowed considerably due to absence of moisture required for metabolic activities (Imam and Gordon, 2002). If devices are wet for prolonged periods, anaerobic conditions set in, thereby slowing down the biodegradation processes. Therefore, the intermittent drying of vegetative SuDS devices provides these devices with aerobic conditions which encourage biodegradation of pollutants (Jefferies *et al*., 2008; Bernal, Alburquerque, and Moral, 2009). These studies collectively showed that biodegradation of pollutants in vegetative SuDS soils are more effective in devices that have intermittent dry periods and less effective in devices that are constantly submerged e.g. wetlands and ponds.

Although measurements of the biodegradation of added pollutants was not carried out in this study, findings from literature and compost's microbial activity in this study showed that compost could carry out effective biodegradation in the varying vegetative SuDS conditions studied compared to T, which had the lowest microbial activity in all the tested conditions. This therefore suggests that compost would fare better in pollutant biodegradation in vegetative SuDS (da Silva, Alves and de França, 2012) at varying levels of moisture, compared to topsoil whose biodegradation efficacy was dependent on moisture levels (Jefferies *et al*., 2008).

It must, however, be noted that though compost could carry out effective biodegradation at varying levels of moisture, organic matter content and pollutant type being treated would determine levels of efficacy because studies by Kukade *et al*. (2011) showed that compost with higher organic matter content (which was MC in this study) recorded lower levels of biodegradation of pollutants, which is contrary to known facts from literature, i.e higher organic matter content is synonymous with higher microbial activity and hence biodegradation (FAO, 2005). This occurrence was explained by Kukade *et al*. (2011) as being due to the preferential utilisation of organic carbon from compost by microorganisms, instead of carbon from the pollutants. The consequence of this occurrence is that biodegradation of pollutants is slowed down while organic carbon from organic matter is being preferentially utilized, and therefore the higher the organic matter, the slower biodegradation will be. This trend suggests that accessibility of microorganisms to organic carbon is crucial to the biodegradation of pollutants because microorganisms find some pollutants, such as pesticides and polychlorinated biphenyls (PCBs), difficult to breakdown easily either due to pollutant toxicity (Fogg *et al*., 2003; Coppola, Pilar-Castillo and Vischetti, 2011) or complexity in pollutant structure (Michel Jr., Quensen and Reddy, 2001).

Therefore, the use of compost would be better at biodegrading run off pollutants compared to T, thereby improving sustainability. But compost derived from garden waste (GC) may be more effective at pollutant biodegradation due to its lower organic matter content, compared to compost derived from a combination of kitchen and garden waste (MC) which has a higher organic matter content. Apart from microbial biodegradation of pollutants, another method of runoff treatment in vegetative SuDS is the use of vegetation as discussed in the next section.

5.4 Grass development in vegetative SuDS

The third objective of this study was to assess grass development in test profiles consisting of compost and recycled aggregates under simulated swale conditions, and this was achieved by monitoring grass growth through biomass measurements in plant pots containing compost and recycled aggregates. Findings showed that GC closely followed by MCT consistently produced the highest grass biomass over time compared to T, and L produced the highest

grass biomass yield, irrespective of the growth media type, compared to G. The results of grass biomass yield in the test profiles were consistent with the physical properties of the growth media samples as described in section 4.2. The presence of organic matter as well as nutrients and microorganisms in GC improved and increased plant growth (Amlinger *et al.*, 2003; Fichtner *et al*., 2004), however, its organic matter content, microbial populations and diversity was much lower than MC, though higher than T. It was therefore expected that MC would be the best growth media to support grass biomass development but this was not the case as GC encouraged grass biomass development significantly compared to the other media. This is because GC's physical properties were closest to ideal while MC was highly porous with large particle sizes making it susceptible to leaching of nutrients. T's physical properties were less than ideal as its organic matter content was very low and its bulk density was high making it susceptible to water-logging and anaerobic conditions which could all be detrimental to microbial activity required for plant growth.

The success of GC in sustaining vegetative growth for five months without the addition of any nutrients can be attributed not only to its organic matter content and microbial populations/diversity but also to its initial nutrient content which was probably released slowly, thereby maintaining vegetative growth for prolonged periods (Alexander, 2004). The main nutrients in compost that help improve plant growth are phosphorus (P), nitrogen (N) and potassium (K) (Walker and Bernal, 2008). However, field experiments by DEFRA (2011[b]) have shown that the concentration of nutrients in GC is usually less than MC as seen in table 5.2.

Table 5.2: Typical total nutrient contents (fresh weight basis). (DEFRA, 2011[b]).

Nevertheless due to the leaching properties of MC of coarser grades (>25mm), as used in this study, nutrient would be more concentrated in GC than MC, hence explaining the higher grass biomass yield in GC. Studies by Rivenshield and Bassuk (2007) showed that the addition of 33% compost to sandy loam soil decreased bulk density by 20% in non-compacted soil and 17% in compacted soil, while the addition of the same volume of compost to clay loam soils actually increased bulk density in compacted soil by 11%, and this increase in bulk density did not decrease until compost volume was increased to 50%. This increase in bulk density was attributed to sand in the compost binding with clay particles in the soil thereby increasing the bulk density. This study by Rivenshield and Bassuk (2007) explains the ability of MCT to produce high grass yields, as it is attributed to the presence of T in MCT which reduced the porosity of MC, increased its bulk density and reduced the leaching of nutrients, thereby making the nutrients available for grass growth, as table 5.2 has shown that MC typically contains more nutrients than GC.

The ability of profiles containing L to consistently produce high biomass yield was explained by the fact that the calcium and magnesium carbonate content of L increases soil pH thereby providing more conducive conditions for grass growth, especially in acidic soils (Nord and Mortensen, 2010; College of Agricultural Sciences, 2013). The species of grasses used in this study (listed in section 3.4.2) are grasses that thrive in soils with neutral pH (Jackson, 2000), and considering the fact that T had the lowest pH , the presence of L probably increased its pH to neutral levels that fell within the CSUGE specification of 7.0-8.7 (WRAP, 2003), thereby encouraging the production of the high biomass yields in LT as shown in table 4.11a. LGC, LMC and LGCT produced the least biomass yield of all the profiles containing L, probably due to increased pH in the compost samples above the CSUGE specification which in turn decreased grass yield. This is because pH of GC and MC were already within the specified pH range and the presence of L further increased their pH to levels less conducive for grass growth.

These results therefore imply that GC can be applied to vegetative SuDS that require dense vegetative cover; and for devices that require improved soil structure, MCT would be

appropriate because the T in MCT would reduce the porosity and increase the bulk density of MC (VanDerZanden and Cook, 2011). For the aggregates, results suggested that L would be a suitable replacement to G in terms of grass biomass production because it produced the highest biomass in all media types.

The development of grasses in vegetative SuDS is essential to the treatment of storm water runoff because vegetation provides flow attenuation of surface runoff which enables the treatment processes, described in section 2.16, to be carried out (Highways Agency, 2006; Woods-Ballard *et al*., 2007). Apart from treatment of runoff, vegetation provides amenity value and creates green spaces in cities (Wilby and Perry, 2006) which help to combat urban heat island effects and provide positive psychological effects on people by creating serene environments close to nature (European Commission, 2012). For vegetation to be established in vegetative SuDS, growth media in the form of T is required but based on findings from this study and discussions in sections 5.2 and 5.3, compost can be used in place of T, because of its ability to improve sustainability and water quality. In terms of the establishment of consistent dense vegetative cover, which is required for effective runoff treatment (Jurries, 2003; MacDonald and Jefferies, 2003[b]; Ellis, 2012), GC was found to be the most appropriate, followed by MCT (Faucette *et al*., 2005). The application of L, a recycled material, in vegetative SuDS would also be sustainable because it can replace G, a natural resource.

Nevertheless, as successful as compost and RA might be in grass development, the addition of compost to vegetative SuDS can pose a risk of leaching of nutrient into runoff and groundwater thereby compromising water quality (Cabrera, Diaz and Madrid, 1989; Faucette *et al*., 2004; Sherman, 2005). Therefore, a balance has to be attained between plant growth and water quality such that the growth media has enough nutrients to sustain plant growth but not in excess to cause leaching (Sherman, 2005). This balance can be attained by carrying out appropriate assessments and the methods of application, timing and amount of compost and RA added to soil can be adjusted to reduce accumulation of nutrients in soil, thereby reducing their leaching (Gilley and Eghball, 2002).

The prolific growth of grasses in vegetative SuDS devices to which compost and L are applied could pose problems if vegetation is not properly maintained, thereby defeating the purpose for which the devices were established (MacDonald and Jefferies, 2003[b]; Pittner and Allerton, 2009). This is because overgrown grasses can extend retention time of surface runoff leading to stagnancy and anaerobic conditions, which reduces biodegradation of pollutants and ultimately results in flooding. Also, amenity value is lost when the devices become unkempt and unsightly (Pittner and Allerton, 2009). Therefore, regular maintenance of grasses by mowing with light machinery (to prevent compaction of soil), especially in devices such as swales, filter strips and detention basins, is necessary (MacDonald and Jefferies, 2003[b]; Ellis, 2012). Vegetative wastes generated from the maintenance of vegetative SuDS could be used on site as wildlife piles or composted and re-used in other SuDS devices or as mulch on farmlands; or if polluted, probably due to heavy uptake of pollutants by grasses, the vegetative wastes can be disposed of into a licensed landfill so as to prevent phytoavailabilty of pollutants as the plants decay (Hyun *et al*., 1998; Susdrain, 2012).

The use of compost alone as a growth medium in vegetative SuDS has raised concerns of phytotoxicity and inhibition of plant germination and growth due to toxic compounds released by fresh, unstable compost (Chaney and Ryan, 1993; Barker and Bryson, 2002; Sæbø and Ferrini, 2006), and therefore Watson (2003) suggested that compost should not be used alone as a pure growing media except where analysis was carried out on the compost to determine its suitability as a growth medium. Sæbø and Ferrini (2006) recommended mixing compost with T to improve nutrient quality, physical properties and structure of the growth medium. To fulfil this criteria, findings from this study has shown that MCT (1:1 blend of MC and T) could improve grass development in vegetative SuDS almost as well as GC alone, thereby providing the required soil property and structure. Therefore, for vegetative SuDS devices that require some structure such as swales and filter strips, MCT can be applied; and for devices where soil structure is immaterial such as in green roofs, ponds and wetlands, GC alone can be applied subject to necessary assessments. This section has discussed the advantages of applying compost and RA in vegetative SuDS for grass development, the next section looks at how well these recycled materials can remediate pollutants in vegetative SuDS.

5.5 Remediation of pollutants in vegetative SuDS

The fourth objective of this study was to determine the efficacy of compost and RA in remediating pollutants in simulated swale conditions and this was achieved by carrying out heavy metal and motor oil analyses of test samples and leachates derived from test profiles dosed with heavy metals and motor oil. Findings from leachate analysis showed that concentrations of all heavy metals used in this study were below WHO (2011) potable water standards and toxicity levels for freshwater organisms, with ≤2% of heavy metal concentrations being leached. About 98% of heavy metals concentrations were retained within components of the test profiles in the following order: growth media>grass roots>grass shoots. Oil concentrations were below limits of detection in all leachate samples whilst Napier *et al*. (2008) have shown that motor oil retained within the test profiles would degrade to at least 81% of the added motor oil concentrations.

The leaching of metals into solution can be attributed to several factors such as decrease in pH, interactions between heavy metals (Yobouet *et al*., 2010; Urasa and Mwebi, 2011), soil moisture (Han, Banin and Triplett, 2001), and soil organic matter content (Dahrazma and Mulligan, 2006). However, any of the above mentioned factors could not have been the case because then leaching would have occurred in all the different media types and not just predominantly in GC and T as shown in table 4.17. The susceptibility of GC and T to leaching can in fact be attributed to their lower carbonate content compared to MC as discussed section 3.3.5. The lower carbonate contents in GC and T implies that there was less binding sites for heavy metals hence their bioavailability and possible risk to groundwater. This was supported by the fact that though Fe, Cu and Zn have high affinities for carbonates (Yu *et al.*, 2001; Yobouet *et al*., 2010; Sundaray *et al*., 2011; Urasa and Mwebi, 2011; Sdiri *et al.*, 2012), they were still leached into solution by GC and T (table 4.17) further reflecting the limited amount of available carbonate binding sites.

The high retention of heavy metals within growth media was expected as studies have shown that soils that are rich in organic matter derived from mature compost have high affinities for heavy metals due to the presence of humic acid, because this acid plays a significant role in the sorption of heavy metals (Clemente *et al*., 2003; Song and Greenway, 2004; Davis and

McCuen, 2005; Kocasoy and Güvener, 2009). Also, the precipitation of heavy metals as metal carbonates and hydroxides renders the metals unavailable (Gibert *et al*., 2005). The significant role of aggregates in heavy metal retention within the test profiles was confirmed by Fach and Geiger (2005) who showed that permeable pavements containing bricks have a heavy metal removal efficiency of 99.2% compared with other aggregates.

Oil concentrations were below limits of detection in all test profile leachate and this result was corroborated by Chapman and Horner (2010) who showed that motor oil could be effectively removed from runoff with 92 - 96% efficiency by bio-retention systems. Davis and McCuen (2005) and Napier *et al*. (2008) also showed that soils, such as sand, clay, silt and soils representative of those found in SuDS such as swales and ponds, were <99% efficient in mitigating oil pollution. The fate of motor oil in soil include processes such as evaporation of volatile components present in motor oil, oxidation in the presence of light (photooxidation and photolysis) (Lin and Tjeerdema, 2008), adsorption and biodegradation (Kingston, 2002). However, studies by Napier *et al*. (2008) showed that sorption was the main mode of oil removal in soil and USEPA (2011) stated that natural organic materials, which in this study included topsoil and compost, mitigate oil pollution by adsorption processes. The results obtained showed that both clean and used motor oil were adsorbed by components of the test profiles including aggregates to a lesser extent. The implications of these results show that in terms of the mitigation of oil pollution in vegetative SuDS, any of the test profiles would suffice as confirmed by Napier *et al*. (2008) thereby improving water quality in vegetative SuDS. This experiment was carried out without vegetation on the test profiles but in practice there would be vegetation which would retain some of the motor oil, further reducing pollution risks to groundwater.

The interaction of vegetative SuDS components with pollutants is very vital in pollutant remediation and this study has shown that compost and RA were able to perform as well as conventional G and T in binding and degrading vegetative SuDS pollutants, thereby potentially improving the sustainability of vegetative SuDS by protecting the water environment and conserving natural resources (Napier *et al*., 2008; Charlesworth *et al*., 2012).

Previous studies have shown that compost is effective in the remediation of polluted soils ranging from soils contaminated with petroleum hydrocarbons and VOCs to soils contaminated with herbicides and heavy metals, thereby reducing their bioavailability in soils, groundwater and water bodies (USEPA, 1997; USCC, 2008). Therefore, apart from the treatment of heavy metals and motor oil by recycled materials in swales, as described in this study, compost and RA would be able to remediate other pollutants in swales and other vegetative SuDS devices, mostly by processes described in section 2.16. The applications of compost and RA in vegetative SuDS have been described in section 2.18, but their application has been limited to compost socks and blankets and as substrates in green roofs. Therefore, the success of the application of compost in simulated swales, as described in this study, provides opportunities for compost and RA to be applied to other vegetative SuDS devices such as wetlands, soakaways, detention and infiltration basins and filter strips, all described in section 2.6.

Findings from this study also showed that apart from compost, grasses also remediated pollutants by taking them up, a process known as phytoremediation (Janecka and Fajalkowski, 2007). This is a vital treatment process for surface runoff in vegetative SuDS as shown by the pollutant concentrations observed in grass roots and shoots (Wilson *et al.*, 2004; Woods-Ballard *et al.*, 2007). Care must therefore be taken in disposing of the grasses so that their decay does not re-introduce the pollutants they contain into the environment. Their disposal, after maintenance operations, must be to designated licensed landfills if heavily contaminated.

However, the efficiency of using recycled and/or conventional materials in remediating pollutants in vegetative SuDS can be compromised if these devices become overloaded with contaminants. Napier *et al*. (2008) observed that this occurrence was dependent on the location of the devices because higher concentrations of pollutants were detected in vegetative SuDS devices close to roadsides and sources of runoff, and therefore the use of stormwater management trains (described in section 2.7) were suggested, which would help to prevent overloading by distributing pollutants more evenly thereby, enabling effective treatment. It must, however, be noted that just like in T, not all pollutants are easily degraded in composted

soil and therefore other specific measures need to be applied in treating recalcitrant pollutants in vegetative SuDS. For example, recalcitrant organochlorines, including dioxins and found in pesticides, sometimes found in runoff (Voldner and Li, 1995; Tanabe, 2002; Barker and Bryson, 2002) are effectively degradable mostly in anaerobic conditions (Coker, 2006; Baczynski, Pleissner and Grotenhuis, 2010) which may not be attainable in most vegetative SuDS devices. Therefore, specific treatment may have to include the inoculation of facultative anaerobic organisms into vegetative SuDS devices, which would then degrade such recalcitrant compounds in restricted oxygen conditions which could be attained in vegetative SuDS, especially in regions farther from the top and closer to the base of the device (Barragán-Huerta *et al*., 2007; Farhan *et al*., 2012).

Apart from contaminant overload in vegetative SuDS devices, threat to water quality could arise if the recycled materials to be used were originally contaminated (Hsu and Lo, 2001; Barker and Bryson, 2002). Therefore, feedstock for recycled materials must be assessed and recycled materials themselves analysed for contaminants before use (Barker and Bryson, 2002; Faucette *et al*., 2004). In extreme cases, where recycled materials in vegetative SuDS become overwhelmed with pollutants and disposal is required, a licensed landfill has to be used which are designed to contain pollutants (Susdrain, 2012), and the organic matter in compost will help to bind and stabilise pollutants thereby minimising their leaching (Chefetz, *et al*., 1998).

5.6 Remediation of oil pollutants by recycled aggregates

The fifth objective of this study was to investigate the effect of recycled aggregates on motor oil pollution remediation and this was achieved by carrying out oil absorption studies on oil-dosed RA and their leachates. Findings showed that RA performed better than G in motor oil pollution remediation and hence would improve water quality.

The higher absorption of used and clean oil by old and new bricks is explained by the fact that bricks are known to contain fine capillaries/pores which makes them porous enough to absorb fluids such as oil and water (Khalaf and DeVenny, 2002; Khalaf and DeVenny, 2005;

Chandigarh, 2006; WRAP, 2007[a]; Arsenovic, Lalic and Radojevic, 2010). Unlike gravel which have little or no pores to retain fluids, fluid retention would be by adsorption as reported by Pratt *et al*. (2002). Pratt *et al*. (2002) reported that oil attenuation by gravel-based permeable pavements on a smaller scale (such as the slow seepage of oil from a vehicle over a period of time) is initially achieved by filtration and adsorption and then biodegradation. The implication of these findings is that the RA would perform better in the mitigation of oil pollution compared to G because of their ability to retain oil better, thereby reducing oil pollution over time and improving water quality in vegetative SuDS devices (Pratt *et al*., 2002; Arsenovic, Lalic and Radojevic, 2010).

Some vegetative SuDS devices, e.g. swales and infiltration basins, incorporate gravel drain beds for retaining and treating pollutants in runoff and as an underlay for stability e.g. filter strips (American Rivers, 2004; USEPA, 2012[b]). Therefore, in terms of treating runoff, RA particularly bricks, would fare better as RA drain beds because the pore sizes in bricks makes them porous enough to absorb/retain not only oil but water (Khalaf and DeVenny, 2005; Arsenovic, Lalic and Radojevic, 2010), compared to gravel in G drain beds which have little or no pores for oil/water retention (Pratt *et al*., 2002). High absorption of water by RA has limited its use in the construction industry because it reduces the mechanical/structural performance of construction mixes containing RA (Paranavithana and Mohajerani, 2006; Cement Concrete and Aggregates Australia 2008; Pérez, Pasandín and Gallego, 2012). Therefore, the use of RA in vegetative SuDS as RA drain bed would provide an alternative use for RA because its absorption properties are required in vegetative SuDS for pollutant treatment.

The use of RA in vegetative SuDS will improve its sustainability because its use will help to conserve natural resources and reduce the amount of waste going into landfill. In addition to the economic benefits that can be derived from the use of RA, as discussed in section 5.2, RA would be a suitable alternative to the use of virgin aggregates in vegetative SuDS due to the Aggregate Levy introduced in April 2002. The Aggregate Levy is a levy imposed on primary aggregates extractions such as sand, crushed rock and gravel extraction, for aggregates use in

the UK. This levy contributes to the increased production costs of virgin aggregates thereby making it more expensive compared to RA (Planning4Minerals, 2006). However, just like with compost, RA must be assessed before use in vegetative SuDS to avoid contamination of water.

Based on the findings from this study on compost and RA, the following model structure is recommended for applying these recycled materials in vegetative SuDS devices, compared to the conventional structure described in figure 2.16

Figure 5.1: A conventional swale cross-section, illustrating the vegetative layer made up of vegetation and topsoil, a gravel drain bed for water storage and an under drain pipe system.

Adapted from: Department of Public Utilities, City of Columbus, Ohio (2013)

Figure 5.2 A recommended swale cross-section illustrating the vegetative layer comprising of a green compost layer, mixed compost and topsoil layer, a crushed old brick and recycled limestone drain bed for storage and an under drain pipe system.

Adapted from: Department of Public Utilities, City of Columbus, Ohio (2013)

According to Woods-Ballard *et al*., 2007, the first 100-150mm of conventional swale depth is the seedbed/vegetative layer required for vegetative growth, root development and infiltration in vegetative SuDS devices, such as a swale, while the remaining swale depth carries out the rest of the treatment processes described in section 2.16 (see figure 5.1). Based on the findings of this study, the first 200mm of swale depth can be replaced by a layer of GC (to further encourage grass growth, infiltration and treatment based on GCs qualities, as described in sections 4.2.1 to 4.2.7) followed by a thicker layer of 1:1 blend of MC and T (to carry out remediation of pollutant s that might have leached through the upper GC layer) as shown in figure 5.2. The gravel drain bed can be replaced with a 70:30 blend of crushed old bricks and limestone aggregates, which will encourage water and pollutant retention and vegetative growth respectively, as discussed in sections 5.6 and 5.4 respectively.

5.7 Critique and Recommendations for Further Studies

This research has shown that compost and RA can perform as well as T and G in vegetative SuDS thereby improving water quality and the overall sustainability of vegetative SuDS devices. Compost and RA were chosen as alternatives to T and G because of their availability, certain physical and chemical similarities to the conventional materials, their lower costs when compared to the conventional materials and ultimately their sustainability. However, there are several other recycled/waste materials which could potentially replace and perform as well, and even better than G and T in vegetative SuDS. It is therefore recommended that, apart from compost and RA which was used in this study, other recycled/waste materials can be tested for use in vegetative SuDS devices thereby improving their sustainability and saving landfill space. Table 5.3 identifies some waste and recycled materials that can be tested for use in vegetative SuDS, subject to relevant assessments.

Recycled/waste materials	Properties	Recommended use in vegetative SuDS devices
Olive waste derived from olive oil processing, which poses a disposal problem (Laufenberg, Rosato and Kunz, 2004).	Improves soil fertility and aeration, increases stability of aggregates, improves water retention in soil, increases bio-availability of micro-elements for vegetative growth, abundant and available especially in Mediterranean regions, contains toxic phenolic compounds (Niaounakis and Halvadakis, 2006; Fernández-Bolaños *et al*., 2006; Sturzenberger, 2007) which can detoxified at low costs prior to use (Mandi *et al*., 2009).	Mixed with topsoil or green compost and used as growth media in swales, filter strips, green roofs
Tree bark	Improves the drainage of clayey topsoil thereby aiding infiltration, improves water holding capacity of sandy topsoil thereby aiding runoff attenuation and treatment, relatively inert, non-toxic (Solt, 1997), widely available (Böhm *et al*., 1998), and accumulates environmental pollutants (Pacheco *et al*., 2002; Suzuki, 2006) Qiu and Hites, 2008).	Mixed with topsoil and used as growth media in detention basins, swales, filter strips, soakaways
Coconut husk (coir)	High water retention capacity useful for runoff treatment and vegetative growth, not easily degraded, widely available (Abad *et al*., 2002; Wellenstein and Wellenstein, 2004; Salaverria, 2012), absorber of some runoff pollutants (Manju, Raji and Anirudhan, 1998; Sumathi, Mahimairaja and Naidu, 2005; Olayinka, Alo and Adu, 2007).	Used as blankets (Be-healthy-with-coconuts, 2012) (like the compost blankets) and mixed with topsoil as growth media in swales, filter strips, ponds and wetlands
Cocoa shell waste	High moisture capacity (National Cocoa Shell, 2003), supports vegetative growth (Adeoye, Sridhar and Ipinmoroti, 2001), capacity to accumulate runoff pollutants (Wantanaphong, Mooney, Bailey, 2005; Odoemelam, Iroh and Igwe, 2011).	Mixed with topsoil or green compost and used as growth media in swales, filter strips, green roofs

Recycled/waste materials	Properties	Recommended use in vegetative SuDS devices
Coffee wastes	Capacity to accumulate pollutants (Utomo and Hunter, 2006; Fiol, Escudero and Villaescusa, 2008), contains nutrients that supports plant growth (Pandey *et al*., 2000; Morikawa and Saigusa, 2008; Adi and Noor, 2009).	Mixed with topsoil or green compost and used as growth media in swales, filter strips, green roofs
Blast furnace slag, a by-product from iron and steel manufacturing (Cement Concrete and Aggregates Australia, 2008)	Removal of pollutants and treatment of waste water (Dimitrova, 1996; Grüneberg and Kern, 2001; Oguz, 2004; Korkusuz, Beklioğlu and Demirer, 2005).	Used in place of gravel in swales, wetlands and detention basins
Expended clay, a by-product from firing natural clay at high temperatures (Cement Concrete and Aggregates Australia (2008)	Removal of pollutants and treatment of waste water (Johansson, 1997; Zhu *et al*., 2003; Malakootian, Nouri and Hossaini, 2009), sustains vegetative growth (Wark and Wark, 2003).	Used in place of gravel in swales, wetlands and detention basins; used alone or mixed with topsoil as growth media in green roofs

Table 5.3: Recycled/waste materials, their properties and recommended use in vegetative SuDS.

The vessels used in this research included 11.6cm by 11.8cm by 28.7cm Perspex rigs for CO_2 monitoring, and 13.3cm diameter pots for vegetative growth, as described in figure 3.1 and 3.3, and these vessels were used to ensure replicability of results obtained. These vessels had small surface areas and studies were carried out under simulated vegetative SuDS conditions, as described in section 3.4.2. The smaller surface area of the pots meant that liquids, such as heavy metals and water, had to be carefully added to the centre of the pots so as to prevent the liquids from running down the sides of the vessels, which could comprise the results obtained.

The use of larger vessels with larger surface areas would probably have given better representations of real-life vegetative SuDS scenarios. To further make this research 'real', it is also recommended that field trials be carried out, especially using the model described in figure 5.2, so as to ascertain the practicality of this research in real-life vegetative SuDS devices under real-life conditions. Studies could be carried out on devices that are either retrofitted or are part of a new development with the necessary modifications made e.g. the addition of compost and RA to existing devices or incorporating compost and RA into devices that are about to be constructed and necessary monitoring carried out.

In the course of this research, bacterial and fungal populations in compost and T were enumerated and oil degrading species were identified. Microbial identification was carried out by physical observation of colonies, preliminary tests and biochemical tests (as described in section 3.3.8). These processes were time consuming and laborious due to robust microbial populations present in compost and T and as a result, only a limited number of microorganisms could be identified. It is therefore recommended that the BBL™ Crystal™ identification kits be used for future studies. These kits employ miniaturised all-at-once identification methods described in section 3.3.8, and provide standardised quality control, require little storage space, give rapid results and are easy to use (Holmes *et al*., 1994; Moll *et al*., 1996; Becton Dickinson and Company, 2001; Lo-Ten-Foe, Ververs and Buiting, 2007). These kits are of various types for different groups of organisms and can be used for the identification of aerobic and anaerobic bacteria, as well as gram negative and gram positive bacteria (Becton Dickinson and Company, 2001). The oil degraders tested for in this research were aerobic, gram negative and gram positive bacteria and therefore the aerobic gram negative and positive BBL™ Crystal™ identification kit would be recommended for use to save time.

Findings from this study showed that motor oil pollutants were retained within the test profiles throughout the test period because leachate analysis showed that motor oil concentrations in leachate were below limits of detection. However, components of the test profiles, i.e. growth media and aggregates, were not analysed for their motor oil content at the end of the

experiment in this study. This would have given an indication of the concentrations of oil degraded within the profiles relative to the initial concentrations of oil added. Napier *et al*. (2008) reported that 71-81% of added motor oil pollutants were degraded and therefore knowledge of the concentrations of motor oil degraded in this study would have given an indication of which of the test profile degraded oil best, thereby impacting water quality positively. Therefore, it is recommended that for future oil degradation studies, apart from oil leachate analysis, the test profiles should be analysed for residual motor oil concentrations at the end of the experiment so as to determine the concentrations of motor oil degraded. This analysis can be carried out by measuring the total petroleum hydrocarbons (TPH) present in the different components of the test profiles using a gas chromatograph/flame ionisation detector (GC/FID), preceded by ultrasonic enhanced solvent (hexane/acetone) extraction, as reported by Napier *et al*. (2008). Ultrasonic extraction would extract hydrocarbons from the pulverised test components, the gas chromatograph would separate out the different hydrocarbon fractions and the FID would detect and identify the various hydrocarbons present in the growth media and aggregates (USEPA, 2007[b]). The results of this analysis would give the concentration of motor oil retained within the profiles at the end of the experiment and the amount of oil degraded can be determined by mass balance calculations i.e.:

Total concentration of oil added = Total concentration of oil leached + concentration of oil retained within profiles + concentration of oil degraded (Napier *et al*., 2008)

5.8 Conclusion

This study has shown that overall, compost and recycled aggregates were able to perform as well as G and T in vegetative SuDS in terms of characterisation, biofilm development, vegetative development, and remediation of pollutants, thereby fulfilling the aims and objectives of this study. Sourcing and characterisation of compost and RA was shown to be less expensive, less time consuming (except for RA), and more sustainable in terms of conserving natural resources and improving water quality. To improve water quality and

reduce pollutant load in stormwater runoff, biodegradation, which is directly associated with microbial activity, is essential. Compost was shown to possess higher microbial activities and hence higher biodegradation in varying vegetative SuDS conditions compared to T. It was therefore deduced that compared to T, compost would be better able to biodegrade organic pollutants in vegetative SuDS in varying conditions, especially in devices that are intermittently dry, as alternating wetness and dryness would enhance effective biodegradation.

Vegetation development, which is an important factor in the efficacy of vegetative SuDS as stormwater attenuators and pollutant remediators, was greater in profiles containing compost compared to T, further enhancing the treatment of stormwater runoff in vegetative SuDS by phytoremediation. Vegetative wastes from these devices can be utilised as wildlife piles, composted and reused in other SuDS devices, as mulch on farmlands or as a source of income to commercial composters. In the remediation of pollutants in vegetative SuDS, this study showed that compost and RA performed as well as G and T with >98% of pollutants being retained within components of the device. Pollutants were retained mostly within the growth media and RA, and least by grass shoots and G, indicating that most pollutants are treated within the growth media of vegetative SuDs devices.

However, before compost and RA can be applied to vegetative SuDS, they must meet specified guidelines e.g. PAS 100, so as to avoid contamination of groundwater and water bodies because the pollutant content of some recycled materials may be high enough to affect water quality adversely. Also, the efficacy of compost in treating stormwater runoff may depend on moisture content, organic matter content and the pollutant types being treated, all of which can be determined by carrying out analyses on compost samples as well as the runoff itself. For grass development, lack of proper maintenance of vegetation could compromise their efficacy in treatment of runoff and therefore proper maintenance is essential. Proper disposal of contaminated vegetative parts must be carried out in licensed landfills so as to prevent the release of the pollutants they contain back into the environment as the plants decay. The use of stormwater management trains have been recommended to distribute pollutants in vegetative SuDS devices so as to prevent them from being overwhelmed and

therefore ineffective. Also, not all pollutants can be treated by compost in vegetative SuDS and treatment of recalcitrant pollutants would have to be specific.

The use of compost and RA would improve water quality and the overall sustainability of vegetative SuDS, however, further studies would be needed to further verify these results in real-life scenarios which can be accomplished by carrying out large-scale experiments and field trials which incorporate these recycled materials. Also, the incorporation of other waste/recycled materials will further increase the sustainability of vegetative SuDS devices and provide alternative uses of these materials thereby saving landfill space.

Chapter Six: Conclusion and Evidence of Originality and Innovations of Thesis

6.1 Conclusion

At the start of this thesis, the issue of flooding, as a result of increased impermeable surfaces and climate change scenarios in urbanised areas, and the attendant effect on water quality were raised. As a result, the need to re-evaluate conventional drainage systems responses to increased flooding in more sustainable ways, were highlighted. Through extensive literature review, Sustainable Drainage Systems (SuDS) were identified as worthy replacements for conventional drainage systems, not only as flood control, but also as control of stormwater pollution and maintenance of ground and surface water quality.

However, the sustainability of vegetative SuDS components was questioned because their constituent materials are derived from natural resources i.e. topsoil and gravel, whose use have significant social, economic and environmental impacts, the three objectives of sustainable development. Replacing or supplementing topsoil and gravel in vegetative SuDS devices with waste/recycled materials can help improve sustainability based on these three objectives, in addition to fulfilling the EU Waste Framework Directive and the Landfill Directive. Compost and recycled aggregates (RA) were employed as alternatives to the original materials in this research because of their similarities in terms of their physical properties, cheaper costs and sustainability. The potential of compost and RA to perform at least as well as the original materials thereby, improving the sustainability of vegetative SuDS, was the main thrust of this research, and formed the basis for the experimental design. Conclusions, based the research's aims and objectives, are presented in this chapter, as well as a summary of recommendations and evidence of originality and innovation.

6.1.1 Characterisation of compost and recycled aggregates

The first objective was to characterise the properties of compost and RA which would help to determine if these materials could perform at least as well as topsoil and virgin aggregate, in vegetative SuDS. Objectives were achieved by comparing derived baseline data with existing requirements and guidelines and the following conclusions were made:

- Compared to compost, characterisation of topsoil was found to be more costly, time consuming and unsustainable, based on the three objectives of sustainable development; while characterisation of RA was found to be more time consuming compared to virgin aggregates.
- Compost and recycled aggregates were found to have some beneficial baseline properties compared to the virgin materials, which could be useful in remediating pollutants in vegetative SuDS.

6.1.2 Biofilm development in compost

The second objective was to monitor biofilm development in compost as this is necessary for pollutant degradation. This was achieved by monitoring microbial activity in compost in conditions similar to those found in vegetative SuDS and the ensuing conclusions were made:

- Microbial activity would decrease over time during periods of no rainfall or runoff in vegetative SuDS due to the shortage of moisture for microbial activities, with a decline in microbial activity being more pronounced in topsoil compared to compost.
- During periods of rainfall, and in the presence of runoff, and in the upper parts of the vegetative layer and sub-bases of vegetative SuDS devices, increased microbial activity would occur over time due to the availability of moisture and oxygen for microbial activities, with activity being more prolific in the presence of light than in the absence of light. Increase in microbial activity was more pronounced in compost compared to topsoil.
- It was therefore concluded that compost would fare better than topsoil in biodegradation of most pollutants in vegetative SuDS at varying levels of moisture, though efficacy and speed of biodegradation would depend on organic matter content, which is required for microbial metabolism, and pollutant type because not all pollutants are easily degraded in composted soil due to pollutant toxicity and complexity.

6.1.3 Grass development in vegetative SuDS

The third objective of this research was to assess grass development in test profiles consisting of compost and RA under simulated swale conditions, as dense vegetation is required for runoff attenuation and phytoremediation of pollutants. This was achieved by monitoring grass growth through biomass measurements in plant pots containing compost and RA and the following conclusions were made:

- Compost and RA, particularly green compost (GC), a 1:1 blend of mixed compost and topsoil (MCT), and recycled limestone aggregates, produced the highest grass biomass yield over time, compared to the original materials and can therefore be applied to vegetative SuDS.
- To avoid the risk of phytotoxicity, inhibition of plant growth and subsidence associated with the use of GC alone, the use of MCT could improve not only grass development in vegetative SuDS but prevent phytotoxicity, and provide the soil structure necessary for stability.
- In applying compost and RA to vegetative SuDS, regular maintenance of vegetation would be necessary due to the prolific grass growth observed in profiles to which they were applied. This is because a lack of maintenance could pose risks of runoff stagnancy leading to flooding and compromised water quality, due to reduction in biodegradation of pollutants caused by anaerobic conditions.

6.1.4 Remediation of pollutants in vegetative SuDS

The fourth objective of this research was to determine the efficacy of compost and RA in remediating pollutants in simulated swale conditions. This was achieved by carrying out heavy metal and motor oil analyses of test samples and leachates derived from test profiles and the following deductions were made:

- Profiles containing compost remediated heavy metals as well as profiles with topsoil in vegetative SuDS devices, thereby improving water quality. Leaching of heavy metals by compost, though low (≤2%), was encouraged by GC and similar in performance to topsoil, thereby posing some risk to groundwater.

- Profiles containing compost also performed as well as profiles with topsoil, with vegetative SuDS components taking up ≥98% of heavy metals present in runoff in the following order: growth media>grass roots>grass shoots.
- Compost profiles mitigated oil pollution as well as conventional materials (i.e. G and T).
- It was therefore concluded that apart from the treatment of pollutants by recycled materials in compost socks and blankets and as substrates in green roofs, compost and RA would be able to remediate pollutants in other vegetative SuDS devices such as swales, thereby reducing pollutant availability in soils, groundwater and water bodies.

6.1.5 Remediation of oil pollutants by recycled aggregates

The fifth objective of this study was to investigate the effect of RA on motor oil pollution remediation. This was achieved by carrying out oil absorption studies on oil-dosed RA and their leachates.

- RA performed better than virgin aggregates in absorbing motor oil due to larger pore sizes which made them porous enough to absorb/retain not only motor oil but water.
- High absorption of water by RA has limited its use in the construction industry because it reduces the mechanical/structural performance of construction mixes. Therefore, its use in vegetative SuDS would provide an alternative use because its absorption properties may assist vegetative SuDS in pollutant treatment

6.1.6 Recommendations

- Apart from compost and RA, other recycled/waste materials can be tested to determine their potential in replacing topsoil and virgin aggregates in vegetative SuDS thereby further improving their sustainability. Alternatives include olive waste, tree bark, coconut husk (coir), cocoa shell waste, coffee wastes, blast furnace slag, and expended clay.
- Due to the extended time spent in identifying oil-degrading organisms in this study, it is recommended that the BBL™ Crystal™ identification kits be used for microbial identification in future studies as they give rapid results and are easy to use.

- The use of vessels with larger surface areas is recommended as this would probably give better representations of real-life scenarios.
- Alternatively, it is recommended that field trials be carried out so as to ascertain the practicality of this research in real-life vegetative SuDS devices under real-life conditions. Studies could be carried out in devices that are either retrofitted or are part of a new development with the necessary modifications made.
- It is recommended that for future oil studies, components of the test profiles should be analysed for residual motor oil concentrations at the end of the experiment, so as to determine which of the test profile degraded oil best, thereby impacting water quality positively.
- Recommendations for a model vegetative SuDS device based on the findings of this study include the use of compost and RA in a soil matrix consisting of GC and a 1:1 blend of MC and topsoil to further encourage infiltration and treatment, and a 70:30 blend of crushed old bricks and limestone aggregates, which will encourage water and pollutant retention, and vegetative growth respectively.
- It is recommended that a full scale trial be carried out on swales using the model described above.

6.2 Evidence of Originality and Innovations of Thesis

- This thesis, through a review of relevant literature, established that SuDS components, i.e. gravel and topsoil, can be as unsustainable as components of conventional drainage systems in terms of their social, economic and environmental impacts, and that recycled materials could perform just as well as conventional materials especially, in vegetative SuDS devices whilst improving their sustainability.
- This thesis further established that compost and recycled aggregates can be used ‘as they are’ in vegetative SuDS such as swales, subject to relevant assessments, as literature has shown that the use of compost and RA in vegetative SuDS has been limited to compost blankets and socks and substrates for green roofs.

- Other waste materials that can be used in place of topsoil and gravel in vegetative SuDS, thereby improving its sustainability, were recommended.
- An ideal model for the treatment of pollutants in vegetative SuDS which comprised of a vegetative layer of green compost (200mm) followed by a thicker layer of 1:1 blend of mixed compost and topsoil (700mm), and 70:30 blend of crushed old bricks and limestone aggregates as the drain bed layer (300mm) was developed.

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Supervisors: Charlesworth S., Coupe J. and Bennett J.

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Appendix 1: Results of moisture content determination on compost and topsoil samples

	Before drying			After drying @ 80^{o}C					
Samples	Weight of foil trays (g) K	Weight of moist samples + foil trays (g) WWK	Weight of moist sample (g) WWK-K	Weight of dry sample + foil trays (g) DWK	Weight of dry sample (g) DWK-K	Moisture content (g) M	% moisture content w/wt	**% Average moisture content w/wt**	**Standard error**
GC1	6.98	96.13	89.15	57.09	50.11	39.04	43.79	**41.51**	**0.74**
GC2	7.00	93.93	86.93	58.77	51.77	35.16	40.45		
GC3	6.95	80.64	73.69	49.26	42.31	31.38	42.58		
GC4	6.92	87.57	80.65	55.50	48.58	32.07	39.76		
GC5	6.95	88.22	81.27	63.90	47.99	33.28	40.95		
MC1	6.93	33.28	26.35	25.22	18.29	8.06	30.59	**27.82**	**0.81**
MC2	6.96	35.82	28.86	28.01	21.05	7.81	27.06		
MC3	6.91	37.62	30.71	29.65	22.74	7.97	25.95		
MC4	6.90	39.55	32.65	30.22	23.32	9.33	28.58		
MC5	6.92	40.66	33.74	31.57	24.65	9.09	26.94		
T1	6.98	199.37	192.39	170.36	163.38	29.01	15.08	**13.92**	**0.34**
T2	6.97	216.24	209.27	187.69	180.72	28.55	13.64		
T3	6.95	187.17	180.22	162.52	155.57	24.65	13.68		
T4	6.90	209.80	202.90	183.31	176.41	26.49	13.06		
T5	6.92	210.01	203.09	181.25	174.33	28.76	14.16		

GC= green compost
MC= mixed compost
T=topsoil
w/wt= wet weight

One-way ANOVA

Moisture content

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	1902.282	2	951.141	435.900	**.000**
Within Groups	26.184	12	2.182		
Total	1928.466	14			

Post Hoc test

Multiple Comparisons

Dependent Variable: Moisture content

LSD

(I) Media	(J) Media	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	MC	13.68032*	.93424	.000	11.6448	15.7159
	T	27.58435*	.93424	.000	25.5488	29.6199
MC	GC	-13.68032*	.93424	.000	-15.7159	-11.6448
	T	13.90402*	.93424	.000	11.8685	15.9396
T	GC	-27.58435*	.93424	.000	-29.6199	-25.5488
	MC	-13.90402*	.93424	.000	-15.9396	-11.8685

*. The mean difference is significant at the 0.05 level.

Appendix 2: Results of determination of water holding capacity on compost and topsoil samples

Samples	Days																			
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
MC1	100	79	75	85	70	59	54	41	27	23	22	27	25	19	18	26	20	21	8	4
MC2	90	60	45	43	35	40	36	37	36	27	30	25	29	24	25	25	22	24	12	5
MC3	90	58	43	50	41	48	32	30	25	27	23	20	28	23	24	35	20	24	5	4
Average	**93**	**66**	**54**	**59**	**49**	**49**	**41**	**36**	**29**	**26**	**25**	**24**	**27**	**22**	**22**	**29**	**21**	**23**	**8**	**4**
Standard error	3.33	6.69	10.35	12.99	10.81	5.51	6.77	3.21	3.38	1.33	2.52	2.08	1.20	1.53	2.19	3.18	0.67	1.00	2.03	0.33
GC1	100	78	65	63	85	67	65	35	26	28	30	26	27	28	25	24	24	23	19	11
GC2	100	100	95	85	77	52	40	35	25	21	25	24	26	25	24	24	18	24	11	8
GC3	100	100	88	65	61	64	61	45	36	37	34	34	35	36	35	35	32	33	22	12
Average	**100**	**93**	**83**	**71**	**74**	**61**	**55**	**38**	**29**	**29**	**30**	**28**	**29**	**30**	**28**	**28**	**25**	**27**	**17**	**10**
Standard error	0	7.33	9.06	7.02	7.06	4.58	7.75	3.33	3.51	4.63	2.60	3.06	2.85	3.28	3.51	3.67	4.06	3.18	3.28	1.20
T1	100	100	100	100	100	90	74	60	45	31	28	36	30	31	29	30	20	26	15	6
T2	100	100	100	98	75	61	50	43	41	32	28	33	40	36	34	30	18	25	17	9
Average	**100**	**100**	**100**	**99**	**88**	**76**	**62**	**52**	**43**	**32**	**28**	**35**	**35**	**34**	**32**	**30**	**19**	**26**	**16**	**8**
Standard error	0.00	0.00	0.00	0.82	10.21	11.84	9.80	6.94	1.63	0.41	0.00	1.22	4.08	2.04	2.04	0.00	0.82	0.41	0.82	1.22

GC= green compost
MC= mixed compost
T= topsoil

One-way ANOVA

Water holding capacity

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	2596.033	2	1298.017	1.731	**.186**
Within Groups	42754.150	57	750.073		
Total	45350.183	59			

Post Hoc test

Multiple Comparisons

Dependent Variable: Water holding capacity

LSD

(I) Samples	(J) Samples	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	MC	9.250	8.661	.290	-8.09	26.59
	T	-6.800	8.661	.436	-24.14	10.54
MC	GC	-9.250	8.661	.290	-26.59	8.09
	T	-16.050	8.661	.069	-33.39	1.29
T	GC	6.800	8.661	.436	-10.54	24.14
	MC	16.050	8.661	.069	-1.29	33.39

*. The mean difference is significant at the 0.05 level.

Appendix 3: Results of organic matter content determination on compost and topsoil samples

	Before ignition			After igniting @ 500^{o}C					
Samples	Weight of crucible (g) K	Weight of sample [2g] + crucible (g) AK	Weight of dry samples (g) AK-K	Weight of crucible + sample (g) BK	Weight of ignited sample (g) BK-K	LOI (g) (organic matter content)	% LOI (% organic Matter content)	**% Average organic matter content**	**Standard error**
GC1	30.10	32.10	2.00	31.33	1.23	0.77	38.50		
GC2	31.21	33.20	1.99	32.15	0.94	1.05	52.76		
GC3	32.78	34.77	1.99	34.06	1.28	0.71	35.68		
GC4	32.57	34.57	2.00	33.55	0.98	1.02	51.00		
GC5	24.59	26.57	1.98	25.91	1.32	0.66	33.33	**42.26**	**4.02**
MC1	26.28	28.27	1.99	26.89	0.61	1.38	69.35		
MC2	26.88	28.87	1.99	27.57	0.69	1.30	65.33		
MC3	34.47	36.47	2.00	35.11	0.64	1.36	68.00		
MC4	30.10	32.07	1.97	30.79	0.69	1.28	64.97		
MC5	29.40	31.39	1.99	30.09	0.69	1.30	65.33	**66.59**	**0.88**
T1	32.53	34.51	1.98	34.31	1.78	0.20	10.10		
T2	34.90	36.89	1.99	36.72	1.82	0.17	8.54		
T3	30.16	32.15	1.99	31.94	1.78	0.21	10.55		
T4	31.23	33.22	1.99	33.01	1.78	0.21	10.55		
T5	35.01	36.99	1.98	36.79	1.78	0.20	10.10	**9.97**	**0.37**

GC= green compost
MC= mixed compost
T= topsoil
LOI= loss on ignition

One-way ANOVA

Organic matter

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	8109.580	2	4054.790	146.874	**.000**
Within Groups	331.287	12	27.607		
Total	8440.868	14			

Post Hoc test

Multiple Comparisons

Dependent Variable: Organic matter

LSD

(I) Media	(J) Media	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	MC	-24.26200*	3.32309	.000	-31.5024	-17.0216
	T	32.49400*	3.32309	.000	25.2536	39.7344
MC	GC	24.26200*	3.32309	.000	17.0216	31.5024
	T	56.75600*	3.32309	.000	49.5156	63.9964
T	GC	-32.49400*	3.32309	.000	-39.7344	-25.2536
	MC	-56.75600*	3.32309	.000	-63.9964	-49.5156

*. The mean difference is significant at the 0.05 level.

Appendix 4: Results of bulk density determination on compost and topsoil samples

Samples	Mass (g)	Volume (cm^3)	Bulk density M/V (gcm^{-3})	**Average bulk density (gcm^{-3}**	**standard error**
GC1	93.58	200	0.47		
GC2	89.02	200	0.45		
GC3	93.77	200	0.47		
GC4	94.95	200	0.47		
GC5	90.05	200	0.45	**0.46**	**0.0058**
MC1	42.58	200	0.21		
MC2	43.71	200	0.22		
MC3	44.62	200	0.22		
MC4	45.76	200	0.23		
MC5	43.65	200	0.22	**0.22**	**0.0027**
T1	202.68	200	1.01		
T2	203.91	200	1.02		
T3	206.12	200	1.03		
T4	201.79	200	1.01		
T5	200.87	200	1.00	**1.02**	**0.0046**

One-way ANOVA

Bulk density

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	1.656	2	.828	8280.867	**.000**
Within Groups	.001	12	.000		
Total	1.657	14			

Post Hoc test

Multiple Comparisons

Dependent Variable: Bulk density

LSD

(I) Media	(J) Media	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	MC	.24200*	.00632	.000	.2282	.2558
	T	-.55200*	.00632	.000	-.5658	-.5382
MC	GC	-.24200*	.00632	.000	-.2558	-.2282
	T	-.79400*	.00632	.000	-.8078	-.7802
T	GC	.55200*	.00632	.000	.5382	.5658
	MC	.79400*	.00632	.000	.7802	.8078

*. The mean difference is significant at the 0.05 level.

Appendix 5: Results of carbonate content determination on compost and topsoil samples

Ignition @ 950°C									
Samples	Weight of crucible (g) K	Weight of sample + crucible (g) CK	Weight of dry samples at 950°C (g) CK-K	Weight of dry sample at 500°C (g) BK-K	Carbonate content (g) BK-(CK-K)	Weight of dry sample at 50°C (2g) AK	% Carbonate content (CK-K) *100 (BK-K)	**% Carbonate content average**	**standard error**
GC1	30.10	31.29	1.19	1.23	0.04	2.00	2.72		
GC2	31.21	32.11	0.9	0.94	0.04	1.99	2.73		
GC3	32.78	34.03	1.25	1.28	0.03	1.99	2.05		
GC4	32.57	33.52	0.95	0.98	0.03	2.00	2.04		
GC5	24.59	25.87	1.28	1.32	0.04	1.98	2.75	**2.46**	**0.17**
MC1	26.28	26.81	0.53	0.61	0.08	1.99	5.47		
MC2	26.88	27.49	0.61	0.69	0.08	1.99	5.47		
MC3	34.47	35.03	0.56	0.64	0.08	2.00	5.44		
MC4	30.10	30.71	0.61	0.69	0.08	1.97	5.52		
MC5	29.40	30.01	0.61	0.69	0.08	1.99	5.47	**5.47**	**0.01**
T1	32.53	34.26	1.73	1.78	0.05	1.98	3.43		
T2	34.90	36.66	1.76	1.82	0.06	1.99	4.10		
T3	30.16	31.86	1.7	1.78	0.08	1.99	5.47		
T4	31.23	32.94	1.71	1.78	0.07	1.99	4.56		
T5	35.01	36.73	1.72	1.78	0.06	1.98	3.78	**4.27**	**0.35**

GC= green compost
MC= mixed compost
T= topsoil

One-way ANOVA

Carbonate content

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	22.276	2	11.138	45.412	**.000**
Within Groups	2.943	12	.245		
Total	25.220	14			

Post Hoc test

Multiple Comparisons

Dependent Variable: Carbonate content

LSD

(I) Media	(J) Media	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	MC	-2.96800*	.31322	.000	-3.6505	-2.2855
	T	-1.76000*	.31322	.000	-2.4425	-1.0775
MC	GC	2.96800*	.31322	.000	2.2855	3.6505
	T	1.20800*	.31322	.002	.5255	1.8905
T	GC	1.76000*	.31322	.000	1.0775	2.4425
	MC	-1.20800*	.31322	.002	-1.8905	-.5255

*. The mean difference is significant at the 0.05 level.

Appendix 6: Results of Hydrogen ion level (pH) of compost and topsoil samples

Samples	pH	**Average pH**	**standard error**
GC1	7.3		
GC2	7.5		
GC3	7.2		
GC4	7.5		
GC5	7.2	**7.34**	**0.07**
MC1	7.6		
MC2	7.7		
MC3	7.6		
MC4	7.8		
MC5	7.1	**7.56**	**0.12**
T1	6.8		
T2	6.9		
T3	6.6		
T4	7.0		
T5	6.9	**6.84**	**0.07**

One-way ANOVA

pH

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	1.361	2	.681	17.160	**.000**
Within Groups	.476	12	.040		
Total	1.837	14			

Multiple Comparisons

Dependent Variable: pH

LSD

(I) Media	(J) Media	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	MC	-.22000	.12596	.106	-.4944	.0544
	T	.50000*	.12596	.002	.2256	.7744
MC	GC	.22000	.12596	.106	-.0544	.4944
	T	.72000*	.12596	.000	.4456	.9944
T	GC	-.50000*	.12596	.002	-.7744	-.2256
	MC	-.72000*	.12596	.000	-.9944	-.4456

*. The mean difference is significant at the 0.05 level.

Appendix 7: Results of heavy metal analysis on topsoil, compost and aggregates

Data results of background total heavy metal concentrations in compost, topsoil and aggregates

Heavy metal concentration in samples (mgL^{-1})									
Standard and samples	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
Std 0	0	0	0	0	0	0	0	0	0
Std 1	50	0.01	0.1	2	100	5	0.2	15	2
Std 2	100	0.02	0.4	4	150	7	0.4	20	4
Std 3	150	0.03	0.7	6	200	12	0.6	40	7
Std 4	180	0.04	1.2	8	250	15	0.8	70	10
GC1	98.62	0.02	1.98	1.19	215.40	6.32	0.39	2.48	4.59
GC2	92.23	0.02	1.79	1.43	202.90	6.56	0.39	2.87	5.38
GC3	103.10	0.02	2.45	1.30	229.80	6.46	0.56	2.48	4.99
MC1	48.39	0.02	0.34	0.66	93.06	4.31	0.17	1.19	2.36
MC2	42.93	0.03	0.35	0.66	88.76	4.34	0.17	1.23	2.68
MC3	42.37	0.03	0.39	0.68	106.10	4.74	0.18	1.33	2.62
T1	140.20	0.01	0.54	3.89	242.30	7.63	0.44	56.18	3.39
T2	191.90	0.01	0.77	3.71	278.90	7.98	0.45	69.67	3.41
T3	167.90	0.01	0.46	4.07	280.70	8.08	0.45	50.85	3.57
NB1	183.00	0.00	0.71	0.15	102.30	3.72	0.18	1.30	0.17
NB2	211.00	0.00	0.80	0.11	181.80	4.53	0.18	0.47	0.14
NB3	221.80	0.00	0.73	0.11	186.00	4.83	0.16	0.53	0.14
OB1	276.00	0.00	0.78	0.19	240.20	10.00	0.32	0.15	0.59
OB2	354.90	0.00	1.15	0.26	287.70	11.17	0.42	0.15	0.84
OB3	353.40	0.00	1.28	0.27	285.00	12.53	0.46	0.19	0.86
G1	10.50	0.00	0.05	0.06	95.82	1.55	0.05	0.08	0.13
G2	7.97	0.00	0.03	0.05	101.80	0.89	0.03	0.06	0.11
G3	6.15	0.00	0.03	0.04	46.65	0.41	0.02	0.06	0.07
L1	68.51	0.01	0.36	0.16	250.80	7.68	0.29	0.13	0.21
L2	39.96	0.01	0.28	0.07	242.40	7.13	0.16	0.16	0.16
L3	67.84	0.01	0.39	0.16	247.90	7.39	0.32	0.11	0.25
Acid blank 1	0.00	0.00	0.00	0.01	0.16	0.01	0.00	0.00	0.04
Acid blank 2	0.00	0.00	0.00	0.01	0.08	0.01	0.00	0.00	0.05
Acid blank 3	0.00	0.00	0.01	0.00	0.09	0.01	0.00	0.00	0.01
Spike 1	4.34	4.71	4.69	4.94	6.39	5.53	5.06	5.24	5.12
Spike 2	4.47	4.81	4.83	5.09	6.62	5.69	5.25	5.43	5.38
Spike 3	4.28	4.72	4.65	4.99	6.38	5.54	5.05	5.19	5.06

Std = standard calibration

OB= old bricks

NB= new bricks

G= gravel

L= limestone

Blank corrected mean heavy metal concentration ($mgkg^{-1}$)									
Samples	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
GC	4899.2	0.90	103.5	65.4	10796.0	322.0	22.5	130.5	247.6
MC	2228.2	1.30	17.9	33.1	4793.0	222.8	8.5	62.5	125.9
T	8333.3	0.30	29.3	194.1	13359.3	394.7	22.2	2945.0	171.3
NB	10263.3	0.02	37.3	5.7	7829.3	217.7	8.60	38.3	5.72
OB	16405.0	0.00	1.6	11.8	13542.7	561.7	19.9	8.0	36.6
G	410.28	0.02	1.6	2.2	4065.5	47.1	1.6	3.3	3.5
L	2938.5	0.27	17.0	6.3	12345.9	369.6	12.5	6.5	8.6

Data results of background available heavy metal concentrations in compost and topsoil leachates for five weeks

Heavy metal concentration in leachate in Week 1 – (mgL^{-1})									
Standard and samples	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
Std 0	0	0	0	0	0	0	0	0	0
Std 1	100	0.01	1.0	2	100	5	0.2	20	2
Std 2	200	0.02	1.5	4	180	10	0.4	40	4
Std 3	300	0.03	2.0	6	250	15	0.6	60	7
Std 4	400	0.04	2.5	8	300	20	0.8	80	10
GC1	0.000	0.002	0.010	0.153	1.788	0.141	0.018	0.077	0.163
GC2	0.000	0.003	0.014	0.233	3.935	0.227	0.029	0.062	0.287
GC3	0.000	0.002	0.006	0.116	1.225	0.090	0.015	0.090	0.119
MC1	0.000	0.003	0.030	0.302	8.328	0.789	0.067	0.085	0.509
MC2	0.000	0.002	0.016	0.109	3.921	0.437	0.027	0.050	0.334
MC3	0.000	0.002	0.022	0.169	4.658	0.469	0.036	0.052	0.261
T1	0.000	0.001	0.005	0.153	0.341	0.061	0.027	0.021	0.233
T2	0.000	0.001	0.002	0.069	0.066	0.003	0.011	0.008	0.045
Mean leachate heavy metal concentration (mgL^{-1}) - Week 1									
Samples	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
GC	0.000	0.002	0.007	0.163	2.202	0.147	0.020	0.076	0.156
MC	0.000	0.002	0.020	0.189	5.522	0.559	0.043	0.062	0.334
T	0.000	0.001	0.001	0.106	0.090	0.026	0.018	0.015	0.105

Heavy metal concentration in leachate in Week 2 (mgL^{-1})									
Standard and samples	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
Std 0	0	0	0	0	0	0	0	0	0
Std 1	100	0.01	1.0	2	100	5	0.2	20	2
Std 2	200	0.02	1.5	4	180	10	0.4	40	4
Std 3	300	0.03	2.0	6	250	15	0.6	60	7
Std 4	400	0.04	2.5	8	300	20	0.8	80	10
GC1	0.000	0.001	0.004	0.095	1.341	0.082	0.015	0.016	0.108
GC2	0.000	0.002	0.009	0.155	2.587	0.163	0.021	0.032	0.171
GC3	0.000	0.001	0.003	0.061	0.766	0.077	0.010	0.007	0.069
MC1	0.000	0.003	0.036	0.297	9.078	0.907	0.083	0.085	0.571
MC2	0.000	0.002	0.015	0.124	4.369	0.409	0.032	0.055	0.335
MC3	0.000	0.001	0.012	0.082	2.904	0.300	0.018	0.033	0.150
T1	0.000	0.000	0.002	0.078	0.048	0.005	0.014	0.002	0.017
T2	0.000	0.000	0.001	0.057	0.073	0.002	0.008	0.006	0.057
Mean leachate heavy metal concentration (mgL^{-1}) - Week 2									
Samples	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
GC	0.000	0.001	0.003	0.099	1.451	0.102	0.015	0.018	0.082
MC	0.000	0.002	0.018	0.163	5.337	0.533	0.044	0.058	0.318
T	0.000	0.000	0.000	0.063	0.000	0.000	0.010	0.004	0.003

Heavy metal concentration in leachate in Week 3 (mgL^{-1})									
Standard and samples	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
Std 0	0	0	0	0	0	0	0	0	0
Std 1	100	0.01	1.0	2	100	5	0.2	20	2
Std 2	200	0.02	1.5	4	180	10	0.4	40	4
Std 3	300	0.03	2.0	6	250	15	0.6	60	7
Std 4	400	0.04	2.5	8	300	20	0.8	80	10
GC1	0.000	0.002	0.009	0.160	1.735	0.081	0.037	0.012	0.134
GC2	0.000	0.001	0.006	0.078	1.316	0.143	0.016	0.007	0.093
GC3	0.000	0.001	0.004	0.054	1.072	0.214	0.017	0.004	0.065
MC1	0.000	0.002	0.025	0.217	8.010	0.575	0.056	0.075	0.342
MC2	0.000	0.001	0.014	0.109	4.457	0.349	0.027	0.061	0.216
MC3	0.000	0.001	0.010	0.062	2.773	0.235	0.015	0.029	0.117
T1	0.000	0.000	0.001	0.063	0.027	0.116	0.019	0.000	0.006
T2	0.000	0.000	0.001	0.062	0.020	0.010	0.009	0.000	0.041
Mean leachate heavy metal concentration (mgL^{-1}) - Week 3									
Samples	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
GC	0.000	0.001	0.004	0.093	1.261	0.140	0.023	0.008	0.064
MC	0.000	0.001	0.014	0.125	4.966	0.381	0.032	0.055	0.191
T	0.000	0.000	0.000	0.058	0.000	0.057	0.013	0.000	0.000

Heavy metal concentration in leachate in Week 4 (mgL^{-1})									
Standard and samples	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
Std 0	0	0	0	0	0	0	0	0	0
Std 1	100	0.01	1.0	2	100	5	0.2	20	2
Std 2	200	0.02	1.5	4	180	10	0.4	40	4
Std 3	300	0.03	2.0	6	250	15	0.6	60	7
Std 4	400	0.04	2.5	8	300	20	0.8	80	10
GC1	0.000	0.001	0.002	0.048	0.807	0.057	0.010	0.005	0.071
GC2	0.000	0.001	0.007	0.105	1.794	0.182	0.020	0.013	0.115
GC3	0.000	0.001	0.005	0.069	1.247	0.197	0.022	0.006	0.060
MC1	0.000	0.002	0.029	0.248	9.468	0.590	0.061	0.082	0.395
MC2	0.000	0.001	0.013	0.113	4.393	0.298	0.025	0.051	0.208
MC3	0.000	0.001	0.010	0.060	2.514	0.193	0.014	0.036	0.114
T1	0.000	0.000	0.002	0.066	0.048	0.106	0.017	0.001	0.010
T2	0.000	0.000	0.001	0.052	0.028	0.005	0.008	0.000	0.017
Mean leachate heavy metal concentration (mgL^{-1}) - Week 4									
Samples	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
GC	0.000	0.001	0.002	0.069	1.169	0.140	0.017	0.008	0.048
MC	0.000	0.001	0.015	0.136	5.345	0.355	0.033	0.056	0.205
T	0.000	0.000	0.000	0.054	0.000	0.050	0.012	0.001	0.000

Heavy metal concentration in leachate in Week 5 (mgL^{-1})									
Standard and samples	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
Std 0	0	0	0	0	0	0	0	0	0
Std 1	100	0.01	1.0	2	100	5	0.2	20	2
Std 2	200	0.02	1.5	4	180	10	0.4	40	4
Std 3	300	0.03	2.0	6	250	15	0.6	60	7
Std 4	400	0.04	2.5	8	300	20	0.8	80	10
GC1	0.000	0.001	0.004	0.069	1.000	0.060	0.015	0.014	0.074
GC2	0.000	0.001	0.005	0.077	1.500	0.150	0.014	0.008	0.090
GC3	0.000	0.001	0.003	0.033	0.877	0.152	0.011	0.001	0.037
MC1	0.000	0.004	0.044	0.403	13.540	0.623	0.094	0.112	0.592
MC2	0.000	0.001	0.007	0.059	3.080	0.254	0.013	0.042	0.142
MC3	0.000	0.001	0.008	0.057	2.511	0.178	0.013	0.032	0.110
T1	0.000	0.000	0.002	0.070	0.030	0.063	0.017	0.000	0.008
T2	0.000	0.001	0.006	0.246	0.050	0.006	0.036	0.000	0.046
Mean leachate heavy metal concentration (mgL^{-1}) - Week 5									
Samples	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
GC	0.000	0.001	0.001	0.055	1.012	0.115	0.013	0.008	0.033
MC	0.000	0.002	0.017	0.168	6.263	0.346	0.039	0.062	0.248
T	0.000	0.001	0.001	0.153	0.000	0.029	0.026	0.000	0.000

One-way ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
Heavy metals in GC background leachate	Between Groups	8.498	8	1.062	43.042	**.000**
	Within Groups	.888	36	.025		
	Total	9.386	44			
Heavy metals in MC background leachate	Between Groups	128.816	8	16.102	591.912	**.000**
	Within Groups	.979	36	.027		
	Total	129.795	44			
Heavy metals in T background leachate	Between Groups	.030	8	.004	5.513	**.000**
	Within Groups	.025	36	.001		
	Total	.055	44			

Post Hoc test

Multiple Comparisons

LSD

Dependent Variable	(I) heavy metals	(J) heavy metals	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
						Lower Bound	Upper Bound
GC heavy metals in bk leachate	Al	Cd	-.001200	.099356	.990	-.20270	.20030
		Cr	-.003400	.099356	.973	-.20490	.19810
		Cu	-.095800	.099356	.341	-.29730	.10570
		Fe	-1.419000*	.099356	.000	-1.62050	-1.21750
		Mn	-.128800	.099356	.203	-.33030	.07270
		Ni	-.017600	.099356	.860	-.21910	.18390
		Pb	-.023600	.099356	.814	-.22510	.17790
		Zn	-.076600	.099356	.446	-.27810	.12490
	Cd	Al	.001200	.099356	.990	-.20030	.20270
		Cr	-.002200	.099356	.982	-.20370	.19930
		Cu	-.094600	.099356	.347	-.29610	.10690
		Fe	-1.417800*	.099356	.000	-1.61930	-1.21630
		Mn	-.127600	.099356	.207	-.32910	.07390
		Ni	-.016400	.099356	.870	-.21790	.18510
		Pb	-.022400	.099356	.823	-.22390	.17910
		Zn	-.075400	.099356	.453	-.27690	.12610

	Cr	Al	.003400	.099356	.973	-.19810	.20490
		Cd	.002200	.099356	.982	-.19930	.20370
		Cu	-.092400	.099356	.359	-.29390	.10910
		Fe	-1.415600*	.099356	.000	-1.61710	-1.21410
		Mn	-.125400	.099356	.215	-.32690	.07610
		Ni	-.014200	.099356	.887	-.21570	.18730
		Pb	-.020200	.099356	.840	-.22170	.18130
		Zn	-.073200	.099356	.466	-.27470	.12830
	Cu	Al	.095800	.099356	.341	-.10570	.29730
		Cd	.094600	.099356	.347	-.10690	.29610
		Cr	.092400	.099356	.359	-.10910	.29390
		Fe	-1.323200*	.099356	.000	-1.52470	-1.12170
		Mn	-.033000	.099356	.742	-.23450	.16850
		Ni	.078200	.099356	.436	-.12330	.27970
		Pb	.072200	.099356	.472	-.12930	.27370
		Zn	.019200	.099356	.848	-.18230	.22070
	Fe	Al	1.419000*	.099356	.000	1.21750	1.62050
		Cd	1.417800*	.099356	.000	1.21630	1.61930
		Cr	1.415600*	.099356	.000	1.21410	1.61710
		Cu	1.323200*	.099356	.000	1.12170	1.52470
		Mn	1.290200*	.099356	.000	1.08870	1.49170
		Ni	1.401400*	.099356	.000	1.19990	1.60290
		Pb	1.395400*	.099356	.000	1.19390	1.59690
		Zn	1.342400*	.099356	.000	1.14090	1.54390
	Mn	Al	.128800	.099356	.203	-.07270	.33030
		Cd	.127600	.099356	.207	-.07390	.32910
		Cr	.125400	.099356	.215	-.07610	.32690
		Cu	.033000	.099356	.742	-.16850	.23450
		Fe	-1.290200*	.099356	.000	-1.49170	-1.08870
		Ni	.111200	.099356	.270	-.09030	.31270
		Pb	.105200	.099356	.297	-.09630	.30670
		Zn	.052200	.099356	.603	-.14930	.25370
	Ni	Al	.017600	.099356	.860	-.18390	.21910
		Cd	.016400	.099356	.870	-.18510	.21790
		Cr	.014200	.099356	.887	-.18730	.21570
		Cu	-.078200	.099356	.436	-.27970	.12330
		Fe	-1.401400*	.099356	.000	-1.60290	-1.19990
		Mn	-.111200	.099356	.270	-.31270	.09030
		Pb	-.006000	.099356	.952	-.20750	.19550
		Zn	-.059000	.099356	.556	-.26050	.14250

	Pb	Al	.023600	.099356	.814	-.17790	.22510
		Cd	.022400	.099356	.823	-.17910	.22390
		Cr	.020200	.099356	.840	-.18130	.22170
		Cu	-.072200	.099356	.472	-.27370	.12930
		Fe	-1.395400*	.099356	.000	-1.59690	-1.19390
		Mn	-.105200	.099356	.297	-.30670	.09630
		Ni	.006000	.099356	.952	-.19550	.20750
		Zn	-.053000	.099356	.597	-.25450	.14850
	Zn	Al	.076600	.099356	.446	-.12490	.27810
		Cd	.075400	.099356	.453	-.12610	.27690
		Cr	.073200	.099356	.466	-.12830	.27470
		Cu	-.019200	.099356	.848	-.22070	.18230
		Fe	-1.342400*	.099356	.000	-1.54390	-1.14090
		Mn	-.052200	.099356	.603	-.25370	.14930
		Ni	.059000	.099356	.556	-.14250	.26050
		Pb	.053000	.099356	.597	-.14850	.25450
MC heavy metals in bk leachate	Al	Cd	-.001600	.104314	.988	-.21316	.20996
		Cr	-.016800	.104314	.873	-.22836	.19476
		Cu	-.156200	.104314	.143	-.36776	.05536
		Fe	-5.486600*	.104314	.000	-5.69816	-5.27504
		Mn	-.434800*	.104314	.000	-.64636	-.22324
		Ni	-.038200	.104314	.716	-.24976	.17336
		Pb	-.058600	.104314	.578	-.27016	.15296
		Zn	-.259200*	.104314	.018	-.47076	-.04764
	Cd	Al	.001600	.104314	.988	-.20996	.21316
		Cr	-.015200	.104314	.885	-.22676	.19636
		Cu	-.154600	.104314	.147	-.36616	.05696
		Fe	-5.485000*	.104314	.000	-5.69656	-5.27344
		Mn	-.433200*	.104314	.000	-.64476	-.22164
		Ni	-.036600	.104314	.728	-.24816	.17496
		Pb	-.057000	.104314	.588	-.26856	.15456
		Zn	-.257600*	.104314	.018	-.46916	-.04604
	Cr	Al	.016800	.104314	.873	-.19476	.22836
		Cd	.015200	.104314	.885	-.19636	.22676
		Cu	-.139400	.104314	.190	-.35096	.07216
		Fe	-5.469800*	.104314	.000	-5.68136	-5.25824
		Mn	-.418000*	.104314	.000	-.62956	-.20644
		Ni	-.021400	.104314	.839	-.23296	.19016
		Pb	-.041800	.104314	.691	-.25336	.16976
		Zn	-.242400*	.104314	.026	-.45396	-.03084

	Cu	Al	.156200	.104314	.143	-.05536	.36776
		Cd	.154600	.104314	.147	-.05696	.36616
		Cr	.139400	.104314	.190	-.07216	.35096
		Fe	-5.330400*	.104314	.000	-5.54196	-5.11884
		Mn	-.278600*	.104314	.011	-.49016	-.06704
		Ni	.118000	.104314	.265	-.09356	.32956
		Pb	.097600	.104314	.356	-.11396	.30916
		Zn	-.103000	.104314	.330	-.31456	.10856
	Fe	Al	5.486600*	.104314	.000	5.27504	5.69816
		Cd	5.485000*	.104314	.000	5.27344	5.69656
		Cr	5.469800*	.104314	.000	5.25824	5.68136
		Cu	5.330400*	.104314	.000	5.11884	5.54196
		Mn	5.051800*	.104314	.000	4.84024	5.26336
		Ni	5.448400*	.104314	.000	5.23684	5.65996
		Pb	5.428000*	.104314	.000	5.21644	5.63956
		Zn	5.227400*	.104314	.000	5.01584	5.43896
	Mn	Al	.434800*	.104314	.000	.22324	.64636
		Cd	.433200*	.104314	.000	.22164	.64476
		Cr	.418000*	.104314	.000	.20644	.62956
		Cu	.278600*	.104314	.011	.06704	.49016
		Fe	-5.051800*	.104314	.000	-5.26336	-4.84024
		Ni	.396600*	.104314	.001	.18504	.60816
		Pb	.376200*	.104314	.001	.16464	.58776
		Zn	.175600	.104314	.101	-.03596	.38716
	Ni	Al	.038200	.104314	.716	-.17336	.24976
		Cd	.036600	.104314	.728	-.17496	.24816
		Cr	.021400	.104314	.839	-.19016	.23296
		Cu	-.118000	.104314	.265	-.32956	.09356
		Fe	-5.448400*	.104314	.000	-5.65996	-5.23684
		Mn	-.396600*	.104314	.001	-.60816	-.18504
		Pb	-.020400	.104314	.846	-.23196	.19116
		Zn	-.221000*	.104314	.041	-.43256	-.00944
	Pb	Al	.058600	.104314	.578	-.15296	.27016
		Cd	.057000	.104314	.588	-.15456	.26856
		Cr	.041800	.104314	.691	-.16976	.25336
		Cu	-.097600	.104314	.356	-.30916	.11396
		Fe	-5.428000*	.104314	.000	-5.63956	-5.21644
		Mn	-.376200*	.104314	.001	-.58776	-.16464
		Ni	.020400	.104314	.846	-.19116	.23196
		Zn	-.200600	.104314	.062	-.41216	.01096

	Zn	Al	.259200*	.104314	.018	.04764	.47076
		Cd	.257600*	.104314	.018	.04604	.46916
		Cr	.242400*	.104314	.026	.03084	.45396
		Cu	.103000	.104314	.330	-.10856	.31456
		Fe	-5.227400*	.104314	.000	-5.43896	-5.01584
		Mn	-.175600	.104314	.101	-.38716	.03596
		Ni	.221000*	.104314	.041	.00944	.43256
		Pb	.200600	.104314	.062	-.01096	.41216
T heavy metals in bk leachate	Al	Cd	-.000400	.016583	.981	-.03403	.03323
		Cr	-.000400	.016583	.981	-.03403	.03323
		Cu	-.086800*	.016583	.000	-.12043	-.05317
		Fe	-.018000	.016583	.285	-.05163	.01563
		Mn	-.032400	.016583	.059	-.06603	.00123
		Ni	-.015800	.016583	.347	-.04943	.01783
		Pb	-.004000	.016583	.811	-.03763	.02963
		Zn	-.021600	.016583	.201	-.05523	.01203
	Cd	Al	.000400	.016583	.981	-.03323	.03403
		Cr	.000000	.016583	1.000	-.03363	.03363
		Cu	-.086400*	.016583	.000	-.12003	-.05277
		Fe	-.017600	.016583	.296	-.05123	.01603
		Mn	-.032000	.016583	.062	-.06563	.00163
		Ni	-.015400	.016583	.359	-.04903	.01823
		Pb	-.003600	.016583	.829	-.03723	.03003
		Zn	-.021200	.016583	.209	-.05483	.01243
	Cr	Al	.000400	.016583	.981	-.03323	.03403
		Cd	.000000	.016583	1.000	-.03363	.03363
		Cu	-.086400*	.016583	.000	-.12003	-.05277
		Fe	-.017600	.016583	.296	-.05123	.01603
		Mn	-.032000	.016583	.062	-.06563	.00163
		Ni	-.015400	.016583	.359	-.04903	.01823
		Pb	-.003600	.016583	.829	-.03723	.03003
		Zn	-.021200	.016583	.209	-.05483	.01243
	Cu	Al	.086800*	.016583	.000	.05317	.12043
		Cd	.086400*	.016583	.000	.05277	.12003
		Cr	.086400*	.016583	.000	.05277	.12003
		Fe	.068800*	.016583	.000	.03517	.10243
		Mn	.054400*	.016583	.002	.02077	.08803
		Ni	.071000*	.016583	.000	.03737	.10463
		Pb	.082800*	.016583	.000	.04917	.11643
		Zn	.065200*	.016583	.000	.03157	.09883

	Fe	Al	.018000	.016583	.285	-.01563	.05163
		Cd	.017600	.016583	.296	-.01603	.05123
		Cr	.017600	.016583	.296	-.01603	.05123
		Cu	-.068800*	.016583	.000	-.10243	-.03517
		Mn	-.014400	.016583	.391	-.04803	.01923
		Ni	.002200	.016583	.895	-.03143	.03583
		Pb	.014000	.016583	.404	-.01963	.04763
		Zn	-.003600	.016583	.829	-.03723	.03003
	Mn	Al	.032400	.016583	.059	-.00123	.06603
		Cd	.032000	.016583	.062	-.00163	.06563
		Cr	.032000	.016583	.062	-.00163	.06563
		Cu	-.054400*	.016583	.002	-.08803	-.02077
		Fe	.014400	.016583	.391	-.01923	.04803
		Ni	.016600	.016583	.324	-.01703	.05023
		Pb	.028400	.016583	.095	-.00523	.06203
		Zn	.010800	.016583	.519	-.02283	.04443
	Ni	Al	.015800	.016583	.347	-.01783	.04943
		Cd	.015400	.016583	.359	-.01823	.04903
		Cr	.015400	.016583	.359	-.01823	.04903
		Cu	-.071000*	.016583	.000	-.10463	-.03737
		Fe	-.002200	.016583	.895	-.03583	.03143
		Mn	-.016600	.016583	.324	-.05023	.01703
		Pb	.011800	.016583	.481	-.02183	.04543
		Zn	-.005800	.016583	.729	-.03943	.02783
	Pb	Al	.004000	.016583	.811	-.02963	.03763
		Cd	.003600	.016583	.829	-.03003	.03723
		Cr	.003600	.016583	.829	-.03003	.03723
		Cu	-.082800*	.016583	.000	-.11643	-.04917
		Fe	-.014000	.016583	.404	-.04763	.01963
		Mn	-.028400	.016583	.095	-.06203	.00523
		Ni	-.011800	.016583	.481	-.04543	.02183
		Zn	-.017600	.016583	.296	-.05123	.01603
	Zn	Al	.021600	.016583	.201	-.01203	.05523
		Cd	.021200	.016583	.209	-.01243	.05483
		Cr	.021200	.016583	.209	-.01243	.05483
		Cu	-.065200*	.016583	.000	-.09883	-.03157
		Fe	.003600	.016583	.829	-.03003	.03723
		Mn	-.010800	.016583	.519	-.04443	.02283
		Ni	.005800	.016583	.729	-.02783	.03943
		Pb	.017600	.016583	.296	-.01603	.05123

*. The mean difference is significant at the 0.05 level.

Data results of background available heavy metal concentrations in leachates from aggregates for three weeks

Heavy metal concentration in leachate - Week 1 (mgL^{-1})									
Samples	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
Std 0	0	0	0	0	0	0	0	0	0
Std 1	100	0.01	1.0	2	100	5	0.2	20	2
Std 2	200	0.02	1.5	4	180	10	0.4	40	4
Std 3	300	0.03	2.0	6	250	15	0.6	60	7
Std 4	400	0.04	2.5	8	300	20	0.8	80	10
OB1	0.000	0.000	0.001	0.005	0.002	0.002	0.005	0.000	0.019
OB2	0.000	0.000	0.002	0.004	0.005	0.001	0.004	0.000	0.004
OB3	0.000	0.000	0.001	0.005	0.005	0.001	0.003	0.000	0.003
NB1	0.000	0.000	0.002	0.003	0.001	0.001	0.003	0.000	0.019
NB2	0.000	0.000	0.001	0.004	0.001	0.001	0.008	0.000	0.015
NB3	0.000	0.000	0.000	0.003	0.001	0.000	0.003	0.000	0.008
L1	0.000	0.000	0.000	0.003	0.007	0.001	0.004	0.000	0.021
L2	0.000	0.000	0.001	0.003	0.004	0.003	0.004	0.000	0.113
G1	0.000	0.000	0.001	0.005	0.003	0.001	0.010	0.000	0.022
G2	0.000	0.000	0.001	0.003	0.002	0.001	0.004	0.000	0.031
Mean heavy metal concentration in leachate - Week 1 (mgL^{-1})									
Samples	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
OB	0.000	0.000	0.001	0.005	0.004	0.001	0.004	0.000	0.009
NB	0.000	0.000	0.001	0.003	0.001	0.001	0.005	0.000	0.014
L	0.000	0.000	0.001	0.003	0.006	0.002	0.004	0.000	0.067
G	0.000	0.000	0.001	0.004	0.003	0.001	0.007	0.000	0.027

Heavy metal concentration in leachate - Week 2 (mgL^{-1})									
Samples	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
Std 0	0	0	0	0	0	0	0	0	0
Std 1	100	0.01	1.0	2	100	5	0.2	20	2
Std 2	200	0.02	1.5	4	180	10	0.4	40	4
Std 3	300	0.03	2.0	6	250	15	0.6	60	7
Std 4	400	0.04	2.5	8	300	20	0.8	80	10
OB1	0.000	0.000	0.003	0.009	0.103	0.009	0.005	0.036	0.012
OB2	0.000	0.000	0.002	0.006	0.035	0.004	0.004	0.012	0.012
OB3	0.000	0.000	0.006	0.013	0.026	0.004	0.006	0.007	0.007
NB1	0.000	0.000	0.002	0.006	0.002	0.002	0.003	0.000	0.021
NB2	0.000	0.000	0.001	0.007	0.002	0.003	0.003	0.004	0.026
NB3	0.000	0.000	0.001	0.006	0.001	0.003	0.003	0.000	0.031
L1	0.000	0.000	0.000	0.005	0.002	0.003	0.005	0.000	0.035
L2	0.000	0.000	0.001	0.014	0.007	0.010	0.007	0.000	0.032
G1	0.000	0.000	0.001	0.004	0.002	0.003	0.003	0.000	0.074
G2	0.000	0.000	0.000	0.004	0.001	0.005	0.003	0.000	0.108
Mean heavy metal concentration in leachate - Week 2 (mgL^{-1})									
Samples	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
OB	0.000	0.000	0.004	0.009	0.055	0.006	0.005	0.018	0.010
NB	0.000	0.000	0.001	0.006	0.002	0.003	0.003	0.001	0.026
L	0.000	0.000	0.001	0.010	0.005	0.007	0.006	0.000	0.034
G	0.000	0.000	0.001	0.004	0.002	0.004	0.003	0.000	0.091

Heavy metal concentration in leachate - Week 3 (mgL^{-1})									
Samples	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
Std 0	0	0	0	0	0	0	0	0	0
Std 1	100	0.01	1.0	2	100	5	0.2	20	2
Std 2	200	0.02	1.5	4	180	10	0.4	40	4
Std 3	300	0.03	2.0	6	250	15	0.6	60	7
Std 4	400	0.04	2.5	8	300	20	0.8	80	10
OB1	0.000	0.000	0.001	0.010	0.016	0.007	0.005	0.000	0.030
OB2	0.000	0.000	0.001	0.009	0.010	0.008	0.004	0.000	0.063
OB3	0.000	0.000	0.002	0.008	0.007	0.008	0.003	0.001	0.057
NB1	0.000	0.000	0.001	0.005	0.002	0.011	0.003	0.000	0.083
NB2	0.000	0.000	0.000	0.007	0.002	0.008	0.005	0.000	0.123
NB3	0.000	0.000	0.001	0.005	0.004	0.006	0.003	0.000	0.060
L1	0.000	0.000	0.001	0.006	0.004	0.007	0.003	0.000	0.049
L2	0.000	0.000	0.001	0.006	0.005	0.008	0.005	0.000	0.119
G1	0.000	0.000	0.001	0.004	0.003	0.006	0.003	0.000	0.158
G2	0.000	0.001	0.001	0.006	0.002	0.006	0.010	0.000	0.408

Mean heavy metal concentration in leachate - Week 3 (mgL^{-1})									
Samples	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
OB	0.000	0.000	0.001	0.009	0.011	0.008	0.004	0.000	0.050
NB	0.000	0.000	0.001	0.006	0.003	0.008	0.004	0.000	0.089
L	0.000	0.000	0.001	0.006	0.005	0.008	0.004	0.000	0.084
G	0.000	0.001	0.001	0.005	0.003	0.006	0.007	0.000	0.283

Appendix 8: Results for CO_2 monitoring of Microbial Respiration

Microbial activity in low moisture, restricted oxygen conditions in the presence of light (ppm)									
Samples	Weeks								
	1	2	3	4	5	6	7	8	9
GC1	51000	70500	57000	55500	63000	55500	28500	57000	30000
GC2	34500	57000	42000	30000	42000	42000	33000	27000	12000
GC3	32250	33000	15000	12000	19500	9000	30000	30000	7500
GC Average	**39250**	**53500**	**38000**	**32500**	**41500**	**35500**	**30500**	**38000**	**16500**
standard error	5911	10966	12288	12619	12560	13811	1323	9539	6874
MC1	22500	30000	21000	12000	24000	24000	16500	22500	13500
MC2	45000	67500	30000	24000	27000	30000	18000	34500	18000
MC3	51000	87000	45000	42000	63000	63000	31500	34500	9000
MC average	**39500**	**61500**	**32000**	**26000**	**38000**	**39000**	**22000**	**30500**	**13500**
standard error	8675	16726	7000	8718	12530	12124	4770	4000	2598
T1	10500	18000	6000	3000	10500	9000	4500	4500	3000
T2	10500	18000	13500	7500	10500	9000	7500	10500	1500
T average	**10500**	**18000**	**9750**	**5250**	**10500**	**9000**	**6000**	**7500**	**2250**
standard error	0	0	3750	2250	0	0	1500	3000	750

Anova: Single Factor, microbial activity in low moisture conditions restricted oxygen conditions in the presence of light

SUMMARY

Groups	*Count*	*Sum*	*Average*	*Variance*
GC	9	325250	36138.889	97236111
MC	9	302000	33555.556	1.84E+08

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	30031250	1	30031250	0.213735	**0.65008**	4.493998
Within Groups	2.248E+09	16	140506944			
Total	2.278E+09	17				

Microbial activity in saturated aerobic conditions, in the absence of light (ppm)									
Samples	Weeks								
	1	2	3	4	5	6	7	8	9
GC1	33000	22500	12000	16500	42000	49500	72000	60000	58500
GC2	37500	19500	19500	24000	36000	30000	55500	57000	54000
GC3	10500	12000	9000	12000	22500	34500	40500	51000	52500
GC Average	**27000**	**18000**	**13500**	**17500**	**33500**	**38000**	**56000**	**56000**	**55000**
standard error	8352	3122	3123	3500	5766	5895	9097	2646	1803
MC1	31500	24000	25500	25500	31500	39000	49500	49500	46500
MC2	6000	6000	4500	7500	12000	12000	13500	16500	15000
MC3	25500	39000	30000	33000	36000	40500	45000	45000	43500
MC Average	**21000**	**23000**	**20000**	**22000**	**26500**	**30500**	**36000**	**37000**	**35000**
standard error	7697	9539	7858	7566	7366	9260	11325	10332	10037
T1	4500	4500	3000	6000	7500	7500	9000	9000	9000
T2	3000	3000	1500	4500	4500	6000	10500	10500	9000
T Average	**3750**	**3750**	**2250**	**5250**	**6000**	**6750**	**9750**	**9750**	**9000**
standard error	750	750	750	750	1500	750	750	750	0

Anova: Single Factor, microbial activity in saturated aerobic conditions, in the absence of light

SUMMARY

Groups	*Count*	*Sum*	*Average*	*Variance*
GC	9	314500	34944.44	3.02E+08
MC	9	251000	27888.89	47048611

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	2.24E+08	1	2.24E+08	1.284156	**0.273823**	4.493998
Within Groups	2.79E+09	16	1.74E+08			
Total	3.02E+09	17				

Microbial activity in saturated aerobic conditions, in the presence of light (ppm)									
Samples	Weeks								
	1	2	3	4	5	6	7	8	9
GC1	36000	45000	37500	45000	52500	60000	64500	70500	73500
GC2	34500	34500	40500	43500	51000	57000	61500	63000	61500
GC3	7500	15000	6000	27000	36000	40500	45000	45000	46500
GC average	**26000**	**31500**	**28000**	**38500**	**46500**	**52500**	**57000**	**59500**	**60500**
standard error	9260	8789	11034	5766	5268	6062	6062	7566	7810
MC1	30000	37500	31500	48000	57000	67500	69000	70500	70500
MC2	10500	24000	15000	27000	30000	43500	52500	55500	60000
MC3	30000	43500	34500	49500	54000	61500	66000	67500	69000
MC average	**23500**	**35000**	**27000**	**41500**	**47000**	**57500**	**62500**	**64500**	**66500**
standard error	6500	5766	6062	7263	8544	7211	5075	4583	3279
TP1	4500	4500	6000	6000	9000	10500	13500	15000	15000
TP2	3000	6000	7500	6000	7500	12000	15000	16500	15000
T average	**3750**	**5250**	**6750**	**6000**	**8250**	**11250**	**14250**	**15750**	**15000**
standard error	750	750	750	0	750	750	750	750	0

Anova: Single Factor, microbial activity in saturated aerobic conditions, in the presence of light

SUMMARY

Groups	*Count*	*Sum*	*Average*	*Variance*
GC	9	400000	44444.44	1.91E+08
MC	9	425000	47222.22	2.7E+08

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	34722222	1	34722222	0.150648	**0.703033**	4.493998
Within Groups	3.69E+09	16	2.3E+08			
Total	3.72E+09	17				

Appendix 9: Results of grass biomass yield obtained from profiles combining green compost with sub-base aggregates of crushed old and new bricks, gravel and recycled limestone aggregates

Mean grass biomass yield	Months				
	1	2	3	4	5
OBGC	42.56	43.98	45.21	48.31	49.16
NBGC	40.82	44.07	42.92	49.66	56.19
GGC	44.54	44.38	44.70	47.74	56.86
LGC	40.37	42.60	49.79	55.24	65.39
Average	**42.07**	**43.76**	**45.65**	**50.24**	**56.90**
Standard error					
OBGC	1.09	1.25	1.36	1.71	1.56
NBGC	1.19	1.74	1.71	0.87	1.10
GGC	4.24	1.84	1.66	1.22	1.40
LGC	0.81	0.78	5.89	0.78	3.50

Anova: Single Factor, grass biomass yield for green compost +aggregates for five months

SUMMARY

Groups	*Count*	*Sum*	*Average*	*Variance*
OBGC	5	229.2215	45.8442962	7.930144
NBGC	5	233.6662	46.7332316	38.66993
GGC	5	238.2121	47.6424271	28.47238
LGC	5	253.3896	50.6779128	102.3594

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	66.235814	3	22.0786046	0.497737	**0.689039**	3.238872
Within Groups	709.72741	16	44.3579634			
Total	775.96323	19				

Appendix 10: Results of grass biomass yield obtained from profiles combining mixed compost with sub-base aggregates of crushed old and new bricks, gravel and recycled limestone aggregates

Mean grass biomass yield	Months				
	1	2	3	4	5
OBMC	54.17	49.29	60.87	54.83	42.29
NBMC	48.52	43.34	42.94	51.37	41.94
GMC	45.54	49.20	53.75	60.19	63.49
LMC	45.04	46.59	66.30	62.26	69.80
Average	**48.32**	**47.11**	**55.97**	**57.16**	**54.38**
Standard error					
OBMC	0.64	0.97	10.36	1.08	0.55
NBMC	2.79	1.00	0.78	0.97	2.36
GMC	1.45	2.02	1.97	2.38	4.50
LMC	2.55	1.09	7.06	0.81	5.27

Anova: Single Factor, grass biomass yield for mixed compost + aggregates for five months

SUMMARY

Groups	*Count*	*Sum*	*Average*	*Variance*
OBMC	5	261.4544	52.29088	48.16616
NBMC	5	228.1145	45.6229	16.86139
GMC	5	272.167	54.43341	55.52559
LMC	5	289.9908	57.99816	131.1501

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	406.3813	3	135.4604	2.1527	**0.133614**	3.238872
Within Groups	1006.813	16	62.92581			
Total	1413.194	19				

Appendix 11: Results of grass biomass yield obtained from profiles combining green compost + topsoil with sub-base aggregates of crushed old and new bricks, gravel and recycled limestone aggregates

Mean grass biomass yield	Months				
	1	2	3	4	5
OBGCT	43.37	43.73	44.23	52.06	49.03
NBGCT	40.17	41.32	40.53	51.19	52.58
GGCT	67.62	46.07	42.16	53.35	63.87
LGCT	58.57	45.56	48.16	59.02	68.65
Average	**52.43**	**44.17**	**43.77**	**53.90**	**58.53**
Standard error					
OBGCT	1.31	1.95	1.39	0.97	3.09
NBGCT	1.44	1.39	1.57	1.51	2.19
GGCT	2.46	1.96	1.62	0.98	4.62
LGCT	2.49	1.64	2.32	0.57	1.79

Anova: Single Factor, grass biomass yield for green compost + topsoil + aggregates for five months

SUMMARY

Groups	*Count*	*Sum*	*Average*	*Variance*
OBGCT	5	232.4179	46.48358	14.97183
NBGCT	5	225.7867	45.15734	38.11057
GGCT	5	273.0759	54.61518	121.1002
LGCT	5	279.9536	55.99071	86.59226

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	458.7154	3	152.9051	2.345397	**0.111427**	3.238872
Within Groups	1043.099	16	65.1937			
Total	1501.815	19				

Appendix 12: Results of grass biomass yield obtained from profiles combining mixed compost + topsoil with sub-base aggregates of crushed old and new bricks, gravel and recycled limestone aggregates

Grass biomass yield	Months				
	1	2	3	4	5
OBMCT	43.96	44.31	46.07	45.79	47.97
NBMCT	39.53	37.52	39.97	48.55	50.82
GMCT	54.02	41.32	38.35	51.77	65.02
LMCT	50.01	42.50	57.71	57.29	78.64
Average	**46.88**	**41.41**	**45.52**	**50.85**	**60.61**
Standard error					
OBMCT	2.16	1.96	1.94	4.21	2.70
NBMCT	0.80	0.86	0.97	1.42	2.43
GMCT	3.49	2.19	1.31	1.34	4.34
LMCT	2.03	1.36	6.26	0.78	3.01

Anova: Single Factor, grass biomass yield for mixed compost + topsoil + aggregates for five months

SUMMARY

Groups	*Count*	*Sum*	*Average*	*Variance*
OBMCT	5	228.0943	45.61886	2.554436
NBMCT	5	216.3983	43.27966	35.71402
GMCT	5	250.4821	50.09642	113.9947
LMCT	5	286.1411	57.22823	181.8781

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	565.2393	3	188.4131	2.255491	**0.121232**	3.238872
Within Groups	1336.565	16	83.5353			
Total	1901.804	19				

Appendix 13: Results of grass biomass yield obtained from profiles combining topsoil with sub-base aggregates of crushed old and new bricks, gravel and recycled limestone aggregates

Mean grass biomass yield	Months				
	1	2	3	4	5
OBT	47.48	45.66	49.73	64.25	60.10
NBT	36.55	38.70	43.34	64.05	58.33
GT	62.46	46.30	41.02	60.94	66.31
LT	72.20	43.45	46.29	71.55	79.39
Average	**54.67**	**43.53**	**45.10**	**65.20**	**66.04**
Standard error					
OBT	4.02	1.58	2.85	3.28	2.75
NBT	2.04	0.59	0.49	1.16	0.61
GT	6.24	2.05	0.49	2.44	1.47
LT	0.93	1.76	1.41	4.00	4.13

Anova: Single Factor, grass biomass yield for topsoil + aggregates for five months

SUMMARY

Groups	*Count*	*Sum*	*Average*	*Variance*
OBT	5	267.2314	53.44627	67.79385
NBT	5	240.9678	48.19356	150.8236
GT	5	277.0252	55.40504	122.3332
LT	5	312.8916	62.57831	271.7321

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	531.5056	3	177.1685	1.156674	**0.356841**	3.2388715
Within Groups	2450.731	16	153.1707			
Total	2982.236	19				

Appendix 14: Two-way Anova test for total grass biomass yield for five months

Tests of Between-Subjects Effects

Dependent Variable: Total Grass Biomass

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	78.331[a]	19	4.123	9.738	.000
Intercept	24521.355	1	24521.355	57922.578	.000
Medium	19.282	4	4.821	11.387	**.000**
Aggregate	42.831	3	14.277	33.724	**.000**
Medium * Aggregate	16.218	12	1.352	3.192	**.001**
Error	25.401	60	.423		
Total	24625.086	80			
Corrected Total	103.732	79			

a. R Squared = .755 (Adjusted R Squared = .678)

Post Hoc test for growth media

Multiple Comparisons

LSD: Total Grass Biomass

(I) Medium	(J) Medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	.8763*	.23004	.000	.4161	1.3364
	MC	1.2263*	.23004	.000	.7661	1.6864
	MCT	.5681*	.23004	.016	.1080	1.0283
	T	1.3694*	.23004	.000	.9092	1.8295
GCT	GC	-.8763*	.23004	.000	-1.3364	-.4161
	MC	.3500	.23004	.133	-.1101	.8101
	MCT	-.3081	.23004	.185	-.7683	.1520
	T	.4931*	.23004	.036	.0330	.9533
MC	GC	-1.2263*	.23004	.000	-1.6864	-.7661
	GCT	-.3500	.23004	.133	-.8101	.1101
	MCT	-.6581*	.23004	.006	-1.1183	-.1980
	T	.1431	.23004	.536	-.3170	.6033
MCT	GC	-.5681*	.23004	.016	-1.0283	-.1080
	GCT	.3081	.23004	.185	-.1520	.7683
	MC	.6581*	.23004	.006	.1980	1.1183
	T	.8012*	.23004	.001	.3411	1.2614
T	GC	-1.3694*	.23004	.000	-1.8295	-.9092
	GCT	-.4931*	.23004	.036	-.9533	-.0330
	MC	-.1431	.23004	.536	-.6033	.3170
	MCT	-.8012*	.23004	.001	-1.2614	-.3411

Based on observed means.

The error term is Mean Square (Error) = .423.

*. The mean difference is significant at the 0.05 level.

Post Hoc test for aggregates

Multiple Comparisons

LSD: Total Grass Biomass

(I) Aggregate	(J) Aggregate	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	.8790*	.20575	.000	.4674	1.2906
	NB	-1.0955*	.20575	.000	-1.5071	-.6839
	OB	-.5680*	.20575	.008	-.9796	-.1564
L	G	-.8790*	.20575	.000	-1.2906	-.4674
	NB	-1.9745*	.20575	.000	-2.3861	-1.5629
	OB	-1.4470*	.20575	.000	-1.8586	-1.0354
NB	G	1.0955*	.20575	.000	.6839	1.5071
	L	1.9745*	.20575	.000	1.5629	2.3861
	OB	.5275*	.20575	.013	.1159	.9391
OB	G	.5680*	.20575	.008	.1564	.9796
	L	1.4470*	.20575	.000	1.0354	1.8586
	NB	-.5275*	.20575	.013	-.9391	-.1159

Based on observed means.

The error term is Mean Square (Error) = .423.

*. The mean difference is significant at the 0.05 level.

Appendix 15: Results for Aluminium concentrations in profile leachate over an eight-week spiking period

Standard calibration	Heavy metals concentrations (mgL^{-1})								
	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
0	0	0	0	0	0	0	0	0	0
1	10	10	10	10	10	10	10	10	10
2	20	20	20	20	20	20	20	20	20
3	35	35	35	35	35	35	35	35	35
4	50	50	50	50	50	50	50	50	50

Al concentrations in profile leachates (mgL^{-1})

Samples	Weeks									
	bk	bk	1	2	3	4	5	6	7	8
OBGC1	0.000	0.000	0.036	0.000	0.057	0.000	0.258	0.691	0.000	0.000
OBGC2	0.000	0.000	0.216	0.000	0.000	0.000	0.273	0.283	0.000	0.000
OBGC3	0.000	0.000	0.077	0.000	0.000	0.000	0.127	0.348	0.000	0.000
OGBG4	0.000	0.000	0.798	0.000	0.000	0.000	0.305	0.598	0.000	0.000
OBMC1	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
OBMC2	0.000	0.000	0.110	0.000	0.000	0.000	0.000	0.000	0.000	0.000
OBMC3	0.000	0.000	0.120	0.000	0.000	0.000	0.029	0.000	0.000	0.000
OBMC4	0.000	0.000	0.004	0.000	0.000	0.000	0.012	0.000	0.000	0.000
OBT1	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.055	0.000	0.000
OBT2	0.000	0.000	0.603	0.073	0.000	0.000	0.000	0.325	0.000	0.868
OBT3	0.000	0.000	0.026	0.000	0.000	0.000	0.000	0.295	0.000	0.235
OBT4	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.464	0.000	0.809
OBGCT1	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.080	0.000	0.000
OBGCT2	0.000	0.000	0.020	0.000	0.000	0.000	0.000	0.032	0.000	0.000
OBGCT3	0.000	0.000	1.571	0.000	0.000	0.000	0.000	0.099	0.000	0.000
OBGCT4	0.000	0.000	0.093	0.000	0.000	0.000	0.000	0.461	0.000	0.000
OBMCT1	0.000	0.000	0.040	0.000	0.000	0.000	0.000	0.268	0.000	0.000
OBMCT2	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
OBMCT3	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.104	0.000	0.000
OBMCT4	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
NBGC1	0.000	0.000	0.335	0.000	0.000	0.000	0.000	0.416	0.000	0.000
NBGC2	0.000	0.000	0.100	0.000	0.000	0.000	0.000	0.025	0.000	0.000
NBGC3	0.000	0.000	0.751	0.000	0.000	0.000	0.000	0.104	0.000	0.000
NBGC4	0.000	0.000	0.137	0.000	0.000	0.000	0.000	0.016	0.000	0.000
NBMC1	0.000	0.000	0.058	0.000	0.000	0.000	0.000	0.040	0.000	0.000
NBMC2	0.000	0.000	0.017	0.000	0.000	0.000	0.000	0.000	0.000	0.000
NBMC3	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
NBMC4	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
NBT1	0.000	0.000	0.000	0.017	0.000	0.000	0.000	3.402	0.000	0.975
NBT2	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.268	0.000	0.420

Samples	Weeks									
	bk	bk	1	2	3	4	5	6	7	8
NBT3	0.000	0.000	0.000	0.115	0.000	0.000	0.000	1.253	0.000	0.000
NBT4	0.000	0.000	0.000	0.000	0.000	0.000	0.000	1.652	0.000	0.227
NBGCT1	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.343	0.000	0.000
NBGCT2	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.677	0.000	0.000
NBGCT3	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
NBGCT4	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.189	0.000	0.000
NBMCT1	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
NBMCT2	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
NBMCT3	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.056	0.000	0.000
NBMCT4	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
GGC1	0.000	0.000	0.471	0.000	0.000	0.000	0.000	0.827	0.000	0.000
GGC2	0.000	0.000	0.077	0.000	0.000	0.000	0.000	0.312	0.000	0.000
GGC3	0.000	0.000	0.154	0.000	0.000	0.000	0.000	0.558	0.000	0.000
GGC4	0.000	0.000	0.300	0.000	0.000	0.000	0.000	0.865	0.000	0.000
GMC1	0.000	0.000	0.244	0.000	0.000	0.000	0.000	0.166	0.000	0.000
GMC2	0.000	0.000	0.547	0.000	0.000	0.000	0.000	0.137	0.000	0.000
GMC3	0.000	0.000	0.061	0.000	0.000	0.000	0.000	0.130	0.000	0.000
GMC4	0.000	0.000	0.113	0.000	0.000	0.000	0.000	0.179	0.000	0.000
GT1	0.000	0.000	0.523	0.000	0.000	0.000	0.000	0.046	0.000	0.000
GT2	0.000	0.000	0.364	0.200	0.000	0.000	0.000	0.078	0.000	0.000
GT3	0.000	0.000	0.134	0.000	0.000	0.000	0.000	0.065	0.000	0.000
GT4	0.000	0.000	0.324	0.614	0.000	0.000	0.000	2.544	0.000	0.805
GGCT1	5.987	0.000	0.159	0.000	0.000	0.000	0.000	0.207	0.000	0.000
GGCT2	0.000	0.000	0.081	0.000	0.000	0.000	0.000	0.119	0.000	0.000
GGCT3	2.763	0.377	1.432	0.000	0.000	0.000	0.000	0.268	0.000	0.000
GGCT4	18.420	0.201	0.058	0.000	0.000	0.000	0.000	0.417	0.000	0.000
GMCT1	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.648	0.000	0.000
GMCT2	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.038	0.000	0.000
GMCT3	0.000	0.000	0.012	0.000	0.000	0.000	0.000	0.055	0.000	0.000
GMCT4	0.000	0.000	0.008	0.000	0.000	0.000	0.000	0.183	0.000	0.000
LGC1	0.000	0.000	0.879	0.000	0.000	0.000	0.000	0.806	0.000	0.000
LGC2	0.000	0.000	0.333	0.000	0.000	0.000	0.000	0.485	0.000	0.000
LGC3	0.000	0.000	0.169	0.000	0.000	0.000	0.000	0.427	0.000	0.000
LGC4	0.000	0.000	0.339	0.000	0.000	0.000	0.000	0.496	0.000	0.000
LMC1	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.009	0.000	0.000
LMC2	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.085	0.000	0.000
LMC3	0.000	0.000	0.012	0.000	0.000	0.000	0.000	0.307	0.000	0.000
LMC4	0.000	0.000	0.059	0.000	0.000	0.000	0.000	0.166	0.000	0.000
LT1	0.000	0.000	0.503	1.422	0.000	0.000	0.000	0.069	0.000	0.124
LT2	0.000	0.000	0.000	0.166	0.000	0.000	0.000	0.000	0.000	0.000
LT3	0.000	0.000	0.027	0.153	0.000	0.000	0.000	0.000	0.000	0.000
LT4	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000

Samples	Weeks									
	bk	bk	1	2	3	4	5	6	7	8
LGCT1	1.706	0.000	0.000	0.000	0.000	0.000	0.000	0.043	0.000	0.000
LGCT2	1.957	2.652	0.155	0.021	0.000	0.000	0.000	0.065	0.000	0.000
LGCT3	0.700	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
LGCT4	0.000	1.345	0.000	0.000	0.000	0.000	0.000	0.173	0.000	0.000
LMCT1	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.032	0.000	0.000
LMCT2	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
LMCT3	0.000	0.000	1.783	0.000	0.000	0.000	0.000	0.000	0.000	0.000
LMCT4	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000

bk = background concentrations

Standard error of Al concentrations in profile leachates (mgL^{-1})

Standard error	Weeks									
	bk	bk	1	2	3	4	5	6	7	8
OBGC	0.000	0.000	0.176	0.000	0.014	0.000	0.039	0.098	0.000	0.000
OBMC	0.000	0.000	0.033	0.000	0.000	0.000	0.007	0.000	0.000	0.000
OBT	0.000	0.000	0.149	0.018	0.000	0.000	0.000	0.085	0.000	0.214
OBGCT	0.000	0.000	0.384	0.000	0.000	0.000	0.000	0.099	0.000	0.000
OBMCT	0.000	0.000	0.010	0.000	0.000	0.000	0.000	0.063	0.000	0.000
NBGC	0.000	0.000	0.149	0.000	0.000	0.000	0.000	0.094	0.000	0.000
NBMC	0.000	0.000	0.014	0.000	0.000	0.000	0.000	0.010	0.000	0.000
NBT	0.000	0.000	0.000	0.028	0.000	0.000	0.000	0.654	0.000	0.208
NBGCT	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.143	0.000	0.000
NBMCT	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.014	0.000	0.000
GGC	0.000	0.000	0.087	0.000	0.000	0.000	0.000	0.129	0.000	0.000
GMC	0.000	0.000	0.109	0.000	0.000	0.000	0.000	0.012	0.000	0.000
GT	0.000	0.000	0.080	0.145	0.000	0.000	0.000	0.620	0.000	0.201
GGCT	4.064	0.091	0.334	0.000	0.000	0.000	0.000	0.063	0.000	0.000
GMCT	0.000	0.000	0.003	0.000	0.000	0.000	0.000	0.143	0.000	0.000
LGC	0.000	0.000	0.155	0.000	0.000	0.000	0.000	0.086	0.000	0.000
LMC	0.000	0.000	0.014	0.000	0.000	0.000	0.000	0.064	0.000	0.000
LT	0.000	0.000	0.124	0.331	0.000	0.000	0.000	0.017	0.000	0.031
LGCT	0.454	0.636	0.039	0.005	0.000	0.000	0.000	0.037	0.000	0.000
LMCT	0.000	0.000	0.446	0.000	0.000	0.000	0.000	0.008	0.000	0.000

Mean Al concentrations in profile leachates (mgL^{-1})

Samples	Weeks									
	bk 1	bk 2	1	2	3	4	5	6	7	8
OBGC	0.000	0.000	0.282	0.000	0.014	0.000	0.241	0.480	0.000	0.000
OBMC	0.000	0.000	0.059	0.000	0.000	0.000	0.000	0.000	0.000	0.000
OBT	0.000	0.000	0.157	0.000	0.000	0.000	0.000	0.285	0.000	0.478
OBGCT	0.000	0.000	0.421	0.000	0.000	0.000	0.000	0.168	0.000	0.000
OBMCT	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.093	0.000	0.000
NBGC	0.000	0.000	0.331	0.000	0.000	0.000	0.000	0.140	0.000	0.000
NBMC	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
NBT	0.000	0.000	0.000	0.033	0.000	0.000	0.000	1.644	0.000	0.406
NBGCT	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.302	0.000	0.000
NBMCT	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
GGC	0.000	0.000	0.251	0.000	0.000	0.000	0.000	0.641	0.000	0.000
GMC	0.000	0.000	0.241	0.000	0.000	0.000	0.000	0.153	0.000	0.000
GT	0.000	0.000	0.336	0.204	0.000	0.000	0.000	0.683	0.000	0.201
GGCT	6.793	0.145	0.433	0.000	0.000	0.000	0.000	0.253	0.000	0.000
GMCT	0.000	0.000	0.005	0.000	0.000	0.000	0.000	0.231	0.000	0.000
LGC	0.000	0.000	0.430	0.000	0.000	0.000	0.000	0.554	0.000	0.000
LMC	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.142	0.000	0.000
LT	0.000	0.000	0.133	0.435	0.000	0.000	0.000	0.000	0.000	0.031
LGCT	1.091	0.999	0.039	0.005	0.000	0.000	0.000	0.070	0.000	0.000
LMCT	0.000	0.000	0.446	0.000	0.000	0.000	0.000	0.000	0.000	0.000
*WHO (2011)	0.200	0.200	0.200	0.200	0.200	0.200	0.200	0.200	0.200	0.200

* WHO (2011) potable water guideline

Two-way Anova test for Al concentrations in profile leachates

Tests of Between-Subjects Effects

Dependent Variable: Al conc. in leachate

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	21.301[a]	19	1.121	2.069	.017
Intercept	26.781	1	26.781	49.426	.000
Medium	14.068	4	3.517	6.491	**.000**
Aggregate	.717	3	.239	.441	.725
Medium * aggregate	6.516	12	.543	1.002	.458
Error	32.510	60	.542		
Total	80.592	80			
Corrected Total	53.811	79			

a. R Squared = .396 (Adjusted R Squared = .205)

Post Hoc test for Al concentrations in profile leachate

Multiple Comparisons

Al conc.

LSD: Growth media

(I) Medium	(J) Medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	.417875	.2602498	.114	-.102702	.938452
	MC	.677750*	.2602498	.012	.157173	1.198327
	MCT	.638875*	.2602498	.017	.118298	1.159452
	T	-.424625	.2602498	.108	-.945202	.095952
GCT	GC	-.417875	.2602498	.114	-.938452	.102702
	MC	.259875	.2602498	.322	-.260702	.780452
	MCT	.221000	.2602498	.399	-.299577	.741577
	T	-.842500*	.2602498	.002	-1.363077	-.321923
MC	GC	-.677750*	.2602498	.012	-1.198327	-.157173
	GCT	-.259875	.2602498	.322	-.780452	.260702
	MCT	-.038875	.2602498	.882	-.559452	.481702
	T	-1.102375*	.2602498	.000	-1.622952	-.581798
MCT	GC	-.638875*	.2602498	.017	-1.159452	-.118298
	GCT	-.221000	.2602498	.399	-.741577	.299577
	MC	.038875	.2602498	.882	-.481702	.559452
	T	-1.063500*	.2602498	.000	-1.584077	-.542923
T	GC	.424625	.2602498	.108	-.095952	.945202
	GCT	.842500*	.2602498	.002	.321923	1.363077
	MC	1.102375*	.2602498	.000	.581798	1.622952
	MCT	1.063500*	.2602498	.000	.542923	1.584077

Based on observed means.

The error term is Mean Square (Error) = .542.

Appendix 16: Results for Cadmium concentrations in profile leachate over an eight-week spiking period

Cd concentrations in profile leachates (mgL^{-1})

Samples	Weeks									
	bk	bk	1	2	3	4	5	6	7	8
OBGC1	0.000	0.000	0.040	0.039	0.529	0.030	0.008	0.025	0.010	0.015
OBGC2	0.001	0.002	0.021	0.013	0.014	0.006	0.004	0.007	0.004	0.003
OBGC3	0.000	0.000	0.009	0.008	0.004	0.003	0.002	0.006	0.004	0.002
OGBG4	0.001	0.001	0.010	0.003	0.002	0.001	0.006	0.005	0.004	0.002
OBMC1	0.000	0.000	0.003	0.002	0.001	0.001	0.000	0.001	0.003	0.001
OBMC2	0.000	0.000	0.140	0.003	0.002	0.001	0.001	0.003	0.003	0.001
OBMC3	0.000	0.000	0.016	0.002	0.001	0.000	0.000	0.003	0.003	0.001
OBMC4	0.000	0.000	0.024	0.010	0.003	0.003	0.002	0.004	0.003	0.001
OBT1	0.000	0.000	0.002	0.001	0.001	0.000	0.000	0.001	0.002	0.001
OBT2	0.001	0.000	0.004	0.002	0.001	0.000	0.000	0.001	0.002	0.001
OBT3	0.001	0.001	0.002	0.001	0.001	0.000	0.000	0.000	0.002	0.001
OBT4	0.001	0.001	0.006	0.001	0.001	0.000	0.000	0.001	0.001	0.000
OBGCT1	0.001	0.000	0.002	0.001	0.001	0.000	0.000	0.001	0.002	0.001
OBGCT2	0.003	0.001	0.002	0.001	0.001	0.000	0.000	0.001	0.001	0.000
OBGCT3	0.001	0.000	0.009	0.000	0.000	0.000	0.000	0.000	0.001	0.000
OBGCT4	0.001	0.001	0.002	0.000	0.001	0.000	0.001	0.003	0.002	0.000
OBMCT1	0.001	0.000	0.008	0.000	0.001	0.000	0.000	0.002	0.002	0.001
OBMCT2	0.001	0.000	0.002	0.001	0.001	0.000	0.000	0.001	0.002	0.001
OBMCT3	0.000	0.000	0.001	0.001	0.000	0.000	0.000	0.001	0.001	0.000
OBMCT4	0.001	0.000	0.002	0.003	0.001	0.000	0.000	0.001	0.002	0.001
NBGC1	0.000	0.001	0.005	0.005	0.016	0.024	0.034	0.034	0.015	0.018
NBGC2	0.000	0.000	0.004	0.002	0.005	0.006	0.009	0.009	0.004	0.004
NBGC3	0.001	0.000	0.136	0.005	0.004	0.003	0.007	0.006	0.004	0.003
NBGC4	0.001	0.000	0.005	0.003	0.002	0.002	0.003	0.003	0.002	0.003
NBMC1	0.000	0.000	0.004	0.002	0.002	0.001	0.004	0.006	0.003	0.002
NBMC2	0.000	0.000	0.004	0.001	0.001	0.001	0.004	0.003	0.002	0.001
NBMC3	0.001	0.000	0.020	0.006	0.002	0.002	0.002	0.002	0.002	0.001
NBMC4	0.001	0.000	0.003	0.002	0.001	0.001	0.004	0.004	0.001	0.001
NBT1	0.001	0.000	0.003	0.002	0.001	0.001	0.001	0.001	0.001	0.001
NBT2	0.002	0.001	0.002	0.002	0.001	0.001	0.001	0.001	0.001	0.001
NBT3	0.000	0.000	0.002	0.001	0.001	0.001	0.001	0.001	0.001	0.001
NBT4	0.000	0.000	0.002	0.001	0.001	0.001	0.001	0.001	0.001	0.001
NBGCT1	0.001	0.000	0.001	0.001	0.000	0.001	0.001	0.001	0.001	0.001
NBGCT2	0.001	0.001	0.002	0.001	0.001	0.001	0.001	0.001	0.001	0.001
NBGCT3	0.001	0.000	0.002	0.001	0.001	0.001	0.001	0.001	0.001	0.001
NBGCT4	0.001	0.000	0.003	0.001	0.000	0.001	0.001	0.002	0.001	0.001
NBMCT1	0.001	0.000	0.004	0.001	0.000	0.000	0.000	0.001	0.000	0.001
NBMCT2	0.001	0.001	0.023	0.004	0.002	0.001	0.001	0.001	0.000	0.001

Samples	Weeks									
	bk	bk	1	2	3	4	5	6	7	8
NBMCT3	0.001	0.000	0.006	0.002	0.000	0.001	0.002	0.002	0.001	0.001
NBMCT4	0.001	0.001	0.001	0.002	0.000	0.001	0.001	0.001	0.001	0.001
GGC1	0.001	0.001	0.003	0.038	0.021	0.028	0.012	0.031	0.007	0.015
GGC2	0.000	0.001	0.003	0.015	0.005	0.007	0.003	0.006	0.003	0.004
GGC3	0.000	0.001	0.003	0.007	0.002	0.003	0.006	0.007	0.002	0.002
GGC4	0.000	0.001	0.003	0.005	0.001	0.002	0.004	0.007	0.002	0.003
GMC1	0.001	0.000	0.005	0.004	0.001	0.002	0.004	0.005	0.001	0.002
GMC2	0.001	0.001	0.008	0.004	0.001	0.002	0.005	0.004	0.001	0.002
GMC3	0.001	0.001	0.010	0.004	0.002	0.002	0.005	0.003	0.001	0.001
GMC4	0.001	0.001	0.002	0.002	0.001	0.002	0.006	0.004	0.001	0.001
GT1	0.001	0.000	0.003	0.001	0.001	0.001	0.001	0.001	0.000	0.001
GT2	0.001	0.000	0.002	0.001	0.000	0.001	0.001	0.001	0.000	0.001
GT3	0.001	0.000	0.002	0.001	0.000	0.001	0.001	0.000	0.000	0.001
GT4	0.001	0.000	0.002	0.001	0.000	0.001	0.001	0.001	0.000	0.002
GGCT1	0.003	0.001	0.005	0.001	0.001	0.001	0.002	0.002	0.000	0.001
GGCT2	0.002	0.002	0.004	0.001	0.001	0.001	0.003	0.002	0.001	0.001
GGCT3	0.001	0.001	0.493	0.005	0.001	0.001	0.001	0.002	0.001	0.001
GGCT4	0.002	0.001	0.005	0.002	0.000	0.001	0.001	0.002	0.001	0.001
GMCT1	0.001	0.000	0.002	0.001	0.001	0.001	0.001	0.002	0.001	0.001
GMCT2	0.001	0.000	0.002	0.001	0.000	0.000	0.001	0.001	0.000	0.000
GMCT3	0.002	0.000	0.004	0.001	0.000	0.001	0.001	0.001	0.000	0.001
GMCT4	0.001	0.001	0.005	0.001	0.001	0.001	0.002	0.002	0.000	0.001
LGC1	0.001	0.002	0.046	0.039	0.032	0.022	0.019	0.028	0.010	0.011
LGC2	0.000	0.001	0.012	0.015	0.005	0.005	0.006	0.010	0.004	0.004
LGC3	0.001	0.001	0.006	0.008	0.002	0.003	0.005	0.011	0.003	0.003
LGC4	0.001	0.000	0.003	0.005	0.001	0.002	0.004	0.006	0.002	0.002
LMC1	0.000	0.001	0.038	0.014	0.003	0.004	0.003	0.005	0.002	0.001
LMC2	0.000	0.001	0.003	0.004	0.001	0.002	0.003	0.003	0.001	0.001
LMC3	0.001	0.000	0.005	0.004	0.001	0.002	0.004	0.006	0.001	0.001
LMC4	0.001	0.000	0.000	0.003	0.001	0.001	0.004	0.004	0.001	0.001
LT1	0.001	0.000	0.000	0.002	0.001	0.001	0.000	0.001	0.001	0.001
LT2	0.001	0.000	0.000	0.001	0.001	0.001	0.000	0.001	0.001	0.000
LT3	0.001	0.000	0.000	0.001	0.000	0.000	0.001	0.001	0.001	0.001
LT4	0.001	0.000	0.000	0.001	0.001	0.001	0.000	0.001	0.001	0.000
LGCT1	0.001	0.000	0.000	0.001	0.000	0.000	0.001	0.002	0.001	0.000
LGCT2	0.002	0.001	0.000	0.002	0.000	0.001	0.001	0.001	0.001	0.000
LGCT3	0.002	0.001	0.000	0.001	0.001	0.000	0.001	0.001	0.001	0.001
LGCT4	0.001	0.001	0.000	0.001	0.000	0.001	0.001	0.001	0.001	0.000
LMCT1	0.002	0.001	0.000	0.001	0.000	0.001	0.000	0.001	0.000	0.000
LMCT2	0.001	0.001	0.000	0.001	0.000	0.001	0.001	0.001	0.001	0.000
LMCT3	0.001	0.000	1.273	0.003	0.000	0.001	0.001	0.001	0.001	0.000
LMCT4	0.001	0.001	0.003	0.001	0.000	0.001	0.001	0.001	0.001	0.001

Standard error of Cd concentrations in profile leachates (mgL^{-1})

standard error	Weeks									
	bk	bk	1	2	3	4	5	6	7	8
OBGC	0.000	0.000	0.007	0.008	0.131	0.007	0.001	0.005	0.002	0.003
OBMC	0.000	0.000	0.032	0.002	0.000	0.001	0.000	0.001	0.000	0.000
OBT	0.000	0.000	0.001	0.000	0.000	0.000	0.000	0.000	0.000	0.000
OBGCT	0.001	0.000	0.002	0.000	0.000	0.000	0.000	0.001	0.000	0.000
OBMCT	0.000	0.000	0.002	0.001	0.000	0.000	0.000	0.000	0.000	0.000
NBGC	0.000	0.000	0.033	0.001	0.003	0.005	0.007	0.007	0.003	0.004
NBMC	0.000	0.000	0.004	0.001	0.000	0.000	0.001	0.001	0.000	0.000
NBT	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
NBGCT	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
NBMCT	0.000	0.000	0.005	0.001	0.001	0.000	0.000	0.000	0.000	0.000
GGC	0.000	0.000	0.000	0.008	0.005	0.006	0.002	0.006	0.001	0.003
GMC	0.000	0.000	0.002	0.001	0.000	0.000	0.000	0.000	0.000	0.000
GT	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
GGCT	0.000	0.000	0.122	0.001	0.000	0.000	0.000	0.000	0.000	0.000
GMCT	0.000	0.000	0.001	0.000	0.000	0.000	0.000	0.000	0.000	0.000
LGC	0.000	0.000	0.010	0.008	0.007	0.005	0.004	0.005	0.002	0.002
LMC	0.000	0.000	0.009	0.003	0.001	0.001	0.000	0.001	0.000	0.000
LT	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
LGCT	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
LMCT	0.000	0.000	0.318	0.001	0.000	0.000	0.000	0.000	0.000	0.000

Mean Cd concentrations in profile leachates (mgL^{-1})

Samples	Weeks									
	bk1	bk2	1	2	3	4	5	6	7	8
OBGC	0.000	0.000	0.020	0.016	0.137	0.010	0.005	0.011	0.006	0.006
OBMC	0.000	0.000	0.046	0.004	0.000	0.000	0.000	0.003	0.003	0.000
OBT	0.000	0.000	0.004	0.000	0.000	0.000	0.000	0.000	0.000	0.000
OBGCT	0.000	0.000	0.004	0.000	0.000	0.000	0.000	0.000	0.000	0.000
OBMCT	0.000	0.000	0.003	0.000	0.000	0.000	0.000	0.000	0.000	0.000
NBGC	0.000	0.000	0.038	0.004	0.007	0.009	0.013	0.013	0.006	0.007
NBMC	0.000	0.000	0.008	0.003	0.000	0.000	0.004	0.004	0.000	0.000
NBT	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
NBGCT	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
NBMCT	0.000	0.000	0.009	0.000	0.000	0.000	0.000	0.000	0.000	0.000
GGC	0.000	0.000	0.003	0.016	0.007	0.010	0.006	0.013	0.004	0.006
GMC	0.000	0.000	0.006	0.004	0.000	0.000	0.005	0.004	0.000	0.000
GT	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
GGCT	0.000	0.000	0.127	0.000	0.000	0.000	0.000	0.000	0.000	0.000
GMCT	0.000	0.000	0.003	0.000	0.000	0.000	0.000	0.000	0.000	0.000
LGC	0.000	0.000	0.017	0.017	0.010	0.008	0.009	0.014	0.005	0.005
LMC	0.000	0.000	0.012	0.006	0.000	0.000	0.004	0.005	0.000	0.000
LT	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
LGCT	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
LMCT	0.000	0.000	0.319	0.000	0.000	0.000	0.000	0.000	0.000	0.000
WHO (2011)	0.003	0.003	0.003	0.003	0.003	0.003	0.003	0.003	0.003	0.003

Two-way Anova test for Cd concentrations in profile leachates

Tests of Between-Subjects Effects

Dependent Variable: Cd conc. in leachate

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.518[a]	19	.027	.919	.563
Intercept	.262	1	.262	8.824	.004
Medium	.119	4	.030	1.005	.412
aggregate	.037	3	.012	.417	.741
Medium * aggregate	.361	12	.030	1.016	.447
Error	1.779	60	.030		
Total	2.558	80			
Corrected Total	2.297	79			

a. R Squared = .225 (Adjusted R Squared = -.020)

Post Hoc test for Cd concentrations in profile leachate

Multiple Comparisons

Cd conc.

LSD: Growth media

(I) Medium	(J) Medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	.073938	.0608791	.229	-.047839	.195714
	MC	.078500	.0608791	.202	-.043276	.200276
	MCT	.024063	.0608791	.694	-.097714	.145839
	T	.105688	.0608791	.088	-.016089	.227464
GCT	GC	-.073938	.0608791	.229	-.195714	.047839
	MC	.004562	.0608791	.941	-.117214	.126339
	MCT	-.049875	.0608791	.416	-.171651	.071901
	T	.031750	.0608791	.604	-.090026	.153526
MC	GC	-.078500	.0608791	.202	-.200276	.043276
	GCT	-.004562	.0608791	.941	-.126339	.117214
	MCT	-.054437	.0608791	.375	-.176214	.067339
	T	.027187	.0608791	.657	-.094589	.148964
MCT	GC	-.024063	.0608791	.694	-.145839	.097714
	GCT	.049875	.0608791	.416	-.071901	.171651
	MC	.054437	.0608791	.375	-.067339	.176214
	T	.081625	.0608791	.185	-.040151	.203401
T	GC	-.105688	.0608791	.088	-.227464	.016089
	GCT	-.031750	.0608791	.604	-.153526	.090026
	MC	-.027187	.0608791	.657	-.148964	.094589
	MCT	-.081625	.0608791	.185	-.203401	.040151

Based on observed means.

The error term is Mean Square (Error) = .030.

Multiple Comparisons

Cd conc.

LSD: Aggregates

(I) aggregate	(J) aggregate	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	-.041600	.0544519	.448	-.150520	.067320
	NB	.017650	.0544519	.747	-.091270	.126570
	OB	-.010800	.0544519	.843	-.119720	.098120
L	G	.041600	.0544519	.448	-.067320	.150520
	NB	.059250	.0544519	.281	-.049670	.168170
	OB	.030800	.0544519	.574	-.078120	.139720
NB	G	-.017650	.0544519	.747	-.126570	.091270
	L	-.059250	.0544519	.281	-.168170	.049670
	OB	-.028450	.0544519	.603	-.137370	.080470
OB	G	.010800	.0544519	.843	-.098120	.119720
	L	-.030800	.0544519	.574	-.139720	.078120
	NB	.028450	.0544519	.603	-.080470	.137370

Based on observed means.

The error term is Mean Square (Error) = .030.

Appendix 17: Results for Chromium concentrations in profile leachate over an eight-week spiking period

Cr concentrations in profile leachates (mgL^{-1})

Samples	Weeks									
	bk	bk	1	2	3	4	5	6	7	8
OBGC1	0.002	0.002	0.029	0.034	0.463	0.030	0.010	0.033	0.011	0.018
OBGC2	0.002	0.001	0.018	0.011	0.015	0.006	0.007	0.013	0.004	0.005
OBGC3	0.002	0.000	0.007	0.009	0.003	0.004	0.003	0.012	0.003	0.002
OGBG4	0.001	0.003	0.022	0.003	0.002	0.003	0.009	0.013	0.004	0.003
OBMC1	0.002	0.002	0.002	0.002	0.001	0.002	0.001	0.003	0.002	0.001
OBMC2	0.001	0.003	0.068	0.008	0.001	0.003	0.003	0.009	0.003	0.001
OBMC3	0.001	0.004	0.014	0.002	0.001	0.002	0.004	0.005	0.002	0.001
OBMC4	0.000	0.002	0.028	0.008	0.001	0.002	0.003	0.005	0.002	0.001
OBT1	0.003	0.002	0.000	0.001	0.001	0.002	0.001	0.003	0.002	0.001
OBT2	0.002	0.003	0.004	0.003	0.001	0.002	0.001	0.003	0.002	0.002
OBT3	0.003	0.003	0.000	0.000	0.001	0.001	0.000	0.001	0.000	0.001
OBT4	0.003	0.003	0.001	0.002	0.001	0.002	0.000	0.003	0.001	0.002
OBGCT1	0.003	0.002	0.001	0.002	0.000	0.001	0.000	0.002	0.001	0.001
OBGCT2	0.003	0.002	0.001	0.001	0.000	0.001	0.000	0.001	0.001	0.000
OBGCT3	0.002	0.003	0.011	0.001	0.000	0.001	0.000	0.003	0.001	0.001
OBGCT4	0.002	0.001	0.000	0.001	0.000	0.001	0.007	0.005	0.001	0.000
OBMCT1	0.002	0.002	0.010	0.000	0.000	0.001	0.006	0.004	0.001	0.001
OBMCT2	0.002	0.002	0.001	0.000	0.000	0.001	0.001	0.003	0.001	0.000
OBMCT3	0.002	0.004	0.001	0.002	0.001	0.001	0.000	0.002	0.001	0.000
OBMCT4	0.003	0.004	0.034	0.004	0.001	0.002	0.000	0.001	0.001	0.001
NBGC1	0.002	0.018	0.007	0.007	0.017	0.025	0.038	0.036	0.017	0.022
NBGC2	0.001	0.005	0.005	0.002	0.005	0.007	0.012	0.010	0.060	0.005
NBGC3	0.002	0.022	0.158	0.006	0.005	0.004	0.009	0.007	0.004	0.003
NBGC4	0.003	0.009	0.004	0.003	0.002	0.002	0.005	0.005	0.003	0.002
NBMC1	0.003	0.008	0.006	0.001	0.003	0.002	0.009	0.008	0.003	0.001
NBMC2	0.002	0.010	0.002	0.001	0.002	0.002	0.012	0.006	0.002	0.001
NBMC3	0.005	0.014	0.026	0.004	0.002	0.001	0.007	0.003	0.002	0.001
NBMC4	0.001	0.005	0.002	0.001	0.001	0.002	0.016	0.005	0.003	0.001
NBT1	0.004	0.005	0.000	0.002	0.002	0.003	0.002	0.006	0.002	0.002
NBT2	0.004	0.014	0.000	0.003	0.002	0.002	0.002	0.002	0.001	0.002
NBT3	0.003	0.007	0.001	0.001	0.002	0.001	0.002	0.003	0.002	0.001
NBT4	0.004	0.033	0.000	0.000	0.002	0.002	0.002	0.003	0.001	0.002
NBGCT1	0.004	0.014	0.000	0.002	0.001	0.001	0.002	0.001	0.001	0.001
NBGCT2	0.006	0.025	0.001	0.001	0.001	0.001	0.003	0.004	0.001	0.000
NBGCT3	0.003	0.013	0.001	0.002	0.001	0.000	0.002	0.000	0.000	0.001
NBGCT4	0.005	0.014	0.002	0.001	0.001	0.001	0.005	0.004	0.002	0.000
NBMCT1	0.005	0.006	0.003	0.001	0.000	0.001	0.001	0.001	0.001	0.000
NBMCT2	0.003	0.008	0.026	0.004	0.001	0.001	0.001	0.000	0.001	0.000
NBMCT3	0.004	0.018	0.002	0.001	0.001	0.001	0.004	0.003	0.001	0.000

Samples	Weeks									
	bk	bk	1	2	3	4	5	6	7	8
NBMCT4	0.006	0.019	0.000	0.000	0.001	0.000	0.004	0.002	0.001	0.001
GGC1	0.001	0.004	0.002	0.034	0.021	0.027	0.017	0.035	0.008	0.020
GGC2	0.001	0.003	0.000	0.013	0.005	0.007	0.007	0.013	0.004	0.005
GGC3	0.002	0.004	0.003	0.005	0.003	0.003	0.009	0.013	0.003	0.003
GGC4	0.001	0.003	0.003	0.004	0.001	0.003	0.007	0.018	0.003	0.003
GMC1	0.002	0.006	0.008	0.004	0.001	0.002	0.012	0.009	0.002	0.002
GMC2	0.001	0.004	0.016	0.004	0.002	0.003	0.016	0.007	0.002	0.001
GMC3	0.001	0.004	0.007	0.003	0.003	0.001	0.011	0.006	0.002	0.002
GMC4	0.002	0.008	0.003	0.003	0.003	0.002	0.014	0.011	0.003	0.001
GT1	0.003	0.002	0.005	0.001	0.002	0.002	0.003	0.002	0.001	0.001
GT2	0.005	0.002	0.003	0.001	0.000	0.001	0.002	0.002	0.001	0.001
GT3	0.003	0.003	0.002	0.001	0.000	0.001	0.002	0.001	0.001	0.002
GT4	0.002	0.002	0.002	0.001	0.001	0.002	0.002	0.007	0.001	0.003
GGCT1	0.017	0.010	0.006	0.001	0.001	0.000	0.002	0.002	0.001	0.001
GGCT2	0.008	0.011	0.004	0.000	0.001	0.001	0.004	0.003	0.001	0.001
GGCT3	0.012	0.009	0.157	0.008	0.002	0.002	0.003	0.003	0.001	0.001
GGCT4	0.032	0.009	0.004	0.000	0.000	0.000	0.003	0.003	0.001	0.001
GMCT1	0.001	0.002	0.000	0.001	0.001	0.001	0.005	0.004	0.001	0.002
GMCT2	0.007	0.008	0.001	0.000	0.000	0.000	0.003	0.002	0.000	0.001
GMCT3	0.014	0.007	0.003	0.000	0.000	0.000	0.003	0.002	0.000	0.001
GMCT4	0.008	0.008	0.010	0.000	0.000	0.000	0.004	0.003	0.001	0.000
LGC1	0.004	0.006	0.043	0.034	0.030	0.021	0.026	0.035	0.014	0.016
LGC2	0.001	0.005	0.008	0.012	0.005	0.004	0.009	0.018	0.005	0.005
LGC3	0.005	0.005	0.004	0.006	0.003	0.004	0.007	0.017	0.004	0.005
LGC4	0.003	0.006	0.003	0.004	0.001	0.003	0.007	0.013	0.003	0.003
LMC1	0.001	0.007	0.035	0.007	0.002	0.003	0.006	0.007	0.003	0.002
LMC2	0.001	0.007	0.002	0.003	0.002	0.002	0.007	0.008	0.002	0.002
LMC3	0.002	0.006	0.004	0.003	0.001	0.002	0.007	0.011	0.003	0.002
LMC4	0.001	0.004	0.000	0.002	0.001	0.002	0.010	0.011	0.002	0.002
LT1	0.003	0.002	0.000	0.004	0.002	0.002	0.002	0.002	0.002	0.002
LT2	0.002	0.002	0.000	0.001	0.001	0.001	0.001	0.001	0.002	0.001
LT3	0.002	0.003	0.000	0.000	0.001	0.001	0.001	0.001	0.002	0.001
LT4	0.003	0.002	0.000	0.000	0.001	0.001	0.001	0.001	0.001	0.001
LGCT1	0.010	0.007	0.000	0.000	0.001	0.001	0.002	0.002	0.001	0.001
LGCT2	0.002	0.013	0.000	0.001	0.001	0.000	0.001	0.001	0.001	0.001
LGCT3	0.012	0.008	0.000	0.001	0.001	0.001	0.001	0.001	0.001	0.001
LGCT4	0.008	0.013	0.000	0.001	0.001	0.001	0.002	0.001	0.001	0.001
LMCT1	0.001	0.010	0.000	0.000	0.001	0.001	0.001	0.001	0.001	0.001
LMCT2	0.005	0.009	0.000	0.000	0.000	0.001	0.001	0.001	0.001	0.001
LMCT3	0.005	0.009	0.604	0.010	0.001	0.002	0.002	0.002	0.001	0.001
LMCT4	0.009	0.006	0.003	0.001	0.001	0.001	0.003	0.002	0.001	0.001

Standard error of Cr concentrations in profile leachates (mgL^{-1})

standard error	Weeks									
	bk 1	bk 2	1	2	3	4	5	6	7	8
OBGC	0.000	0.001	0.005	0.007	0.114	0.006	0.002	0.005	0.002	0.004
OBMC	0.000	0.000	0.014	0.002	0.000	0.000	0.001	0.001	0.000	0.000
OBT	0.000	0.000	0.001	0.001	0.000	0.000	0.000	0.001	0.000	0.000
OBGCT	0.000	0.000	0.003	0.000	0.000	0.000	0.002	0.001	0.000	0.000
OBMCT	0.000	0.001	0.008	0.001	0.000	0.000	0.001	0.001	0.000	0.000
NBGC	0.000	0.004	0.038	0.001	0.003	0.005	0.007	0.007	0.013	0.005
NBMC	0.001	0.002	0.006	0.001	0.000	0.000	0.002	0.001	0.000	0.000
NBT	0.000	0.006	0.000	0.001	0.000	0.000	0.000	0.001	0.000	0.000
NBGCT	0.001	0.003	0.000	0.000	0.000	0.000	0.001	0.001	0.000	0.000
NBMCT	0.001	0.003	0.006	0.001	0.000	0.000	0.001	0.001	0.000	0.000
GGC	0.000	0.000	0.001	0.007	0.005	0.006	0.002	0.005	0.001	0.004
GMC	0.000	0.001	0.003	0.000	0.000	0.000	0.001	0.001	0.000	0.000
GT	0.001	0.000	0.001	0.000	0.000	0.000	0.000	0.001	0.000	0.000
GGCT	0.005	0.000	0.038	0.002	0.000	0.000	0.000	0.000	0.000	0.000
GMCT	0.003	0.001	0.002	0.000	0.000	0.000	0.000	0.000	0.000	0.000
LGC	0.001	0.000	0.010	0.007	0.007	0.004	0.005	0.005	0.003	0.003
LMC	0.000	0.001	0.008	0.001	0.000	0.000	0.001	0.001	0.000	0.000
LT	0.000	0.000	0.000	0.001	0.000	0.000	0.000	0.000	0.000	0.000
LGCT	0.002	0.002	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
LMCT	0.002	0.001	0.151	0.002	0.000	0.000	0.000	0.000	0.000	0.000

Mean Cr concentrations in profile leachates (mgL^{-1})

Samples	Weeks									
	BK1	Bk2	1	2	3	4	5	6	7	8
OBGC	0.000	0.000	0.019	0.014	0.121	0.011	0.007	0.018	0.000	0.000
OBMC	0.000	0.000	0.028	0.000	0.000	0.000	0.000	0.000	0.000	0.000
OBT	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
OBGCT	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
OBMCT	0.000	0.000	0.012	0.000	0.000	0.000	0.000	0.000	0.000	0.000
NBGC	0.000	0.014	0.044	0.000	0.007	0.010	0.016	0.015	0.021	0.008
NBMC	0.000	0.009	0.009	0.000	0.000	0.000	0.011	0.000	0.000	0.000
NBT	0.000	0.015	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
NBGCT	0.000	0.017	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
NBMCT	0.000	0.013	0.008	0.000	0.000	0.000	0.000	0.000	0.000	0.000
GGC	0.000	0.000	0.000	0.014	0.008	0.010	0.010	0.020	0.000	0.008
GMC	0.000	0.000	0.009	0.000	0.000	0.000	0.013	0.008	0.000	0.000
GT	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
GGCT	0.017	0.010	0.043	0.000	0.000	0.000	0.000	0.000	0.000	0.000
GMCT	0.008	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
LGC	0.000	0.000	0.015	0.014	0.010	0.008	0.012	0.021	0.007	0.007
LMC	0.000	0.000	0.010	0.000	0.000	0.000	0.008	0.009	0.003	0.000

Samples	Weeks									
	bk	bk	1	2	3	4	5	6	7	8
LT	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.002	0.000
LGCT	0.008	0.010	0.000	0.000	0.000	0.000	0.000	0.000	0.001	0.000
LMCT	0.000	0.009	0.152	0.000	0.000	0.000	0.000	0.000	0.001	0.000
WHO (2011) Potable Water guideline	0.050	0.050	0.050	0.050	0.050	0.050	0.050	0.050	0.050	0.050

Two-way Anova test for Cr concentrations in profile leachates

Tests of Between-Subjects Effects

Dependent Variable: Cr conc. In leachate

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.232[a]	19	.012	1.219	.274
Intercept	.201	1	.201	19.999	.000
Medium	.125	4	.031	3.105	**.022**
aggregate	.009	3	.003	.284	.837
Medium * aggregate	.099	12	.008	.825	.625
Error	.602	60	.010		
Total	1.035	80			
Corrected Total	.835	79			

a. R Squared = .279 (Adjusted R Squared = .050)

Post Hoc test for Cr concentrations in profile leachate

Multiple Comparisons

Cr conc.

LSD: Growth media

(I) Medium	(J) Medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	.102688*	.0354184	.005	.031840	.173535
	MC	.083063*	.0354184	.022	.012215	.153910
	MCT	.071125*	.0354184	.049	.000278	.141972
	T	.111438*	.0354184	.003	.040590	.182285
GCT	GC	-.102688*	.0354184	.005	-.173535	-.031840
	MC	-.019625	.0354184	.582	-.090472	.051222
	MCT	-.031563	.0354184	.376	-.102410	.039285
	T	.008750	.0354184	.806	-.062097	.079597
MC	GC	-.083063*	.0354184	.022	-.153910	-.012215
	GCT	.019625	.0354184	.582	-.051222	.090472
	MCT	-.011937	.0354184	.737	-.082785	.058910
	T	.028375	.0354184	.426	-.042472	.099222
MCT	GC	-.071125*	.0354184	.049	-.141972	-.000278
	GCT	.031563	.0354184	.376	-.039285	.102410
	MC	.011937	.0354184	.737	-.058910	.082785
	T	.040313	.0354184	.260	-.030535	.111160
T	GC	-.111438*	.0354184	.003	-.182285	-.040590
	GCT	-.008750	.0354184	.806	-.079597	.062097
	MC	-.028375	.0354184	.426	-.099222	.042472
	MCT	-.040313	.0354184	.260	-.111160	.030535

Based on observed means.

The error term is Mean Square (Error) = .010.

*.The mean difference is significant at the 0.05 level.

Multiple Comparisons

Cr conc.

LSD: Aggregates

(I) aggregate	(J) aggregate	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	-.022450	.0316792	.481	-.085818	.040918
	NB	-.000450	.0316792	.989	-.063818	.062918
	OB	-.019050	.0316792	.550	-.082418	.044318
L	G	.022450	.0316792	.481	-.040918	.085818
	NB	.022000	.0316792	.490	-.041368	.085368
	OB	.003400	.0316792	.915	-.059968	.066768
NB	G	.000450	.0316792	.989	-.062918	.063818
	L	-.022000	.0316792	.490	-.085368	.041368
	OB	-.018600	.0316792	.559	-.081968	.044768
OB	G	.019050	.0316792	.550	-.044318	.082418
	L	-.003400	.0316792	.915	-.066768	.059968
	NB	.018600	.0316792	.559	-.044768	.081968

Based on observed means.

The error term is Mean Square (Error) = .010.

Appendix 18: Results for Copper concentrations in profile leachate over an eight-week spiking period

Cu concentrations in profile leachates (mgL^{-1})

Samples	Weeks									
	bk	bk	1	2	3	4	5	6	7	8
OBGC1	0.083	0.035	0.038	0.042	0.474	0.035	0.041	0.083	0.190	0.027
OBGC2	0.035	0.025	0.087	0.020	0.032	0.021	0.039	0.048	0.011	0.010
OBGC3	0.044	0.013	0.038	0.056	0.017	0.013	0.036	0.066	0.012	0.010
OGBG4	0.084	0.094	0.231	0.018	0.016	0.016	0.056	0.086	0.017	0.010
OBMC1	0.063	0.033	0.012	0.010	0.013	0.009	0.019	0.016	0.008	0.009
OBMC2	0.031	0.046	0.104	0.019	0.016	0.012	0.023	0.017	0.010	0.014
OBMC3	0.034	0.054	0.103	0.011	0.012	0.010	0.025	0.022	0.010	0.009
OBMC4	0.056	0.029	0.056	0.027	0.017	0.013	0.023	0.028	0.012	0.010
OBT1	0.312	0.135	0.027	0.018	0.018	0.016	0.031	0.036	0.013	0.011
OBT2	0.214	0.126	0.431	0.307	0.074	0.041	0.056	0.073	0.026	0.032
OBT3	0.357	0.254	0.102	0.042	0.037	0.023	0.044	0.061	0.025	0.027
OBT4	0.337	0.245	1.016	0.149	0.089	0.069	0.067	0.105	0.045	0.042
OBGCT1	0.308	0.102	0.027	0.045	0.018	0.015	0.026	0.035	0.012	0.017
OBGCT2	0.300	0.100	0.054	0.019	0.014	0.012	0.028	0.038	0.012	0.011
OBGCT3	0.117	0.111	0.962	0.027	0.014	0.016	0.023	0.025	0.012	0.011
OBGCT4	0.122	0.089	0.052	0.014	0.010	0.011	0.030	0.048	0.011	0.009
OBMCT1	0.283	0.138	0.351	0.010	0.018	0.023	0.037	0.043	0.017	0.012
OBMCT2	0.171	0.145	0.060	0.015	0.010	0.011	0.025	0.025	0.011	0.008
OBMCT3	0.235	0.344	0.030	0.019	0.014	0.014	0.041	0.034	0.01	0.008
OBMCT4	0.307	0.212	0.047	0.051	0.025	0.015	0.023	0.042	0.022	0.009
NBGC1	0.043	0.060	0.094	0.040	0.026	0.028	0.079	0.090	0.025	0.026
NBGC2	0.059	0.015	0.075	0.011	0.010	0.015	0.038	0.050	0.018	0.011
NBGC3	0.038	0.042	0.308	0.016	0.013	0.012	0.052	0.052	0.016	0.011
NBGC4	0.102	0.022	0.064	0.025	0.015	0.010	0.036	0.051	0.013	0.013
NBMC1	0.077	0.026	0.036	0.011	0.020	0.013	0.023	0.045	0.016	0.011
NBMC2	0.024	0.038	0.017	0.011	0.010	0.010	0.022	0.033	0.016	0.009
NBMC3	0.048	0.020	0.045	0.016	0.009	0.009	0.017	0.016	0.017	0.010
NBMC4	0.032	0.015	0.021	0.011	0.009	0.015	0.026	0.033	0.016	0.011
NBT1	0.147	0.063	0.255	0.099	0.100	0.050	0.066	0.112	0.053	0.042
NBT2	0.245	0.157	0.115	0.235	0.057	0.049	0.042	0.054	0.037	0.030
NBT3	0.206	0.138	0.114	0.087	0.046	0.029	0.058	0.127	0.044	0.037
NBT4	0.142	0.153	0.219	0.071	0.053	0.039	0.051	0.088	0.043	0.033
NBGCT1	0.203	0.109	0.013	0.036	0.012	0.011	0.028	0.048	0.018	0.011
NBGCT2	0.203	0.135	0.048	0.029	0.019	0.014	0.038	0.056	0.020	0.015
NBGCT3	0.227	0.153	0.031	0.016	0.018	0.011	0.025	0.034	0.015	0.012
NBGCT4	0.160	0.124	0.202	0.033	0.026	0.015	0.033	0.047	0.023	0.014
NBMCT1	0.282	0.136	0.319	0.032	0.015	0.016	0.032	0.037	0.015	0.009
NBMCT2	0.668	0.518	0.328	0.091	0.033	0.039	0.026	0.039	0.022	0.016

Samples	Weeks									
	bk	bk	1	2	3	4	5	6	7	8
NBMCT3	0.193	0.138	0.193	0.035	0.016	0.011	0.034	0.050	0.015	0.007
NBMCT4	0.259	0.284	0.033	0.030	0.020	0.014	0.032	0.043	0.017	0.010
GGC1	0.031	0.042	0.031	0.038	0.069	0.039	0.118	0.485	0.033	0.027
GGC2	0.038	0.061	0.015	0.030	0.059	0.021	0.054	0.075	0.018	0.014
GGC3	0.048	0.067	0.027	0.019	0.046	0.015	0.061	0.099	0.020	0.012
GGC4	0.033	0.054	0.046	0.018	0.020	0.018	0.056	0.150	0.021	0.014
GMC1	0.154	0.103	0.056	0.018	0.013	0.017	0.022	0.032	0.019	0.008
GMC2	0.037	0.042	0.124	0.030	0.027	0.020	0.046	0.038	0.016	0.011
GMC3	0.061	0.057	0.045	0.013	0.040	0.014	0.046	0.033	0.015	0.012
GMC4	0.062	0.080	0.026	0.019	0.040	0.019	0.047	0.045	0.023	0.012
GT1	0.237	0.121	0.488	0.064	0.057	0.025	0.014	0.032	0.021	0.014
GT2	0.191	0.096	0.416	0.047	0.026	0.032	0.032	0.033	0.023	0.015
GT3	0.582	0.195	0.143	0.019	0.022	0.021	0.038	0.032	0.025	0.013
GT4	0.186	0.141	0.303	0.116	0.064	0.074	0.069	0.095	0.038	0.035
GGCT1	0.379	0.378	0.251	0.015	0.048	0.013	0.042	0.039	0.017	0.015
GGCT2	0.366	0.583	0.127	0.017	0.031	0.017	0.033	0.038	0.015	0.010
GGCT3	0.235	0.247	0.405	0.032	0.033	0.016	0.029	0.029	0.018	0.010
GGCT4	0.557	0.208	0.098	0.016	0.012	0.014	0.030	0.043	0.016	0.009
GMCT1	0.103	0.114	0.047	0.023	0.031	0.023	0.066	0.062	0.028	0.019
GMCT2	0.209	0.285	0.050	0.016	0.014	0.014	0.045	0.033	0.017	0.009
GMCT3	0.508	0.232	0.195	0.023	0.015	0.014	0.031	0.033	0.012	0.009
GMCT4	0.276	0.229	0.244	0.020	0.013	0.018	0.048	0.043	0.017	0.012
LGC1	0.091	0.090	0.224	0.045	0.043	0.026	0.060	0.073	0.023	0.020
LGC2	0.133	0.247	0.072	0.028	0.019	0.015	0.039	0.099	0.019	0.010
LGC3	0.774	0.226	0.063	0.019	0.016	0.012	0.040	0.102	0.019	0.008
LGC4	0.054	0.075	0.064	0.012	0.009	0.009	0.060	0.097	0.020	0.010
LMC1	0.095	0.230	0.076	0.027	0.012	0.011	0.036	0.057	0.013	0.010
LMC2	0.017	0.088	0.030	0.013	0.008	0.013	0.026	0.040	0.008	0.008
LMC3	0.046	0.074	0.024	0.013	0.006	0.010	0.029	0.059	0.013	0.009
LMC4	0.011	0.036	0.031	0.017	0.006	0.012	0.044	0.560	0.010	0.008
LT1	0.227	0.087	0.096	0.160	0.050	0.058	0.028	0.048	0.032	0.027
LT2	0.230	0.081	0.104	0.035	0.033	0.017	0.023	0.033	0.017	0.012
LT3	0.458	0.215	0.118	0.032	0.032	0.024	0.031	0.026	0.018	0.015
LT4	0.276	0.212	0.396	0.039	0.025	0.024	0.024	0.038	0.017	0.015
LGCT1	0.167	0.181	0.075	0.010	0.009	0.007	0.016	0.025	0.011	0.009
LGCT2	0.992	0.508	0.404	0.094	0.020	0.010	0.027	0.029	0.016	0.011
LGCT3	0.402	0.368	0.047	0.011	0.018	0.007	0.013	0.020	0.012	0.009
LGCT4	0.491	0.419	0.015	0.025	0.023	0.017	0.027	0.032	0.019	0.015
LMCT1	0.534	0.414	0.004	0.010	0.025	0.008	0.024	0.023	0.011	0.007
LMCT2	0.199	0.553	0.072	0.019	0.007	0.009	0.022	0.028	0.016	0.017
LMCT3	0.174	0.413	1.046	0.022	0.012	0.010	0.026	0.025	0.011	0.008
LMCT4	0.416	0.325	0.140	0.023	0.015	0.011	0.023	0.029	0.016	0.012

Standard error of Cu concentrations in profile leachates (mgL^{-1})

standard error	Weeks									
	bk	bk	1	2	3	4	5	6	7	8
OBGC	0.013	0.018	0.046	0.009	0.113	0.005	0.004	0.009	0.044	0.004
OBMC	0.008	0.006	0.022	0.004	0.001	0.001	0.001	0.003	0.001	0.001
OBT	0.032	0.034	0.225	0.066	0.016	0.012	0.008	0.014	0.007	0.006
OBGCT	0.053	0.005	0.229	0.007	0.002	0.001	0.001	0.005	0.000	0.002
OBMCT	0.028	0.033	0.035	0.035	0.035	0.035	0.035	0.035	0.035	0.033
NBGC	0.015	0.010	0.058	0.006	0.003	0.004	0.010	0.010	0.003	0.004
NBMC	0.012	0.005	0.007	0.001	0.003	0.001	0.002	0.006	0.000	0.000
NBT	0.025	0.022	0.036	0.038	0.012	0.005	0.005	0.016	0.003	0.003
NBGCT	0.014	0.009	0.043	0.004	0.003	0.001	0.003	0.005	0.002	0.001
NBMCT	0.108	0.090	0.069	0.015	0.004	0.006	0.002	0.003	0.002	0.002
GGC	0.004	0.005	0.006	0.005	0.011	0.005	0.015	0.096	0.003	0.003
GMC	0.026	0.013	0.021	0.004	0.006	0.001	0.006	0.003	0.002	0.001
GT	0.095	0.021	0.075	0.020	0.011	0.012	0.011	0.016	0.004	0.005
GGCT	0.066	0.085	0.070	0.004	0.007	0.001	0.003	0.003	0.001	0.001
GMCT	0.086	0.036	0.050	0.002	0.004	0.002	0.007	0.007	0.003	0.002
LGC	0.171	0.045	0.039	0.007	0.007	0.004	0.006	0.007	0.001	0.003
LMC	0.019	0.042	0.012	0.003	0.001	0.001	0.004	0.127	0.001	0.000
LT	0.055	0.037	0.073	0.031	0.005	0.009	0.002	0.005	0.004	0.003
LGCT	0.174	0.069	0.090	0.020	0.003	0.002	0.004	0.003	0.002	0.001
LMCT	0.087	0.047	0.245	0.003	0.004	0.001	0.001	0.001	0.001	0.002

Mean Cu concentrations in profile leachates (mgL^{-1})

Samples	Weeks									
	bk1	bk2	1	2	3	4	5	6	7	8
OBGC	0.062	0.042	0.099	0.034	0.135	0.021	0.043	0.071	0.058	0.014
OBMC	0.046	0.041	0.069	0.017	0.015	0.011	0.023	0.021	0.010	0.011
OBT	0.305	0.190	0.394	0.129	0.055	0.037	0.050	0.069	0.027	0.028
OBGCT	0.212	0.101	0.274	0.026	0.014	0.014	0.027	0.037	0.012	0.012
OBMCT	0.249	0.210	0.122	0.024	0.017	0.016	0.032	0.036	0.015	0.000
NBGC	0.061	0.035	0.135	0.023	0.016	0.016	0.051	0.061	0.018	0.015
NBMC	0.045	0.025	0.030	0.012	0.012	0.012	0.022	0.032	0.016	0.010
NBT	0.185	0.128	0.176	0.123	0.064	0.042	0.054	0.095	0.044	0.036
NBGCT	0.198	0.130	0.074	0.029	0.019	0.013	0.031	0.046	0.019	0.013
NBMCT	0.351	0.269	0.218	0.047	0.021	0.020	0.031	0.042	0.017	0.011
GGC	0.038	0.056	0.030	0.026	0.049	0.023	0.072	0.202	0.023	0.017
GMC	0.079	0.071	0.063	0.020	0.030	0.018	0.040	0.037	0.018	0.011
GT	0.299	0.138	0.338	0.062	0.042	0.038	0.038	0.048	0.027	0.019
GGCT	0.384	0.354	0.220	0.020	0.031	0.015	0.034	0.037	0.017	0.011
GMCT	0.274	0.215	0.134	0.021	0.018	0.017	0.048	0.043	0.019	0.012
LGC	0.263	0.160	0.106	0.026	0.022	0.016	0.050	0.093	0.020	0.012

Samples	Weeks									
	bk1	bk2	1	2	3	4	5	6	7	8
LMC	0.042	0.107	0.040	0.018	0.000	0.012	0.034	0.179	0.011	0.000
LT	0.298	0.149	0.179	0.067	0.035	0.031	0.027	0.036	0.021	0.017
LGCT	0.513	0.369	0.135	0.035	0.018	0.010	0.021	0.027	0.015	0.011
LMCT	0.331	0.426	0.316	0.019	0.015	0.000	0.024	0.026	0.014	0.011
WHO (2011)	2.000	2.000	2.000	2.000	2.000	2.000	2.000	2.000	2.000	2.000

Two-way Anova test for Cu concentrations in profile leachates

Tests of Between-Subjects Effects

Dependent Variable: Cu conc. in leachate

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	5.990[a]	19	.315	3.861	.000
Intercept	45.301	1	45.301	554.771	.000
medium	3.521	4	.880	10.781	**.000**
Aggregate	.223	3	.074	.911	.441
medium * Aggregate	2.246	12	.187	2.292	**.018**
Error	4.899	60	.082		
Total	56.190	80			
Corrected Total	10.890	79			

a. R Squared = .550 (Adjusted R Squared = .408)

Post Hoc test for Cu concentrations in profile leachate

Multiple Comparisons

Cu conc

LSD: Growth media

(I) medium	(J) medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	-.009562	.1010298	.925	-.211652	.192527
	MC	.148187	.1010298	.148	-.053902	.350277
	MCT	-.060188	.1010298	.554	-.262277	.141902
	T	-.475312*	.1010298	.000	-.677402	-.273223
GCT	GC	.009562	.1010298	.925	-.192527	.211652
	MC	.157750	.1010298	.124	-.044340	.359840
	MCT	-.050625	.1010298	.618	-.252715	.151465
	T	-.465750*	.1010298	.000	-.667840	-.263660
MC	GC	-.148187	.1010298	.148	-.350277	.053902
	GCT	-.157750	.1010298	.124	-.359840	.044340
	MCT	-.208375*	.1010298	.043	-.410465	-.006285
	T	-.623500*	.1010298	.000	-.825590	-.421410
MCT	GC	.060188	.1010298	.554	-.141902	.262277
	GCT	.050625	.1010298	.618	-.151465	.252715
	MC	.208375*	.1010298	.043	.006285	.410465
	T	-.415125*	.1010298	.000	-.617215	-.213035
T	GC	.475312*	.1010298	.000	.273223	.677402
	GCT	.465750*	.1010298	.000	.263660	.667840
	MC	.623500*	.1010298	.000	.421410	.825590
	MCT	.415125*	.1010298	.000	.213035	.617215

Based on observed means.

The error term is Mean Square (Error) = .082.

*.The mean difference is significant at the 0.05 level.

Multiple Comparisons

Cu conc.

LSD: Aggregates

(I) Aggregate	(J) Aggregate	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	.096100	.0903638	.292	-.084655	.276855
	NB	.050100	.0903638	.581	-.130655	.230855
	OB	-.044600	.0903638	.623	-.225355	.136155
L	G	-.096100	.0903638	.292	-.276855	.084655
	NB	-.046000	.0903638	.613	-.226755	.134755
	OB	-.140700	.0903638	.125	-.321455	.040055
NB	G	-.050100	.0903638	.581	-.230855	.130655
	L	.046000	.0903638	.613	-.134755	.226755
	OB	-.094700	.0903638	.299	-.275455	.086055
OB	G	.044600	.0903638	.623	-.136155	.225355
	L	.140700	.0903638	.125	-.040055	.321455
	NB	.094700	.0903638	.299	-.086055	.275455

Based on observed means.

The error term is Mean Square (Error) = .082.

Appendix 19: Results for Iron concentrations in profile leachate over an eight-week spiking period

Fe concentrations in profile leachates (mgL^{-1})

Samples	Weeks								
		1	2	3	4	5	6	7	8
OBGC1	0.170	0.218	0.231	0.100	0.534	0.064	0.487	1.121	0.050
OBGC2	0.099	0.296	1.107	0.191	0.076	0.099	0.467	0.526	0.026
OBGC3	0.137	0.142	0.600	0.448	0.076	0.057	0.441	1.010	0.055
OGBG4	0.233	0.815	3.893	0.092	0.103	0.052	0.775	1.197	0.039
OBMC1	0.076	0.114	0.052	0.017	0.009	0.008	0.042	0.024	0.008
OBMC2	0.144	0.367	0.379	0.022	0.016	0.015	0.087	0.065	0.012
OBMC3	0.141	0.520	0.636	0.034	0.017	0.015	0.220	0.178	0.016
OBMC4	0.059	0.221	0.183	0.066	0.019	0.030	0.116	0.100	0.014
OBT1	1.023	0.407	0.042	0.030	0.027	0.053	0.071	0.111	0.059
OBT2	0.094	0.148	0.488	0.306	0.236	0.145	0.453	0.290	0.044
OBT3	0.123	0.046	0.079	0.061	0.124	0.061	0.143	0.252	0.033
OBT4	0.118	0.049	0.062	0.078	0.244	0.091	0.104	0.375	0.028
OBGCT1	0.381	0.290	0.046	0.057	0.034	0.023	0.054	0.124	0.013
OBGCT2	0.443	0.184	0.101	0.042	0.040	0.018	0.152	0.276	0.015
OBGCT3	0.150	0.156	2.284	0.092	0.054	0.047	0.065	0.132	0.015
OBGCT4	0.228	0.227	0.206	0.018	0.028	0.039	0.144	0.372	0.067
OBMCT1	0.195	0.167	0.211	0.015	0.032	0.101	0.131	0.240	0.099
OBMCT2	0.223	0.170	0.060	0.026	0.012	0.015	0.052	0.061	0.005
OBMCT3	0.211	0.248	0.045	0.025	0.013	0.021	0.154	0.151	0.006
OBMCT4	0.232	0.230	0.020	0.031	0.016	0.011	0.019	0.047	0.014
NBGC1	0.117	0.766	1.959	0.300	0.058	0.056	0.642	0.959	0.043
NBGC2	0.064	0.228	0.772	0.059	0.029	0.034	0.289	0.477	0.018
NBGC3	0.059	0.584	1.158	0.089	0.032	0.026	0.365	0.554	0.022
NBGC4	0.189	0.148	1.169	0.268	0.042	0.031	0.311	0.523	0.023
NBMC1	0.166	0.422	0.537	0.077	0.027	0.033	0.276	0.524	0.018
NBMC2	0.195	0.624	0.239	0.048	0.018	0.023	0.171	0.196	0.007
NBMC3	0.104	0.149	0.132	0.040	0.007	0.013	0.051	0.047	0.008
NBMC4	0.073	0.358	0.224	0.060	0.014	0.016	0.178	0.235	0.013
NBT1	0.212	0.089	0.062	0.159	0.143	0.079	0.227	2.174	0.037
NBT2	0.152	0.034	0.016	0.121	0.086	0.074	0.093	0.275	0.030
NBT3	0.103	0.161	0.034	0.231	0.132	0.055	0.361	0.908	0.016
NBT4	0.077	0.121	0.024	0.065	0.099	0.056	0.086	1.099	0.011
NBGCT1	0.213	0.196	0.023	0.039	0.013	0.009	0.047	0.292	0.009
NBGCT2	0.229	0.160	0.032	0.043	0.028	0.016	0.139	0.507	0.015
NBGCT3	0.305	0.194	0.027	0.023	0.020	0.010	0.063	0.088	0.015
NBGCT4	0.219	0.252	0.118	0.090	0.024	0.018	0.053	0.211	0.012
NBMCT1	0.191	0.185	0.091	0.038	0.014	0.012	0.044	0.088	0.015
NBMCT2	0.139	0.170	0.278	0.046	0.016	0.013	0.037	0.027	0.023

Samples	Weeks								
		1	2	3	4	5	6	7	8
NBMCT3	0.222	0.192	0.088	0.061	0.020	0.012	0.103	0.141	0.011
NBMCT4	0.306	0.298	0.017	0.045	0.013	0.010	0.030	0.057	0.007
GGC1	0.211	0.972	0.935	0.126	0.286	0.072	1.930	2.048	0.063
GGC2	0.169	1.085	0.339	0.142	0.207	0.062	0.716	1.107	0.032
GGC3	0.192	1.151	0.519	0.123	0.114	0.043	0.847	1.562	0.060
GGC4	0.165	0.836	1.021	0.155	0.098	0.054	1.168	2.431	0.078
GMC1	0.161	1.107	0.754	0.116	0.026	0.037	0.323	0.525	0.012
GMC2	0.149	0.436	2.516	0.205	0.046	0.040	0.772	0.537	0.023
GMC3	0.130	0.595	0.481	0.084	0.066	0.023	0.726	0.423	0.023
GMC4	0.205	1.521	0.611	0.126	0.077	0.036	0.588	0.571	0.026
GT1	1.399	0.519	0.405	0.127	0.036	0.025	0.027	0.059	0.009
GT2	0.906	0.254	0.323	0.272	0.026	0.074	0.020	0.074	0.009
GT3	0.763	0.408	0.135	0.033	0.026	0.045	0.045	0.070	0.007
GT4	0.127	0.141	0.335	0.603	0.174	0.341	0.167	1.641	0.121
GGCT1	8.169	3.585	0.382	0.033	0.089	0.039	0.096	0.167	0.012
GGCT2	2.669	3.861	0.250	0.044	0.039	0.033	0.059	0.098	0.009
GGCT3	5.741	4.324	1.937	0.086	0.053	0.030	0.099	0.210	0.015
GGCT4	17.260	3.976	0.212	0.038	0.015	0.012	0.111	0.280	0.038
GMCT1	0.152	0.180	0.071	0.052	0.082	0.084	0.340	0.438	0.026
GMCT2	0.932	0.851	0.070	0.044	0.014	0.016	0.072	0.061	0.010
GMCT3	2.129	0.580	0.149	0.023	0.009	0.016	0.091	0.790	0.009
GMCT4	0.966	0.978	0.205	0.035	0.015	0.021	0.149	0.158	0.008
LGC1	0.909	1.973	2.996	0.218	0.135	0.070	0.780	0.921	0.047
LGC2	0.077	1.110	1.090	0.118	0.116	0.080	0.474	1.319	0.070
LGC3	1.184	1.750	1.047	0.118	0.086	0.063	0.564	1.504	0.055
LGC4	0.622	1.732	1.127	0.104	0.058	0.049	0.829	1.453	0.074
LMC1	0.292	1.394	0.191	0.093	0.024	0.029	0.243	0.260	0.013
LMC2	0.229	0.978	0.271	0.054	0.026	0.027	0.429	0.461	0.012
LMC3	0.391	0.693	0.372	0.124	0.037	0.056	0.521	1.013	0.022
LMC4	0.249	0.748	0.600	0.091	0.018	0.032	0.874	0.944	0.025
LT1	0.806	0.942	0.350	0.921	0.065	0.261	0.037	0.108	0.020
LT2	0.360	0.549	0.064	0.228	0.022	0.055	0.019	0.043	0.005
LT3	0.584	1.518	0.105	0.219	0.032	0.044	0.026	0.040	0.006
LT4	0.543	0.418	0.087	0.057	0.013	0.069	0.012	0.052	0.010
LGCT1	5.048	2.947	0.104	0.020	0.012	0.014	0.030	0.091	0.006
LGCT2	5.348	5.801	0.390	0.126	0.031	0.037	0.095	0.103	0.011
LGCT3	4.679	2.193	0.058	0.035	0.029	0.021	0.040	0.062	0.008
LGCT4	2.142	5.192	0.052	0.084	0.039	0.064	0.052	0.155	0.013
LMCT1	1.479	0.612	0.020	0.033	0.019	0.018	0.095	0.065	0.019
LMCT2	0.987	0.864	0.068	0.024	0.015	0.026	0.050	0.033	0.013
LMCT3	0.970	1.190	1.960	0.039	0.016	0.017	0.029	0.047	0.005
LMCT4	1.437	0.555	0.139	0.025	0.014	0.015	0.031	0.052	0.007

Standard error of Fe concentrations in profile leachates (mgL^{-1})

standard error	Weeks									
	bk	bk	1	2	3	4	5	6	7	8
OBGC	0.028	0.152	0.831	0.083	0.112	0.011	0.078	0.151	0.006	0.003
OBMC	0.022	0.088	0.127	0.011	0.002	0.005	0.038	0.033	0.002	0.001
OBT	0.228	0.085	0.107	0.063	0.051	0.021	0.088	0.055	0.007	0.143
OBGCT	0.068	0.029	0.543	0.015	0.006	0.007	0.026	0.060	0.013	0.010
OBMCT	0.008	0.021	0.043	0.003	0.005	0.021	0.032	0.045	0.023	0.041
NBGC	0.030	0.082	0.123	0.121	0.007	0.007	0.082	0.111	0.006	0.004
NBMC	0.028	0.098	0.088	0.008	0.004	0.004	0.046	0.100	0.003	0.003
NBT	0.030	0.027	0.010	0.035	0.013	0.006	0.065	0.395	0.006	0.173
NBGCT	0.021	0.019	0.023	0.014	0.003	0.002	0.021	0.088	0.001	0.009
NBMCT	0.035	0.029	0.056	0.005	0.002	0.001	0.017	0.024	0.003	0.012
GGC	0.011	0.069	0.164	0.007	0.044	0.006	0.272	0.288	0.010	0.006
GMC	0.016	0.248	0.478	0.026	0.011	0.004	0.101	0.032	0.003	0.001
GT	0.262	0.083	0.058	0.125	0.036	0.074	0.034	0.393	0.028	0.156
GGCT	3.142	0.153	0.416	0.012	0.015	0.006	0.011	0.038	0.007	0.006
GMCT	0.407	0.176	0.033	0.006	0.017	0.017	0.061	0.164	0.004	0.033
LGC	0.237	0.185	0.477	0.026	0.017	0.007	0.085	0.132	0.006	0.006
LMC	0.036	0.159	0.089	0.014	0.004	0.007	0.132	0.184	0.003	0.005
LT	0.092	0.247	0.067	0.192	0.011	0.052	0.005	0.016	0.003	0.050
LGCT	0.734	0.868	0.081	0.024	0.006	0.011	0.014	0.019	0.002	0.005
LMCT	0.139	0.145	0.472	0.004	0.001	0.002	0.015	0.007	0.003	0.017

Mean Fe concentrations in profile leachates (mgL^{-1})

Samples	Weeks									
	bk1	bk2	1	2	3	4	5	6	7	8
OBGC	0.160	0.368	1.458	0.208	0.197	0.068	0.543	0.964	0.043	0.050
OBMC	0.105	0.306	0.313	0.035	0.015	0.017	0.116	0.092	0.013	0.011
OBT	0.340	0.163	0.168	0.119	0.158	0.088	0.193	0.257	0.041	0.519
OBGCT	0.301	0.214	0.659	0.052	0.039	0.032	0.104	0.226	0.028	0.074
OBMCT	0.215	0.204	0.084	0.024	0.018	0.037	0.089	0.125	0.031	0.081
NBGC	0.107	0.432	1.265	0.179	0.040	0.037	0.402	0.628	0.027	0.017
NBMC	0.135	0.388	0.283	0.056	0.017	0.021	0.169	0.251	0.012	0.014
NBT	0.136	0.101	0.034	0.144	0.115	0.066	0.192	1.114	0.024	0.490
NBGCT	0.242	0.201	0.050	0.049	0.021	0.013	0.076	0.275	0.013	0.038
NBMCT	0.215	0.211	0.119	0.048	0.016	0.012	0.054	0.078	0.014	0.046
GGC	0.184	1.011	0.704	0.137	0.176	0.058	1.165	1.787	0.058	0.034
GMC	0.161	0.915	1.091	0.133	0.054	0.034	0.602	0.514	0.021	0.013
GT	0.799	0.331	0.300	0.259	0.066	0.121	0.065	0.461	0.037	0.235
GGCT	8.460	3.937	0.695	0.050	0.049	0.029	0.091	0.189	0.019	0.035
GMCT	1.045	0.647	0.124	0.039	0.030	0.034	0.163	0.362	0.013	0.074
LGC	0.698	1.641	1.565	0.140	0.099	0.066	0.662	1.299	0.062	0.039

Samples	Weeks									
	bk1	bk2	1	2	3	4	5	6	7	8
LMC	0.290	0.953	0.359	0.091	0.026	0.036	0.517	0.670	0.018	0.022
LT	0.573	0.857	0.152	0.356	0.033	0.107	0.024	0.061	0.010	0.095
LGCT	4.304	4.033	0.151	0.066	0.028	0.034	0.054	0.103	0.010	0.031
LMCT	1.218	0.805	0.547	0.030	0.016	0.019	0.051	0.049	0.011	0.037
WHO (2011) Potable water guideline	2.000	2.000	2.000	2.000	2.000	2.000	2.000	2.000	2.000	2.000

Two-way Anova test for Fe concentrations in profile leachates

Tests of Between-Subjects Effects

Dependent Variable: Fe conc. in leachate

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	105.938[a]	19	5.576	6.221	.000
Intercept	201.698	1	201.698	225.034	.000
Medium	85.603	4	21.401	23.877	**.000**
Aggregate	5.700	3	1.900	2.120	.107
Medium * Aggregate	14.634	12	1.220	1.361	.210
Error	53.778	60	.896		
Total	361.414	80			
Corrected Total	159.716	79			

a. R Squared = .663 (Adjusted R Squared = .557)

Post Hoc test for Fe concentrations in profile leachate

Multiple Comparisons

Fe conc.

LSD: Growth media

(I) Medium	(J) Medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	2.697688*	.3347205	.000	2.028147	3.367228
	MC	2.135000*	.3347205	.000	1.465459	2.804541
	MCT	2.924625*	.3347205	.000	2.255084	3.594166
	T	2.018188*	.3347205	.000	1.348647	2.687728
GCT	GC	-2.697688*	.3347205	.000	-3.367228	-2.028147
	MC	-.562688	.3347205	.098	-1.232228	.106853
	MCT	.226937	.3347205	.500	-.442603	.896478
	T	-.679500*	.3347205	.047	-1.349041	-.009959
MC	GC	-2.135000*	.3347205	.000	-2.804541	-1.465459
	GCT	.562688	.3347205	.098	-.106853	1.232228
	MCT	.789625*	.3347205	.022	.120084	1.459166
	T	-.116812	.3347205	.728	-.786353	.552728
MCT	GC	-2.924625*	.3347205	.000	-3.594166	-2.255084
	GCT	-.226937	.3347205	.500	-.896478	.442603
	MC	-.789625*	.3347205	.022	-1.459166	-.120084
	T	-.906438*	.3347205	.009	-1.575978	-.236897
T	GC	-2.018188*	.3347205	.000	-2.687728	-1.348647
	GCT	.679500*	.3347205	.047	.009959	1.349041
	MC	.116812	.3347205	.728	-.552728	.786353
	MCT	.906438*	.3347205	.009	.236897	1.575978

Based on observed means.

The error term is Mean Square (Error) = .896.

*.The mean difference is significant at the 0.05 level.

Fe conc.

LSD: Aggregates

(I) Aggregate	(J) Aggregate	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	.474900	.2993832	.118	-.123955	1.073755
	NB	.720900*	.2993832	.019	.122045	1.319755
	OB	.546650	.2993832	.073	-.052205	1.145505
L	G	-.474900	.2993832	.118	-1.073755	.123955
	NB	.246000	.2993832	.415	-.352855	.844855
	OB	.071750	.2993832	.811	-.527105	.670605
NB	G	-.720900*	.2993832	.019	-1.319755	-.122045
	L	-.246000	.2993832	.415	-.844855	.352855
	OB	-.174250	.2993832	.563	-.773105	.424605
OB	G	-.546650	.2993832	.073	-1.145505	.052205
	L	-.071750	.2993832	.811	-.670605	.527105
	NB	.174250	.2993832	.563	-.424605	.773105

Based on observed means.

The error term is Mean Square (Error) = .896

*.The mean difference is significant at the 0.05 level.

Appendix 20: Results for Manganese concentrations in profile leachate over an eight-week spiking period

Mn concentrations in profile leachates (mgL^{-1})

Samples	Weeks									
	bk	bk	1	2	3	4	5	6	7	8
OBGC1	0.035	0.006	0.045	0.049	0.517	0.042	0.014	0.046	0.008	0.015
OBGC2	0.043	0.016	0.092	0.027	0.027	0.017	0.011	0.018	0.004	0.005
OBGC3	0.030	0.005	0.031	0.078	0.015	0.013	0.005	0.022	0.004	0.003
OGBG4	0.038	0.018	0.234	0.023	0.017	0.015	0.012	0.036	0.005	0.003
OBMC1	0.061	0.004	0.007	0.010	0.003	0.005	0.000	0.002	0.005	0.003
OBMC2	0.047	0.004	0.265	0.012	0.002	0.003	0.001	0.002	0.003	0.002
OBMC3	0.104	0.009	0.036	0.009	0.004	0.004	0.003	0.005	0.003	0.002
OBMC4	0.069	0.005	0.051	0.007	0.007	0.003	0.005	0.007	0.003	0.003
OBT1	0.018	0.007	0.005	0.003	0.003	0.002	0.001	0.004	0.003	0.003
OBT2	0.083	0.028	0.008	0.014	0.005	0.004	0.004	0.005	0.002	0.007
OBT3	0.115	0.055	0.002	0.002	0.003	0.003	0.000	0.002	0.002	0.004
OBT4	0.069	0.036	0.015	0.004	0.006	0.003	0.000	0.005	0.001	0.007
OBGCT1	0.114	0.021	0.005	0.019	0.007	0.005	0.000	0.003	0.002	0.003
OBGCT2	0.081	0.016	0.007	0.004	0.003	0.003	0.000	0.004	0.001	0.002
OBGCT3	0.046	0.019	0.049	0.009	0.005	0.005	0.000	0.002	0.001	0.001
OBGCT4	0.042	0.022	0.004	0.003	0.002	0.003	0.001	0.006	0.002	0.002
OBMCT1	0.124	0.017	0.023	0.006	0.004	0.005	0.000	0.006	0.003	0.003
OBMCT2	0.096	0.008	0.007	0.003	0.007	0.005	0.000	0.007	0.001	0.001
OBMCT3	0.059	0.014	0.005	0.003	0.003	0.004	0.000	0.004	0.001	0.001
OBMCT4	0.095	0.009	0.007	0.005	0.003	0.003	0.000	0.002	0.001	0.001
NBGC1	0.027	0.015	0.137	0.076	0.030	0.042	0.038	0.051	0.014	0.017
NBGC2	0.024	0.008	0.054	0.028	0.013	0.015	0.013	0.020	0.006	0.005
NBGC3	0.018	0.013	1.094	0.026	0.009	0.012	0.007	0.013	0.004	0.003
NBGC4	0.051	0.060	0.074	0.060	0.013	0.011	0.007	0.015	0.004	0.003
NBMC1	0.030	0.013	0.048	0.025	0.009	0.008	0.007	0.009	0.003	0.002
NBMC2	0.041	0.010	0.028	0.017	0.006	0.008	0.005	0.009	0.002	0.002
NBMC3	0.044	0.002	0.011	0.006	0.003	0.004	0.002	0.003	0.002	0.002
NBMC4	0.029	0.006	0.021	0.015	0.005	0.005	0.008	0.016	0.002	0.002
NBT1	0.091	0.014	0.007	0.004	0.004	0.004	0.002	0.016	0.002	0.007
NBT2	0.109	0.007	0.003	0.004	0.003	0.003	0.002	0.003	0.002	0.006
NBT3	0.061	0.028	0.004	0.007	0.008	0.008	0.003	0.007	0.002	0.002
NBT4	0.035	0.009	0.005	0.002	0.004	0.003	0.002	0.008	0.002	0.005
NBGCT1	0.041	0.012	0.005	0.008	0.002	0.003	0.001	0.004	0.001	0.002
NBGCT2	0.062	0.030	0.008	0.004	0.003	0.004	0.003	0.008	0.001	0.002
NBGCT3	0.045	0.018	0.007	0.004	0.006	0.006	0.001	0.002	0.001	0.001
NBGCT4	0.030	0.016	0.011	0.007	0.005	0.004	0.002	0.006	0.001	0.002
NBMCT1	0.117	0.009	0.008	0.002	0.002	0.002	0.001	0.003	0.001	0.001
NBMCT2	0.074	0.022	0.087	0.006	0.008	0.007	0.002	0.003	0.002	0.002

Samples	Weeks									
	bk	bk	1	2	3	4	5	6	7	8
NBMCT3	0.036	0.020	0.010	0.003	0.007	0.005	0.004	0.005	0.001	0.001
NBMCT4	0.068	0.016	0.004	0.003	0.005	0.003	0.002	0.002	0.001	0.002
GGC1	0.017	0.026	0.032	0.053	0.062	0.045	0.029	0.094	0.007	0.013
GGC2	0.022	0.019	0.013	0.030	0.036	0.016	0.010	0.023	0.003	0.004
GGC3	0.033	0.020	0.042	0.025	0.022	0.010	0.010	0.037	0.003	0.003
GGC4	0.023	0.020	0.038	0.023	0.008	0.007	0.021	0.048	0.006	0.004
GMC1	0.041	0.014	0.095	0.027	0.017	0.020	0.009	0.022	0.002	0.003
GMC2	0.022	0.008	0.182	0.066	0.031	0.023	0.012	0.012	0.002	0.004
GMC3	0.065	0.012	0.059	0.024	0.036	0.012	0.021	0.013	0.003	0.003
GMC4	0.038	0.015	0.050	0.033	0.052	0.017	0.014	0.015	0.001	0.002
GT1	0.030	0.007	0.013	0.004	0.008	0.003	0.002	0.002	0.001	0.002
GT2	0.063	0.016	0.020	0.011	0.005	0.004	0.002	0.002	0.001	0.002
GT3	0.030	0.014	0.033	0.007	0.004	0.003	0.002	0.002	0.001	0.002
GT4	0.078	0.040	0.018	0.015	0.007	0.010	0.003	0.014	0.002	0.007
GGCT1	0.150	0.059	0.027	0.011	0.013	0.006	0.002	0.004	0.001	0.002
GGCT2	0.121	0.076	0.029	0.015	0.013	0.006	0.002	0.003	0.001	0.001
GGCT3	0.056	0.053	1.079	0.020	0.006	0.003	0.001	0.005	0.001	0.001
GGCT4	0.188	0.048	0.012	0.007	0.003	0.002	0.002	0.004	0.002	0.002
GMCT1	0.088	0.210	0.007	0.003	0.004	0.004	0.004	0.006	0.001	0.004
GMCT2	0.075	0.055	0.004	0.002	0.002	0.001	0.001	0.002	0.001	0.001
GMCT3	0.180	0.065	0.008	0.003	0.002	0.003	0.002	0.003	0.001	0.002
GMCT4	0.094	0.052	0.011	0.001	0.003	0.003	0.002	0.003	0.001	0.001
LGC1	0.041	0.037	0.181	0.059	0.052	0.032	0.033	0.060	0.014	0.014
LGC2	0.038	0.025	0.060	0.027	0.021	0.016	0.011	0.027	0.005	0.005
LGC3	0.061	0.030	0.035	0.022	0.010	0.010	0.009	0.032	0.003	0.003
LGC4	0.041	0.021	0.029	0.011	0.009	0.009	0.015	0.027	0.003	0.004
LMC1	0.020	0.014	0.062	0.007	0.006	0.004	0.008	0.010	0.002	0.002
LMC2	0.029	0.011	0.013	0.005	0.004	0.005	0.013	0.015	0.002	0.002
LMC3	0.037	0.009	0.024	0.011	0.003	0.004	0.023	0.044	0.002	0.004
LMC4	0.020	0.014	0.029	0.009	0.004	0.004	0.033	0.034	0.003	0.003
LT1	0.018	0.008	0.002	0.009	0.002	0.003	0.001	0.002	0.001	0.003
LT2	0.025	0.006	0.000	0.003	0.003	0.002	0.001	0.001	0.001	0.002
LT3	0.020	0.020	0.000	0.003	0.003	0.002	0.002	0.002	0.001	0.002
LT4	0.017	0.013	0.003	0.001	0.003	0.002	0.001	0.003	0.001	0.001
LGCT1	0.068	0.034	0.001	0.005	0.003	0.003	0.001	0.002	0.001	0.002
LGCT2	0.183	0.081	0.003	0.016	0.003	0.002	0.001	0.003	0.001	0.002
LGCT3	0.119	0.041	0.002	0.002	0.004	0.002	0.001	0.003	0.001	0.002
LGCT4	0.071	0.101	0.000	0.003	0.004	0.002	0.002	0.004	0.001	0.002
LMCT1	0.150	0.075	0.000	0.001	0.002	0.001	0.001	0.002	0.001	0.001
LMCT2	0.055	0.065	0.000	0.001	0.001	0.001	0.001	0.001	0.001	0.002
LMCT3	0.071	0.064	1.729	0.015	0.002	0.003	0.001	0.002	0.001	0.001
LMCT4	0.086	0.056	0.006	0.003	0.003	0.001	0.001	0.002	0.001	0.001

Standard error of Mn concentrations in profile leachates (mgL^{-1})

standard error	Weeks									
	bk	bk	1	2	3	4	5	6	7	8
OBGC	0.003	0.003	0.046	0.013	0.124	0.007	0.002	0.006	0.001	0.003
OBMC	0.012	0.001	0.059	0.001	0.001	0.000	0.001	0.001	0.001	0.000
OBT	0.020	0.010	0.003	0.003	0.001	0.000	0.001	0.001	0.000	0.001
OBGCT	0.017	0.001	0.011	0.004	0.001	0.001	0.000	0.001	0.000	0.000
OBMCT	0.013	0.002	0.004	0.001	0.001	0.000	0.000	0.001	0.001	0.001
NBGC	0.007	0.012	0.252	0.012	0.005	0.007	0.007	0.009	0.002	0.003
NBMC	0.004	0.002	0.008	0.004	0.001	0.001	0.001	0.003	0.000	0.000
NBT	0.016	0.005	0.001	0.001	0.001	0.001	0.000	0.003	0.000	0.001
NBGCT	0.007	0.004	0.001	0.001	0.001	0.001	0.000	0.001	0.000	0.000
NBMCT	0.017	0.003	0.020	0.001	0.001	0.001	0.001	0.001	0.000	0.000
GGC	0.003	0.002	0.006	0.007	0.012	0.009	0.005	0.015	0.001	0.002
GMC	0.009	0.002	0.030	0.010	0.007	0.002	0.003	0.002	0.000	0.000
GT	0.012	0.007	0.004	0.002	0.001	0.002	0.000	0.003	0.000	0.001
GGCT	0.028	0.006	0.264	0.003	0.003	0.001	0.000	0.000	0.000	0.000
GMCT	0.024	0.038	0.001	0.000	0.000	0.001	0.001	0.001	0.000	0.001
LGC	0.005	0.003	0.036	0.010	0.010	0.005	0.005	0.008	0.003	0.003
LMC	0.004	0.001	0.011	0.001	0.001	0.000	0.006	0.008	0.000	0.000
LT	0.002	0.003	0.001	0.002	0.000	0.000	0.000	0.000	0.000	0.000
LGCT	0.027	0.016	0.001	0.003	0.000	0.000	0.000	0.000	0.000	0.000
LMCT	0.021	0.004	0.432	0.003	0.000	0.001	0.000	0.000	0.000	0.000

Mean Mn concentrations in profile leachates (mgL^{-1})

Samples		Weeks								
	bk1	bk2	1	2	3	4	5	6	7	8
OBGC	0.037	0.011	0.101	0.044	0.144	0.022	0.011	0.031	0.005	0.007
OBMC	0.070	0.006	0.090	0.010	0.004	0.004	0.002	0.004	0.004	0.003
OBT	0.071	0.032	0.008	0.006	0.004	0.003	0.000	0.004	0.002	0.005
OBGCT	0.071	0.020	0.016	0.009	0.004	0.004	0.000	0.004	0.002	0.002
OBMCT	0.094	0.012	0.011	0.004	0.004	0.004	0.000	0.005	0.002	0.002
NBGC	0.030	0.024	0.340	0.048	0.016	0.020	0.016	0.025	0.007	0.007
NBMC	0.036	0.008	0.027	0.016	0.006	0.006	0.006	0.009	0.002	0.002
NBT	0.074	0.015	0.005	0.004	0.005	0.005	0.002	0.009	0.002	0.005
NBGCT	0.045	0.019	0.008	0.006	0.004	0.004	0.002	0.005	0.000	0.002
NBMCT	0.074	0.017	0.027	0.004	0.006	0.004	0.002	0.003	0.000	0.002
GGC	0.024	0.021	0.031	0.033	0.032	0.020	0.018	0.051	0.005	0.006
GMC	0.042	0.012	0.097	0.038	0.034	0.018	0.014	0.016	0.002	0.003
GT	0.050	0.019	0.021	0.009	0.006	0.005	0.002	0.005	0.000	0.003
GGCT	0.129	0.059	0.287	0.013	0.009	0.004	0.002	0.004	0.000	0.002
GMCT	0.109	0.096	0.008	0.002	0.003	0.003	0.002	0.004	0.000	0.002
LGC	0.045	0.028	0.076	0.030	0.023	0.017	0.017	0.037	0.006	0.007
LMC	0.027	0.012	0.032	0.008	0.004	0.004	0.019	0.026	0.002	0.003

Samples		Weeks								
	bk1	bk2	1	2	3	4	5	6	7	8
LT	0.020	0.012	0.000	0.004	0.003	0.002	0.000	0.002	0.000	0.002
LGCT	0.110	0.064	0.002	0.007	0.004	0.002	0.000	0.003	0.000	0.002
LMCT	0.091	0.065	0.434	0.000	0.002	0.002	0.000	0.002	0.000	0.000
WHO (2011) Potable water guideline	0.400	0.400	0.400	0.400	0.400	0.400	0.400	0.400	0.400	0.400

Two-way Anova test for Mn concentrations in profile leachates

Tests of Between-Subjects Effects

Dependent Variable: Mn conc. in leachate

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	1.709[a]	19	.090	1.295	.221
Intercept	1.640	1	1.640	23.620	.000
Medium	.674	4	.168	2.425	.058
Aggregate	.028	3	.009	.133	.940
Medium * Aggregate	1.008	12	.084	1.209	.298
Error	4.167	60	.069		
Total	7.517	80			
Corrected Total	5.877	79			

a. R Squared = .291 (Adjusted R Squared = .066)

Post Hoc test for Mn concentrations in profile leachate

Multiple Comparisons

Mn conc.

LSD: Growth media

(I) Medium	(J) Medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	.208375*	.0931768	.029	.021994	.394756
	MC	.184000	.0931768	.053	-.002381	.370381
	MCT	.174250	.0931768	.066	-.012131	.360631
	T	.277375*	.0931768	.004	.090994	.463756
GCT	GC	-.208375*	.0931768	.029	-.394756	-.021994
	MC	-.024375	.0931768	.795	-.210756	.162006
	MCT	-.034125	.0931768	.715	-.220506	.152256
	T	.069000	.0931768	.462	-.117381	.255381
MC	GC	-.184000	.0931768	.053	-.370381	.002381
	GCT	.024375	.0931768	.795	-.162006	.210756
	MCT	-.009750	.0931768	.917	-.196131	.176631
	T	.093375	.0931768	.320	-.093006	.279756
MCT	GC	-.174250	.0931768	.066	-.360631	.012131
	GCT	.034125	.0931768	.715	-.152256	.220506
	MC	.009750	.0931768	.917	-.176631	.196131
	T	.103125	.0931768	.273	-.083256	.289506
T	GC	-.277375*	.0931768	.004	-.463756	-.090994
	GCT	-.069000	.0931768	.462	-.255381	.117381
	MC	-.093375	.0931768	.320	-.279756	.093006
	MCT	-.103125	.0931768	.273	-.289506	.083256

Based on observed means.

The error term is Mean Square (Error) = .069.

*.The mean difference is significant at the 0.05 level.

Mn conc
LSD: Aggregates

(I) Aggregate	(J) Aggregate	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	.003600	.0833398	.966	-.163104	.170304
	NB	.029000	.0833398	.729	-.137704	.195704
	OB	.045200	.0833398	.590	-.121504	.211904
L	G	-.003600	.0833398	.966	-.170304	.163104
	NB	.025400	.0833398	.762	-.141304	.192104
	OB	.041600	.0833398	.619	-.125104	.208304
NB	G	-.029000	.0833398	.729	-.195704	.137704
	L	-.025400	.0833398	.762	-.192104	.141304
	OB	.016200	.0833398	.847	-.150504	.182904
OB	G	-.045200	.0833398	.590	-.211904	.121504
	L	-.041600	.0833398	.619	-.208304	.125104
	NB	-.016200	.0833398	.847	-.182904	.150504

Based on observed means. The error term is Mean Square (Error) = .069.

Appendix 21: Results for Nickel concentrations in profile leachate over an eight-week spiking period

Ni concentrations in profile leachates (mgL^{-1})

Samples	Weeks									
	bk1	bk2	1	2	3	4	5	6	7	8
OBGC1	0.005	0.003	0.029	0.031	0.408	0.026	0.008	0.027	0.007	0.013
OBGC2	0.003	0.003	0.025	0.011	0.014	0.005	0.006	0.009	0.004	0.004
OBGC3	0.006	0.001	0.011	0.012	0.003	0.003	0.005	0.011	0.003	0.003
OGBG4	0.006	0.008	0.029	0.003	0.004	0.002	0.012	0.012	0.004	0.002
OBMC1	0.004	0.003	0.005	0.004	0.002	0.001	0.000	0.002	0.003	0.002
OBMC2	0.006	0.009	0.118	0.007	0.000	0.000	0.004	0.005	0.003	0.000
OBMC3	0.009	0.013	0.035	0.002	0.002	0.003	0.006	0.007	0.003	0.001
OBMC4	0.005	0.005	0.049	0.019	0.004	0.005	0.004	0.008	0.003	0.001
OBT1	0.020	0.014	0.004	0.003	0.002	0.001	0.005	0.005	0.003	0.000
OBT2	0.018	0.014	0.047	0.032	0.006	0.004	0.007	0.009	0.003	0.004
OBT3	0.026	0.023	0.010	0.004	0.002	0.002	0.002	0.005	0.003	0.003
OBT4	0.031	0.032	0.131	0.021	0.007	0.008	0.005	0.012	0.004	0.003
OBGCT1	0.028	0.011	0.005	0.007	0.003	0.002	0.001	0.002	0.001	0.003
OBGCT2	0.021	0.010	0.006	0.003	0.001	0.001	0.001	0.004	0.002	0.000
OBGCT3	0.011	0.013	0.094	0.004	0.001	0.000	0.000	0.002	0.002	0.001
OBGCT4	0.014	0.010	0.007	0.000	0.002	0.001	0.008	0.009	0.002	0.001
OBMCT1	0.021	0.019	0.063	0.001	0.001	0.001	0.009	0.010	0.003	0.002
OBMCT2	0.021	0.020	0.013	0.003	0.002	0.001	0.006	0.004	0.001	0.001
OBMCT3	0.014	0.037	0.005	0.002	0.002	0.000	0.000	0.004	0.001	0.001
OBMCT4	0.029	0.032	0.014	0.014	0.003	0.002	0.003	0.006	0.001	0.001
NBGC1	0.004	0.009	0.015	0.010	0.013	0.022	0.042	0.039	0.011	0.016
NBGC2	0.003	0.001	0.013	0.002	0.003	0.007	0.013	0.013	0.003	0.002
NBGC3	0.004	0.008	0.240	0.011	0.004	0.003	0.012	0.010	0.001	0.003
NBGC4	0.008	0.003	0.011	0.008	0.001	0.004	0.007	0.011	0.001	0.003
NBMC1	0.006	0.005	0.013	0.003	0.002	0.004	0.008	0.012	0.002	0.003
NBMC2	0.005	0.014	0.008	0.005	0.000	0.003	0.009	0.007	0.001	0.002
NBMC3	0.006	0.007	0.045	0.011	0.000	0.004	0.003	0.004	0.001	0.003
NBMC4	0.004	0.006	0.007	0.003	0.001	0.003	0.010	0.009	0.000	0.002
NBT1	0.015	0.009	0.032	0.011	0.007	0.007	0.006	0.012	0.002	0.006
NBT2	0.023	0.021	0.016	0.029	0.003	0.006	0.005	0.006	0.002	0.005
NBT3	0.020	0.017	0.018	0.010	0.004	0.004	0.007	0.012	0.002	0.004
NBT4	0.016	0.018	0.027	0.009	0.003	0.005	0.006	0.009	0.002	0.005
NBGCT1	0.016	0.012	0.002	0.005	0.001	0.002	0.004	0.005	0.000	0.001
NBGCT2	0.017	0.017	0.009	0.005	0.001	0.003	0.006	0.007	0.001	0.003
NBGCT3	0.016	0.015	0.007	0.005	0.001	0.001	0.005	0.005	0.000	0.002
NBGCT4	0.016	0.015	0.032	0.005	0.002	0.002	0.008	0.009	0.001	0.003
NBMCT1	0.035	0.019	0.050	0.005	0.001	0.002	0.004	0.005	0.000	0.001
NBMCT2	0.020	0.024	0.177	0.020	0.002	0.006	0.004	0.003	0.001	0.002

Samples	Weeks									
	bk1	bk2	1	2	3	4	5	6	7	8
NBMCT3	0.020	0.019	0.038	0.006	0.001	0.002	0.008	0.008	0.001	0.002
NBMCT4	0.031	0.043	0.005	0.004	0.001	0.003	0.006	0.005	0.001	0.002
GGC1	0.006	0.006	0.005	0.029	0.022	0.024	0.026	0.036	0.005	0.013
GGC2	0.004	0.007	0.004	0.013	0.008	0.007	0.007	0.014	0.001	0.004
GGC3	0.006	0.009	0.006	0.006	0.004	0.003	0.013	0.015	0.001	0.003
GGC4	0.004	0.008	0.007	0.005	0.002	0.003	0.010	0.023	0.002	0.002
GMC1	0.008	0.017	0.014	0.004	0.001	0.002	0.007	0.009	0.001	0.002
GMC2	0.005	0.011	0.029	0.007	0.003	0.002	0.015	0.012	0.001	0.003
GMC3	0.007	0.012	0.020	0.006	0.005	0.001	0.014	0.008	0.000	0.001
GMC4	0.006	0.020	0.008	0.003	0.004	0.002	0.015	0.011	0.001	0.002
GT1	0.021	0.012	0.049	0.007	0.004	0.003	0.004	0.003	0.000	0.001
GT2	0.048	0.015	0.036	0.004	0.002	0.003	0.002	0.004	0.000	0.003
GT3	0.048	0.024	0.018	0.002	0.001	0.001	0.006	0.002	0.001	0.002
GT4	0.090	0.025	0.033	0.012	0.004	0.006	0.010	0.010	0.001	0.004
GGCT1	0.046	0.034	0.028	0.001	0.003	0.000	0.007	0.004	0.000	0.002
GGCT2	0.043	0.054	0.019	0.002	0.003	0.002	0.006	0.007	0.000	0.001
GGCT3	0.035	0.024	0.418	0.010	0.003	0.000	0.005	0.004	0.000	0.001
GGCT4	0.057	0.021	0.016	0.001	0.000	0.000	0.006	0.005	0.000	0.001
GMCT1	0.012	0.014	0.007	0.001	0.003	0.001	0.009	0.006	0.000	0.002
GMCT2	0.044	0.046	0.008	0.002	0.000	0.001	0.006	0.003	0.001	0.001
GMCT3	0.103	0.039	0.029	0.002	0.001	0.002	0.005	0.003	0.000	0.003
GMCT4	0.043	0.037	0.046	0.001	0.001	0.001	0.009	0.005	0.000	0.001
LGC1	0.007	0.011	0.056	0.032	0.030	0.019	0.025	0.031	0.009	0.009
LGC2	0.018	0.023	0.015	0.011	0.004	0.003	0.010	0.019	0.002	0.001
LGC3	0.014	0.013	0.011	0.007	0.003	0.003	0.010	0.021	0.003	0.002
LGC4	0.007	0.009	0.010	0.005	0.001	0.002	0.010	0.016	0.002	0.001
LMC1	0.003	0.027	0.067	0.016	0.003	0.003	0.007	0.011	0.002	0.000
LMC2	0.001	0.028	0.010	0.005	0.001	0.001	0.009	0.012	0.001	0.001
LMC3	0.007	0.016	0.013	0.006	0.001	0.002	0.010	0.018	0.003	0.001
LMC4	0.005	0.010	0.006	0.004	0.001	0.002	0.012	0.013	0.001	0.000
LT1	0.021	0.008	0.009	0.018	0.006	0.005	0.003	0.005	0.003	0.001
LT2	0.022	0.007	0.008	0.003	0.002	0.002	0.003	0.003	0.002	0.001
LT3	0.026	0.017	0.008	0.003	0.002	0.001	0.004	0.004	0.002	0.002
LT4	0.020	0.021	0.037	0.003	0.001	0.003	0.003	0.005	0.001	0.000
LGCT1	0.016	0.016	0.010	0.001	0.000	0.001	0.002	0.003	0.001	0.001
LGCT2	0.070	0.038	0.034	0.006	0.001	0.002	0.003	0.005	0.001	0.000
LGCT3	0.037	0.035	0.005	0.001	0.002	0.001	0.002	0.003	0.001	0.001
LGCT4	0.032	0.035	0.000	0.003	0.003	0.000	0.006	0.003	0.000	0.000
LMCT1	0.084	0.072	0.000	0.002	0.002	0.001	0.005	0.003	0.002	0.000
LMCT2	0.028	0.071	0.011	0.004	0.001	0.002	0.002	0.002	0.000	0.000
LMCT3	0.024	0.057	1.038	0.008	0.001	0.001	0.040	0.003	0.000	0.000
LMCT4	0.050	0.046	0.026	0.003	0.002	0.000	0.006	0.004	0.001	0.000

Standard error of Ni concentrations in profile leachates (mgL^{-1})

standard error	Weeks									
	bk	bk	1	2	3	4	5	6	7	8
OBGC	0.001	0.001	0.004	0.006	0.100	0.006	0.002	0.004	0.001	0.003
OBMC	0.001	0.002	0.024	0.004	0.001	0.001	0.001	0.001	0.000	0.000
OBT	0.003	0.004	0.029	0.007	0.001	0.002	0.001	0.002	0.000	0.001
OBGCT	0.004	0.001	0.022	0.001	0.000	0.000	0.002	0.002	0.000	0.001
OBMCT	0.003	0.004	0.013	0.003	0.000	0.000	0.002	0.001	0.001	0.000
NBGC	0.001	0.002	0.057	0.002	0.003	0.004	0.008	0.007	0.002	0.003
NBMC	0.000	0.002	0.009	0.002	0.000	0.000	0.002	0.002	0.000	0.000
NBT	0.002	0.003	0.004	0.005	0.001	0.001	0.000	0.001	0.000	0.000
NBGCT	0.000	0.001	0.007	0.000	0.000	0.000	0.001	0.001	0.000	0.000
NBMCT	0.004	0.006	0.038	0.004	0.000	0.001	0.001	0.001	0.000	0.000
GGC	0.001	0.001	0.001	0.006	0.005	0.005	0.004	0.005	0.001	0.003
GMC	0.001	0.002	0.004	0.001	0.001	0.000	0.002	0.001	0.000	0.000
GT	0.014	0.003	0.006	0.002	0.001	0.001	0.002	0.002	0.000	0.001
GGCT	0.005	0.007	0.099	0.002	0.001	0.001	0.000	0.001	0.000	0.000
GMCT	0.019	0.007	0.009	0.000	0.001	0.000	0.001	0.001	0.000	0.000
LGC	0.003	0.003	0.011	0.006	0.007	0.004	0.004	0.003	0.002	0.002
LMC	0.001	0.004	0.014	0.003	0.001	0.000	0.001	0.002	0.000	0.000
LT	0.001	0.003	0.007	0.004	0.001	0.001	0.000	0.000	0.000	0.000
LGCT	0.011	0.005	0.008	0.001	0.001	0.000	0.001	0.001	0.000	0.000
LMCT	0.014	0.006	0.256	0.001	0.000	0.000	0.009	0.000	0.000	0.000

Mean Ni concentrations in profile leachates (mgL^{-1})

Samples	Weeks									
	bk1	bk2	1	2	3	4	5	6	7	8
OBGC	0.000	0.000	0.024	0.000	0.107	0.000	0.000	0.015	0.000	0.000
OBMC	0.000	0.000	0.052	0.000	0.000	0.000	0.000	0.000	0.000	0.000
OBT	0.024	0.021	0.048	0.015	0.000	0.000	0.000	0.000	0.000	0.000
OBGCT	0.019	0.000	0.028	0.000	0.000	0.000	0.000	0.000	0.000	0.000
OBMCT	0.021	0.027	0.024	0.000	0.000	0.000	0.000	0.000	0.000	0.000
NBGC	0.000	0.000	0.070	0.000	0.000	0.000	0.019	0.018	0.000	0.000
NBMC	0.000	0.000	0.018	0.000	0.000	0.000	0.000	0.000	0.000	0.000
NBT	0.019	0.016	0.023	0.015	0.000	0.000	0.000	0.000	0.000	0.000
NBGCT	0.016	0.015	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
NBMCT	0.027	0.026	0.068	0.000	0.000	0.000	0.000	0.000	0.000	0.000
GGC	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.022	0.000	0.000
GMC	0.000	0.015	0.018	0.000	0.000	0.000	0.000	0.000	0.000	0.000
GT	0.052	0.019	0.034	0.000	0.000	0.000	0.000	0.000	0.000	0.000
GGCT	0.045	0.033	0.120	0.000	0.000	0.000	0.000	0.000	0.000	0.000
GMCT	0.051	0.034	0.023	0.000	0.000	0.000	0.000	0.000	0.000	0.000
LGC	0.000	0.000	0.023	0.000	0.000	0.000	0.000	0.022	0.000	0.000
LMC	0.000	0.020	0.024	0.000	0.000	0.000	0.000	0.000	0.000	0.000

Samples	Weeks									
	bk1	bk2	1	2	3	4	5	6	7	8
LT	0.022	0.000	0.016	0.000	0.000	0.000	0.000	0.000	0.000	0.000
LGCT	0.039	0.031	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
LMCT	0.047	0.062	0.269	0.000	0.000	0.000	0.000	0.000	0.000	0.000
WHO (2011)	0.070	0.070	0.070	0.070	0.070	0.070	0.070	0.070	0.070	0.070

Two-way Anova test for Ni concentrations in profile leachates

Tests of Between-Subjects Effects

Dependent Variable: Ni conc in leachate

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.307[a]	19	.016	.760	.742
Intercept	.586	1	.586	27.560	.000
Medium	.070	4	.017	.820	.517
Aggregate	.010	3	.003	.153	.927
Medium * Aggregate	.227	12	.019	.891	.561
Error	1.277	60	.021		
Total	2.170	80			
Corrected Total	1.584	79			

a. R Squared = .194 (Adjusted R Squared = -.061)

Post Hoc test for Ni concentrations in profile leachate

Multiple Comparisons

Ni conc

LSD: Growth media

(I) Medium	(J) Medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	.064375	.0515697	.217	-.038780	.167530
	MC	.065937	.0515697	.206	-.037217	.169092
	MCT	.007500	.0515697	.885	-.095655	.110655
	T	.061000	.0515697	.242	-.042155	.164155
GCT	GC	-.064375	.0515697	.217	-.167530	.038780
	MC	.001563	.0515697	.976	-.101592	.104717
	MCT	-.056875	.0515697	.274	-.160030	.046280
	T	-.003375	.0515697	.948	-.106530	.099780
MC	GC	-.065937	.0515697	.206	-.169092	.037217
	GCT	-.001563	.0515697	.976	-.104717	.101592
	MCT	-.058437	.0515697	.262	-.161592	.044717
	T	-.004937	.0515697	.924	-.108092	.098217
MCT	GC	-.007500	.0515697	.885	-.110655	.095655
	GCT	.056875	.0515697	.274	-.046280	.160030
	MC	.058437	.0515697	.262	-.044717	.161592
	T	.053500	.0515697	.304	-.049655	.156655
T	GC	-.061000	.0515697	.242	-.164155	.042155
	GCT	.003375	.0515697	.948	-.099780	.106530
	MC	.004937	.0515697	.924	-.098217	.108092
	MCT	-.053500	.0515697	.304	-.156655	.049655

Based on observed means.

The error term is Mean Square (Error) = .021.

Ni conc
LSD: Aggregates

(I) Aggregate	(J) Aggregate	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	-.028050	.0461253	.545	-.120314	.064214
	NB	-.002700	.0461253	.954	-.094964	.089564
	OB	-.013900	.0461253	.764	-.106164	.078364
L	G	.028050	.0461253	.545	-.064214	.120314
	NB	.025350	.0461253	.585	-.066914	.117614
	OB	.014150	.0461253	.760	-.078114	.106414
NB	G	.002700	.0461253	.954	-.089564	.094964
	L	-.025350	.0461253	.585	-.117614	.066914
	OB	-.011200	.0461253	.809	-.103464	.081064
OB	G	.013900	.0461253	.764	-.078364	.106164
	L	-.014150	.0461253	.760	-.106414	.078114
	NB	.011200	.0461253	.809	-.081064	.103464

Based on observed means.
The error term is Mean Square (Error) = .021.

Appendix 22: Results for Lead concentrations in profile leachate over an eight-week spiking period

Pb concentrations in profile leachates (mgL^{-1})

Samples	Weeks									
	bk1	bk2	1	2	3	4	5	6	7	8
OBGC1	0.044	0.021	0.042	0.029	0.369	0.035	0.018	0.050	0.011	0.015
OBGC2	0.024	0.009	0.057	0.009	0.014	0.017	0.011	0.012	0.003	0.004
OBGC3	0.021	0.009	0.027	0.009	0.006	0.009	0.011	0.017	0.012	0.004
OGBG4	0.021	0.021	0.080	0.000	0.003	0.013	0.019	0.019	0.009	0.003
OBMC1	0.009	0.007	0.005	0.000	0.000	0.014	0.007	0.000	0.008	0.003
OBMC2	0.012	0.008	0.095	0.006	0.000	0.010	0.006	0.003	0.002	0.000
OBMC3	0.006	0.015	0.026	0.000	0.002	0.014	0.005	0.001	0.009	0.003
OBMC4	0.002	0.003	0.025	0.000	0.001	0.008	0.003	0.001	0.010	0.000
OBT1	0.042	0.020	0.011	0.001	0.000	0.010	0.005	0.011	0.008	0.010
OBT2	0.015	0.015	0.060	0.031	0.022	0.030	0.053	0.034	0.015	0.057
OBT3	0.021	0.011	0.020	0.003	0.008	0.018	0.014	0.029	0.014	0.025
OBT4	0.017	0.011	0.031	0.004	0.026	0.017	0.016	0.360	0.016	0.057
OBGCT1	0.030	0.039	0.015	0.004	0.004	0.010	0.008	0.016	0.014	0.014
OBGCT2	0.038	0.020	0.021	0.000	0.005	0.012	0.014	0.028	0.013	0.005
OBGCT3	0.017	0.016	0.221	0.003	0.015	0.020	0.014	0.010	0.006	0.009
OBGCT4	0.018	0.025	0.028	0.000	0.000	0.007	0.028	0.040	0.012	0.005
OBMCT1	0.024	0.030	0.040	0.000	0.000	0.012	0.017	0.019	0.013	0.010
OBMCT2	0.017	0.018	0.013	0.000	0.003	0.014	0.014	0.015	0.004	0.003
OBMCT3	0.035	0.032	0.018	0.000	0.001	0.005	0.030	0.024	0.005	0.005
OBMCT4	0.029	0.030	0.015	0.000	0.002	0.014	0.008	0.003	0.012	0.006
NBGC1	0.013	0.010	0.050	0.022	0.020	0.025	0.041	0.055	0.014	0.011
NBGC2	0.002	0.006	0.017	0.000	0.005	0.010	0.016	0.018	0.004	0.001
NBGC3	0.006	0.008	0.098	0.004	0.010	0.007	0.017	0.020	0.001	0.004
NBGC4	0.010	0.011	0.026	0.009	0.004	0.002	0.013	0.016	0.005	0.004
NBMC1	0.006	0.007	0.018	0.007	0.003	0.004	0.006	0.015	0.000	0.005
NBMC2	0.004	0.007	0.012	0.004	0.000	0.002	0.005	0.009	0.005	0.000
NBMC3	0.004	0.004	0.019	0.000	0.000	0.008	0.001	0.006	0.005	0.000
NBMC4	0.005	0.003	0.012	0.003	0.002	0.005	0.011	0.010	0.001	0.006
NBT1	0.028	0.018	0.016	0.026	0.016	0.013	0.025	0.171	0.009	0.059
NBT2	0.016	0.005	0.018	0.029	0.011	0.013	0.022	0.031	0.012	0.046
NBT3	0.014	0.016	0.018	0.024	0.019	0.013	0.045	0.093	0.007	0.008
NBT4	0.005	0.016	0.015	0.020	0.014	0.011	0.017	0.102	0.001	0.033
NBGCT1	0.025	0.020	0.008	0.006	0.004	0.000	0.010	0.044	0.002	0.000
NBGCT2	0.015	0.020	0.012	0.003	0.002	0.001	0.018	0.059	0.003	0.007
NBGCT3	0.027	0.019	0.013	0.007	0.005	0.000	0.012	0.014	0.003	0.002
NBGCT4	0.032	0.027	0.017	0.008	0.010	0.002	0.011	0.035	0.003	0.002
NBMCT1	0.024	0.031	0.024	0.008	0.005	0.004	0.022	0.028	0.009	0.003
NBMCT2	0.019	0.034	0.047	0.003	0.006	0.004	0.006	0.002	0.002	0.000

Samples	Weeks									
	bk1	bk2	1	2	3	4	5	6	7	8
NBMCT3	0.017	0.013	0.015	0.015	0.006	0.006	0.013	0.018	0.001	0.008
NBMCT4	0.022	0.035	0.004	0.013	0.005	0.006	0.010	0.016	0.008	0.003
GGC1	0.001	0.012	0.021	0.030	0.032	0.028	0.049	0.055	0.008	0.013
GGC2	0.006	0.011	0.011	0.015	0.014	0.010	0.013	0.028	0.005	0.002
GGC3	0.012	0.019	0.015	0.009	0.008	0.003	0.020	0.032	0.003	0.006
GGC4	0.008	0.014	0.032	0.003	0.000	0.006	0.024	0.046	0.000	0.005
GMC1	0.015	0.019	0.020	0.002	0.008	0.004	0.005	0.006	0.001	0.002
GMC2	0.000	0.006	0.057	0.005	0.007	0.004	0.018	0.007	0.002	0.001
GMC3	0.011	0.012	0.012	0.001	0.007	0.000	0.013	0.006	0.001	0.001
GMC4	0.008	0.019	0.020	0.002	0.004	0.003	0.008	0.011	0.003	0.000
GT1	0.054	0.026	0.028	0.005	0.000	0.004	0.004	0.009	0.002	0.004
GT2	0.046	0.015	0.045	0.012	0.003	0.013	0.009	0.011	0.005	0.005
GT3	0.041	0.018	0.021	0.002	0.007	0.007	0.007	0.005	0.003	0.004
GT4	0.009	0.020	0.050	0.052	0.019	0.037	0.034	0.159	0.016	0.054
GGCT1	0.243	0.137	0.031	0.000	0.017	0.001	0.017	0.019	0.002	0.008
GGCT2	0.078	0.151	0.028	0.003	0.006	0.001	0.015	0.010	0.000	0.003
GGCT3	0.179	0.162	0.801	0.039	0.014	0.009	0.015	0.020	0.004	0.002
GGCT4	0.514	0.153	0.026	0.008	0.000	0.004	0.013	0.026	0.003	0.003
GMCT1	0.018	0.023	0.017	0.005	0.015	0.014	0.038	0.042	0.003	0.008
GMCT2	0.048	0.047	0.008	0.000	0.001	0.000	0.006	0.010	0.003	0.003
GMCT3	0.126	0.050	0.023	0.002	0.005	0.000	0.017	0.009	0.000	0.004
GMCT4	0.062	0.060	0.024	0.003	0.007	0.002	0.017	0.017	0.000	0.004
LGC1	0.003	0.017	0.090	0.034	0.028	0.023	0.027	0.037	0.014	0.008
LGC2	0.003	0.013	0.024	0.020	0.007	0.009	0.010	0.033	0.005	0.007
LGC3	0.011	0.020	0.019	0.007	0.004	0.003	0.004	0.046	0.007	0.006
LGC4	0.000	0.010	0.020	0.009	0.000	0.009	0.017	0.031	0.005	0.000
LMC1	0.008	0.018	0.018	0.003	0.000	0.010	0.004	0.012	0.005	0.003
LMC2	0.000	0.012	0.004	0.000	0.000	0.003	0.003	0.007	0.000	0.001
LMC3	0.004	0.005	0.009	0.006	0.002	0.006	0.009	0.016	0.005	0.002
LMC4	0.005	0.013	0.007	0.008	0.000	0.004	0.006	0.016	0.006	0.008
LT1	0.034	0.035	0.010	0.040	0.005	0.019	0.000	0.140	0.007	0.006
LT2	0.012	0.027	0.007	0.008	0.001	0.005	0.000	0.005	0.000	0.000
LT3	0.032	0.055	0.006	0.011	0.003	0.005	0.000	0.007	0.000	0.003
LT4	0.025	0.023	0.007	0.008	0.006	0.008	0.000	0.008	0.004	0.003
LGCT1	0.157	0.111	0.007	0.005	0.006	0.005	0.003	0.004	0.000	0.006
LGCT2	0.177	0.226	0.022	0.008	0.001	0.004	0.001	0.004	0.002	0.004
LGCT3	0.159	0.079	0.003	0.001	0.003	0.010	0.000	0.010	0.000	0.006
LGCT4	0.078	0.192	0.000	0.000	0.001	0.007	0.000	0.009	0.004	0.003
LMCT1	0.078	0.049	0.000	0.000	0.001	0.006	0.001	0.007	0.003	0.000
LMCT2	0.052	0.066	0.000	0.005	0.003	0.001	0.000	0.003	0.007	0.006
LMCT3	0.051	0.070	1.833	0.009	0.001	0.005	0.000	0.004	0.002	0.002
LMCT4	0.073	0.044	0.016	0.004	0.000	0.003	0.000	0.010	0.004	0.000

Standard error of Pb concentrations in profile leachates (mgL^{-1})

standard error	Weeks									
	bk	bk	1	2	3	4	5	6	7	8
OBGC	0.006	0.003	0.011	0.006	0.090	0.006	0.002	0.009	0.002	0.003
OBMC	0.002	0.002	0.020	0.002	0.000	0.002	0.001	0.001	0.002	0.001
OBT	0.006	0.002	0.011	0.007	0.006	0.004	0.011	0.084	0.002	0.012
OBGCT	0.005	0.005	0.050	0.001	0.003	0.003	0.004	0.007	0.002	0.002
OBMCT	0.004	0.003	0.006	0.000	0.001	0.002	0.005	0.004	0.002	0.001
NBGC	0.002	0.001	0.018	0.005	0.004	0.005	0.006	0.009	0.003	0.002
NBMC	0.000	0.001	0.002	0.001	0.001	0.001	0.002	0.002	0.001	0.002
NBT	0.005	0.003	0.001	0.002	0.002	0.001	0.006	0.029	0.002	0.011
NBGCT	0.004	0.002	0.002	0.001	0.002	0.000	0.002	0.009	0.000	0.001
NBMCT	0.002	0.005	0.009	0.003	0.000	0.001	0.003	0.005	0.002	0.002
GGC	0.002	0.002	0.005	0.006	0.007	0.006	0.008	0.006	0.002	0.002
GMC	0.003	0.003	0.010	0.001	0.001	0.001	0.003	0.001	0.000	0.000
GT	0.010	0.002	0.007	0.012	0.004	0.007	0.007	0.038	0.003	0.012
GGCT	0.093	0.005	0.193	0.009	0.004	0.002	0.001	0.003	0.001	0.001
GMCT	0.023	0.008	0.004	0.001	0.003	0.003	0.007	0.008	0.001	0.001
LGC	0.002	0.002	0.017	0.006	0.006	0.004	0.005	0.003	0.002	0.002
LMC	0.002	0.003	0.003	0.002	0.001	0.002	0.001	0.002	0.001	0.002
LT	0.005	0.007	0.001	0.008	0.001	0.003	0.000	0.033	0.002	0.001
LGCT	0.022	0.034	0.005	0.002	0.001	0.001	0.001	0.002	0.001	0.001
LMCT	0.007	0.006	0.457	0.002	0.001	0.001	0.000	0.002	0.001	0.001

Mean Pb concentrations in profile leachates (mgL^{-1})

Samples	Weeks									
	bK1	bk2	1	2	3	4	5	6	7	8
OBGC	0.028	0.015	0.052	0.012	0.098	0.019	0.015	0.025	0.009	0.007
OBMC	0.007	0.008	0.038	0.002	0.001	0.012	0.005	0.001	0.007	0.002
OBT	0.024	0.014	0.031	0.010	0.014	0.019	0.022	0.109	0.013	0.037
OBGCT	0.026	0.025	0.071	0.002	0.006	0.012	0.016	0.024	0.011	0.008
OBMCT	0.026	0.028	0.022	0.000	0.002	0.011	0.017	0.015	0.009	0.006
NBGC	0.008	0.009	0.048	0.009	0.010	0.011	0.022	0.027	0.006	0.005
NBMC	0.005	0.005	0.015	0.004	0.001	0.005	0.006	0.010	0.003	0.003
NBT	0.016	0.014	0.017	0.025	0.015	0.013	0.027	0.099	0.007	0.037
NBGCT	0.025	0.022	0.013	0.006	0.005	0.001	0.013	0.038	0.003	0.003
NBMCT	0.021	0.028	0.023	0.010	0.006	0.005	0.013	0.016	0.005	0.004
GGC	0.007	0.014	0.020	0.014	0.014	0.012	0.027	0.040	0.004	0.007
GMC	0.009	0.014	0.027	0.003	0.007	0.003	0.011	0.008	0.002	0.001
GT	0.038	0.020	0.036	0.018	0.007	0.015	0.014	0.046	0.007	0.017
GGCT	0.254	0.151	0.222	0.013	0.009	0.004	0.015	0.019	0.002	0.004
GMCT	0.064	0.045	0.018	0.003	0.007	0.004	0.020	0.020	0.002	0.005
LGC	0.004	0.015	0.038	0.018	0.010	0.011	0.015	0.037	0.008	0.005

Samples	Weeks									
	bK1	bk2	1	2	3	4	5	6	7	8
LMC	0.004	0.012	0.010	0.004	0.001	0.006	0.006	0.013	0.004	0.004
LT	0.026	0.035	0.008	0.017	0.004	0.009	0.000	0.040	0.003	0.003
LGCT	0.143	0.152	0.008	0.004	0.003	0.007	0.001	0.007	0.002	0.005
LMCT	0.064	0.057	0.462	0.005	0.001	0.004	0.000	0.006	0.004	0.002
WHO (2011)	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01

Two-way Anova test for Pb concentrations in profile leachates

Tests of Between-Subjects Effects

Dependent Variable: Pb conc in leachate

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.921[a]	19	.048	.823	.672
Intercept	1.656	1	1.656	28.116	.000
Medium	.180	4	.045	.765	.552
Aggregate	.022	3	.007	.126	.944
Medium * Aggregate	.718	12	.060	1.016	.446
Error	3.534	60	.059		
Total	6.111	80			
Corrected Total	4.455	79			

a. R Squared = .207 (Adjusted R Squared = -.045)

Post Hoc test for Ni concentrations in profile leachate

Multiple Comparisons

Pb conc

LSD: Growth medium

(I) Medium	(J) Medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	.024000	.0858036	.781	-.147633	.195633
	MC	.107500	.0858036	.215	-.064133	.279133
	MCT	-.018313	.0858036	.832	-.189945	.153320
	T	-.021625	.0858036	.802	-.193258	.150008
GCT	GC	-.024000	.0858036	.781	-.195633	.147633
	MC	.083500	.0858036	.334	-.088133	.255133
	MCT	-.042313	.0858036	.624	-.213945	.129320
	T	-.045625	.0858036	.597	-.217258	.126008
MC	GC	-.107500	.0858036	.215	-.279133	.064133
	GCT	-.083500	.0858036	.334	-.255133	.088133
	MCT	-.125813	.0858036	.148	-.297445	.045820
	T	-.129125	.0858036	.138	-.300758	.042508
MCT	GC	.018313	.0858036	.832	-.153320	.189945
	GCT	.042313	.0858036	.624	-.129320	.213945
	MC	.125813	.0858036	.148	-.045820	.297445
	T	-.003312	.0858036	.969	-.174945	.168320
T	GC	.021625	.0858036	.802	-.150008	.193258
	GCT	.045625	.0858036	.597	-.126008	.217258
	MC	.129125	.0858036	.138	-.042508	.300758
	MCT	.003312	.0858036	.969	-.168320	.174945

Based on observed means.

The error term is Mean Square (Error) = .059.

Pb conc
LSD: Aggregates

(I) Aggregate	(J) Aggregate	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	-.013750	.0767451	.858	-.167263	.139763
	NB	.027250	.0767451	.724	-.126263	.180763
	OB	-.013400	.0767451	.862	-.166913	.140113
L	G	.013750	.0767451	.858	-.139763	.167263
	NB	.041000	.0767451	.595	-.112513	.194513
	OB	.000350	.0767451	.996	-.153163	.153863
NB	G	-.027250	.0767451	.724	-.180763	.126263
	L	-.041000	.0767451	.595	-.194513	.112513
	OB	-.040650	.0767451	.598	-.194163	.112863
OB	G	.013400	.0767451	.862	-.140113	.166913
	L	-.000350	.0767451	.996	-.153863	.153163
	NB	.040650	.0767451	.598	-.112863	.194163

Based on observed means.
The error term is Mean Square (Error) = .059.

Appendix 23: Results for Zinc concentrations in profile leachate over an eight-week spiking period

Zn concentrations in profile leachates (mgL^{-1})

Samples	Weeks									
	bk	bk	1	2	3	4	5	6	7	8
OBGC1	0.036	0.039	0.048	0.053	0.422	0.038	0.043	0.121	0.015	0.019
OBGC2	0.027	0.045	0.670	0.063	0.036	0.030	0.048	0.058	0.008	0.009
OBGC3	0.037	0.062	0.069	0.112	0.019	0.016	0.040	0.095	0.010	0.008
OGBG4	0.085	0.127	0.314	0.032	0.022	0.019	0.067	0.113	0.012	0.008
OBMC1	0.024	0.040	0.011	0.010	0.005	0.004	0.005	0.009	0.008	0.006
OBMC2	0.056	0.066	0.170	0.012	0.003	0.005	0.008	0.013	0.009	0.006
OBMC3	0.038	0.083	0.083	0.014	0.006	0.007	0.016	0.015	0.006	0.005
OBMC4	0.056	0.042	0.041	0.037	0.010	0.014	0.015	0.016	0.008	0.005
OBT1	0.050	0.006	0.005	0.005	0.004	0.004	0.005	0.007	0.005	0.006
OBT2	0.028	0.031	0.016	0.020	0.011	0.008	0.012	0.015	0.009	0.018
OBT3	0.154	0.019	0.051	0.024	0.012	0.018	0.003	0.009	0.006	0.010
OBT4	0.047	0.015	0.005	0.008	0.015	0.005	0.005	0.016	0.006	0.016
OBGCT1	0.081	0.026	0.009	0.020	0.008	0.008	0.050	0.012	0.006	0.006
OBGCT2	0.122	0.021	0.020	0.013	0.007	0.007	0.009	0.019	0.005	0.005
OBGCT3	0.063	0.027	0.175	0.028	0.009	0.008	0.005	0.007	0.004	0.004
OBGCT4	0.163	0.431	0.109	0.014	0.006	0.005	0.008	0.017	0.006	0.004
OBMCT1	0.099	0.063	0.059	0.012	0.008	0.011	0.004	0.012	0.006	0.006
OBMCT2	0.109	0.062	0.009	0.013	0.006	0.009	0.002	0.007	0.005	0.003
OBMCT3	0.092	0.076	0.007	0.011	0.004	0.004	0.005	0.008	0.003	0.002
OBMCT4	0.145	0.067	0.006	0.017	0.008	0.006	0.002	0.007	0.005	0.004
NBGC1	0.072	0.103	0.140	0.088	0.034	0.035	0.086	0.130	0.021	0.021
NBGC2	0.058	0.029	0.088	0.017	0.011	0.012	0.038	0.063	0.009	0.006
NBGC3	0.021	0.064	0.346	0.014	0.009	0.007	0.037	0.062	0.009	0.006
NBGC4	0.049	0.028	0.091	0.047	0.016	0.009	0.035	0.061	0.008	0.007
NBMC1	0.087	0.062	0.045	0.021	0.016	0.013	0.023	0.057	0.009	0.008
NBMC2	0.048	0.141	0.023	0.014	0.013	0.012	0.020	0.027	0.005	0.008
NBMC3	0.068	0.043	0.029	0.020	0.007	0.009	0.009	0.010	0.007	0.007
NBMC4	0.038	0.032	0.022	0.019	0.007	0.007	0.018	0.026	0.006	0.005
NBT1	0.056	0.014	0.003	0.005	0.009	0.004	0.006	0.033	0.007	0.015
NBT2	0.093	0.002	0.003	0.008	0.012	0.011	0.007	0.008	0.006	0.012
NBT3	0.036	0.022	0.002	0.014	0.016	0.017	0.060	0.018	0.003	0.006
NBT4	0.024	0.012	0.004	0.007	0.012	0.011	0.003	0.022	0.005	0.011
NBGCT1	0.075	0.019	0.003	0.019	0.005	0.006	0.004	0.012	0.004	0.004
NBGCT2	0.049	0.021	0.005	0.016	0.007	0.006	0.007	0.026	0.005	0.006
NBGCT3	0.040	0.027	0.007	0.011	0.009	0.007	0.004	0.010	0.004	0.004
NBGCT4	0.073	0.034	0.016	0.020	0.010	0.005	0.005	0.014	0.004	0.005
NBMCT1	0.075	0.049	0.016	0.010	0.005	0.003	0.006	0.008	0.003	0.004
NBMCT2	0.032	0.055	0.020	0.011	0.007	0.004	0.004	0.006	0.004	0.005

Samples	Weeks									
	bk	bk	1	2	3	4	5	6	7	8
NBMCT3	0.054	0.045	0.012	0.013	0.011	0.006	0.060	0.014	0.003	0.005
NBMCT4	0.086	0.071	0.003	0.013	0.013	0.006	0.005	0.007	0.005	0.006
GGC1	0.043	0.080	0.062	0.050	0.075	0.034	0.155	0.176	0.012	0.017
GGC2	0.036	0.096	0.030	0.059	0.063	0.019	0.063	0.105	0.007	0.008
GGC3	0.087	0.124	0.045	0.041	0.046	0.014	0.075	0.137	0.009	0.007
GGC4	0.036	0.095	0.085	0.036	0.028	0.016	0.083	0.200	0.011	0.010
GMC1	0.039	0.133	0.083	0.037	0.012	0.013	0.022	0.041	0.004	0.007
GMC2	0.030	0.066	0.143	0.046	0.025	0.013	0.048	0.051	0.006	0.005
GMC3	0.077	0.156	0.047	0.029	0.040	0.010	0.047	0.037	0.006	0.006
GMC4	0.065	0.149	0.037	0.024	0.030	0.010	0.040	0.042	0.006	0.006
GT1	0.049	0.008	0.032	0.011	0.014	0.006	0.005	0.005	0.002	0.007
GT2	0.036	0.012	0.043	0.022	0.009	0.011	0.006	0.007	0.002	0.007
GT3	0.139	0.016	0.021	0.022	0.006	0.010	0.006	0.006	0.002	0.005
GT4	0.064	0.034	0.021	0.029	0.015	0.030	0.010	0.031	0.007	0.015
GGCT1	0.174	0.099	0.066	0.013	0.028	0.009	0.014	0.019	0.004	0.005
GGCT2	0.101	0.160	0.055	0.015	0.022	0.008	0.010	0.013	0.004	0.004
GGCT3	0.165	0.121	0.849	0.032	0.016	0.006	0.010	0.019	0.004	0.004
GGCT4	0.324	0.096	0.039	0.038	0.005	0.006	0.007	0.015	0.004	0.004
GMCT1	0.039	0.040	0.015	0.015	0.013	0.013	0.009	0.016	0.005	0.008
GMCT2	0.116	0.127	0.019	0.032	0.010	0.014	0.008	0.013	0.004	0.005
GMCT3	0.384	0.109	0.031	0.022	0.004	0.007	0.010	0.011	0.003	0.005
GMCT4	0.177	0.073	0.054	0.015	0.006	0.007	0.009	0.015	0.002	0.005
LGC1	0.067	0.117	0.247	0.067	0.048	0.031	0.085	0.094	0.015	0.014
LGC2	0.028	0.087	0.091	0.031	0.023	0.015	0.045	0.111	0.014	0.008
LGC3	0.104	0.174	0.085	0.030	0.017	0.001	0.052	0.139	0.010	0.006
LGC4	0.042	0.073	0.099	0.025	0.012	0.010	0.072	0.137	0.013	0.008
LMC1	0.033	0.165	0.044	0.032	0.010	0.010	0.022	0.029	0.004	0.004
LMC2	0.018	0.124	0.024	0.015	0.004	0.008	0.032	0.039	0.005	0.004
LMC3	0.043	0.084	0.039	0.014	0.004	0.005	0.028	0.055	0.003	0.002
LMC4	0.015	0.054	0.039	0.017	0.004	0.005	0.049	0.052	0.004	0.003
LT1	0.016	0.015	0.002	0.018	0.002	0.005	0.002	0.003	0.003	0.005
LT2	0.032	0.008	0.003	0.015	0.004	0.003	0.001	0.001	0.002	0.002
LT3	0.041	0.024	0.004	0.011	0.003	0.004	0.008	0.003	0.003	0.003
LT4	0.020	0.013	0.011	0.005	0.003	0.004	0.002	0.004	0.003	0.003
LGCT1	0.115	0.074	0.023	0.007	0.005	0.004	0.004	0.007	0.003	0.002
LGCT2	0.165	0.128	0.036	0.023	0.002	0.003	0.006	0.006	0.002	0.001
LGCT3	0.202	0.072	0.005	0.009	0.004	0.002	0.006	0.006	0.004	0.004
LGCT4	0.110	0.128	0.002	0.017	0.006	0.008	0.006	0.009	0.003	0.003
LMCT1	0.257	0.110	0.001	0.010	0.004	0.003	0.005	0.009	0.002	0.002
LMCT2	0.100	0.114	0.006	0.011	0.001	0.002	0.004	0.007	0.003	0.006
LMCT3	0.085	0.149	1.429	0.013	0.003	0.002	0.003	0.005	0.002	0.001
LMCT4	0.142	0.089	0.020	0.009	0.003	0.002	0.002	0.005	0.002	0.001

Standard error of Zn concentrations in profile leachates (mgL^{-1})

standard error	Weeks									
	bk	bk	1	2	3	4	5	6	7	8
OBGC	0.013	0.020	0.145	0.017	0.099	0.005	0.006	0.014	0.001	0.003
OBMC	0.008	0.010	0.035	0.006	0.001	0.002	0.003	0.002	0.001	0.000
OBT	0.029	0.005	0.011	0.005	0.002	0.003	0.002	0.002	0.001	0.003
OBGCT	0.022	0.102	0.039	0.003	0.001	0.001	0.011	0.003	0.000	0.000
OBMCT	0.012	0.003	0.013	0.001	0.001	0.002	0.001	0.001	0.001	0.001
NBGC	0.011	0.018	0.061	0.017	0.006	0.006	0.012	0.017	0.003	0.004
NBMC	0.011	0.025	0.005	0.002	0.002	0.001	0.003	0.010	0.001	0.001
NBT	0.015	0.004	0.000	0.002	0.001	0.003	0.014	0.005	0.001	0.002
NBGCT	0.009	0.003	0.003	0.002	0.001	0.000	0.001	0.004	0.000	0.000
NBMCT	0.012	0.006	0.004	0.001	0.002	0.001	0.014	0.002	0.000	0.000
GGC	0.012	0.009	0.012	0.005	0.010	0.005	0.021	0.021	0.001	0.002
GMC	0.011	0.021	0.024	0.005	0.006	0.001	0.006	0.003	0.001	0.000
GT	0.023	0.006	0.005	0.004	0.002	0.005	0.001	0.006	0.001	0.002
GGCT	0.047	0.015	0.199	0.006	0.005	0.001	0.001	0.002	0.000	0.000
GMCT	0.074	0.019	0.009	0.004	0.002	0.002	0.000	0.001	0.001	0.001
LGC	0.017	0.022	0.039	0.010	0.008	0.006	0.009	0.011	0.001	0.002
LMC	0.007	0.024	0.004	0.004	0.002	0.001	0.006	0.006	0.000	0.000
LT	0.006	0.003	0.002	0.003	0.000	0.000	0.002	0.001	0.000	0.001
LGCT	0.022	0.016	0.008	0.004	0.001	0.001	0.001	0.001	0.000	0.001
LMCT	0.039	0.012	0.355	0.001	0.001	0.000	0.001	0.001	0.000	0.001

Mean Zn concentrations in profile leachates (mgL^{-1})

Samples	Weeks									
	bK1	bk2	1	2	3	4	5	6	7	8
OBGC	0.046	0.068	0.275	0.065	0.125	0.026	0.050	0.097	0.011	0.011
OBMC	0.044	0.058	0.076	0.018	0.006	0.008	0.011	0.013	0.008	0.000
OBT	0.070	0.018	0.019	0.014	0.011	0.009	0.006	0.012	0.007	0.013
OBGCT	0.107	0.126	0.078	0.019	0.008	0.007	0.018	0.014	0.000	0.000
OBMCT	0.111	0.067	0.020	0.013	0.007	0.008	0.000	0.009	0.000	0.000
NBGC	0.050	0.056	0.166	0.042	0.018	0.016	0.049	0.079	0.012	0.010
NBMC	0.060	0.070	0.030	0.019	0.011	0.010	0.018	0.030	0.007	0.007
NBT	0.052	0.013	0.000	0.009	0.012	0.011	0.019	0.020	0.000	0.011
NBGCT	0.059	0.025	0.008	0.017	0.008	0.006	0.000	0.016	0.000	0.000
NBMCT	0.062	0.055	0.013	0.012	0.009	0.000	0.019	0.009	0.000	0.000
GGC	0.051	0.099	0.056	0.047	0.053	0.021	0.094	0.155	0.010	0.011
GMC	0.053	0.126	0.078	0.034	0.027	0.012	0.039	0.043	0.000	0.006
GT	0.072	0.018	0.029	0.021	0.011	0.014	0.007	0.012	0.000	0.009
GGCT	0.191	0.119	0.252	0.025	0.018	0.007	0.010	0.017	0.000	0.000
GMCT	0.179	0.087	0.030	0.021	0.008	0.010	0.009	0.014	0.000	0.000
LGC	0.060	0.113	0.131	0.038	0.025	0.014	0.064	0.120	0.013	0.009

Samples	Weeks									
	bK1	bk2	1	2	3	4	5	6	7	8
LMC	0.027	0.107	0.037	0.020	0.000	0.007	0.033	0.044	0.000	0.000
LT	0.027	0.015	0.000	0.012	0.000	0.000	0.000	0.000	0.000	0.000
LGCT	0.148	0.101	0.017	0.014	0.000	0.000	0.000	0.007	0.000	0.000
LMCT	0.146	0.116	0.364	0.011	0.000	0.000	0.000	0.007	0.000	0.000
WHO (2011) Potable water guideline	3.000	3.000	3.000	3.000	3.000	3.000	3.000	3.000	3.000	3.000

Two-way Anova test for Zn concentrations in profile leachates

Tests of Between-Subjects Effects

Dependent Variable: Zn conc in leachate

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	2.252[a]	19	.119	2.987	.001
Intercept	3.452	1	3.452	86.991	.000
Medium	1.529	4	.382	9.632	**.000**
Aggregate	.100	3	.033	.840	.477
Medium * Aggregate	.624	12	.052	1.309	.237
Error	2.381	60	.040		
Total	8.085	80			
Corrected Total	4.633	79			

a. R Squared = .486 (Adjusted R Squared = .323)

Post Hoc test for Zn concentrations in profile leachate

Multiple Comparisons

Zn conc

LSD: Growth medium

(I) Medium	(J) Medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	.323438*	.0704290	.000	.182558	.464317
	MC	.309188*	.0704290	.000	.168308	.450067
	MCT	.317563*	.0704290	.000	.176683	.458442
	T	.396500*	.0704290	.000	.255621	.537379
GCT	GC	-.323438*	.0704290	.000	-.464317	-.182558
	MC	-.014250	.0704290	.840	-.155129	.126629
	MCT	-.005875	.0704290	.934	-.146754	.135004
	T	.073063	.0704290	.304	-.067817	.213942
MC	GC	-.309188*	.0704290	.000	-.450067	-.168308
	GCT	.014250	.0704290	.840	-.126629	.155129
	MCT	.008375	.0704290	.906	-.132504	.149254
	T	.087313	.0704290	.220	-.053567	.228192
MCT	GC	-.317563*	.0704290	.000	-.458442	-.176683
	GCT	.005875	.0704290	.934	-.135004	.146754
	MC	-.008375	.0704290	.906	-.149254	.132504
	T	.078938	.0704290	.267	-.061942	.219817
T	GC	-.396500*	.0704290	.000	-.537379	-.255621
	GCT	-.073063	.0704290	.304	-.213942	.067817
	MC	-.087313	.0704290	.220	-.228192	.053567
	MCT	-.078938	.0704290	.267	-.219817	.061942

Based on observed means.

The error term is Mean Square (Error) = .040.

*.The mean difference is significant at the 0.05 level.

Zn conc

LSD: Aggregates

(I) Aggregate	(J) Aggregate	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	.035650	.0629936	.574	-.090356	.161656
	NB	.095750	.0629936	.134	-.030256	.221756
	OB	.023300	.0629936	.713	-.102706	.149306
L	G	-.035650	.0629936	.574	-.161656	.090356
	NB	.060100	.0629936	.344	-.065906	.186106
	OB	-.012350	.0629936	.845	-.138356	.113656
NB	G	-.095750	.0629936	.134	-.221756	.030256
	L	-.060100	.0629936	.344	-.186106	.065906
	OB	-.072450	.0629936	.255	-.198456	.053556
OB	G	-.023300	.0629936	.713	-.149306	.102706
	L	.012350	.0629936	.845	-.113656	.138356
	NB	.072450	.0629936	.255	-.053556	.198456

Based on observed means. The error term is Mean Square (Error) = .040.

Appendix 24: Results for Al concentrations in harvested grass shoots, growth media and grass roots

Samples	Blank corrected mean Al concentrations (mg/kg)						
	bk	1st harvest	2nd harvest	3rd harvest	cumulative grass harvest	growth media	Grass roots
OBGC	162.65	46.45	7.24	26.05	242.39	4129.45	162.65
OBMC	30.35	16.23	17.48	14.81	78.86	3265.95	30.35
OBT	137.96	50.09	95.83	88.69	372.56	6947.70	137.96
OBGCT	64.79	18.48	13.98	1039.75	1136.99	7560.20	64.79
OBMCT	71.34	28.08	10.53	381.50	491.44	6337.70	71.34
NBGC	43.08	10.51	4.20	13.78	71.56	4127.20	43.08
NBMC	87.01	46.81	14.33	16.91	165.06	5152.70	87.01
NBT	100.85	18.61	34.66	165.88	320.00	7395.20	100.85
NBGCT	71.34	21.68	26.44	65.73	185.18	5019.70	71.34
NBMCT	138.38	28.05	25.36	356.49	548.28	4748.20	138.38
GGC	106.45	19.64	28.85	44.48	199.41	2931.45	106.45
GMC	75.68	30.68	84.61	203.15	394.11	3164.20	75.68
GT	140.68	27.73	63.70	360.75	592.85	5795.20	140.68
GGCT	67.58	131.51	46.90	104.50	350.49	4735.45	67.58
GMCT	216.81	116.91	28.96	64.41	427.10	5902.70	216.81
LGC	90.63	20.40	7.16	120.69	238.88	3528.70	90.63
LMC	102.84	40.34	33.24	83.38	259.79	5129.70	102.84
LT	53.10	75.14	24.81	67.51	220.56	5910.20	53.10
LGCT	101.35	96.25	49.39	109.96	356.95	5291.20	101.35
LMCT	183.08	116.56	21.15	54.21	375.00	5670.20	183.08

Two-way Anova test for Al concentrations in grass shoots

Tests of Between-Subjects Effects

Dependent Variable: Al conc.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	3201.777[a]	19	168.515	17.683	.000
Intercept	3970.515	1	3970.515	416.654	.000
Medium	932.711	4	233.178	24.469	**.000**
aggregate	413.926	3	137.975	14.479	**.000**
Medium * aggregate	1855.141	12	154.595	16.223	**.000**
Error	190.591	20	9.530		
Total	7362.882	40			
Corrected Total	3392.368	39			

a. R Squared = .944 (Adjusted R Squared = .890)

Post Hoc test for Al concentrations in grass shoots

Multiple Comparisons

Al conc.

LSD: growth media

(I) Medium	(J) Medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	-13.751125*	1.5434967	.000	-16.970803	-10.531447
	MC	-2.525125	1.5434967	.117	-5.744803	.694553
	MCT	-8.827750*	1.5434967	.000	-12.047428	-5.608072
	T	-7.239500*	1.5434967	.000	-10.459178	-4.019822
GCT	GC	13.751125*	1.5434967	.000	10.531447	16.970803
	MC	11.226000*	1.5434967	.000	8.006322	14.445678
	MCT	4.923375*	1.5434967	.005	1.703697	8.143053
	T	6.511625*	1.5434967	.000	3.291947	9.731303
MC	GC	2.525125	1.5434967	.117	-.694553	5.744803
	GCT	-11.226000*	1.5434967	.000	-14.445678	-8.006322
	MCT	-6.302625*	1.5434967	.001	-9.522303	-3.082947
	T	-4.714375*	1.5434967	.006	-7.934053	-1.494697
MCT	GC	8.827750*	1.5434967	.000	5.608072	12.047428
	GCT	-4.923375*	1.5434967	.005	-8.143053	-1.703697
	MC	6.302625*	1.5434967	.001	3.082947	9.522303
	T	1.588250	1.5434967	.316	-1.631428	4.807928
T	GC	7.239500*	1.5434967	.000	4.019822	10.459178
	GCT	-6.511625*	1.5434967	.000	-9.731303	-3.291947
	MC	4.714375*	1.5434967	.006	1.494697	7.934053
	MCT	-1.588250	1.5434967	.316	-4.807928	1.631428

Based on observed means.

The error term is Mean Square (Error) = 9.530.

*. The mean difference is significant at the 0.05 level.

Al grass conc.

LSD: Aggregates

(I) aggregate	(J) aggregate	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	3.492700*	1.3805455	.020	.612933	6.372467
	NB	4.058800*	1.3805455	.008	1.179033	6.938567
	OB	-3.987000*	1.3805455	.009	-6.866767	-1.107233
L	G	-3.492700*	1.3805455	.020	-6.372467	-.612933
	NB	.566100	1.3805455	.686	-2.313667	3.445867
	OB	-7.479700*	1.3805455	.000	-10.359467	-4.599933
NB	G	-4.058800*	1.3805455	.008	-6.938567	-1.179033
	L	-.566100	1.3805455	.686	-3.445867	2.313667
	OB	-8.045800*	1.3805455	.000	-10.925567	-5.166033
OB	G	3.987000*	1.3805455	.009	1.107233	6.866767
	L	7.479700*	1.3805455	.000	4.599933	10.359467
	NB	8.045800*	1.3805455	.000	5.166033	10.925567

Based on observed means. The error term is Mean Square (Error) = 9.530.

*. The mean difference is significant at the 0.05 level.

Two-way Anova test for Al concentrations in growth media

Tests of Between-Subjects Effects

Dependent Variable: Al conc.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	27857.832[a]	19	1466.202	9.745	.000
Intercept	422244.962	1	422244.962	2806.474	.000
medium	17531.995	4	4382.999	29.132	**.000**
aggregates	2734.729	3	911.576	6.059	**.004**
medium * aggregates	7591.108	12	632.592	4.205	**.002**
Error	3009.078	20	150.454		
Total	453111.872	40			
Corrected Total	30866.910	39			

a. R Squared = .903 (Adjusted R Squared = .810)

Post Hoc test for Al concentrations in growth media

Multiple Comparisons

Al conc.

LSD: Growth media

(I) medium	(J) medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	-39.4488*	6.13298	.000	-52.2419	-26.6556
	MC	-9.9787	6.13298	.119	-22.7719	2.8144
	MCT	-39.7100*	6.13298	.000	-52.5032	-26.9168
	T	-56.6575*	6.13298	.000	-69.4507	-43.8643
GCT	GC	39.4488*	6.13298	.000	26.6556	52.2419
	MC	29.4700*	6.13298	.000	16.6768	42.2632
	MCT	-.2612	6.13298	.966	-13.0544	12.5319
	T	-17.2088*	6.13298	.011	-30.0019	-4.4156
MC	GC	9.9787	6.13298	.119	-2.8144	22.7719
	GCT	-29.4700*	6.13298	.000	-42.2632	-16.6768
	MCT	-29.7312*	6.13298	.000	-42.5244	-16.9381
	T	-46.6788*	6.13298	.000	-59.4719	-33.8856
MCT	GC	39.7100*	6.13298	.000	26.9168	52.5032
	GCT	.2612	6.13298	.966	-12.5319	13.0544
	MC	29.7312*	6.13298	.000	16.9381	42.5244
	T	-16.9475*	6.13298	.012	-29.7407	-4.1543
T	GC	56.6575*	6.13298	.000	43.8643	69.4507
	GCT	17.2088*	6.13298	.011	4.4156	30.0019
	MC	46.6788*	6.13298	.000	33.8856	59.4719
	MCT	16.9475*	6.13298	.012	4.1543	29.7407

Based on observed means.

The error term is Mean Square (Error) = 150.454.

*. The mean difference is significant at the 0.05 level.

Al conc.
LSD: Aggregates

(I) aggregates	(J) aggregates	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	-12.0040*	5.48551	.041	-23.4466	-.5614
	NB	-15.6560*	5.48551	.010	-27.0986	-4.2134
	OB	-22.8480*	5.48551	.000	-34.2906	-11.4054
L	G	12.0040*	5.48551	.041	.5614	23.4466
	NB	-3.6520	5.48551	.513	-15.0946	7.7906
	OB	-10.8440	5.48551	.062	-22.2866	.5986
NB	G	15.6560*	5.48551	.010	4.2134	27.0986
	L	3.6520	5.48551	.513	-7.7906	15.0946
	OB	-7.1920	5.48551	.205	-18.6346	4.2506
OB	G	22.8480*	5.48551	.000	11.4054	34.2906
	L	10.8440	5.48551	.062	-.5986	22.2866
	NB	7.1920	5.48551	.205	-4.2506	18.6346

Based on observed means.
The error term is Mean Square (Error) = 150.454.
*. The mean difference is significant at the 0.05 level.

Two-way Anova test for Al concentrations in grass roots

Tests of Between-Subjects Effects
Dependent Variable: Al conc.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	9266.870[a]	19	487.730	9.963	.000
Intercept	39537.223	1	39537.223	807.660	.000
Medium	4647.307	4	1161.827	23.734	**.000**
aggregate	720.066	3	240.022	4.903	**.019**
Medium * aggregate	2695.998	12	224.666	4.589	**.007**
Error	587.434	12	48.953		
Total	54780.085	32			
Corrected Total	9854.304	31			

a. R Squared = .940 (Adjusted R Squared = .846)

Post Hoc test for Al concentrations in grass roots

Multiple Comparisons

Al conc.

LSD: Growth media

(I) Medium	(J) Medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	-15.882857*	3.7398552	.001	-24.031302	-7.734413
	MC	-7.638286	4.0968061	.087	-16.564459	1.287888
	MCT	-19.359286*	3.8925647	.000	-27.840456	-10.878116
	T	-39.157143*	3.7398552	.000	-47.305587	-31.008698
GCT	GC	15.882857*	3.7398552	.001	7.734413	24.031302
	MC	8.244571	4.0968061	.067	-.681602	17.170745
	MCT	-3.476429	3.8925647	.389	-11.957598	5.004741
	T	-23.274286*	3.7398552	.000	-31.422730	-15.125841
MC	GC	7.638286	4.0968061	.087	-1.287888	16.564459
	GCT	-8.244571	4.0968061	.067	-17.170745	.681602
	MCT	-11.721000*	4.2366689	.017	-20.951908	-2.490092
	T	-31.518857*	4.0968061	.000	-40.445031	-22.592684
MCT	GC	19.359286*	3.8925647	.000	10.878116	27.840456
	GCT	3.476429	3.8925647	.389	-5.004741	11.957598
	MC	11.721000*	4.2366689	.017	2.490092	20.951908
	T	-19.797857*	3.8925647	.000	-28.279027	-11.316687
T	GC	39.157143*	3.7398552	.000	31.008698	47.305587
	GCT	23.274286*	3.7398552	.000	15.125841	31.422730
	MC	31.518857*	4.0968061	.000	22.592684	40.445031
	MCT	19.797857*	3.8925647	.000	11.316687	28.279027

Based on observed means.

The error term is Mean Square (Error) = 48.953.

*. The mean difference is significant at the 0.05 level.

Al grass root conc.

LSD: Aggregates

(I) aggregate	(J) aggregate	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	2.982250	3.9886919	.469	-5.708363	11.672863
	NB	-.250750	3.3187922	.941	-7.481777	6.980277
	OB	9.385139*	3.3997503	.017	1.977719	16.792558
L	G	-2.982250	3.9886919	.469	-11.672863	5.708363
	NB	-3.233000	3.8322112	.415	-11.582671	5.116671
	OB	6.402889	3.9025328	.127	-2.100000	14.905778
NB	G	.250750	3.3187922	.941	-6.980277	7.481777
	L	3.233000	3.8322112	.415	-5.116671	11.582671
	OB	9.635889*	3.2147289	.011	2.631596	16.640181
OB	G	-9.385139*	3.3997503	.017	-16.792558	-1.977719
	L	-6.402889	3.9025328	.127	-14.905778	2.100000
	NB	-9.635889*	3.2147289	.011	-16.640181	-2.631596

Based on observed means.

The error term is Mean Square (Error) = 48.953

*. The mean difference is significant at the 0.05 level.

Appendix 25: Results for Cd concentrations in harvested grass shoots, growth media and grass roots

Samples	Blank corrected mean Cd concentration ($mgkg^{-1}$)						
	Bk	1st harvest	2nd harvest	3rd harvest	cumulative grass harvest	growth media	Grass root
OBGC	0.00	0.90	0.79	0.29	1.98	6.80	4.93
OBMC	0.00	1.44	1.25	0.68	3.36	9.80	6.75
OBT	0.00	0.85	1.00	0.79	2.64	1.50	8.25
OBGCT	0.00	1.03	1.26	0.86	3.15	4.08	5.05
OBMCT	0.00	1.11	1.51	0.68	3.30	1.45	5.53
NBGC	0.00	1.24	1.54	0.40	3.18	6.88	6.78
NBMC	0.00	1.48	1.64	0.79	3.90	10.98	7.15
NBT	0.01	1.31	1.26	0.90	3.49	2.15	10.50
NBGCT	0.00	1.24	1.66	0.54	3.44	2.55	6.78
NBMCT	0.00	1.20	1.38	0.55	3.13	3.23	4.53
GGC	0.00	0.46	1.50	0.40	2.36	6.05	4.35
GMC	0.00	1.85	3.71	0.60	6.16	8.88	6.65
GT	0.00	0.75	1.93	0.88	3.55	2.00	7.35
GGCT	0.00	1.10	1.56	0.56	3.23	2.58	5.48
GMCT	0.00	0.84	1.85	0.49	3.18	4.50	5.20
LGC	0.00	0.90	1.49	0.44	2.83	8.58	5.45
LMC	0.00	1.04	1.63	1.00	3.66	6.43	11.85
LT	0.05	0.58	1.21	0.73	2.56	2.35	5.65
LGCT	0.00	0.58	1.25	0.61	2.44	4.60	4.70
LMCT	0.10	0.90	1.93	0.68	3.61	2.63	6.75

Two-way Anova test for Cd concentrations in grass shoots

Tests of Between-Subjects Effects
Dependent Variable: Cd conc.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.043[a]	19	.002	100.898	.000
Intercept	.680	1	.680	30095.858	.000
Medium	.020	4	.005	221.674	.000
aggregate	.007	3	.002	101.448	.000
Medium * aggregate	.016	12	.001	60.502	.000
Error	.000	20	2.260E-5		
Total	.724	40			
Corrected Total	.044	39			

a. R Squared = .990 (Adjusted R Squared = .980)

Post Hoc test for Cd concentrations in grass shoots

Multiple Comparisons

Cd conc.

LSD: Growth media

(I) Medium	(J) Medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	-.019125*	.0023770	.000	-.024083	-.014167
	MC	-.067500*	.0023770	.000	-.072458	-.062542
	MCT	-.027625*	.0023770	.000	-.032583	-.022667
	T	-.018375*	.0023770	.000	-.023333	-.013417
GCT	GC	.019125*	.0023770	.000	.014167	.024083
	MC	-.048375*	.0023770	.000	-.053333	-.043417
	MCT	-.008500*	.0023770	.002	-.013458	-.003542
	T	.000750	.0023770	.756	-.004208	.005708
MC	GC	.067500*	.0023770	.000	.062542	.072458
	GCT	.048375*	.0023770	.000	.043417	.053333
	MCT	.039875*	.0023770	.000	.034917	.044833
	T	.049125*	.0023770	.000	.044167	.054083
MCT	GC	.027625*	.0023770	.000	.022667	.032583
	GCT	.008500*	.0023770	.002	.003542	.013458
	MC	-.039875*	.0023770	.000	-.044833	-.034917
	T	.009250*	.0023770	.001	.004292	.014208
T	GC	.018375*	.0023770	.000	.013417	.023333
	GCT	-.000750	.0023770	.756	-.005708	.004208
	MC	-.049125*	.0023770	.000	-.054083	-.044167
	MCT	-.009250*	.0023770	.001	-.014208	-.004292

Based on observed means.

The error term is Mean Square (Error) = 2.26E-005.

*. The mean difference is significant at the 0.05 level.

Cd conc.

LSD: Aggregates

(I) aggregate	(J) aggregate	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	.028300*	.0021260	.000	.023865	.032735
	NB	.010900*	.0021260	.000	.006465	.015335
	OB	.032400*	.0021260	.000	.027965	.036835
L	G	-.028300*	.0021260	.000	-.032735	-.023865
	NB	-.017400*	.0021260	.000	-.021835	-.012965
	OB	.004100	.0021260	.068	-.000335	.008535
NB	G	-.010900*	.0021260	.000	-.015335	-.006465
	L	.017400*	.0021260	.000	.012965	.021835
	OB	.021500*	.0021260	.000	.017065	.025935
OB	G	-.032400*	.0021260	.000	-.036835	-.027965
	L	-.004100	.0021260	.068	-.008535	.000335
	NB	-.021500*	.0021260	.000	-.025935	-.017065

Based on observed means. The error term is Mean Square (Error) = 2.26E-005.

*.The mean difference is significant at the 0.05 level.

Two-way Anova test for Cd concentrations in growth media

Tests of Between-Subjects Effects

Dependent Variable: Cd conc.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.134[a]	19	.007	106.278	.000
Intercept	.384	1	.384	5797.871	.000
medium	.115	4	.029	434.998	**.000**
aggregates	.000	3	.000	2.131	.128
medium * aggregates	.018	12	.002	22.741	**.000**
Error	.001	20	6.623E-5		
Total	.519	40			
Corrected Total	.135	39			

a. R Squared = .990 (Adjusted R Squared = .981)

Post Hoc test for Cd concentrations in growth media

Multiple Comparisons

Cd conc.

LSD: Growth media

(I) medium	(J) medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	.0725*	.00407	.000	.0640	.0810
	MC	-.0389*	.00407	.000	-.0474	-.0304
	MCT	.0825*	.00407	.000	.0740	.0910
	T	.1015*	.00407	.000	.0930	.1100
GCT	GC	-.0725*	.00407	.000	-.0810	-.0640
	MC	-.1114*	.00407	.000	-.1199	-.1029
	MCT	.0100*	.00407	.023	.0015	.0185
	T	.0290*	.00407	.000	.0205	.0375
MC	GC	.0389*	.00407	.000	.0304	.0474
	GCT	.1114*	.00407	.000	.1029	.1199
	MCT	.1214*	.00407	.000	.1129	.1299
	T	.1404*	.00407	.000	.1319	.1489
MCT	GC	-.0825*	.00407	.000	-.0910	-.0740
	GCT	-.0100*	.00407	.023	-.0185	-.0015
	MC	-.1214*	.00407	.000	-.1299	-.1129
	T	.0190*	.00407	.000	.0105	.0275
T	GC	-.1015*	.00407	.000	-.1100	-.0930
	GCT	-.0290*	.00407	.000	-.0375	-.0205
	MC	-.1404*	.00407	.000	-.1489	-.1319
	MCT	-.0190*	.00407	.000	-.0275	-.0105

Based on observed means.

The error term is Mean Square (Error) = 6.62E-005.

*. The mean difference is significant at the 0.05 level.

Cd conc.
LSD: Aggregates

(I) aggregates	(J) aggregates	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	-.0023	.00364	.535	-.0099	.0053
	NB	-.0071	.00364	.065	-.0147	.0005
	OB	.0015	.00364	.685	-.0061	.0091
L	G	.0023	.00364	.535	-.0053	.0099
	NB	-.0048	.00364	.202	-.0124	.0028
	OB	.0038	.00364	.309	-.0038	.0114
NB	G	.0071	.00364	.065	-.0005	.0147
	L	.0048	.00364	.202	-.0028	.0124
	OB	.0086*	.00364	.028	.0010	.0162
OB	G	-.0015	.00364	.685	-.0091	.0061
	L	-.0038	.00364	.309	-.0114	.0038
	NB	-.0086*	.00364	.028	-.0162	-.0010

Based on observed means.
The error term is Mean Square (Error) = 6.62E-005.
*. The mean difference is significant at the 0.05 level.

Two-way Anova test for Cd concentrations in grass roots

Tests of Between-Subjects Effects
Dependent Variable: Cd conc.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.043[a]	19	.002	25.715	.000
Intercept	.399	1	.399	4579.988	.000
Medium	.018	4	.004	51.170	**.000**
aggregate	.004	3	.001	14.799	**.000**
Medium * aggregate	.017	12	.001	16.219	**.000**
Error	.001	12	8.708E-5		
Total	.485	32			
Corrected Total	.044	31			

a. R Squared = .976 (Adjusted R Squared = .938)

Post Hoc test for Cd concentrations in grass roots

Multiple Comparisons

Cd conc.

LSD: Growth media

(I) Medium	(J) Medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	-.005000	.0049881	.336	-.015868	.005868
	MC	-.050914*	.0054642	.000	-.062820	-.039009
	MCT	.000452	.0051918	.932	-.010859	.011764
	T	-.058000*	.0049881	.000	-.068868	-.047132
GCT	GC	.005000	.0049881	.336	-.005868	.015868
	MC	-.045914*	.0054642	.000	-.057820	-.034009
	MCT	.005452	.0051918	.314	-.005859	.016764
	T	-.053000*	.0049881	.000	-.063868	-.042132
MC	GC	.050914*	.0054642	.000	.039009	.062820
	GCT	.045914*	.0054642	.000	.034009	.057820
	MCT	.051367*	.0056507	.000	.039055	.063679
	T	-.007086	.0054642	.219	-.018991	.004820
MCT	GC	-.000452	.0051918	.932	-.011764	.010859
	GCT	-.005452	.0051918	.314	-.016764	.005859
	MC	-.051367*	.0056507	.000	-.063679	-.039055
	T	-.058452*	.0051918	.000	-.069764	-.047141
T	GC	.058000*	.0049881	.000	.047132	.068868
	GCT	.053000*	.0049881	.000	.042132	.063868
	MC	.007086	.0054642	.219	-.004820	.018991
	MCT	.058452*	.0051918	.000	.047141	.069764

Based on observed means.

The error term is Mean Square (Error) = 8.71E-005.

*.The mean difference is significant at the 0.05 level.

Cd conc.

LSD: Aggregates

(I) aggregate	(J) aggregate	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	-.022100*	.0053200	.001	-.033691	-.010509
	NB	-.027400*	.0044265	.000	-.037044	-.017756
	OB	-.005056	.0045345	.287	-.014935	.004824
L	G	.022100*	.0053200	.001	.010509	.033691
	NB	-.005300	.0051113	.320	-.016436	.005836
	OB	.017044*	.0052051	.007	.005704	.028385
NB	G	.027400*	.0044265	.000	.017756	.037044
	L	.005300	.0051113	.320	-.005836	.016436
	OB	.022344*	.0042877	.000	.013002	.031687
OB	G	.005056	.0045345	.287	-.004824	.014935
	L	-.017044*	.0052051	.007	-.028385	-.005704
	NB	-.022344*	.0042877	.000	-.031687	-.013002

Based on observed means. The error term is Mean Square (Error) = 8.71E-005.

*.The mean difference is significant at the 0.05 level.

Appendix 26: Results for Cr concentrations in harvested grass shoots, growth media and grass roots

Samples	Blank corrected mean Cr concentration (mg/kg)						
	Bk	1st harvest	2nd harvest	3rd harvest	cumulative grass harvest	growth media	Grass root
OBGC	2.00	0.78	1.4	1.3	5.48	41.7	7.7
OBMC	0.00	0.89	2.1	1.5	4.49	42.1	7.3
OBT	0.20	0.56	1.5	1.9	4.11	38.0	9.8
OBGCT	0.18	0.64	1.5	3.4	5.65	102.9	6.8
OBMCT	0.19	0.83	2.0	2.5	5.59	29.5	7.6
NBGC	0.21	0.85	2.2	1.0	4.25	36.4	10.8
NBMC	0.39	1.03	1.8	1.0	4.16	40.0	8.8
NBT	0.45	0.68	1.5	1.7	4.31	38.5	11.7
NBGCT	0.23	0.89	2.0	1.1	4.18	61.2	9.7
NBMCT	0.63	0.78	2.1	1.6	5.13	114.5	8.1
GGC	0.46	0.19	1.9	1.4	3.99	206.0	8.1
GMC	0.15	1.03	4.2	1.6	6.91	64.4	9.7
GT	0.30	0.41	2.5	3.1	6.28	29.0	13.2
GGCT	0.28	0.88	2.3	1.4	4.84	60.4	8.3
GMCT	0.55	1.04	2.4	1.3	5.25	77.8	10.2
LGC	0.31	1.08	2.1	2.0	5.40	40.0	10.3
LMC	0.26	0.95	1.9	1.6	4.71	68.1	9.4
LT	0.15	0.25	1.8	2.0	4.21	42.1	10.3
LGCT	0.33	0.64	2.0	1.8	4.74	57.5	9.5
LMCT	0.93	0.89	2.2	1.3	5.35	33.1	7.4

Two-way Anova test for Cr concentrations in grass shoots

Tests of Between-Subjects Effects

Dependent Variable: Cr conc

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.048[a]	19	.003	56.437	.000
Intercept	.822	1	.822	18347.520	.000
Medium	.021	4	.005	116.210	**.000**
aggregate	.007	3	.002	49.937	**.000**
Medium * aggregate	.021	12	.002	38.138	**.000**
Error	.001	20	4.480E-5		
Total	.871	40			
Corrected Total	.049	39			

a. R Squared = .982 (Adjusted R Squared = .964)

Post Hoc test for Cr concentrations in grass shoots

Multiple Comparisons

Cr grass conc.

LSD: Growth media

(I) Medium	(J) Medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	-.004625	.0033466	.182	-.011606	.002356
	MC	-.044625*	.0033466	.000	-.051606	-.037644
	MCT	-.025375*	.0033466	.000	-.032356	-.018394
	T	.022250*	.0033466	.000	.015269	.029231
GCT	GC	.004625	.0033466	.182	-.002356	.011606
	MC	-.040000*	.0033466	.000	-.046981	-.033019
	MCT	-.020750*	.0033466	.000	-.027731	-.013769
	T	.026875*	.0033466	.000	.019894	.033856
MC	GC	.044625*	.0033466	.000	.037644	.051606
	GCT	.040000*	.0033466	.000	.033019	.046981
	MCT	.019250*	.0033466	.000	.012269	.026231
	T	.066875*	.0033466	.000	.059894	.073856
MCT	GC	.025375*	.0033466	.000	.018394	.032356
	GCT	.020750*	.0033466	.000	.013769	.027731
	MC	-.019250*	.0033466	.000	-.026231	-.012269
	T	.047625*	.0033466	.000	.040644	.054606
T	GC	-.022250*	.0033466	.000	-.029231	-.015269
	GCT	-.026875*	.0033466	.000	-.033856	-.019894
	MC	-.066875*	.0033466	.000	-.073856	-.059894
	MCT	-.047625*	.0033466	.000	-.054606	-.040644

Based on observed means.

The error term is Mean Square (Error) = 4.48E-005.

*.The mean difference is significant at the 0.05 level.

Cr Conc.
LSD: Aggregates

(I) aggregate	(J) aggregate	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	.021800*	.0029933	.000	.015556	.028044
	NB	.019600*	.0029933	.000	.013356	.025844
	OB	.036400*	.0029933	.000	.030156	.042644
L	G	-.021800*	.0029933	.000	-.028044	-.015556
	NB	-.002200	.0029933	.471	-.008444	.004044
	OB	.014600*	.0029933	.000	.008356	.020844
NB	G	-.019600*	.0029933	.000	-.025844	-.013356
	L	.002200	.0029933	.471	-.004044	.008444
	OB	.016800*	.0029933	.000	.010556	.023044
OB	G	-.036400*	.0029933	.000	-.042644	-.030156
	L	-.014600*	.0029933	.000	-.020844	-.008356
	NB	-.016800*	.0029933	.000	-.023044	-.010556

Based on observed means. The error term is Mean Square (Error) = 4.48E-005.
*.The mean difference is significant at the 0.05 level.

Two-way Anova test for Cr concentrations in growth media

Tests of Between-Subjects Effects
Dependent Variable: Cr conc.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	25.919[a]	19	1.364	6.865	.000
Intercept	59.817	1	59.817	301.044	.000
medium	3.627	4	.907	4.564	**.009**
aggregates	3.916	3	1.305	6.569	**.003**
medium * aggregates	18.375	12	1.531	7.707	**.000**
Error	3.974	20	.199		
Total	89.710	40			
Corrected Total	29.893	39			

a. R Squared = .867 (Adjusted R Squared = .741)

Post Hoc test for Cr concentrations in growth media

Multiple Comparisons

Cr conc.

LSD: Growth media

(I) medium	(J) medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	.2104	.22288	.356	-.2545	.6753
	MC	.5474*	.22288	.023	.0825	1.0123
	MCT	.3465	.22288	.136	-.1184	.8114
	T	.8826*	.22288	.001	.4177	1.3475
GCT	GC	-.2104	.22288	.356	-.6753	.2545
	MC	.3370	.22288	.146	-.1279	.8019
	MCT	.1361	.22288	.548	-.3288	.6010
	T	.6722*	.22288	.007	.2073	1.1372
MC	GC	-.5474*	.22288	.023	-1.0123	-.0825
	GCT	-.3370	.22288	.146	-.8019	.1279
	MCT	-.2009	.22288	.378	-.6658	.2640
	T	.3352	.22288	.148	-.1297	.8002
MCT	GC	-.3465	.22288	.136	-.8114	.1184
	GCT	-.1361	.22288	.548	-.6010	.3288
	MC	.2009	.22288	.378	-.2640	.6658
	T	.5361*	.22288	.026	.0712	1.0010
T	GC	-.8826*	.22288	.001	-1.3475	-.4177
	GCT	-.6722*	.22288	.007	-1.1372	-.2073
	MC	-.3352	.22288	.148	-.8002	.1297
	MCT	-.5361*	.22288	.026	-1.0010	-.0712

Based on observed means.

The error term is Mean Square (Error) = .199.

*. The mean difference is significant at the 0.05 level.

Cr conc.
LSD: Aggregates

(I) aggregates	(J) aggregates	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	.7868*	.19935	.001	.3710	1.2026
	NB	.5877*	.19935	.008	.1719	1.0035
	OB	.7336*	.19935	.001	.3178	1.1494
L	G	-.7868*	.19935	.001	-1.2026	-.3710
	NB	-.1991	.19935	.330	-.6149	.2167
	OB	-.0532	.19935	.792	-.4690	.3626
NB	G	-.5877*	.19935	.008	-1.0035	-.1719
	L	.1991	.19935	.330	-.2167	.6149
	OB	.1459	.19935	.473	-.2699	.5617
OB	G	-.7336*	.19935	.001	-1.1494	-.3178
	L	.0532	.19935	.792	-.3626	.4690
	NB	-.1459	.19935	.473	-.5617	.2699

Based on observed means.
The error term is Mean Square (Error) = .199.
*. The mean difference is significant at the 0.05 level.

Two-way Anova test for Cr concentrations in grass roots

Tests of Between-Subjects Effects
Dependent Variable: Cr grass root conc.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.036[a]	19	.002	3.015	.027
Intercept	.884	1	.884	1408.308	.000
Medium	.014	4	.003	5.434	.010
Aggregate	.009	3	.003	5.012	.018
Medium * aggregate	.009	12	.001	1.160	.401
Error	.008	12	.001		
Total	1.033	32			
Corrected Total	.043	31			

a. R Squared = .827 (Adjusted R Squared = .553)

Post Hoc test for Cr concentrations in grass roots

Multiple Comparisons

Cr conc.

LSD: Growth media

(I) Medium	(J) Medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	.012714	.0133902	.361	-.016460	.041889
	MC	.005486	.0146682	.715	-.026474	.037445
	MCT	.018286	.0139370	.214	-.012080	.048652
	T	-.046143*	.0133902	.005	-.075318	-.016968
GCT	GC	-.012714	.0133902	.361	-.041889	.016460
	MC	-.007229	.0146682	.631	-.039188	.024731
	MCT	.005571	.0139370	.696	-.024795	.035937
	T	-.058857*	.0133902	.001	-.088032	-.029682
MC	GC	-.005486	.0146682	.715	-.037445	.026474
	GCT	.007229	.0146682	.631	-.024731	.039188
	MCT	.012800	.0151690	.415	-.020250	.045850
	T	-.051629*	.0146682	.004	-.083588	-.019669
MCT	GC	-.018286	.0139370	.214	-.048652	.012080
	GCT	-.005571	.0139370	.696	-.035937	.024795
	MC	-.012800	.0151690	.415	-.045850	.020250
	T	-.064429*	.0139370	.001	-.094795	-.034063
T	GC	.046143*	.0133902	.005	.016968	.075318
	GCT	.058857*	.0133902	.001	.029682	.088032
	MC	.051629*	.0146682	.004	.019669	.083588
	MCT	.064429*	.0139370	.001	.034063	.094795

Based on observed means.

The error term is Mean Square (Error) = .001.

*.The mean difference is significant at the 0.05 level.

Cr conc.

LSD: Aggregates

(I) aggregate	(J) aggregate	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	.011200	.0142811	.448	-.019916	.042316
	NB	.001600	.0118826	.895	-.024290	.027490
	OB	.040667*	.0121725	.006	.014145	.067188
L	G	-.011200	.0142811	.448	-.042316	.019916
	NB	-.009600	.0137209	.497	-.039495	.020295
	OB	.029467	.0139727	.057	-.000977	.059910
NB	G	-.001600	.0118826	.895	-.027490	.024290
	L	.009600	.0137209	.497	-.020295	.039495
	OB	.039067*	.0115100	.005	.013988	.064145
OB	G	-.040667*	.0121725	.006	-.067188	-.014145
	L	-.029467	.0139727	.057	-.059910	.000977
	NB	-.039067*	.0115100	.005	-.064145	-.013988

Based on observed means.

The error term is Mean Square (Error) = .001.

*.The mean difference is significant at the 0.05 level.

Appendix 27: Results for Cu concentrations in harvested grass shoots, growth media and grass roots

Samples	Blank corrected mean Cu concentration (mg/kg)						
	bk	1st harvest	2nd harvest	3rd harvest	cumulative grass harvest	growth media	Grass root
OBGC	17.81	9.36	7.11	6.19	40.48	84.83	1007.25
OBMC	9.54	8.69	7.33	7.51	33.06	58.75	41.00
OBT	19.06	16.39	12.70	10.95	59.10	207.23	158.45
OBGCT	11.85	12.06	9.46	17.50	50.88	170.05	109.95
OBMCT	12.65	12.91	12.30	13.45	51.31	175.20	125.70
NBGC	8.65	8.96	8.84	7.20	33.65	91.15	59.50
NBMC	8.73	9.08	9.24	8.83	35.86	77.50	38.55
NBT	16.34	13.71	11.55	13.63	55.23	194.28	143.55
NBGCT	11.84	10.14	11.38	9.79	43.14	331.63	127.78
NBMCT	14.00	10.86	11.65	8.75	45.26	169.33	112.90
GGC	10.50	5.11	8.81	8.01	32.44	71.95	47.83
GMC	8.20	8.79	16.06	7.36	40.41	64.93	46.10
GT	14.98	12.39	11.15	14.58	53.09	335.78	143.80
GGCT	12.36	12.08	9.75	9.56	43.75	125.78	108.08
GMCT	13.14	12.24	10.45	8.24	44.06	1783.55	61.40
LGC	9.48	8.00	7.98	8.76	34.21	89.58	63.40
LMC	9.31	7.51	7.41	8.66	32.90	63.10	44.80
LT	12.31	11.96	9.49	11.14	44.90	150.90	124.10
LGCT	12.09	10.16	10.46	14.33	47.04	140.63	109.35
LMCT	18.94	11.66	10.61	12.01	53.23	150.83	107.60

Two-way Anova test for Cu concentrations in grass shoots

Tests of Between-Subjects Effects

Dependent Variable: Cu conc.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	2.242[a]	19	.118	13.928	.000
Intercept	61.996	1	61.996	7317.453	.000
Medium	1.702	4	.425	50.221	**.000**
Aggregate	.066	3	.022	2.616	.079
medium * Aggregate	.474	12	.039	4.659	**.001**
Error	.169	20	.008		
Total	64.408	40			
Corrected Total	2.412	39			

a. R Squared = .930 (Adjusted R Squared = .863)

Post Hoc test for Cu concentrations in grass shoots

Multiple Comparisons

Cu conc.

LSD: Growth media

(I) medium	(J) medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	-.423250*	.0460227	.000	-.519252	-.327248
	MC	-.121250*	.0460227	.016	-.217252	-.025248
	MCT	-.408000*	.0460227	.000	-.504002	-.311998
	T	-.552875*	.0460227	.000	-.648877	-.456873
GCT	GC	.423250*	.0460227	.000	.327248	.519252
	MC	.302000*	.0460227	.000	.205998	.398002
	MCT	.015250	.0460227	.744	-.080752	.111252
	T	-.129625*	.0460227	.011	-.225627	-.033623
MC	GC	.121250*	.0460227	.016	.025248	.217252
	GCT	-.302000*	.0460227	.000	-.398002	-.205998
	MCT	-.286750*	.0460227	.000	-.382752	-.190748
	T	-.431625*	.0460227	.000	-.527627	-.335623
MCT	GC	.408000*	.0460227	.000	.311998	.504002
	GCT	-.015250	.0460227	.744	-.111252	.080752
	MC	.286750*	.0460227	.000	.190748	.382752
	T	-.144875*	.0460227	.005	-.240877	-.048873
T	GC	.552875*	.0460227	.000	.456873	.648877
	GCT	.129625*	.0460227	.011	.033623	.225627
	MC	.431625*	.0460227	.000	.335623	.527627
	MCT	.144875*	.0460227	.005	.048873	.240877

Based on observed means.

The error term is Mean Square (Error) = .008.

*.The mean difference is significant at the 0.05 level.

Cu conc.
LSD: Aggregates

(I) Aggregate	(J) Aggregate	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	.035400	.0411639	.400	-.050466	.121266
	NB	.007900	.0411639	.850	-.077966	.093766
	OB	-.074700	.0411639	.085	-.160566	.011166
L	G	-.035400	.0411639	.400	-.121266	.050466
	NB	-.027500	.0411639	.512	-.113366	.058366
	OB	-.110100*	.0411639	.015	-.195966	-.024234
NB	G	-.007900	.0411639	.850	-.093766	.077966
	L	.027500	.0411639	.512	-.058366	.113366
	OB	-.082600	.0411639	.059	-.168466	.003266
OB	G	.074700	.0411639	.085	-.011166	.160566
	L	.110100*	.0411639	.015	.024234	.195966
	NB	.082600	.0411639	.059	-.003266	.168466

Based on observed means. The error term is Mean Square (Error) = .008.
*.The mean difference is significant at the 0.05 level.

Two-way Anova test for Cu concentrations in growth media

Tests of Between-Subjects Effects
Dependent Variable: Cu conc.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	2136.821[a]	19	112.464	126.517	.000
Intercept	823.348	1	823.348	926.228	.000
medium	527.838	4	131.959	148.448	**.000**
aggregates	338.033	3	112.678	126.757	**.000**
medium * aggregates	1270.951	12	105.913	119.147	**.000**
Error	17.779	20	.889		
Total	2977.947	40			
Corrected Total	2154.600	39			

a. R Squared = .992 (Adjusted R Squared = .984)

Post Hoc test for Cu concentrations in growth media

Multiple Comparisons

Cu conc.

LSD: Growth media

(I) medium	(J) medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	-2.1529*	.47141	.000	-3.1362	-1.1695
	MC	.3661	.47141	.446	-.6172	1.3495
	MCT	-9.7070*	.47141	.000	-10.6904	-8.7236
	T	-2.7534*	.47141	.000	-3.7367	-1.7700
GCT	GC	2.1529*	.47141	.000	1.1695	3.1362
	MC	2.5190*	.47141	.000	1.5356	3.5024
	MCT	-7.5541*	.47141	.000	-8.5375	-6.5708
	T	-.6005	.47141	.217	-1.5839	.3829
MC	GC	-.3661	.47141	.446	-1.3495	.6172
	GCT	-2.5190*	.47141	.000	-3.5024	-1.5356
	MCT	-10.0731*	.47141	.000	-11.0565	-9.0898
	T	-3.1195*	.47141	.000	-4.1029	-2.1361
MCT	GC	9.7070*	.47141	.000	8.7236	10.6904
	GCT	7.5541*	.47141	.000	6.5708	8.5375
	MC	10.0731*	.47141	.000	9.0898	11.0565
	T	6.9536*	.47141	.000	5.9703	7.9370
T	GC	2.7534*	.47141	.000	1.7700	3.7367
	GCT	.6005	.47141	.217	-.3829	1.5839
	MC	3.1195*	.47141	.000	2.1361	4.1029
	MCT	-6.9536*	.47141	.000	-7.9370	-5.9703

Based on observed means.

The error term is Mean Square (Error) = .889.

*. The mean difference is significant at the 0.05 level.

Cu conc.
LSD: Aggregates

(I) aggregates	(J) aggregates	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	7.1478*	.42165	.000	6.2683	8.0273
	NB	6.0724*	.42165	.000	5.1929	6.9519
	OB	6.7437*	.42165	.000	5.8642	7.6232
L	G	-7.1478*	.42165	.000	-8.0273	-6.2683
	NB	-1.0754*	.42165	.019	-1.9549	-.1959
	OB	-.4041	.42165	.349	-1.2836	.4754
NB	G	-6.0724*	.42165	.000	-6.9519	-5.1929
	L	1.0754*	.42165	.019	.1959	1.9549
	OB	.6713	.42165	.127	-.2082	1.5508
OB	G	-6.7437*	.42165	.000	-7.6232	-5.8642
	L	.4041	.42165	.349	-.4754	1.2836
	NB	-.6713	.42165	.127	-1.5508	.2082

Based on observed means.
The error term is Mean Square (Error) = .889.
*. The mean difference is significant at the 0.05 level.

Two-way Anova test for Cu concentrations in grass roots

Tests of Between-Subjects Effects
Dependent Variable: Cu conc.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	638.470[a]	19	33.604	2.468	.056
Intercept	219.158	1	219.158	16.093	.002
medium	82.829	4	20.707	1.521	.258
Aggregate	95.659	3	31.886	2.341	.125
medium * Aggregate	393.725	12	32.810	2.409	.071
Error	163.417	12	13.618		
Total	1107.673	32			
Corrected Total	801.887	31			

a. R Squared = .796 (Adjusted R Squared = .474)

Post Hoc test for Cu concentrations in grass roots

Multiple Comparisons

Cu conc.

LSD: Growth media

(I) medium	(J) medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	4.261714	1.9725313	.052	-.036062	8.559491
	MC	5.714143*	2.1607997	.021	1.006165	10.422121
	MCT	4.396143	2.0530756	.053	-.077125	8.869410
	T	3.648143	1.9725313	.089	-.649634	7.945919
GCT	GC	-4.261714	1.9725313	.052	-8.559491	.036062
	MC	1.452429	2.1607997	.514	-3.255550	6.160407
	MCT	.134429	2.0530756	.949	-4.338839	4.607696
	T	-.613571	1.9725313	.761	-4.911348	3.684205
MC	GC	-5.714143*	2.1607997	.021	-10.422121	-1.006165
	GCT	-1.452429	2.1607997	.514	-6.160407	3.255550
	MCT	-1.318000	2.2345683	.566	-6.186706	3.550706
	T	-2.066000	2.1607997	.358	-6.773978	2.641978
MCT	GC	-4.396143	2.0530756	.053	-8.869410	.077125
	GCT	-.134429	2.0530756	.949	-4.607696	4.338839
	MC	1.318000	2.2345683	.566	-3.550706	6.186706
	T	-.748000	2.0530756	.722	-5.221268	3.725268
T	GC	-3.648143	1.9725313	.089	-7.945919	.649634
	GCT	.613571	1.9725313	.761	-3.684205	4.911348
	MC	2.066000	2.1607997	.358	-2.641978	6.773978
	MCT	.748000	2.0530756	.722	-3.725268	5.221268

Based on observed means.

The error term is Mean Square (Error) = 13.618.

*.The mean difference is significant at the 0.05 level.

Cu conc.

LSD: Aggregates

(I) Aggregate	(J) Aggregate	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	-.029750	2.1037765	.989	-4.613485	4.553985
	NB	-.161850	1.7504479	.928	-3.975748	3.652048
	OB	-4.552083*	1.7931480	.026	-8.459017	-.645150
L	G	.029750	2.1037765	.989	-4.553985	4.613485
	NB	-.132100	2.0212431	.949	-4.536010	4.271810
	OB	-4.522333*	2.0583332	.048	-9.007056	-.037611
NB	G	.161850	1.7504479	.928	-3.652048	3.975748
	L	.132100	2.0212431	.949	-4.271810	4.536010
	OB	-4.390233*	1.6955612	.024	-8.084544	-.695923
OB	G	4.552083*	1.7931480	.026	.645150	8.459017
	L	4.522333*	2.0583332	.048	.037611	9.007056
	NB	4.390233*	1.6955612	.024	.695923	8.084544

Based on observed means.

The error term is Mean Square (Error) = 13.618.

*.The mean difference is significant at the 0.05 level.

Appendix 28: Results for Fe concentrations in harvested grass shoots, growth media and grass roots

Samples	Blank corrected Mean Fe concentrations (mg/kg)						
	bk	1st harvest	2nd harvest	3rd harvest	cumulative grass harvest	growth media	Grass root
OBGC	369.71	119.24	63.88	78.31	631.14	12370.00	1325.25
OBMC	96.26	76.50	73.35	71.34	317.45	6687.50	1304.00
OBT	207.30	95.00	148.54	116.71	567.55	16022.50	3007.00
OBGCT	136.41	69.49	58.69	1228.61	1493.20	14600.00	1728.00
OBMCT	144.66	74.26	55.39	409.11	683.43	11747.50	2356.50
NBGC	128.68	72.55	57.53	85.76	344.51	9262.50	2499.50
NBMC	150.46	109.28	72.61	94.84	427.19	10915.00	1867.25
NBT	155.68	60.34	78.73	212.10	506.84	12507.50	3738.00
NBGCT	134.86	72.00	81.75	127.96	416.58	12170.00	2570.25
NBMCT	195.35	79.71	75.26	608.31	958.64	11412.50	2955.25
GGC	216.66	52.01	102.69	117.31	488.68	9370.00	1741.00
GMC	132.66	86.45	186.91	183.80	589.83	8030.00	1869.50
GT	191.81	70.11	100.86	426.11	788.90	12542.50	4038.75
GGCT	142.36	181.39	88.71	187.75	600.21	10967.50	2212.25
GMCT	309.89	153.21	74.73	127.91	665.74	11810.00	2619.00
LGC	150.40	84.10	75.36	223.69	533.55	10407.50	2744.00
LMC	191.00	97.29	114.35	183.54	586.18	10352.50	1677.50
LT	93.94	142.53	64.68	122.33	423.46	12260.00	2758.50
LGCT	185.09	141.40	97.31	166.06	589.86	10907.50	2840.50
LMCT	429.74	150.84	73.41	119.99	773.98	11877.50	1986.00

Two-way Anova test for Fe concentrations in grass shoot

Tests of Between-Subjects Effects

Dependent Variable: Fe conc.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	3960.652[a]	19	208.455	8.200	.000
Intercept	12186.802	1	12186.802	479.384	.000
Medium	942.519	4	235.630	9.269	**.000**
Aggregate	320.363	3	106.788	4.201	**.019**
Medium * Aggregate	2697.769	12	224.814	8.843	**.000**
Error	508.436	20	25.422		
Total	16655.889	40			
Corrected Total	4469.088	39			

a. R Squared = .886 (Adjusted R Squared = .778)

Post Hoc test for Fe concentrations in growth media

Multiple Comparisons

Fe grass conc.

LSD: Growth media

(I) Medium	(J) Medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	-13.687000*	2.5210023	.000	-18.945719	-8.428281
	MC	-2.178250	2.5210023	.398	-7.436969	3.080469
	MCT	-8.694000*	2.5210023	.003	-13.952719	-3.435281
	T	-5.056000	2.5210023	.059	-10.314719	.202719
GCT	GC	13.687000*	2.5210023	.000	8.428281	18.945719
	MC	11.508750*	2.5210023	.000	6.250031	16.767469
	MCT	4.993000	2.5210023	.062	-.265719	10.251719
	T	8.631000*	2.5210023	.003	3.372281	13.889719
MC	GC	2.178250	2.5210023	.398	-3.080469	7.436969
	GCT	-11.508750*	2.5210023	.000	-16.767469	-6.250031
	MCT	-6.515750*	2.5210023	.018	-11.774469	-1.257031
	T	-2.877750	2.5210023	.267	-8.136469	2.380969
MCT	GC	8.694000*	2.5210023	.003	3.435281	13.952719
	GCT	-4.993000	2.5210023	.062	-10.251719	.265719
	MC	6.515750*	2.5210023	.018	1.257031	11.774469
	T	3.638000	2.5210023	.164	-1.620719	8.896719
T	GC	5.056000	2.5210023	.059	-.202719	10.314719
	GCT	-8.631000*	2.5210023	.003	-13.889719	-3.372281
	MC	2.877750	2.5210023	.267	-2.380969	8.136469
	MCT	-3.638000	2.5210023	.164	-8.896719	1.620719

Based on observed means.

The error term is Mean Square (Error) = 25.422.

*.The mean difference is significant at the 0.05 level.

Fe conc.

LSD: Aggregates

(I) Aggregate	(J) Aggregate	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	2.267300	2.2548530	.327	-2.436241	6.970841
	NB	2.009900	2.2548530	.383	-2.693641	6.713441
	OB	-4.787600*	2.2548530	.046	-9.491141	-.084059
L	G	-2.267300	2.2548530	.327	-6.970841	2.436241
	NB	-.257400	2.2548530	.910	-4.960941	4.446141
	OB	-7.054900*	2.2548530	.005	-11.758441	-2.351359
NB	G	-2.009900	2.2548530	.383	-6.713441	2.693641
	L	.257400	2.2548530	.910	-4.446141	4.960941
	OB	-6.797500*	2.2548530	.007	-11.501041	-2.093959
OB	G	4.787600*	2.2548530	.046	.084059	9.491141
	L	7.054900*	2.2548530	.005	2.351359	11.758441
	NB	6.797500*	2.2548530	.007	2.093959	11.501041

Based on observed means.

The error term is Mean Square (Error) = 25.422.

*.The mean difference is significant at the 0.05 level.

Two-way Anova test for Fe concentrations in growth media

Tests of Between-Subjects Effects

Dependent Variable: Fe conc.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	65418.304[a]	19	3443.069	7.403	.000
Intercept	2047019.536	1	2047019.536	4401.237	.000
Medium	35998.096	4	8999.524	19.350	**.000**
Aggregates	6254.982	3	2084.994	4.483	**.015**
medium * aggregates	23165.225	12	1930.435	4.151	**.003**
Error	9302.020	20	465.101		
Total	2121739.860	40			
Corrected Total	74720.324	39			

a. R Squared = .876 (Adjusted R Squared = .757)

Post Hoc test for Fe concentrations in growth media

Multiple Comparisons

Fe conc.

LSD: Growth media

(I) medium	(J) medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	-36.1750*	10.78310	.003	-58.6682	-13.6818
	MC	27.1250*	10.78310	.021	4.6318	49.6182
	MCT	-27.1875*	10.78310	.020	-49.6807	-4.6943
	T	-59.6125*	10.78310	.000	-82.1057	-37.1193
GCT	GC	36.1750*	10.78310	.003	13.6818	58.6682
	MC	63.3000*	10.78310	.000	40.8068	85.7932
	MCT	8.9875	10.78310	.414	-13.5057	31.4807
	T	-23.4375*	10.78310	.042	-45.9307	-.9443
MC	GC	-27.1250*	10.78310	.021	-49.6182	-4.6318
	GCT	-63.3000*	10.78310	.000	-85.7932	-40.8068
	MCT	-54.3125*	10.78310	.000	-76.8057	-31.8193
	T	-86.7375*	10.78310	.000	-109.2307	-64.2443
MCT	GC	27.1875*	10.78310	.020	4.6943	49.6807
	GCT	-8.9875	10.78310	.414	-31.4807	13.5057
	MC	54.3125*	10.78310	.000	31.8193	76.8057
	T	-32.4250*	10.78310	.007	-54.9182	-9.9318
T	GC	59.6125*	10.78310	.000	37.1193	82.1057
	GCT	23.4375*	10.78310	.042	.9443	45.9307
	MC	86.7375*	10.78310	.000	64.2443	109.2307
	MCT	32.4250*	10.78310	.007	9.9318	54.9182

Based on observed means.

The error term is Mean Square (Error) = 465.101.

*. The mean difference is significant at the 0.05 level.

Fe conc.

LSD: Aggregates

(I) aggregates	(J) aggregates	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	-12.3400	9.64470	.215	-32.4585	7.7785
	NB	-14.1900	9.64470	.157	-34.3085	5.9285
	OB	-34.8300*	9.64470	.002	-54.9485	-14.7115
L	G	12.3400	9.64470	.215	-7.7785	32.4585
	NB	-1.8500	9.64470	.850	-21.9685	18.2685
	OB	-22.4900*	9.64470	.030	-42.6085	-2.3715
NB	G	14.1900	9.64470	.157	-5.9285	34.3085
	L	1.8500	9.64470	.850	-18.2685	21.9685
	OB	-20.6400*	9.64470	.045	-40.7585	-.5215
OB	G	34.8300*	9.64470	.002	14.7115	54.9485
	L	22.4900*	9.64470	.030	2.3715	42.6085
	NB	20.6400*	9.64470	.045	.5215	40.7585

Based on observed means.

The error term is Mean Square (Error) = 465.101.

*. The mean difference is significant at the 0.05 level.

Two-way Anova test for Fe concentrations in grass roots

Tests of Between-Subjects Effects

Dependent Variable: Fe conc.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	7094.104[a]	19	373.374	4.899	.004
Intercept	65326.219	1	65326.219	857.158	.000
Medium	3712.324	4	928.081	12.178	**.000**
Aggregate	1143.974	3	381.325	5.003	**.018**
Medium * Aggregate	1130.614	12	94.218	1.236	.360
Error	914.551	12	76.213		
Total	83752.034	32			
Corrected Total	8008.655	31			

a. R Squared = .886 (Adjusted R Squared = .705)

Post Hoc test for Fe concentrations in grass roots

Multiple Comparisons

Fe conc.

LSD: Growth media

(I) Medium	(J) Medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	-5.674286	4.6663708	.247	-15.841434	4.492863
	MC	5.302286	5.1117531	.320	-5.835268	16.439839
	MCT	-11.117381*	4.8569127	.041	-21.699685	-.535077
	T	-29.858571*	4.6663708	.000	-40.025720	-19.691423
GCT	GC	5.674286	4.6663708	.247	-4.492863	15.841434
	MC	10.976571	5.1117531	.053	-.160982	22.114125
	MCT	-5.443095	4.8569127	.284	-16.025399	5.139209
	T	-24.184286*	4.6663708	.000	-34.351434	-14.017137
MC	GC	-5.302286	5.1117531	.320	-16.439839	5.835268
	GCT	-10.976571	5.1117531	.053	-22.114125	.160982
	MCT	-16.419667*	5.2862657	.009	-27.937450	-4.901883
	T	-35.160857*	5.1117531	.000	-46.298410	-24.023304
MCT	GC	11.117381*	4.8569127	.041	.535077	21.699685
	GCT	5.443095	4.8569127	.284	-5.139209	16.025399
	MC	16.419667*	5.2862657	.009	4.901883	27.937450
	T	-18.741190*	4.8569127	.002	-29.323494	-8.158887
T	GC	29.858571*	4.6663708	.000	19.691423	40.025720
	GCT	24.184286*	4.6663708	.000	14.017137	34.351434
	MC	35.160857*	5.1117531	.000	24.023304	46.298410
	MCT	18.741190*	4.8569127	.002	8.158887	29.323494

Based on observed means.

The error term is Mean Square (Error) = 76.213.

*.The mean difference is significant at the 0.05 level.

Fe conc.

LSD: Aggregates

(I) Aggregate	(J) Aggregate	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	3.155250	4.9768546	.538	-7.688385	13.998885
	NB	-3.339750	4.1409933	.436	-12.362199	5.682699
	OB	10.875694*	4.2420080	.025	1.633153	20.118236
L	G	-3.155250	4.9768546	.538	-13.998885	7.688385
	NB	-6.495000	4.7816072	.199	-16.913227	3.923227
	OB	7.720444	4.8693504	.139	-2.888959	18.329848
NB	G	3.339750	4.1409933	.436	-5.682699	12.362199
	L	6.495000	4.7816072	.199	-3.923227	16.913227
	OB	14.215444*	4.0111492	.004	5.475901	22.954988
OB	G	-10.875694*	4.2420080	.025	-20.118236	-1.633153
	L	-7.720444	4.8693504	.139	-18.329848	2.888959
	NB	-14.215444*	4.0111492	.004	-22.954988	-5.475901

Based on observed means.

The error term is Mean Square (Error) = 76.213.

*.The mean difference is significant at the 0.05 level.

Appendix 29: Results for Mn concentrations in harvested grass shoots, growth media and grass roots

Samples	Blank corrected mean Mn concentration (mg/kg)						
	Bk	1st harvest	2nd harvest	3rd harvest	cumulative grass harvest	growth media	Grass root
OBGC	102.54	152.75	194.56	118.34	568.19	341.55	177.30
OBMC	40.91	96.39	199.20	102.50	439.00	311.53	128.15
OBT	22.50	28.66	43.70	43.09	137.95	387.78	101.50
OBGCT	35.03	43.31	65.40	80.30	224.04	384.23	73.83
OBMCT	34.85	47.93	61.53	55.70	200.00	359.25	94.00
NBGC	112.64	163.65	226.74	137.01	640.04	323.95	233.98
NBMC	39.83	55.11	113.55	81.65	290.14	395.75	193.15
NBT	20.59	34.89	71.94	62.76	190.18	414.20	156.35
NBGCT	32.05	46.09	73.58	54.71	206.43	336.20	132.95
NBMCT	38.35	55.05	68.39	65.56	227.35	348.13	139.38
GGC	116.96	91.69	200.75	111.43	520.83	284.00	197.98
GMC	40.65	61.85	244.96	82.55	430.01	355.98	172.50
GT	28.00	34.40	50.31	48.40	161.11	392.70	145.10
GGCT	35.41	48.70	58.44	47.20	189.75	361.73	101.20
GMCT	44.64	46.16	52.83	37.99	181.61	409.73	195.40
LGC	104.13	156.06	206.85	118.64	585.68	336.08	218.05
LMC	41.48	67.51	142.54	65.93	317.45	384.68	167.20
LT	28.91	34.24	37.84	40.70	141.69	455.13	115.20
LGCT	34.86	36.76	51.74	37.29	160.65	417.73	126.25
LMCT	57.91	43.24	55.20	30.35	186.70	416.63	99.80

Two-way Anova test for Mn concentrations in grass shoots

Tests of Between-Subjects Effects

Dependent Variable: Mn conc.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	1191.142[a]	19	62.692	549.722	.000
Intercept	3978.509	1	3978.509	34886.180	.000
Medium	1089.338	4	272.335	2388.008	**.000**
Aggregate	17.463	3	5.821	51.043	**.000**
Medium * Aggregate	84.341	12	7.028	61.630	**.000**
Error	2.281	20	.114		
Total	5171.932	40			
Corrected Total	1193.423	39			

a. R Squared = .998 (Adjusted R Squared = .996)

Post Hoc test for Mn concentrations in grass shoots

Multiple Comparisons

Mn conc.

LSD: Growth media

(I) Medium	(J) Medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	12.349500*	.1688509	.000	11.997283	12.701717
	MC	5.647250*	.1688509	.000	5.295033	5.999467
	MCT	12.585500*	.1688509	.000	12.233283	12.937717
	T	13.475375*	.1688509	.000	13.123158	13.827592
GCT	GC	-12.349500*	.1688509	.000	-12.701717	-11.997283
	MC	-6.702250*	.1688509	.000	-7.054467	-6.350033
	MCT	.236000	.1688509	.178	-.116217	.588217
	T	1.125875*	.1688509	.000	.773658	1.478092
MC	GC	-5.647250*	.1688509	.000	-5.999467	-5.295033
	GCT	6.702250*	.1688509	.000	6.350033	7.054467
	MCT	6.938250*	.1688509	.000	6.586033	7.290467
	T	7.828125*	.1688509	.000	7.475908	8.180342
MCT	GC	-12.585500*	.1688509	.000	-12.937717	-12.233283
	GCT	-.236000	.1688509	.178	-.588217	.116217
	MC	-6.938250*	.1688509	.000	-7.290467	-6.586033
	T	.889875*	.1688509	.000	.537658	1.242092
T	GC	-13.475375*	.1688509	.000	-13.827592	-13.123158
	GCT	-1.125875*	.1688509	.000	-1.478092	-.773658
	MC	-7.828125*	.1688509	.000	-8.180342	-7.475908
	MCT	-.889875*	.1688509	.000	-1.242092	-.537658

Based on observed means.

The error term is Mean Square (Error) = .114.

*.The mean difference is significant at the 0.05 level.

Mn conc.
LSD: Aggregates

(I) Aggregate	(J) Aggregate	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	.742200*	.1510249	.000	.427168	1.057232
	NB	-.744200*	.1510249	.000	-1.059232	-.429168
	OB	-.925600*	.1510249	.000	-1.240632	-.610568
L	G	-.742200*	.1510249	.000	-1.057232	-.427168
	NB	-1.486400*	.1510249	.000	-1.801432	-1.171368
	OB	-1.667800*	.1510249	.000	-1.982832	-1.352768
NB	G	.744200*	.1510249	.000	.429168	1.059232
	L	1.486400*	.1510249	.000	1.171368	1.801432
	OB	-.181400	.1510249	.244	-.496432	.133632
OB	G	.925600*	.1510249	.000	.610568	1.240632
	L	1.667800*	.1510249	.000	1.352768	1.982832
	NB	.181400	.1510249	.244	-.133632	.496432

Based on observed means.
The error term is Mean Square (Error) = .114.
*.The mean difference is significant at the 0.05 level.

Two-way Anova test for Mn concentrations in growth media

Tests of Between-Subjects Effects
Dependent Variable: Mn growth media conc.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	27.220[a]	19	1.433	5.287	.000
Intercept	2200.416	1	2200.416	8120.052	.000
medium	14.177	4	3.544	13.079	**.000**
aggregates	5.284	3	1.761	6.500	**.003**
medium * aggregates	7.758	12	.647	2.386	**.041**
Error	5.420	20	.271		
Total	2233.056	40			
Corrected Total	32.640	39			

a. R Squared = .834 (Adjusted R Squared = .676)

Post Hoc test for Mn concentrations in growth media

Multiple Comparisons

Mn conc.

LSD: growth media

(I) medium	(J) medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	-1.0715*	.26028	.001	-1.6144	-.5286
	MC	-.8118*	.26028	.005	-1.3547	-.2688
	MCT	-1.2408*	.26028	.000	-1.7837	-.6978
	T	-1.8211*	.26028	.000	-2.3641	-1.2782
GCT	GC	1.0715*	.26028	.001	.5286	1.6144
	MC	.2598	.26028	.330	-.2832	.8027
	MCT	-.1692	.26028	.523	-.7122	.3737
	T	-.7496*	.26028	.009	-1.2926	-.2067
MC	GC	.8118*	.26028	.005	.2688	1.3547
	GCT	-.2598	.26028	.330	-.8027	.2832
	MCT	-.4290	.26028	.115	-.9719	.1139
	T	-1.0094*	.26028	.001	-1.5523	-.4664
MCT	GC	1.2408*	.26028	.000	.6978	1.7837
	GCT	.1692	.26028	.523	-.3737	.7122
	MC	.4290	.26028	.115	-.1139	.9719
	T	-.5804*	.26028	.037	-1.1233	-.0374
T	GC	1.8211*	.26028	.000	1.2782	2.3641
	GCT	.7496*	.26028	.009	.2067	1.2926
	MC	1.0094*	.26028	.001	.4664	1.5523
	MCT	.5804*	.26028	.037	.0374	1.1233

Based on observed means.

The error term is Mean Square (Error) = .271.

*. The mean difference is significant at the 0.05 level.

Mn conc.

LSD: Aggregates

(I) aggregates	(J) aggregates	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	-.8244*	.23280	.002	-1.3100	-.3388
	NB	-.0564	.23280	.811	-.5420	.4292
	OB	.0792	.23280	.737	-.4064	.5648
L	G	.8244*	.23280	.002	.3388	1.3100
	NB	.7680*	.23280	.004	.2824	1.2536
	OB	.9036*	.23280	.001	.4180	1.3892
NB	G	.0564	.23280	.811	-.4292	.5420
	L	-.7680*	.23280	.004	-1.2536	-.2824
	OB	.1356	.23280	.567	-.3500	.6212
OB	G	-.0792	.23280	.737	-.5648	.4064
	L	-.9036*	.23280	.001	-1.3892	-.4180
	NB	-.1356	.23280	.567	-.6212	.3500

Based on observed means.

The error term is Mean Square (Error) = .271.

*. The mean difference is significant at the 0.05 level.

Two-way Anova test for Mn concentrations in grass roots

Tests of Between-Subjects Effects

Dependent Variable: Mn conc.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	25.699[a]	19	1.353	7.059	.001
Intercept	251.729	1	251.729	1313.701	.000
Medium	14.795	4	3.699	19.302	**.000**
Aggregate	6.368	3	2.123	11.077	**.001**
Medium * Aggregate	2.586	12	.215	1.125	.421
Error	2.299	12	.192		
Total	305.812	32			
Corrected Total	27.998	31			

a. R Squared = .918 (Adjusted R Squared = .788)

Post Hoc test for Mn concentrations in grass roots

Multiple Comparisons

Mn conc

LSD: Growth media

(I) Medium	(J) Medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	1.983857*	.2339829	.000	1.474052	2.493662
	MC	.687829*	.2563154	.020	.129365	1.246292
	MCT	1.564595*	.2435371	.000	1.033973	2.095217
	T	1.472714*	.2339829	.000	.962909	1.982519
GCT	GC	-1.983857*	.2339829	.000	-2.493662	-1.474052
	MC	-1.296029*	.2563154	.000	-1.854492	-.737565
	MCT	-.419262	.2435371	.111	-.949884	.111360
	T	-.511143*	.2339829	.049	-1.020948	-.001338
MC	GC	-.687829*	.2563154	.020	-1.246292	-.129365
	GCT	1.296029*	.2563154	.000	.737565	1.854492
	MCT	.876767*	.2650659	.006	.299238	1.454296
	T	.784886*	.2563154	.010	.226422	1.343349
MCT	GC	-1.564595*	.2435371	.000	-2.095217	-1.033973
	GCT	.419262	.2435371	.111	-.111360	.949884
	MC	-.876767*	.2650659	.006	-1.454296	-.299238
	T	-.091881	.2435371	.713	-.622503	.438741
T	GC	-1.472714*	.2339829	.000	-1.982519	-.962909
	GCT	.511143*	.2339829	.049	.001338	1.020948
	MC	-.784886*	.2563154	.010	-1.343349	-.226422
	MCT	.091881	.2435371	.713	-.438741	.622503

Based on observed means.

The error term is Mean Square (Error) = .192.

*.The mean difference is significant at the 0.05 level.

Mn conc.

LSD: Aggregates

(I) Aggregate	(J) Aggregate	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	.235125	.2495513	.365	-.308601	.778851
	NB	-.282075	.2076392	.199	-.734482	.170332
	OB	.871347*	.2127043	.001	.407904	1.334790
L	G	-.235125	.2495513	.365	-.778851	.308601
	NB	-.517200	.2397611	.052	-1.039595	.005195
	OB	.636222*	.2441608	.023	.104242	1.168203
NB	G	.282075	.2076392	.199	-.170332	.734482
	L	.517200	.2397611	.052	-.005195	1.039595
	OB	1.153422*	.2011285	.000	.715201	1.591644
OB	G	-.871347*	.2127043	.001	-1.334790	-.407904
	L	-.636222*	.2441608	.023	-1.168203	-.104242
	NB	-1.153422*	.2011285	.000	-1.591644	-.715201

Based on observed means. The error term is Mean Square (Error) = .192.

*.The mean difference is significant at the 0.05 level.

Appendix 30: Results for Ni concentrations in harvested grass shoots, growth media and grass roots

Samples	Blank corrected mean Ni concentration (mg/kg)						
	Bk	1st harvest	2nd harvest	3rd harvest	cumulative grass harvest	growth media	Grass root
OBGC	9.08	1.41	1.44	1.76	13.69	33.83	6.6
OBMC	0.98	2.09	3.19	3.04	9.29	23.23	7.3
OBT	2.79	4.25	6.53	6.20	19.76	23.58	15.8
OBGCT	1.23	1.98	3.21	4.70	11.11	29.13	10.8
OBMCT	3.16	3.15	4.76	5.35	16.43	21.28	14.6
NBGC	0.70	1.53	2.36	1.33	5.91	22.55	9.8
NBMC	0.50	1.93	2.86	2.91	8.20	23.25	8.6
NBT	2.81	5.18	8.56	8.46	25.01	25.43	18.3
NBGCT	1.13	2.34	4.31	2.95	10.73	23.15	13.9
NBMCT	2.11	3.11	4.33	4.60	14.15	25.78	13.5
GGC	0.85	0.59	5.06	3.38	9.88	32.58	7.4
GMC	0.61	1.89	9.78	5.79	18.06	20.70	8.5
GT	2.01	3.54	27.88	7.88	41.30	20.80	15.5
GGCT	0.85	1.98	6.29	5.34	14.45	20.18	11.6
GMCT	1.68	2.08	6.23	3.61	13.59	253.38	9.3
LGC	0.63	1.15	3.15	2.13	7.05	23.53	9.7
LMC	0.80	1.55	3.01	3.85	9.21	22.10	9.2
LT	1.60	3.04	5.15	7.61	17.40	20.78	13.7
LGCT	1.03	1.53	2.75	4.75	10.05	23.68	12.9
LMCT	1.98	2.29	3.95	4.63	12.84	19.85	11.0

Two-way Anova test for Ni concentrations in grass shoots

Tests of Between-Subjects Effects

Dependent Variable: Ni conc.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	3.587[a]	19	.189	249.099	.000
Intercept	10.149	1	10.149	13391.235	.000
Medium	2.162	4	.540	713.048	**.000**
Aggregate	.700	3	.233	307.713	**.000**
Medium * Aggregate	.726	12	.060	79.795	**.000**
Error	.015	20	.001		
Total	13.751	40			
Corrected Total	3.602	39			

a. R Squared = .996 (Adjusted R Squared = .992)

Post Hoc test for Ni concentrations in grass shoots

Multiple Comparisons

Ni conc.

LSD: Growth media

(I) Medium	(J) Medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	-.168375*	.0137645	.000	-.197087	-.139663
	MC	-.166000*	.0137645	.000	-.194712	-.137288
	MCT	-.228000*	.0137645	.000	-.256712	-.199288
	T	-.689875*	.0137645	.000	-.718587	-.661163
GCT	GC	.168375*	.0137645	.000	.139663	.197087
	MC	.002375	.0137645	.865	-.026337	.031087
	MCT	-.059625*	.0137645	.000	-.088337	-.030913
	T	-.521500*	.0137645	.000	-.550212	-.492788
MC	GC	.166000*	.0137645	.000	.137288	.194712
	GCT	-.002375	.0137645	.865	-.031087	.026337
	MCT	-.062000*	.0137645	.000	-.090712	-.033288
	T	-.523875*	.0137645	.000	-.552587	-.495163
MCT	GC	.228000*	.0137645	.000	.199288	.256712
	GCT	.059625*	.0137645	.000	.030913	.088337
	MC	.062000*	.0137645	.000	.033288	.090712
	T	-.461875*	.0137645	.000	-.490587	-.433163
T	GC	.689875*	.0137645	.000	.661163	.718587
	GCT	.521500*	.0137645	.000	.492788	.550212
	MC	.523875*	.0137645	.000	.495163	.552587
	MCT	.461875*	.0137645	.000	.433163	.490587

Based on observed means.

The error term is Mean Square (Error) = .001.

*.The mean difference is significant at the 0.05 level.

Ni conc.

LSD: Aggregates

(I) Aggregate	(J) Aggregate	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	.326000*	.0123114	.000	.300319	.351681
	NB	.276200*	.0123114	.000	.250519	.301881
	OB	.305800*	.0123114	.000	.280119	.331481
L	G	-.326000*	.0123114	.000	-.351681	-.300319
	NB	-.049800*	.0123114	.001	-.075481	-.024119
	OB	-.020200	.0123114	.116	-.045881	.005481
NB	G	-.276200*	.0123114	.000	-.301881	-.250519
	L	.049800*	.0123114	.001	.024119	.075481
	OB	.029600*	.0123114	.026	.003919	.055281
OB	G	-.305800*	.0123114	.000	-.331481	-.280119
	L	.020200	.0123114	.116	-.005481	.045881
	NB	-.029600*	.0123114	.026	-.055281	-.003919

Based on observed means. The error term is Mean Square (Error) = .001.

*.The mean difference is significant at the 0.05 level.

Two-way Anova test for Ni concentrations in growth media

Tests of Between-Subjects Effects

Dependent Variable: Ni conc.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	40.222[a]	19	2.117	144.578	.000
Intercept	20.092	1	20.092	1372.172	.000
medium	8.036	4	2.009	137.214	**.000**
aggregates	6.233	3	2.078	141.900	**.000**
medium * aggregates	25.952	12	2.163	147.703	**.000**
Error	.293	20	.015		
Total	60.607	40			
Corrected Total	40.515	39			

a. R Squared = .993 (Adjusted R Squared = .986)

Post Hoc test for Ni concentrations in growth media

Multiple Comparisons

Ni conc.

LSD: Growth media

(I) medium	(J) medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	.0817	.06050	.192	-.0445	.2080
	MC	.1160	.06050	.070	-.0102	.2422
	MCT	-1.0390*	.06050	.000	-1.1652	-.9128
	T	.1095	.06050	.085	-.0167	.2357
GCT	GC	-.0817	.06050	.192	-.2080	.0445
	MC	.0343	.06050	.578	-.0920	.1605
	MCT	-1.1208*	.06050	.000	-1.2470	-.9945
	T	.0277	.06050	.651	-.0985	.1540
MC	GC	-.1160	.06050	.070	-.2422	.0102
	GCT	-.0343	.06050	.578	-.1605	.0920
	MCT	-1.1550*	.06050	.000	-1.2812	-1.0288
	T	-.0065	.06050	.916	-.1327	.1197
MCT	GC	1.0390*	.06050	.000	.9128	1.1652
	GCT	1.1208*	.06050	.000	.9945	1.2470
	MC	1.1550*	.06050	.000	1.0288	1.2812
	T	1.1485*	.06050	.000	1.0223	1.2747
T	GC	-.1095	.06050	.085	-.2357	.0167
	GCT	-.0277	.06050	.651	-.1540	.0985
	MC	.0065	.06050	.916	-.1197	.1327
	MCT	-1.1485*	.06050	.000	-1.2747	-1.0223

Based on observed means.

The error term is Mean Square (Error) = .015.

*. The mean difference is significant at the 0.05 level.

Ni conc.

LSD: Aggregates

(I) aggregates	(J) aggregates	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	.9508*	.05412	.000	.8379	1.0637
	NB	.9099*	.05412	.000	.7970	1.0228
	OB	.8664*	.05412	.000	.7535	.9793
L	G	-.9508*	.05412	.000	-1.0637	-.8379
	NB	-.0409	.05412	.459	-.1538	.0720
	OB	-.0844	.05412	.135	-.1973	.0285
NB	G	-.9099*	.05412	.000	-1.0228	-.7970
	L	.0409	.05412	.459	-.0720	.1538
	OB	-.0435	.05412	.431	-.1564	.0694
OB	G	-.8664*	.05412	.000	-.9793	-.7535
	L	.0844	.05412	.135	-.0285	.1973
	NB	.0435	.05412	.431	-.0694	.1564

Based on observed means.

The error term is Mean Square (Error) = .015.

*. The mean difference is significant at the 0.05 level.

Two-way Anova test for Ni concentrations in grass roots

Tests of Between-Subjects Effects

Dependent Variable: Ni conc.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.141[a]	19	.007	10.903	.000
Intercept	1.365	1	1.365	2011.085	.000
Medium	.093	4	.023	34.263	**.000**
Aggregate	.011	3	.004	5.418	**.014**
Medium * Aggregate	.016	12	.001	1.961	.129
Error	.008	12	.001		
Total	1.763	32			
Corrected Total	.149	31			

a. R Squared = .945 (Adjusted R Squared = .859)

Post Hoc test for Ni concentrations in grass roots

Multiple Comparisons

Ni conc.

LSD: Growth media

(I) Medium	(J) Medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	-.079714*	.0139241	.000	-.110052	-.049376
	MC	-.004143	.0152531	.791	-.037377	.029091
	MCT	-.090310*	.0144927	.000	-.121886	-.058733
	T	-.158714*	.0139241	.000	-.189052	-.128376
GCT	GC	.079714*	.0139241	.000	.049376	.110052
	MC	.075571*	.0152531	.000	.042338	.108805
	MCT	-.010595	.0144927	.479	-.042172	.020982
	T	-.079000*	.0139241	.000	-.109338	-.048662
MC	GC	.004143	.0152531	.791	-.029091	.037377
	GCT	-.075571*	.0152531	.000	-.108805	-.042338
	MCT	-.086167*	.0157738	.000	-.120535	-.051798
	T	-.154571*	.0152531	.000	-.187805	-.121338
MCT	GC	.090310*	.0144927	.000	.058733	.121886
	GCT	.010595	.0144927	.479	-.020982	.042172
	MC	.086167*	.0157738	.000	.051798	.120535
	T	-.068405*	.0144927	.000	-.099982	-.036828
T	GC	.158714*	.0139241	.000	.128376	.189052
	GCT	.079000*	.0139241	.000	.048662	.109338
	MC	.154571*	.0152531	.000	.121338	.187805
	MCT	.068405*	.0144927	.000	.036828	.099982

Based on observed means.

The error term is Mean Square (Error) = .001.

*.The mean difference is significant at the 0.05 level.

Ni conc.

LSD: Aggregates

(I) Aggregate	(J) Aggregate	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	-.009100	.0148506	.551	-.041457	.023257
	NB	-.039600*	.0123564	.008	-.066522	-.012678
	OB	-.011722	.0126578	.373	-.039301	.015857
L	G	.009100	.0148506	.551	-.023257	.041457
	NB	-.030500	.0142680	.054	-.061587	.000587
	OB	-.002622	.0145298	.860	-.034280	.029035
NB	G	.039600*	.0123564	.008	.012678	.066522
	L	.030500	.0142680	.054	-.000587	.061587
	OB	.027878*	.0119690	.038	.001800	.053956
OB	G	.011722	.0126578	.373	-.015857	.039301
	L	.002622	.0145298	.860	-.029035	.034280
	NB	-.027878*	.0119690	.038	-.053956	-.001800

Based on observed means.

The error term is Mean Square (Error) = .001.

*.The mean difference is significant at the 0.05 level.

Appendix 31: Results for Pb concentrations in harvested grass shoots, growth media and grass roots

Samples	Blank corrected mean Pb concentration (mg/kg)						
	Bk	1st harvest	2nd harvest	3rd harvest	cumulative grass harvest	growth media	Grass root
OBGC	7.96	2.24	1.94	1.48	13.61	144.75	46.05
OBMC	3.81	2.49	1.91	1.56	9.78	99.43	46.90
OBT	15.08	6.83	12.99	6.66	41.55	2394.88	419.68
OBGCT	5.33	3.09	2.63	32.33	43.36	6029.88	223.58
OBMCT	6.88	4.81	3.90	10.14	25.73	6109.88	290.75
NBGC	2.14	1.65	1.84	1.34	6.96	442.23	99.38
NBMC	4.71	2.69	1.79	1.56	10.75	3252.88	56.68
NBT	13.95	4.34	6.98	18.73	43.99	7657.38	516.18
NBGCT	14.51	3.80	5.54	8.03	31.88	2981.13	382.75
NBMCT	12.25	6.45	5.46	25.50	49.66	2045.38	332.15
GGC	3.05	1.50	2.41	2.36	9.33	1337.38	81.80
GMC	2.56	2.28	5.21	2.84	12.89	338.48	90.85
GT	13.54	4.01	8.18	36.06	61.79	7199.88	562.00
GGCT	8.81	7.59	4.08	7.85	28.33	2503.38	278.55
GMCT	18.25	8.06	4.60	6.36	37.28	2102.88	138.30
LGC	1.85	1.65	2.09	7.69	13.28	174.88	130.75
LMC	3.23	1.98	2.39	4.23	11.81	314.88	85.15
LT	7.51	10.30	3.66	6.98	28.45	1789.88	378.15
LGCT	16.54	7.70	4.74	7.20	36.18	2787.13	226.05
LMCT	31.34	9.24	4.09	4.87	49.53	11364.88	208.75

Two-way Anova test for Pb concentrations in grass shoots

Tests of Between-Subjects Effects

Dependent Variable: Pb conc.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	9.152[a]	19	.482	14.176	.000
Intercept	22.240	1	22.240	654.512	.000
Medium	5.944	4	1.486	43.732	**.000**
Aggregate	.205	3	.068	2.014	.144
Medium * Aggregate	3.003	12	.250	7.365	**.000**
Error	.680	20	.034		
Total	32.071	40			
Corrected Total	9.832	39			

a. R Squared = .931 (Adjusted R Squared = .865)

Post Hoc test for Pb concentrations in grass shoots

Multiple Comparisons

Pb conc.

LSD: Growth media

(I) Medium	(J) Medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	-.663750*	.0921672	.000	-.856007	-.471493
	MC	-.027375	.0921672	.770	-.219632	.164882
	MCT	-.653125*	.0921672	.000	-.845382	-.460868
	T	-.975250*	.0921672	.000	-1.167507	-.782993
GCT	GC	.663750*	.0921672	.000	.471493	.856007
	MC	.636375*	.0921672	.000	.444118	.828632
	MCT	.010625	.0921672	.909	-.181632	.202882
	T	-.311500*	.0921672	.003	-.503757	-.119243
MC	GC	.027375	.0921672	.770	-.164882	.219632
	GCT	-.636375*	.0921672	.000	-.828632	-.444118
	MCT	-.625750*	.0921672	.000	-.818007	-.433493
	T	-.947875*	.0921672	.000	-1.140132	-.755618
MCT	GC	.653125*	.0921672	.000	.460868	.845382
	GCT	-.010625	.0921672	.909	-.202882	.181632
	MC	.625750*	.0921672	.000	.433493	.818007
	T	-.322125*	.0921672	.002	-.514382	-.129868
T	GC	.975250*	.0921672	.000	.782993	1.167507
	GCT	.311500*	.0921672	.003	.119243	.503757
	MC	.947875*	.0921672	.000	.755618	1.140132
	MCT	.322125*	.0921672	.002	.129868	.514382

Based on observed means.

The error term is Mean Square (Error) = .034.

*.The mean difference is significant at the 0.05 level.

Pb conc.

LSD: Aggregate

(I) Aggregate	(J) Aggregate	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	.196800*	.0824368	.027	.024840	.368760
	NB	.061700	.0824368	.463	-.110260	.233660
	OB	.067300	.0824368	.424	-.104660	.239260
L	G	-.196800*	.0824368	.027	-.368760	-.024840
	NB	-.135100	.0824368	.117	-.307060	.036860
	OB	-.129500	.0824368	.132	-.301460	.042460
NB	G	-.061700	.0824368	.463	-.233660	.110260
	L	.135100	.0824368	.117	-.036860	.307060
	OB	.005600	.0824368	.947	-.166360	.177560
OB	G	-.067300	.0824368	.424	-.239260	.104660
	L	.129500	.0824368	.132	-.042460	.301460
	NB	-.005600	.0824368	.947	-.177560	.166360

Based on observed means.

The error term is Mean Square (Error) = .034.

*.The mean difference is significant at the 0.05 level.

Two-way Anova test for Pb concentrations in growth media

Tests of Between-Subjects Effects

Dependent Variable: Pb conc.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	144116.431[a]	19	7585.075	28.768	.000
Intercept	149190.957	1	149190.957	565.835	.000
medium	61838.841	4	15459.710	58.634	**.000**
aggregates	962.798	3	320.933	1.217	.329
medium * aggregates	81314.792	12	6776.233	25.700	**.000**
Error	5273.300	20	263.665		
Total	298580.688	40			
Corrected Total	149389.732	39			

a. R Squared = .965 (Adjusted R Squared = .931)

Post Hoc test for Pb concentrations in growth media

Multiple Comparisons

Pb conc.

LSD: growth media

(I) medium	(J) medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	-61.0114*	8.11888	.000	-77.9471	-44.0757
	MC	-9.5321	8.11888	.254	-26.4678	7.4036
	MCT	-97.6189*	8.11888	.000	-114.5546	-80.6832
	T	-84.7139*	8.11888	.000	-101.6496	-67.7782
GCT	GC	61.0114*	8.11888	.000	44.0757	77.9471
	MC	51.4792*	8.11888	.000	34.5436	68.4149
	MCT	-36.6075*	8.11888	.000	-53.5432	-19.6718
	T	-23.7025*	8.11888	.008	-40.6382	-6.7668
MC	GC	9.5321	8.11888	.254	-7.4036	26.4678
	GCT	-51.4792*	8.11888	.000	-68.4149	-34.5436
	MCT	-88.0868*	8.11888	.000	-105.0224	-71.1511
	T	-75.1818*	8.11888	.000	-92.1174	-58.2461
MCT	GC	97.6189*	8.11888	.000	80.6832	114.5546
	GCT	36.6075*	8.11888	.000	19.6718	53.5432
	MC	88.0868*	8.11888	.000	71.1511	105.0224
	T	12.9050	8.11888	.128	-4.0307	29.8407
T	GC	84.7139*	8.11888	.000	67.7782	101.6496
	GCT	23.7025*	8.11888	.008	6.7668	40.6382
	MC	75.1818*	8.11888	.000	58.2461	92.1174
	MCT	-12.9050	8.11888	.128	-29.8407	4.0307

Based on observed means.

The error term is Mean Square (Error) = 263.665.

*. The mean difference is significant at the 0.05 level.

Pb conc.

LSD: aggregates

(I) aggregates	(J) aggregates	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	-11.7986	7.26175	.120	-26.9463	3.3491
	NB	-11.5880	7.26175	.126	-26.7357	3.5597
	OB	-5.1873	7.26175	.483	-20.3350	9.9604
L	G	11.7986	7.26175	.120	-3.3491	26.9463
	NB	.2106	7.26175	.977	-14.9371	15.3583
	OB	6.6113	7.26175	.373	-8.5364	21.7590
NB	G	11.5880	7.26175	.126	-3.5597	26.7357
	L	-.2106	7.26175	.977	-15.3583	14.9371
	OB	6.4007	7.26175	.389	-8.7470	21.5484
OB	G	5.1873	7.26175	.483	-9.9604	20.3350
	L	-6.6113	7.26175	.373	-21.7590	8.5364
	NB	-6.4007	7.26175	.389	-21.5484	8.7470

Based on observed means.

The error term is Mean Square (Error) = 263.665.

Two-way Anova test for Pb concentrations in grass roots

Tests of Between-Subjects Effects

Dependent Variable: Pb conc.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	339.073[a]	19	17.846	31.466	.000
Intercept	587.069	1	587.069	1035.131	.000
Medium	248.802	4	62.201	109.673	**.000**
Aggregate	11.894	3	3.965	6.991	**.**006
Medium * Aggregate	27.724	12	2.310	4.074	**.011**
Error	6.806	12	.567		
Total	1103.564	32			
Corrected Total	345.879	31			

a. R Squared = .980 (Adjusted R Squared = .949)

Post Hoc test for Pb concentrations in grass roots

Multiple Comparisons

Pb conc.

LSD: Growth media

(I) Medium	(J) Medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	-4.030286*	.4025434	.000	-4.907352	-3.153219
	MC	.327000	.4409642	.473	-.633778	1.287778
	MCT	-3.637500*	.4189805	.000	-4.550380	-2.724620
	T	-7.967571*	.4025434	.000	-8.844638	-7.090505
GCT	GC	4.030286*	.4025434	.000	3.153219	4.907352
	MC	4.357286*	.4409642	.000	3.396507	5.318064
	MCT	.392786	.4189805	.367	-.520094	1.305666
	T	-3.937286*	.4025434	.000	-4.814352	-3.060219
MC	GC	-.327000	.4409642	.473	-1.287778	.633778
	GCT	-4.357286*	.4409642	.000	-5.318064	-3.396507
	MCT	-3.964500*	.4560185	.000	-4.958079	-2.970921
	T	-8.294571*	.4409642	.000	-9.255350	-7.333793
MCT	GC	3.637500*	.4189805	.000	2.724620	4.550380
	GCT	-.392786	.4189805	.367	-1.305666	.520094
	MC	3.964500*	.4560185	.000	2.970921	4.958079
	T	-4.330071*	.4189805	.000	-5.242951	-3.417191
T	GC	7.967571*	.4025434	.000	7.090505	8.844638
	GCT	3.937286*	.4025434	.000	3.060219	4.814352
	MC	8.294571*	.4409642	.000	7.333793	9.255350
	MCT	4.330071*	.4189805	.000	3.417191	5.242951

Based on observed means.

The error term is Mean Square (Error) = .567.

*.The mean difference is significant at the 0.05 level.

Pb conc.

LSD: Aggregates

(I) Aggregate	(J) Aggregate	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	1.069225*	.4293272	.028	.133801	2.004649
	NB	-.363875	.3572218	.328	-1.142195	.414445
	OB	.724625	.3659359	.071	-.072681	1.521931
L	G	-1.069225*	.4293272	.028	-2.004649	-.133801
	NB	-1.433100*	.4124843	.005	-2.331826	-.534374
	OB	-.344600	.4200534	.428	-1.259818	.570618
NB	G	.363875	.3572218	.328	-.414445	1.142195
	L	1.433100*	.4124843	.005	.534374	2.331826
	OB	1.088500*	.3460209	.008	.334585	1.842415
OB	G	-.724625	.3659359	.071	-1.521931	.072681
	L	.344600	.4200534	.428	-.570618	1.259818
	NB	-1.088500*	.3460209	.008	-1.842415	-.334585

Based on observed means.

The error term is Mean Square (Error) = .567.

*.The mean difference is significant at the 0.05 level.

Appendix 32: Results for Zn concentrations in harvested grass shoots, growth media and grass roots

Samples	Blank corrected mean Zn concentration (mg/kg)						
	bk	1st harvest	2nd harvest	3rd harvest	cumulative grass harvest	growth media	Grass root
OBGC	126.65	83.21	76.95	57.01	343.83	283.90	843.25
OBMC	64.65	60.93	57.38	62.41	245.36	177.70	300.55
OBT	103.96	138.61	124.16	101.45	468.19	174.45	376.80
OBGCT	74.34	80.41	82.21	89.83	326.79	206.90	314.05
OBMCT	81.71	89.18	102.75	102.09	375.73	165.70	298.68
NBGC	87.58	71.95	73.35	52.64	285.51	318.80	378.13
NBMC	68.31	59.81	54.74	51.56	234.43	218.25	304.65
NBT	93.65	116.84	122.16	102.69	435.34	171.78	337.85
NBGCT	71.81	73.95	89.08	72.00	306.84	201.28	333.13
NBMCT	77.20	83.01	98.14	73.85	332.20	153.28	280.20
GGC	96.28	45.64	83.85	60.00	285.76	257.30	327.93
GMC	59.84	60.98	125.29	60.70	306.80	184.83	256.10
GT	90.19	107.01	94.28	84.93	376.40	182.93	292.25
GGCT	75.18	76.81	70.43	65.56	287.98	183.90	284.48
GMCT	75.28	79.43	82.05	63.90	300.65	195.03	258.00
LGC	79.81	77.96	83.20	68.01	308.99	310.18	280.25
LMC	65.60	59.14	61.13	61.55	247.41	188.40	217.70
LT	75.73	87.25	81.43	72.34	316.74	185.90	296.20
LGCT	72.25	70.89	73.69	79.81	296.64	202.10	271.60
LMCT	89.25	72.59	84.98	83.39	330.21	179.63	275.45

Two-way Anova test for Zn concentrations in grass shoots

Tests of Between-Subjects Effects

Dependent Variable: Zn conc.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	159.301[a]	19	8.384	188.099	.000
Intercept	3659.971	1	3659.971	82110.682	.000
Medium	103.032	4	25.758	577.875	**.000**
Aggregate	12.862	3	4.287	96.185	**.000**
Medium * Aggregate	43.407	12	3.617	81.152	**.000**
Error	.891	20	.045		
Total	3820.163	40			
Corrected Total	160.193	39			

a. R Squared = .994 (Adjusted R Squared = .989)

Post Hoc test for Zn concentrations in grass shoots

Multiple Comparisons

Zn conc.

LSD: Growth media

(I) Medium	(J) Medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	-.908875*	.1055623	.000	-1.129074	-.688676
	MC	.581750*	.1055623	.000	.361551	.801949
	MCT	-1.815625*	.1055623	.000	-2.035824	-1.595426
	T	-3.993625*	.1055623	.000	-4.213824	-3.773426
GCT	GC	.908875*	.1055623	.000	.688676	1.129074
	MC	1.490625*	.1055623	.000	1.270426	1.710824
	MCT	-.906750*	.1055623	.000	-1.126949	-.686551
	T	-3.084750*	.1055623	.000	-3.304949	-2.864551
MC	GC	-.581750*	.1055623	.000	-.801949	-.361551
	GCT	-1.490625*	.1055623	.000	-1.710824	-1.270426
	MCT	-2.397375*	.1055623	.000	-2.617574	-2.177176
	T	-4.575375*	.1055623	.000	-4.795574	-4.355176
MCT	GC	1.815625*	.1055623	.000	1.595426	2.035824
	GCT	.906750*	.1055623	.000	.686551	1.126949
	MC	2.397375*	.1055623	.000	2.177176	2.617574
	T	-2.178000*	.1055623	.000	-2.398199	-1.957801
T	GC	3.993625*	.1055623	.000	3.773426	4.213824
	GCT	3.084750*	.1055623	.000	2.864551	3.304949
	MC	4.575375*	.1055623	.000	4.355176	4.795574
	MCT	2.178000*	.1055623	.000	1.957801	2.398199

Based on observed means.

The error term is Mean Square (Error) = .045.

*. The mean difference is significant at the 0.05 level.

Zn conc.

LSD: Aggregates

(I) Aggregate	(J) Aggregate	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	.348000*	.0944178	.001	.151048	.544952
	NB	-.279400*	.0944178	.008	-.476352	-.082448
	OB	-1.181900*	.0944178	.000	-1.378852	-.984948
L	G	-.348000*	.0944178	.001	-.544952	-.151048
	NB	-.627400*	.0944178	.000	-.824352	-.430448
	OB	-1.529900*	.0944178	.000	-1.726852	-1.332948
NB	G	.279400*	.0944178	.008	.082448	.476352
	L	.627400*	.0944178	.000	.430448	.824352
	OB	-.902500*	.0944178	.000	-1.099452	-.705548
OB	G	1.181900*	.0944178	.000	.984948	1.378852
	L	1.529900*	.0944178	.000	1.332948	1.726852
	NB	.902500*	.0944178	.000	.705548	1.099452

Based on observed means. The error term is Mean Square (Error) = .045.

*. The mean difference is significant at the 0.05 level.

Two-way Anova test for Zn concentrations in growth media

Tests of Between-Subjects Effects

Dependent Variable: Zn conc.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	34.249[a]	19	1.803	40.523	.000
Intercept	686.479	1	686.479	15432.518	.000
medium	30.500	4	7.625	171.417	**.000**
aggregates	.549	3	.183	4.118	**.020**
medium * aggregates	3.199	12	.267	5.993	**.000**
Error	.890	20	.044		
Total	721.617	40			
Corrected Total	35.138	39			

a. R Squared = .975 (Adjusted R Squared = .951)

Post Hoc test for Zn concentrations in growth media

Multiple Comparisons

Zn conc.

LSD: growth media

(I) medium	(J) medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	1.8800*	.10545	.000	1.6600	2.1000
	MC	2.0050*	.10545	.000	1.7850	2.2250
	MCT	2.3827*	.10545	.000	2.1628	2.6027
	T	2.2756*	.10545	.000	2.0557	2.4956
GCT	GC	-1.8800*	.10545	.000	-2.1000	-1.6600
	MC	.1250	.10545	.250	-.0950	.3450
	MCT	.5028*	.10545	.000	.2828	.7227
	T	.3956*	.10545	.001	.1757	.6156
MC	GC	-2.0050*	.10545	.000	-2.2250	-1.7850
	GCT	-.1250	.10545	.250	-.3450	.0950
	MCT	.3777*	.10545	.002	.1578	.5977
	T	.2706*	.10545	.018	.0507	.4906
MCT	GC	-2.3827*	.10545	.000	-2.6027	-2.1628
	GCT	-.5028*	.10545	.000	-.7227	-.2828
	MC	-.3777*	.10545	.002	-.5977	-.1578
	T	-.1071	.10545	.322	-.3271	.1128
T	GC	-2.2756*	.10545	.000	-2.4956	-2.0557
	GCT	-.3956*	.10545	.001	-.6156	-.1757
	MC	-.2706*	.10545	.018	-.4906	-.0507
	MCT	.1071	.10545	.322	-.1128	.3271

Based on observed means.

The error term is Mean Square (Error) = .044.

*.The mean difference is significant at the 0.05 level.

Zn conc.

LSD: aggregates

(I) aggregates	(J) aggregates	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	-.2489*	.09432	.016	-.4457	-.0521
	NB	-.2376*	.09432	.020	-.4344	-.0408
	OB	-.0187	.09432	.845	-.2155	.1781
L	G	.2489*	.09432	.016	.0521	.4457
	NB	.0113	.09432	.906	-.1855	.2081
	OB	.2302*	.09432	.024	.0334	.4270
NB	G	.2376*	.09432	.020	.0408	.4344
	L	-.0113	.09432	.906	-.2081	.1855
	OB	.2189*	.09432	.031	.0221	.4157
OB	G	.0187	.09432	.845	-.1781	.2155
	L	-.2302*	.09432	.024	-.4270	-.0334
	NB	-.2189*	.09432	.031	-.4157	-.0221

Based on observed means.

The error term is Mean Square (Error) = .044.

*.The mean difference is significant at the 0.05 level.

Two-way Anova test for Zn concentrations in grass roots

Tests of Between-Subjects Effects

Dependent Variable: Zn conc.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	232.909[a]	19	12.258	2.823	.035
Intercept	1205.741	1	1205.741	277.695	.000
Medium	56.124	4	14.031	3.232	.051
Aggregate	44.692	3	14.897	3.431	.052
Medium * Aggregate	100.144	12	8.345	1.922	.136
Error	52.104	12	4.342		
Total	1756.272	32			
Corrected Total	285.012	31			

a. R Squared = .817 (Adjusted R Squared = .528)

Post Hoc test for Zn concentrations in grass roots

Multiple Comparisons

Zn conc.

LSD: Growth media

(I) Medium	(J) Medium	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
GC	GCT	3.554143*	1.1138052	.008	1.127370	5.980916
	MC	4.119257*	1.2201125	.006	1.460860	6.777654
	MCT	4.016524*	1.1592852	.005	1.490658	6.542389
	T	3.053857*	1.1138052	.018	.627084	5.480630
GCT	GC	-3.554143*	1.1138052	.008	-5.980916	-1.127370
	MC	.565114	1.2201125	.652	-2.093282	3.223511
	MCT	.462381	1.1592852	.697	-2.063485	2.988247
	T	-.500286	1.1138052	.661	-2.927059	1.926487
MC	GC	-4.119257*	1.2201125	.006	-6.777654	-1.460860
	GCT	-.565114	1.2201125	.652	-3.223511	2.093282
	MCT	-.102733	1.2617665	.936	-2.851886	2.646420
	T	-1.065400	1.2201125	.400	-3.723797	1.592997
MCT	GC	-4.016524*	1.1592852	.005	-6.542389	-1.490658
	GCT	-.462381	1.1592852	.697	-2.988247	2.063485
	MC	.102733	1.2617665	.936	-2.646420	2.851886
	T	-.962667	1.1592852	.423	-3.488532	1.563199
T	GC	-3.053857*	1.1138052	.018	-5.480630	-.627084
	GCT	.500286	1.1138052	.661	-1.926487	2.927059
	MC	1.065400	1.2201125	.400	-1.592997	3.723797
	MCT	.962667	1.1592852	.423	-1.563199	3.488532

Based on observed means.

The error term is Mean Square (Error) = 4.342.

*.The mean difference is significant at the 0.05 level.

Zn conc.

LSD: Aggregate

(I) Aggregate	(J) Aggregate	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
G	L	.443700	1.1879139	.715	-2.144542	3.031942
	NB	-.727300	.9884041	.476	-2.880848	1.426248
	OB	-3.005056*	1.0125151	.012	-5.211136	-.798975
L	G	-.443700	1.1879139	.715	-3.031942	2.144542
	NB	-1.171000	1.1413107	.325	-3.657702	1.315702
	OB	-3.448756*	1.1622540	.012	-5.981089	-.916422
NB	G	.727300	.9884041	.476	-1.426248	2.880848
	L	1.171000	1.1413107	.325	-1.315702	3.657702
	OB	-2.277756*	.9574119	.035	-4.363777	-.191734
OB	G	3.005056*	1.0125151	.012	.798975	5.211136
	L	3.448756*	1.1622540	.012	.916422	5.981089
	NB	2.277756*	.9574119	.035	.191734	4.363777

Based on observed means.

The error term is Mean Square (Error) = 4.342.

*.The mean difference is significant at the 0.05 level.

Standard error for background grass concentrations									
Samples	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
OBGC	0	0	0	0	0	0	0	0	0
OBMC	2.000	0.025	0.000	0.125	3.250	0.075	0.000	0.250	1.250
OBT	10.638	0.000	0.000	0.625	1.337	0.338	0.062	0.163	4.163
OBGCT	0.038	0.000	0.025	1.563	14.450	1.388	0.050	0.737	3.938
OBMCT	15.338	0.000	0.063	1.213	17.575	1.338	0.013	1.363	2.813
NBGC	3.000	0.000	0.013	0.888	8.362	5.050	0.025	0.200	3.575
NBMC	22.688	0.038	0.113	0.763	15.200	1.188	0.554	0.550	0.462
NBT	1.175	0.013	0.025	0.500	1.112	0.050	0.062	0.562	0.825
NBGCT	10.038	0.000	0.025	0.250	0.275	0.638	0.000	0.600	3.663
NBMCT	21.550	0.000	0.150	0.887	3.088	0.512	0.038	0.163	1.075
GGC	23.500	0.000	0.138	2.438	14.200	1.475	0.150	0.288	9.025
GMC	5.475	0.000	0.025	0.763	3.000	0.212	0.013	0.300	0.713
GT	62.600	0.000	0.150	0.288	46.800	0.887	0.062	1.975	1.238
GGCT	19.875	0.000	0.175	1.500	38.400	4.625	0.175	2.500	4.125
GMCT	38.838	0.000	0.100	0.750	39.875	1.925	0.225	2.538	2.125
LGC	17.025	0.000	0.063	0.138	10.388	0.438	0.050	0.288	2.788
LMC	8.938	0.000	0.038	0.075	8.987	0.188	0.000	0.038	0.575
LT	1.850	0.000	0.050	0.650	12.525	3.475	0.275	0.600	7.625
LGCT	15.750	0.000	0.025	0.075	22.875	0.675	0.050	2.025	1.300
LMCT	25.675	0.000	0.025	0.425	105.025	0.375	0.050	5.275	0.850

Standard error for 1st grass harvest									
Samples	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
OBGC	9.700	0.038	0.038	0.175	1.513	1.400	0.038	0.050	0.012
OBMC	0.825	0.025	0.050	0.125	1.675	0.238	0.063	0.075	0.575
OBT	2.663	0.012	0.025	0.225	0.600	0.363	0.050	0.137	3.513
OBGCT	1.350	0.012	0.025	0.325	0.563	0.363	0.025	0.025	0.138
OBMCT	9.850	0.100	0.138	2.350	9.388	3.450	0.200	0.500	5.575
NBGC	5.638	0.025	0.088	0.050	4.925	3.375	0.025	0.138	0.875
NBMC	11.063	0.113	0.063	0.213	9.425	1.263	0.075	0.050	1.013
NBT	2.038	0.050	0.088	0.325	1.263	0.787	0.175	0.075	2.763
NBGCT	3.350	0.050	0.050	1.100	2.475	0.363	0.063	0.113	0.150
NBMCT	5.800	0.063	0.038	1.750	4.013	0.225	0.088	0.238	0.788
GGC	1.738	0.025	0.050	0.500	2.113	4.888	0.088	0.163	2.388
GMC	3.550	0.163	0.088	0.500	3.900	3.625	0.163	0.013	3.625
GT	2.475	0.038	0.000	0.425	1.638	1.150	0.013	0.125	5.137
GGCT	32.313	0.038	0.063	0.213	17.363	0.575	0.000	0.225	1.263
GMCT	25.388	0.025	0.050	0.800	1.612	1.163	0.050	0.650	2.625
LGC	0.875	0.038	0.013	0.213	3.225	1.613	0.000	0.063	1.463
LMC	2.488	0.000	0.013	0.300	1.788	0.288	0.050	0.113	0.988
LT	0.787	0.013	0.012	0.700	0.275	1.863	0.113	0.188	4.825
LGCT	7.925	0.013	0.025	0.600	7.150	0.962	0.025	0.962	1.163
LMCT	2.388	0.038	0.025	0.475	3.438	1.513	0.088	0.375	2.238
Standard error for 2nd grass harvest									
Samples	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
OBGC	0.888	0.025	0.013	0.113	2.775	2.088	0.013	0.413	1.025
OBMC	3.775	0.013	0.062	0.425	2.200	0.925	0.038	0.038	1.400
OBT	16.150	0.013	0.000	0.250	14.538	0.375	0.050	1.363	0.287
OBGCT	2.025	0.000	0.063	0.313	4.213	2.500	0.163	0.050	2.213
OBMCT	1.975	0.050	0.062	0.125	3.563	0.825	0.113	0.050	0.150
NBGC	0.350	0.150	0.063	0.238	0.100	1.262	0.088	0.237	1.475
NBMC	5.350	0.050	0.038	0.013	4.838	1.150	0.113	0.113	0.687
NBT	5.812	0.050	0.050	0.450	6.125	2.388	0.038	0.225	2.612
NBGCT	0.063	0.025	0.063	0.325	1.375	1.925	0.088	0.138	1.300
NBMCT	1.188	0.038	0.037	0.225	0.137	0.212	0.050	0.013	1.362
GGC	3.650	0.013	0.100	0.038	2.063	1.625	0.062	0.063	1.450
GMC	21.838	0.150	0.050	2.987	13.813	2.612	0.100	0.188	0.737
GT	15.600	0.088	0.062	0.350	14.613	1.463	0.900	0.550	0.850
GGCT	5.100	0.125	0.075	0.975	7.363	2.163	0.413	0.375	2.525
GMCT	2.838	0.138	0.038	0.650	0.200	0.450	0.125	0.375	0.325
LGC	0.713	0.050	0.062	0.425	1.288	1.800	0.075	0.062	0.250
LMC	0.287	0.138	0.050	0.363	0.200	3.513	0.163	0.063	0.250
LT	4.838	0.100	0.000	0.313	2.400	0.363	0.000	0.013	1.200
LGCT	1.138	0.063	0.025	0.238	0.788	0.312	0.025	0.013	1.888
LMCT	5.150	0.038	0.088	0.638	4.638	1.625	0.050	0.063	0.225

Standard error for 3rd grass harvest									
Samples	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
OBGC	6.200	0.025	0.038	0.288	4.875	1.688	0.050	0.138	0.475
OBMC	4.713	0.038	0.138	1.338	5.575	4.050	0.025	0.100	0.600
OBT	11.412	0.025	0.075	0.400	9.050	0.837	0.388	0.375	0.312
OBGCT	2.750	0.025	0.025	1.100	70.750	2.725	0.138	2.063	1.088
OBMCT	71.500	0.063	0.213	1.225	57.000	3.525	0.313	1.450	6.850
NBGC	2.500	0.013	0.000	0.775	7.100	1.888	0.013	0.275	0.950
NBMC	5.788	0.175	0.063	1.700	20.900	5.900	0.975	0.250	3.550
NBT	20.675	0.038	0.037	1.375	26.613	2.363	0.250	1.887	0.200
NBGCT	8.200	0.050	0.075	1.538	14.450	1.513	0.038	0.988	2.113
NBMCT	221.263	0.013	0.537	1.850	379.800	18.088	0.563	12.788	1.463
GGC	8.900	0.013	0.088	0.388	6.175	4.575	0.412	0.050	2.788
GMC	88.600	0.013	0.463	1.513	41.613	4.175	0.350	0.275	2.288
GT	32.000	0.013	0.150	0.700	45.750	2.225	0.488	4.250	0.662
GGCT	1.650	0.000	0.138	1.288	20.388	2.775	0.025	1.038	2.175
GMCT	35.088	0.075	0.250	2.188	45.725	6.163	0.550	2.425	4.163
LGC	0.413	0.025	0.125	0.588	11.250	0.038	0.113	0.200	0.850
LMC	13.650	0.012	0.000	1.713	10.225	1.800	0.188	0.013	2.288
LT	5.113	0.088	0.138	2.313	19.612	2.500	0.125	2.788	1.400
LGCT	8.213	0.025	0.075	0.400	3.850	0.163	0.163	0.138	0.350
LMCT	6.638	0.006	0.000	0.488	19.194	1.075	0.106	0.669	2.369

Cumulative grass harvest standard error									
Samples	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
OBGC	16.788	0.088	0.088	0.575	9.163	5.175	0.100	0.600	1.513
OBMC	11.313	0.100	0.250	2.013	12.700	5.287	0.125	0.463	3.825
OBT	40.862	0.050	0.100	1.500	25.525	1.913	0.550	2.038	8.275
OBGCT	6.163	0.038	0.138	3.300	89.975	6.975	0.375	2.875	7.375
OBMCT	98.663	0.213	0.475	4.913	87.525	9.138	0.638	3.363	15.388
NBGC	11.488	0.188	0.163	1.950	20.488	11.575	0.150	0.850	6.875
NBMC	44.888	0.375	0.275	2.688	50.363	9.500	1.716	0.962	5.713
NBT	29.700	0.150	0.200	2.650	35.113	5.588	0.525	2.750	6.400
NBGCT	21.650	0.125	0.213	3.213	18.575	4.438	0.188	1.838	7.225
NBMCT	249.800	0.113	0.762	4.713	387.038	19.038	0.738	13.200	4.687
GGC	37.788	0.050	0.375	3.363	24.550	12.563	0.712	0.562	15.650
GMC	119.463	0.325	0.625	5.762	62.325	10.625	0.625	0.775	7.362
GT	112.675	0.138	0.363	1.763	108.800	5.725	1.463	6.900	7.888
GGCT	58.938	0.163	0.450	3.975	83.513	10.138	0.613	4.137	10.088
GMCT	102.150	0.238	0.437	4.388	87.413	9.700	0.950	5.988	9.238
LGC	19.025	0.113	0.263	1.363	26.150	3.888	0.238	0.613	5.350
LMC	25.363	0.150	0.100	2.450	21.200	5.788	0.400	0.225	4.100
LT	12.588	0.200	0.200	3.975	34.812	8.200	0.513	3.588	15.050
LGCT	33.025	0.100	0.150	1.313	34.663	2.113	0.263	3.138	4.700
LMCT	39.850	0.081	0.138	2.025	132.294	4.588	0.294	6.381	5.681

Growth media standard error									
Samples	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
OBGC	33.75	0.50	0.40	6.98	2635.00	41.85	11.48	3.37	11.73
OBMC	17.75	0.35	1.98	2.85	92.50	10.03	2.18	3.15	5.98
OBT	310.00	0.05	1.03	31.38	1092.50	5.78	0.52	63.50	4.33
OBGCT	1052.50	0.03	22.45	0.10	415.00	10.33	0.93	685.00	1.08
OBMCT	315.00	0.05	3.48	3.95	292.50	0.30	0.08	240.00	2.83
NBGC	401.50	0.33	2.50	3.60	642.50	18.55	1.15	32.90	5.68
NBMC	45.00	0.38	2.33	1.35	555.00	4.20	0.10	1026.00	3.42
NBT	597.50	0.05	0.03	7.92	262.50	18.25	1.28	682.50	6.95
NBGCT	188.00	0.15	2.10	71.03	295.00	7.35	0.65	237.75	6.60
NBMCT	105.00	0.03	5.10	0.72	462.50	1.08	0.43	152.50	2.80
GGC	37.75	0.00	65.13	0.40	70.00	1.65	6.58	509.50	3.57
GMC	146.50	0.23	8.43	2.48	525.00	2.68	0.80	160.60	5.95
GT	62.50	0.05	3.42	9.73	662.50	4.15	0.50	715.00	4.50
GGCT	507.25	0.48	6.35	18.58	52.50	34.73	1.48	283.50	21.52
GMCT	155.00	0.05	1.10	124.50	50.00	6.63	13.18	69.50	0.75
LGC	159.50	0.58	0.67	5.43	352.50	5.67	0.82	9.70	12.40
LMC	1118.00	0.43	4.10	0.95	792.50	6.88	1.45	95.25	6.68
LT	242.50	0.10	3.85	4.40	305.00	25.93	0.62	35.50	0.67
LGCT	496.50	0.45	0.98	6.88	657.50	44.47	1.38	126.25	2.67
LMCT	37.50	0.03	2.38	0.78	362.50	5.13	0.10	1885.00	5.40
Grass roots standard error									
Samples	Al	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
OBGC	68.50	0.33	0.70	451.25	159.75	10.45	0.62	2.10	250.25
OBMC	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
OBT	169.25	0.10	0.28	2.40	65.00	0.10	0.20	0.42	11.60
OBGCT	74.25	0.25	0.33	5.80	112.00	4.23	0.60	15.98	3.65
OBMCT	455.00	0.33	1.30	11.15	513.00	18.35	1.65	42.45	24.88
NBGC	140.50	0.37	0.63	5.70	292.50	14.28	0.88	10.08	15.13
NBMC	194.25	0.55	1.33	4.55	289.75	24.15	1.13	5.68	29.90
NBT	447.50	0.40	1.55	10.90	606.50	6.55	1.10	62.82	3.30
NBGCT	51.75	0.13	0.13	2.68	119.75	6.65	0.13	11.95	19.13
NBMCT	456.25	0.13	1.23	6.10	496.75	37.83	1.25	34.25	12.85
GGC	61.50	0.05	0.38	0.78	138.00	8.32	0.43	2.90	3.13
GMC	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
GT	7.75	0.40	0.63	9.00	116.75	5.85	0.90	24.00	7.80
GGCT	126.50	0.48	0.68	13.53	81.25	1.45	0.93	21.55	1.63
GMCT	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
LGC	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
LMC	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
LT	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
LGCT	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
LMCT	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00

Appendix 33: Results of oil retention experiments on recycled aggregates

Initial weights of recycled aggregates and increase in weight of aggregates + absorbed clean and used motor oil

New bricks	Initial weight (g)	Weight of new bricks + clean oil (g)							
		Weeks							
		1	2	3	4	5	6	7	8
NBC1	38.33	40.10	40.47	40.43	40.49	40.49	40.51	40.53	40.56
NBC2	30.41	32.02	32.04	32.10	32.16	32.16	32.15	32.18	32.11
NBC3	46.31	48.22	48.55	48.60	48.65	48.65	48.65	48.68	48.71
NBC4	24.89	26.93	26.16	26.15	26.16	26.19	26.18	26.19	26.21
NBC5	14.78	15.57	15.59	15.62	15.65	15.64	15.68	15.64	15.68
NBC6	132.13	135.45	136.78	137.62	138.32	138.42	138.49	138.54	138.62
New bricks	Initial weight (g)	Weight of New bricks + used oil (g)							
		Weeks							
		1	2	3	4	5	6	7	8
NBU1	29.31	30.33	30.57	30.68	30.75	30.80	30.74	30.72	30.66
NBU2	32.30	33.17	33.57	33.91	34.06	34.11	34.12	34.12	34.15
NBU3	63.12	64.65	65.21	65.63	65.85	65.93	65.98	66.01	66.11
NBU4	11.12	11.57	11.67	11.73	11.71	11.72	11.72	11.72	11.72
NBU5	7.88	8.33	8.35	8.37	8.29	8.31	8.30	8.31	8.36
NBU6	102.50	104.53	105.30	106.01	106.73	107.03	107.29	107.36	107.48
Old bricks	Initial weight (g)	Weight of old bricks + clean oil (g)							
		Weeks							
		1	2	3	4	5	6	7	8
OBC1	22.88	26.04	26.05	26.24	26.37	26.98	27.00	27.04	27.10
OBC2	29.24	34.00	34.05	34.15	34.88	35.08	35.10	35.11	35.14
OBC3	7.23	8.86	8.90	8.96	8.86	9.00	9.09	9.10	9.10
OBC4	8.02	9.52	9.58	9.52	9.66	9.98	10.01	10.06	10.10
OBC5	38.66	44.71	44.84	44.84	44.98	45.57	45.69	45.80	45.88
OBC6	98.28	113.33	113.67	113.89	113.96	114.07	114.25	114.37	114.47
Old bricks	Initial weight (g)	Weight of old bricks + used oil (g)							
		Weeks							
		1	2	3	4	5	6	7	8
OBU1	26.26	29.67	29.69	29.75	29.97	30.14	30.68	30.86	30.97
OBU2	33.11	38.87	38.87	38.89	39.05	39.89	40.08	40.09	40.10
OBU3	17.24	19.73	19.77	19.84	19.08	19.99	20.06	20.08	20.10
OBU4	12.54	14.96	14.99	15.01	15.99	16.27	16.69	16.88	16.98
OBU5	8.39	9.65	9.66	9.67	10.01	10.66	10.89	10.98	10.01
OBU6	107.34	123.66	123.98	124.22	124.98	125.08	125.88	125.91	125.97

Gravel Aggregates	Initial Weight (g)	**Weight of gravel + clean oil (g)**			
		Weeks			
		1	2	3	4
GC1	22.85	22.88	22.87	22.91	22.91
GC2	12.59	12.64	12.65	12.65	12.66
GC3	15.20	15.26	15.24	15.30	15.26
GC4	13.04	13.05	13.05	13.08	13.09
GC5	13.24	13.29	13.31	13.37	13.29
GC6	11.40	11.48	11.45	11.46	11.45
Gravel Aggregates	Initial Weight (g)	**Weight of gravel + used oil (g)**			
		Weeks			
		1	2	3	4
GU1	17.39	17.46	17.47	17.49	17.45
GU2	15.41	15.47	15.47	15.51	15.48
GU3	20.30	20.36	20.36	20.39	20.39
GU4	13.11	13.15	13.14	13.16	13.15
GU5	19.54	19.64	19.69	19.69	19.66
GU6	18.55	18.58	18.59	18.62	18.59
Limestone Aggregates	Initial Weight (g)	**Weight of limestone + clean oil (g)**			
		Weeks			
		1	2	3	4
LC1	30.38	30.58	30.54	30.55	30.56
LC2	49.00	49.22	49.31	49.27	49.20
LC3	46.21	46.47	46.52	46.58	46.49
LC4	76.80	77.02	77.00	77.06	77.03
LC5	34.94	35.09	35.15	35.17	35.18
LC6	64.28	64.47	64.41	64.52	64.56
Limestone Aggregates	Initial Weight (g)	**Weight of limestone + used oil (g)**			
		Weeks			
		1	2	3	4
LU1	19.48	19.55	19.57	19.59	19.57
LU2	66.36	66.68	66.69	66.75	66.71
LU3	32.14	32.24	32.24	32.29	32.30
LU4	53.66	55.07	55.34	55.51	55.39
LU5	49.11	49.50	49.53	49.70	49.67
LU6	19.29	19.37	19.39	19.40	19.45

Weight of clean and used motor oil absorbed by recycled aggregates

New bricks	Weight of clean oil absorbed by new bricks (g)							
	Weeks							
	1	2	3	4	5	6	7	8
NBC1	4.62	5.58	5.49	5.64	5.64	5.70	5.75	5.82
NBC2	5.31	5.36	5.55	5.75	5.74	5.71	5.83	5.60
NBC3	4.11	4.83	4.93	5.05	5.04	5.04	5.12	5.17
NBC4	8.20	5.10	5.04	5.09	5.23	5.19	5.21	5.28
NBC5	5.39	5.51	5.69	5.88	5.85	6.07	5.82	6.08
NBC6	2.51	3.52	4.16	4.69	4.76	4.81	4.85	4.91
New bricks	Weight of used oil absorbed by new bricks (g)							
	Weeks							
	1	2	3	4	5	6	7	8
NBU1	3.48	4.28	4.66	4.89	5.06	4.87	4.81	4.59
NBU2	2.71	3.94	4.97	5.46	5.59	5.63	5.63	5.71
NBU3	2.42	3.32	3.98	4.33	4.45	4.54	4.59	4.74
NBU4	4.07	4.95	5.48	5.30	5.44	5.41	5.42	5.44
NBU5	5.78	6.07	6.32	5.30	5.49	5.36	5.50	6.10
NBU6	1.98	2.73	3.43	4.13	4.42	4.68	4.74	4.86
Old bricks	Weight of clean oil absorbed by old bricks (g)							
	Weeks							
	1	2	3	4	5	6	7	8
OBC1	13.79	13.84	14.68	15.21	17.89	17.99	18.17	18.41
OBC2	16.29	16.48	16.79	19.29	19.98	20.05	20.07	20.20
OBC3	22.55	23.13	23.98	22.62	24.50	25.71	25.86	25.94
OBC4	18.71	19.42	18.63	20.42	24.37	24.77	25.38	25.88
OBC5	15.66	15.98	15.99	16.34	17.87	18.18	18.47	18.67
OBC6	15.30	15.65	15.88	15.95	16.06	16.24	16.36	16.47
Old bricks	Weight of used oil absorbed by old bricks (g)							
	Weeks							
	1	2	3	4	5	6	7	8
OBU1	12.99	13.07	13.30	14.12	14.78	16.81	17.50	17.92
OBU2	17.39	17.39	17.45	17.92	20.47	21.04	21.07	21.10
OBU3	14.41	14.63	15.07	10.63	15.91	16.31	16.45	16.55
OBU4	19.31	19.56	19.69	27.48	29.71	33.06	34.57	35.36
OBU5	14.96	15.08	15.18	19.26	26.98	29.77	30.81	19.26
OBU6	15.20	15.50	15.72	16.43	16.52	17.27	17.29	17.35

Gravel Aggregates	Weight of clean oil absorbed by gravel (g)			
	Weeks			
	1	2	3	4
GC1	0.15	0.08	0.27	0.27
GC2	0.37	0.48	0.46	0.54
GC3	0.36	0.26	0.64	0.37
GC4	0.12	0.08	0.33	0.41
GC4	0.36	0.48	0.97	0.36
GC5	0.76	0.49	0.55	0.47
Gravel Aggregates	Weight of used oil absorbed by gravel (g)			
	Weeks			
	1	2	3	4
GU1	0.41	0.43	0.56	0.33
GU2	0.43	0.38	0.66	0.47
GU3	0.30	0.30	0.46	0.46
GU4	0.26	0.19	0.37	0.29
GU5	0.53	0.77	0.77	0.62
GU6	0.12	0.22	0.37	0.20
Limestone Aggregates	Weight of clean oil absorbed by limestone (g)			
	Weeks			
	1	2	3	4
LC1	0.64	0.52	0.55	0.58
LC2	0.44	0.62	0.55	0.41
LC3	0.56	0.69	0.81	0.61
LC4	0.29	0.27	0.35	0.31
LC5	0.41	0.60	0.65	0.68
LC6	0.30	0.19	0.37	0.43
Limestone Aggregates	Weight of used oil absorbed by limestone (g)			
	Weeks			
	1	2	3	4
LU1	0.37	0.45	0.56	0.46
LU2	0.48	0.49	0.59	0.53
LU3	0.32	0.30	0.47	0.50
LU4	2.63	3.12	3.45	3.22
LU5	0.80	0.84	1.20	1.14
LU6	0.45	0.54	0.60	0.86

Oil concentrations in leachate over three weeks

Samples	Oil concentration in leachate for Week 1 mgL^{-1}			Average	**Average**	**standard error**
	Rep 1	Rep 2	Rep 3			
OBU1	1.1	1.2	1.2	1.2		
OBU2	1.1	1.2	1.2	1.2		
OBU3	1.0	1.0	1.1	1.0	**1.1**	**0.044**
OBC1	0.7	0.7	0.8	0.7		
OBC2	0.6	0.6	0.7	0.6		
OBC3	0.3	0.3	0.3	0.3	**0.6**	**0.131**
NBU1	1.3	1.7	1.7	1.6		
NBU2	2.5	2.7	2.6	2.6		
NBU3	0.7	0.9	0.9	0.0	**1.4**	**0.757**
NBC1	0.3	0.3	0.3	0.3		
NBC2	0.7	0.9	0.9	0.8		
NBC3	0.7	0.7	0.8	0.7	**0.6**	**0.164**
LU1	1.4	1.6	1.6	1.5		
LU2	1.1	1.3	1.3	1.2	**1.4**	**0.123**
LC1	1.3	1.4	1.4	1.4		
LC2	0.9	0.9	0.9	0.0	**0.7**	**0.204**
GU1	1.7	2.0	2.0	1.9		
GU2	1.3	1.3	1.4	1.3	**1.6**	**0.232**
GC1	1.3	1.3	1.4	1.3		
GC2	1.0	1.0	1.0	1.0	**1.2**	**0.136**

Samples	Oil concentration in leachate for Week 2 mgL^{-1}			Average	**Average**	**standard error**
OBU1	0.1	0.2	0.2	0.2		
OBU2	0.0	0.0	0.0	0.0		
OBU3	0.0	0.0	0.0	0.0	**0.1**	**0.056**
OBC1	0.7	1.0	1.1	0.9		
OBC2	0.5	0.6	0.6	0.6		
OBC3	0.6	0.7	0.8	0.7	**0.7**	**0.107**
NBU1	0.2	0.2	0.2	0.2		
NBU2	0.0	0.0	0.0	0.0		
NBU3	0.0	0.0	0.0	0.0	**0.1**	**0.067**
NBC1	0.0	0.0	0.0	0.0		
NBC2	0.0	0.0	0.0	0.0		
NBC3	0.1	0.1	0.2	0.1	**0.0**	**0.044**
LU1	0.9	1.2	1.2	1.1		
LU2	0.2	0.2	0.2	0.0	**0.6**	**0.450**
LC1	0.6	0.8	0.8	0.0		
LC2	1.5	1.7	1.8	1.7	**0.8**	**0.681**
GU1	1.8	2.1	2.2	2.0		
GU2	0.5	0.6	0.6	0.0	**1.0**	**0.831**
GC1	1.4	1.7	1.7	1.6		
GC2	1.6	1.7	1.8	1.7	**1.7**	**0.041**

Samples	Oil concentration in leachate for Week 3 mgL^{-1}			Average	**Average**	**standard error**
OBU1	0.0	0.0	0.1	0.0		
OBU2	0.1	0.1	0.2	0.1		
OBU3	0.1	0.1	0.2	0.1	**0.1**	**0.033**
OBC1	0.1	0.1	0.1	0.1		
OBC2	0.1	0.1	0.1	0.1		
OBC3	0.0	0.0	0.1	0.0	**0.1**	**0.022**
NBU1	0.4	0.6	0.7	0.6		
NBU2	0.3	0.4	0.4	0.4		
NBU3	0.3	0.4	0.4	0.4	**0.4**	**0.067**
NBC1	0.1	0.1	0.1	0.1		
NBC2	0.0	0.1	0.1	0.1		
NBC3	0.1	0.1	0.1	0.1	**0.1**	**0.011**
LU1	0.7	0.9	1.0	0.9		
LU2	0.7	0.7	0.8	0.7	**0.8**	**0.054**
LC1	0.9	1.1	1.1	1.0		
LC2	0.8	0.8	0.8	0.8	**0.9**	**0.095**
GU1	0.6	0.7	0.7	0.7		
GU2	0.7	0.9	0.9	0.8	**0.8**	**0.068**
GC1	0.8	0.8	0.9	0.8		
GC2	0.6	0.6	0.6	0.6	**0.7**	**0.095**

Ethical Approval form