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Materials for Enhancing Denitrification Performance in Partially Saturated Vertical Flow Wetlands for Marae

A thesis

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at

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Abstract

There are upwards of 773 marae in New Zealand which are responsible for many key events for the indigenous people, Māori, many of which are located rurally and have inadequate on-site wastewater treatment mostly comprising of a septic tank followed by land application. Māori have many strong beliefs about water and subsequently criteria surround the treatment of wastewater due to the effects it can have on people and environment. One proposed solution that meets criteria are treatment wetlands, more specifically partially saturated vertical flow wetlands which require a smaller footprint and produce better effluent than traditional wetland designs. NIWA (National Institute of Water and Atmospheric Research) has experimented with mesocosm scale versions of this design and has encountered issues with denitrification in the saturated zone.

This research is therefore directed at enhancing denitrification in the saturated zone with materials that are able to supplement organic carbon for denitrification. Several woodchip materials (Eastern Cottonwood, Mahogany, Manuka, Eucalyptus, Crack Willow, Princess Tree and Pine) along with two industry by-product materials (Brewers Spent Grain and Slum gum) and Coir (Coconut derivative) were examined for denitrification capabilities and to serve in producing a leachate for dosing the proposed system to supplement organic carbon for denitrification.

The research is split into three phases; the first examines the ability of materials to release organic carbon into water over 60 days, the second examines the same but over three 10 day cycles, and the third quantifies denitrification performance of the materials at lab scale.

Results showed that producing a suitable leachate is not possible without the use of fresh material for each batch. By-product materials had great capacity for releasing organic carbon into water and denitrification (up to 85.5% nitrate removal) but had limited lifespans as media. While woodchip materials did not exhibit as great carbon release, they achieved comparable denitrification (74.2-74.5% nitrate removal) with long lifespans. Therefore, a woodchip and industry by-product mix could be the solution. Alternatively, there are many woodchip species, some which could have better denitrification capacity or suitable industry by-products with longer useful lifespans.

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Firstly, my gratitude goes to my supervisor Mark Lay, not only for his patience, guidance and knowledge in directing this research and the writing of this thesis, but also for being the kind soul in supporting my return to academics in various forms.

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Chapter 1
Introduction

1.1 Background

A key part of New Zealand's indigenous population, Māori, are the tribal meeting grounds (marae) and associated housing (papakāinga). Most maraes are situated in remote rural areas and as a result are not connected to municipal wastewater treatment facilities, therefore are required to manage their wastewater on-site. Current on-site treatment mainly consists of a septic tank leading to land infiltrative disposal via soak pits or trenches. While requiring minimal maintenance, cost and land this approach leaves a lot to be desired in terms of nutrient removal (i.e. nitrogen) upon discharge to land (Tanner et al., 2012).

Maraes are used as a hub for a variety of community events including celebrations (such as birthdays or weddings) and tangihanga (a traditional funeral rite). The result is fluctuating occupancy where the site could hold a few individuals up to a few hundred people during these events. Therefore wastewater productions varies largely compared to a typical domestic household where consistent flow experienced and there can also be periods of inactivity with very little to no wastewater generated. This presents a unique challenge for wastewater treatment requiring a treatment system that can withstand fluctuating loads.

Māori have a cultural and spiritual relationship with water (wai) and regard it as a treasure possession (taonga) which is to be protected for current and future generations. Taonga such as water have become part of New Zealand legislation through its inclusion in the Treaty of Waitangi and the Resource Management Act (RMA) 1991, signifying the importance of these treasures to local tribes (iwi). The discharge of wastewater to bodies of water is considered taboo (tapu) as it can an effect on food provided by the water (kai moana) and the recreational uses of those bodies of water (Simmonds et al., 2019). As a result land-based treatment is the preferred method to restore the spiritual aspect of the wastewater, however, this presents its own challenges in meeting nutrient limits according to resource consent (i.e. phosphorus, nitrogen and faecal coliforms) when discharging effluent to land.

Constructed wetlands (CWs) as a wastewater treatment technology align with cultural values where wastewater passes through land to convert the water from tapu (bad/unsafe/taboo) to noa (good/safe/clean). CWs offer affordable, reliable, simple in design and operation, and are an environmentally friend approach to waste water treatment for small communities and remote locations at the cost of land area (Wu et al., 2015). More recently new designs/and or operational strategies have been developed to reduce the footprint required and improve effluent quality.

One option being pursued is partially saturated vertical flow (PSVF) wetlands consisting of both a saturated and unsaturated zone that aims to provide suitable environmental conditions for nitrification and denitrification. These systems offer various configurations and options for media and can have a reduced footprint compared to classical CWs. One key factor to the performance of PSVF wetlands is the denitrification capacity of the saturated zone, which is dependent on carbon sources being available. The source of this carbon is typically reliant on the media that occupies the saturated zone. To align with cultural values, the use of organic media is preferred over man made carbon sources (which are also costly).

1.2 Project Objectives

This research is directed at selected locally accessible organic media that align with cultural guidelines in this application. These media are evaluated for:

- Ability to release carbon in a single stage and over multiple cycles
- Practicality to produce a leachate that can be dosed to supplement carbon available for denitrification in a saturated zone
- Denitrification performance at a lab scale

The first goal is to understand if there is a correlation in carbon release and denitrification performance of the materials, allowing for quick and easy evaluation of materials for application in PSVF wetlands.

The second goal is to evaluate if producing a leachate to supplement carbon available for denitrification upon dosing the saturated zone in a PSVF wetland is practical.

Finally the third objective is to provide information on media lifetimes and denitrification performance which can be used when selecting media for PSVF wetland units for maraes. This will also provide information for media selection for mesocosm scale CWs set up at the Pukete wastewater treatment plant that serve as part of a larger project for NIWA (National Institute of Water and Atmospheric Research).

1.3 Thesis Structure

Chapter 2 will review information available on;

- Māori, maraes and papakāinga in New Zealand
- Māori values surrounding water
- Current on-site wastewater treatment practices and criteria for future wastewater treatment
- Treatment wetlands and variations as secondary treatment and how they are suitable
- Nitrogen removal and the pathways occurring in treatment wetlands
- Materials that can improve denitrification

Chapter 3 outlines the experimental setups for each phase of research, how parameters are monitored and analysis methods. Chapter 4 provides the results and discussion around each phase of experiments and chapter 5 gives an overview on the conclusions drawn from the results obtained.

Chapter 2

Literature Review

2.1 Māori, Maraes and Papakāinga in New Zealand

Māori are the indigenous people of New Zealand, also referred to as tangata whenua meaning people of the land. Traditionally tangata whenua lived in tribal areas where members of the same iwi (tribe) or hapu (sub-tribe) would live together at their respective marae (meeting grounds) and associated papakāinga (housing). Colonisation by the Bristish in the 1840 resulted in major impacts on the Māori population changing multiple facets of daily life and integration into mostly westernised living (Tapsell, 2002). However, despite these changes Māori still maintain ties with their respective Iwi and a distinct set of customs/values often referred to as tikanga (Garth Harmsworth, 2005).

There are upwards of 773 marae around New Zealand which incorporate a carved meeting house (wharenui), an open space in front (marae ātea), wharekai (dining hall and cooking area), toilets, shower blocks and potentially other facilities such as health clinics and papakāinga for residents/caretakers. Marae are used for hosting meetings, celebrations, funerals (tangi or tangihanga) among various other important tribal events (Tapsell, 2002).

2.2 Māori Values Surrounding Water

Māori have a spiritual and physical relationship with water (wai), thus are deemed the kaitiakitanga (guardians of geographic resources and knowledge) and treat it as a taonga (treasure). Many iwi make enhancing the health and wellbeing of local waterways a priority with Māori often considering personal and economic health to be closely linked with the health of the taonga including local water bodies (Morgan, 2006b). Māori co-habitation with the New Zealand environment in the past resulting in comprehensive knowledge of New Zealand ecosystems and how to sustain them. In conjunction with Māori cultural heritage being considered a backbone of New Zealand identity, the involvement of iwi offer a unique and relevant indigenous perspective around resource management (Ataria et al., 2018). The Treaty of Waitangi (1840) recognises the importance of tangata whenua in many aspects including the management of resources (water), this is further reflected in three key pieces of legislation, Environment Act 1986, Resource Management Act 1991, and Local Government act 2002.

Tangata whenua believe water and all water bodies have their own mauri (life force or essence) and is important to protect these taonga from pollution, degradation and damage. The importance of mauri can be interpreted by a Venn diagram of the mauri model (Figure 1, Morgan, 2006a) where economic, social and cultural health are a result of the environments health. Although, in the mauri model these factors (economic, social and cultural) are redefined as mauri of whanau (family), community and hapu (or more largely iwi) respectively.

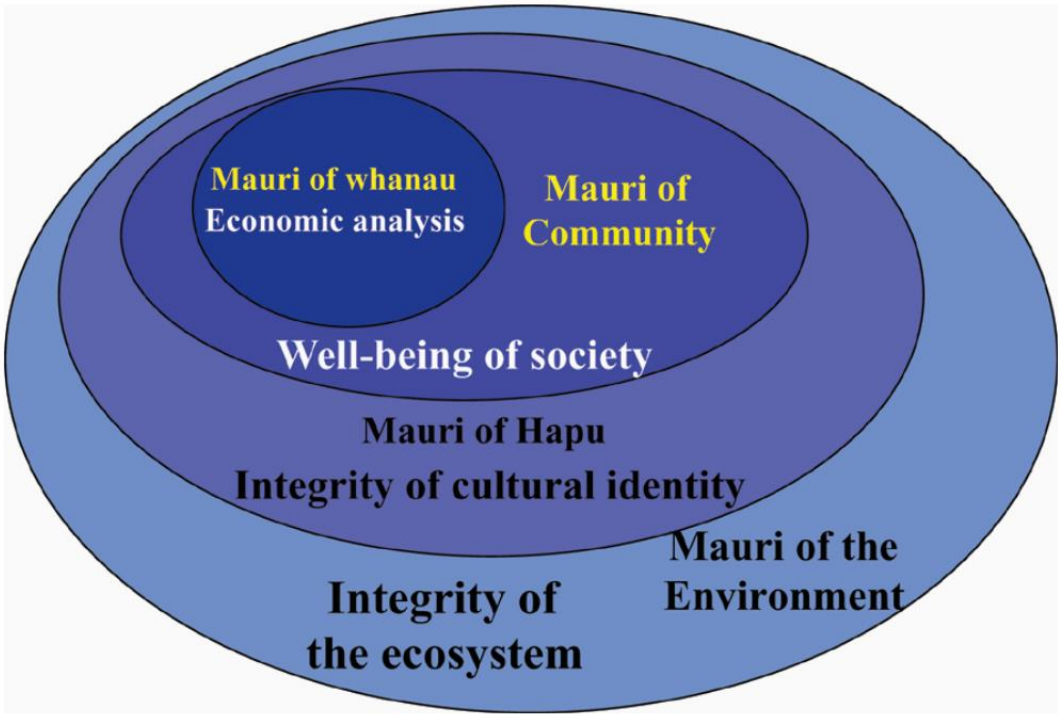

Figure 1: Venn diagram representation of Mauri Model

While there is no qualitative test for mauri, mauri is defined by the water types and sources (Morgan, 2006b). Ferguson et al., 2003 provides a highly simplified view of the traditional Māori water cycle (Figure 2) gives an overview of how mauri is lost through daily uses/pollution and replenished by passing through the earth and sea.

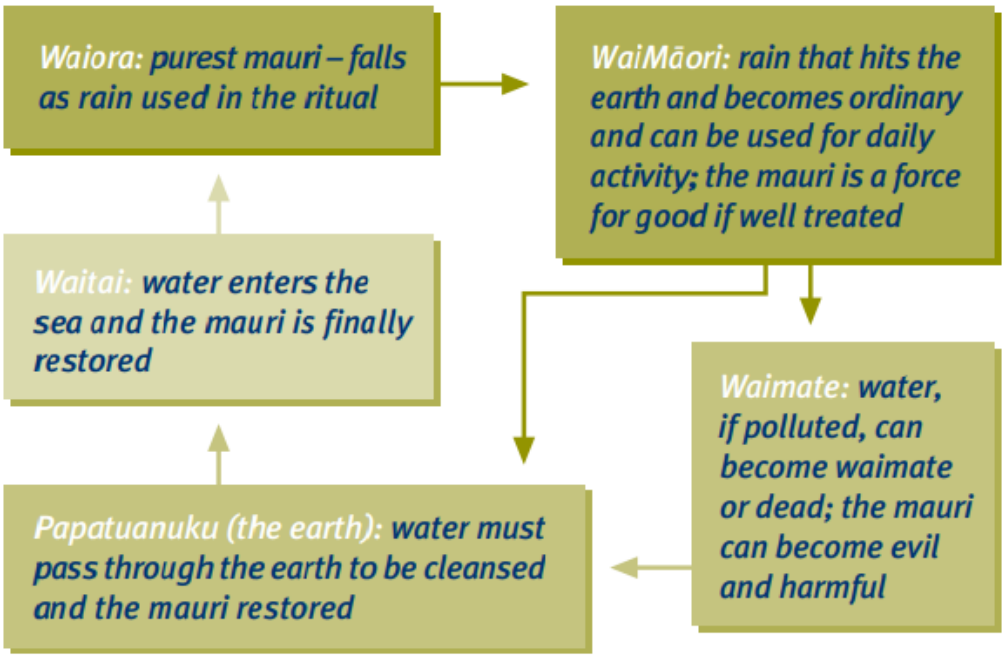

Figure 2: Simple form of water cycle in regards to mauri

The right way of doing things (tikanga), knowledge (mātauranga), values (uara) and principles (mātāpono) can vary between different iwis. However there are some values that are generally shared amongst all (GHD et al., 2020);

- Wai is a taonga and essential to life. It has a mauri and can be a medium for both enchancing and removing tapu
- Papatūānuku (Mother Earth) is a primal parent, the foundation of all life. Is the cleanser and the place where all life returns
- Human waste is considered tapu and needs to be kept separate from where people cook, eat, harvest food, talk and sleep
- Discharge to water, freshwater, recreation areas, marine environment and food crops is strongly disapproved
- A strong preference for land-based discharge/treatment (involving meaningful penetration rather than cursory passing over rocks or shallow ponds
- Higher quality of treatment for all contaminants is desirable
- A preference for centralised and individual local systems over decentralised municipal systems. The idea of using water as a medium for transporting waste is offensive
- Cost, land availability and geo-technical issues are a concern for land based systems

As a result it can be summarised that land application or some form of passage through soils is the desired method for purifying wastes (Tanner & Kloosterman, 1997).

2.3 Issues with Current Treatment methods

Te Puni Kōkiri, 2012 and Williams, 2014 reported the majority of marae using septic tanks draining to soil infiltration fields as the main form of wastewater treatment. Williams, 2014 reported 55% of maraes surveyed having issues with their wastewater systems and (Te Puni Kōkiri, 2012) reporting 24% of marae surveyed deeming their septic systems as inadequate. The need to improve current on-site wastewater processes for marae is clear, however there are some unique challenges summarised below:

- Limited funding available for system maintenance and upgrades
- High incidences of blockages and overflows
- Variability of flows due to events being hosted
- Distance/cost from service centre
- Land slips/movement
- Heavy rains and flooding (high water table and close to surface waters)
- Cannot put hāngi (pit in which food is cooked on heated stones) down in ground due to contaminated ground or high water table
- Existing systems being undersized or not meeting current codes/standards
- Pressure on land and receiving environment

NIWA has developed a ranked list of criteria, from working with 20 marae, for designing future wastewater systems (Williams et al., 2012). Table 1 shows a summary of the criteria. This list of criteria is reflective of the values held by the tangata whenua in wanting to protect the health of the people and environment, while upholding cultural values. In addition to the criteria listed, easy and accessible maintenance is a key issue with most marae preferring a member of the local community to perform the maintenance since most marae are rurally based.

Septic tanks and infiltration beds are only a primary treatment for wastewater, i.e. the preliminary removal of wastewater or sludge constituents, such as oil, grease and various solids (Tilley et al., 2014). They do not reduce the faecal bacteria count or nutrient contaminants in effluent (Tanner et al., 2012) and therefore secondary treatment is required for marae wastewaters before discharging effluent. Various mechanical

secondary treatment systems are available today such as aerated water treatment systems, sequential batch reactors, membrane bioreactors to only name a few. However, the varying load and requirement for low cost, maintenance and energy use make these unsuitable for implementation.

Table 1: List of criteria ranked from most to least important in future designing and implementation of wastewater systems at marae

Criterion
Avoid/minimise human health risk
Avoid/minimise contamination of the environment
System is in harmony with tikanga
Simple to operate and maintain
Reliability
Low energy/power requirements
Provides a resource use
Low Cost
Low odour risk
Minimal/low area requirements

One proposed solution addressing the concern of varying load rates is constructed wetlands to serve as secondary treatment prior to land application. NIWA has previously installed a constructed wetland system as a secondary treatment to a septic tank followed by infiltration beds (Figure 3, Waikato River Authority, 2017) at the Horahora marae in Huntly, New Zealand. The goal with this installation was to achieve higher effluent quality for land application and withstanding the varying load rates of experienced by the marae while requiring minimal maintenance and energy use. Therefore, constructed wetlands seem to be a good option to pursue in enhancing wastewater treatment at marae sites.

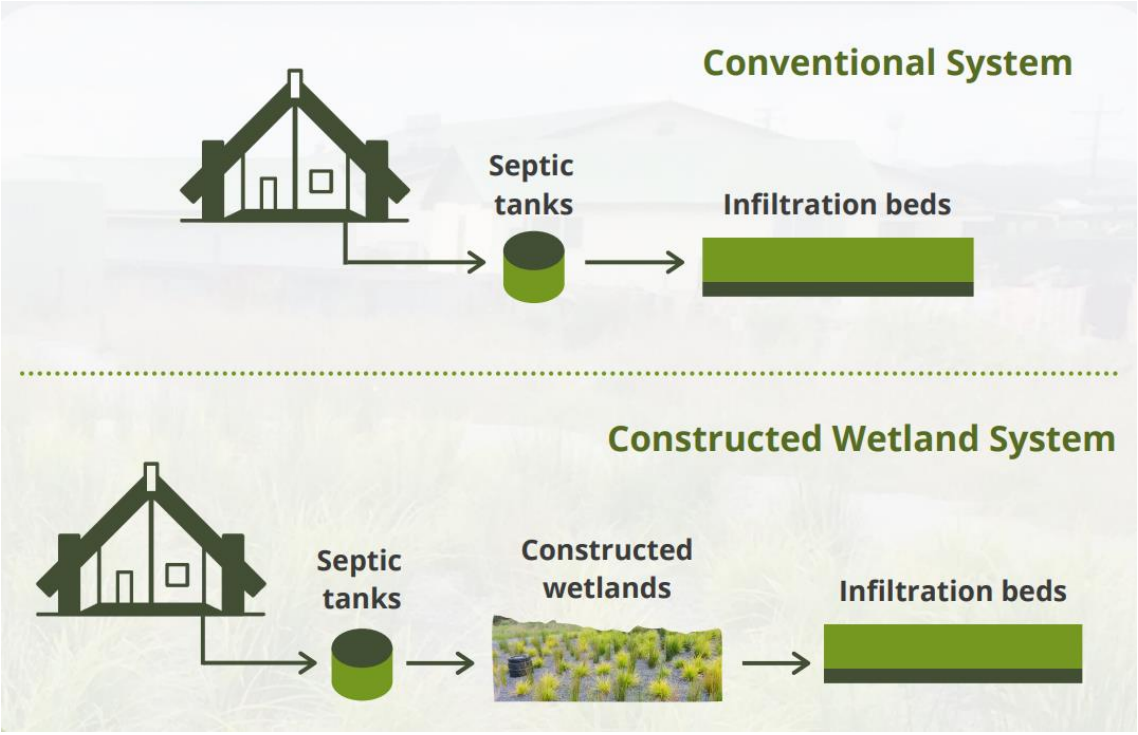

Figure 3: Horahora marae wastewater configuration

2.4 Treatment Wetlands as Secondary Treatment

Treatment wetlands (TWs) are artificially constructed wetland (CW) systems designed to enhance and optimise certain physical, biological and chemical processes that occur in natural wetland environments for the primary purpose of removing contaminants and treating wastewaters (Fonder & Headley, 2010). Treatment wetlands consist of;

- Macrophytic vegetation that typically occur in natural wetlands
- Water-logged or saturated substrate conditions for at least part of the time, and
- Inflow of contaminated waters (containing elements that are to be removed)

Contaminants are removed via processes involving the plants, substrates/soils and associated microbial populations (Vymazal, 2010).

Treatment wetlands meet a lot of the requirements in Table 1, being accepting of variable flow, have minimal power requirements, simple in operation, low maintenance and can be constructed out of locally available materials/resources. However, TWs do typically require a larger land area compared to conventional secondary treatment technologies (Vymazal, 2010).

TWs can be split into two main classifications, surface flow and subsurface flow, and classified even further (Figure 4).

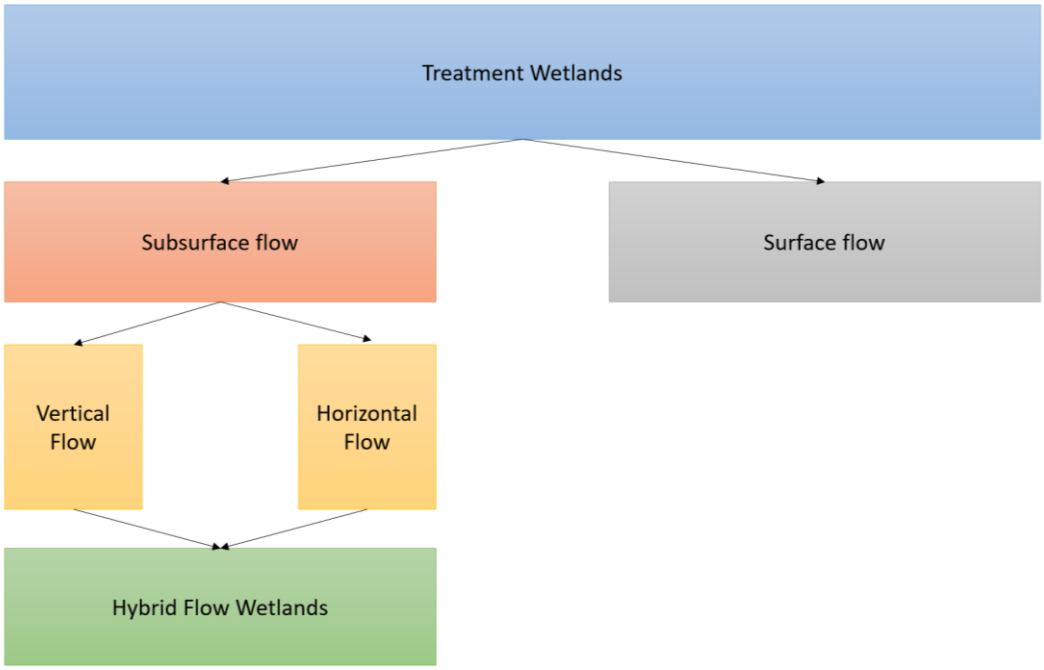

Figure 4: Major classifications of treatment wetlands

Surface flow wetlands are open body of water similar to ponds, with partial to complete coverage of the surface with macrophyte vegetation (Headley & Tanner, 2012). Wastewater flows horizontally over the vegetated soil interacting with the soil, plants and associated biofilm for contaminant removal. A basic example of a surface flow wetland from Vymazal, 2010 is shown in Figure 5. While there are many variations to surface flow wetlands, generally they are not fit for purpose with wastewater mix being exposed to air posing a threat to public and environmental health, requiring a large land area for implementation and potential for high maintenance (weeding excess plant growth). There are also issues concerning flooding and overloading with this type of wetland which would present themselves with high loading rates experienced by marae during the large events hosted.

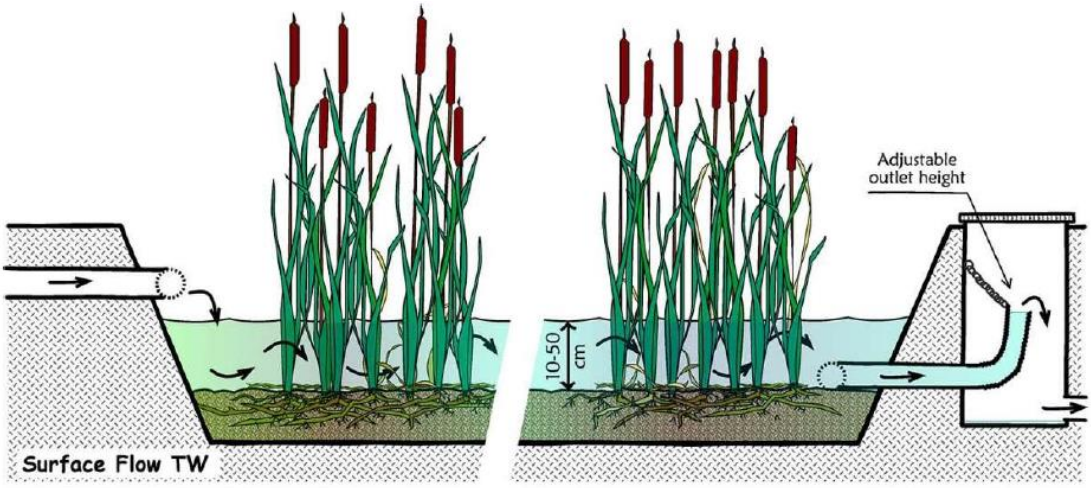

Figure 5: Surface flow treatment wetland

Subsurface treatment wetlands are a design where wastewater is passed through a basin filled with porous granular media (e.g. sand or gravel) planted with emergent vegetation (Tilley et al., 2014). These systems operate on the same principles as surface flow wetlands where contaminant removal is via interaction with the media, plants and associated biofilm. This class of TWs however can be designed for either horizontal (Figure 6, Vymazal, 2010) or vertical (Figure 7, Vymazal, 2010) flow depending on the treatment objectives and process requirements. Subsurface flow TWs are suitable for small (e.g. household) to medium (e.g. village of up to 2000 people) sized applications and good for supplementing existing primary treatment systems (i.e. septic tanks). These factors along with the design not readily exposing wastewater to public make it an ideal fit for implementation at marae.

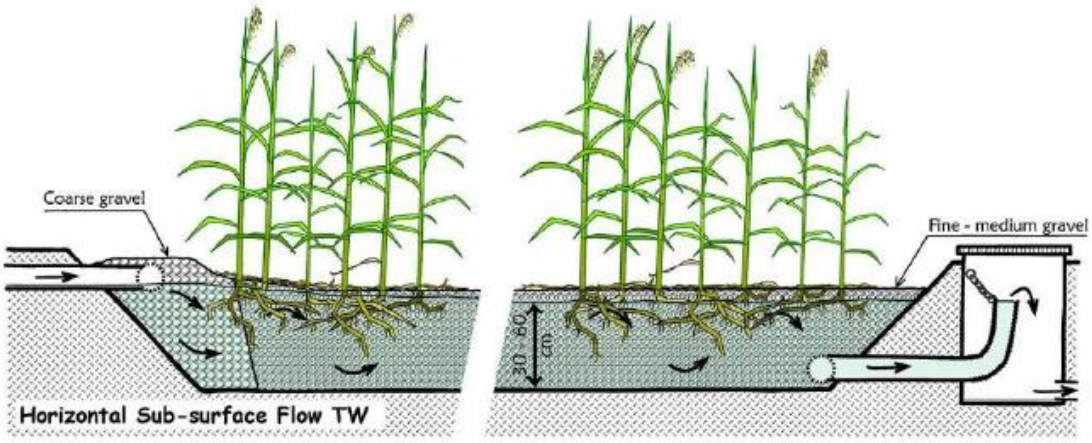

Figure 6: Horizontal flow subsurface treatment wetland

Horizontal flow subsurface wetlands consist of a porous media (typically sand, gravel or soil) filled basin planted with emergent macophytes (commonly *Phragmites australis* reed) (Tilley et al., 2014). As the name suggests, wastewater flows in a relatively horizontal path from the inlet to the outlet passing through the media. However, due to the basin being mostly saturated with a limited area where wastewater is exposed (and therefore limited oxygen mass transfer) microbial degradation of organics are largely due to anaerobic pathways (Vymazal, 2010). This environment therefore is capable of supporting denitrification processes, however struggles with the nitrification step involved in nitrogen removal resulting in total nitrogen removal being poor in this type of subsurface flow wetland. A positive for this type of subsurface flow wetland is that no electrical energy source is required with no requirement for a pump to achieve flow over the porous media.

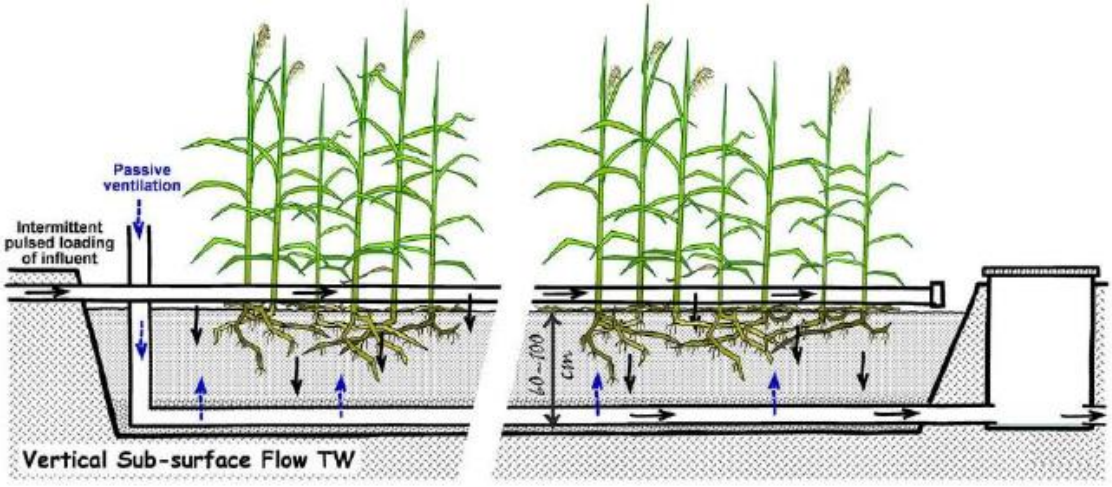

Figure 7: Vertical flow subsurface treatment wetland

On the other hand, vertical flow subsurface treatment wetlands consist of a planted (using emergent macophytes as with horizontal flow subsurface wetlands) filter bed consisting of typically sand or gravel where wastewater flows in a vertical direction. (Vymazal, 2010). The term, vertical flow, encompasses multiple variations of this wetland grouped into three main categories as down flow, up flow, and fill and drain TWs. Up flow and fill and drain variations pose a risk of surface flooding if not operated or designed correctly therefore are not suitable for this purpose. Down flow vertical flow wetlands (Figure 7) are the most common, they are free-draining and remain unsaturated for the majority of time. Typically a network of pipes are used to distribute flow across the surface of the bed in a way that avoids surface flooding. Oxygen mass transfer occurs as the bed drains and air is drawn into the filter bed allowing for aerobic conditions and a

high rate of nitrification. However, this results in poor denitrification performance due to the lack of anaerobic conditions. It is important to note vertical flow wetlands require more maintenance and have an electrical energy demand in the form of a pump (Dotro, 2017; Tilley et al., 2014).

Comparing horizontal and vertical flow subsurface treatment wetlands there are some notable factors. Both designs result in a high reduction of biological oxygen demand (BOD), suspended solids and pathogens with a relatively low operating cost. Despite the requirement for an electrical energy supply, horizontal flow wetlands generally require more land area, clog more frequently, have lower nitrification capabilities (therefore less nitrogen removal) and remove less nutrients than vertical flow wetlands. In light of these factors a vertical flow wetland seems appropriate for purpose however denitrification is very limited due to high oxygenation in porous media (Vymazal, 2007).

Partially saturated vertical flow wetlands (PSVF) are a recent type of design that aims to provide suitable conditions for both nitrification and denitrification to occur in a single vertical flow wetland unit. These are comprised of an unsaturated upper layer known as the free draining zone (FDZ) and a saturated lower layer known as the saturated zone (SZ). In the FDZ between influent loading cycles mass transfer of oxygen is allowed to occur, resulting in aerobic degradation of organic matter and ammonia removal via nitrification. Here suspended solids from influent are also trapped. While in the SZ a fixed outlet level allows for the constant presence of water resulting in limited mass transfer of oxygen causing anaerobic conditions for denitrification to occur. This design is a promising version of the traditional vertical flow wetland allowing for more denitrification (and therefore more total nitrogen removal) while maintaining the small land footprint they are known for. Morvannou et al., 2017 examined a commercialised version known as the Biho-Filter® and gives a visual representation of the configuration of the system (Figure 8).

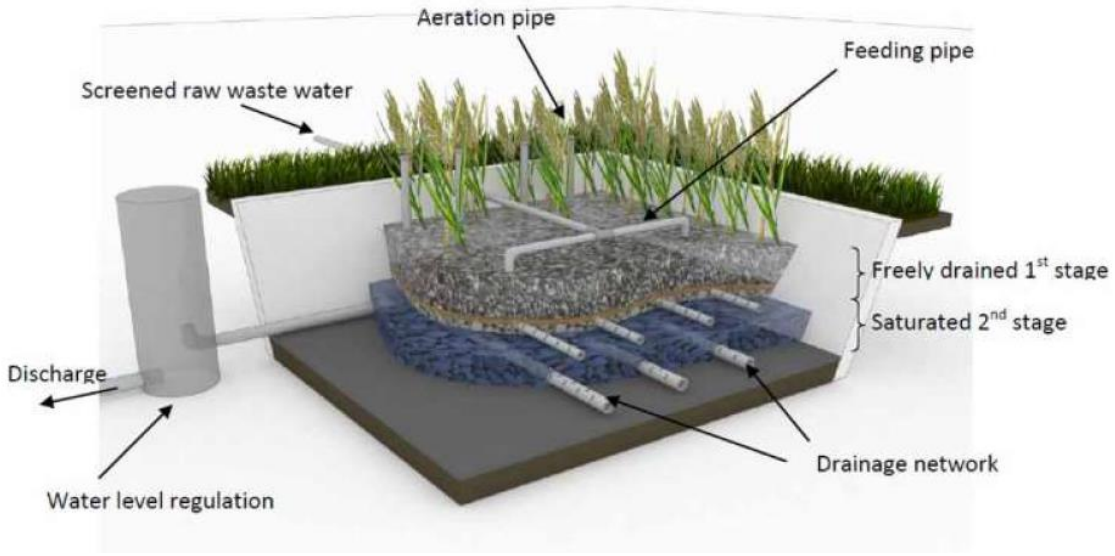

Figure 8: Visual representation of the configuration of a PSVF (Biho-Filter®)

Taking this design of vertical flow wetlands and its promising capabilities, NIWA has designed and implemented mesocosm scale replicates at the Pukete wastewater treatment plant (responsible for processing municipal wastewater from Hamilton, New Zealand) using standard municipal wastewater as the influent. These units used zeolites (crystalline aluminosilicate minerals) instead of the standard sand or gravel in the FDZ in an attempt to achieve high ammonium removal via nitrification. However this results in nitrate concentrations passing into the SZ that could not be sufficiently removed via denitrification.

Intensification of wetlands can result in improved performance of the wetlands and reduced land requirements. This can take place in two forms; use of external energy sources to increase aerobic capacity of the system i.e. mechanical aeration or effluent recirculation (Dotro, 2017) or the use of specific media aimed at better nutrient removal. While the use of external energy sources does not align with the criteria in this case, the idea of finding better media for nutrient removal aligns goal for marae to find uses for available resources. In this particular case where zeolites have been identified as a good material achieving high nitrification in PSVF wetland units the challenge is finding media to enhance denitrification performance in the SZ, and the focus of this research.

2.5 Nitrogen Removal

Nitrogen is one of the contaminants in wastewater that contributes to the eutrophication phenomenon. Eutrophication occurs when too much nitrogen enriches water, causing excessive growth of plants and algae. The result can even cause water bodies to turn bright green or other colours due to a smelly algae called phytoplankton, upon death microbes in the water decompose the phytoplankton. The process of decomposition reduces the dissolved oxygen in the water which can result in a dead zone that does not have adequate oxygen to support the majority of life forms (Aczel, 2019). Forms of nitrogen can also be toxic to aquatic organisms, while with land disposal, groundwater contamination is a risk. Therefore nitrogen and its various forms pose a potential risk to environment if not treated appropriately. Total nitrogen (TN) is a parameter for measuring nutrients in water and thus quality of water, this includes nitrogen in its many forms.

Nitrogen exists in various forms and interrelated processes convert it from one form to another in the nitrogen cycle (Figure 9, Henze et al., 2008). Waste water is comprised of both organic and inorganic forms of nitrogen. Organic forms include amino acids, urea, uric acid, purine and pyrimidines. Inorganic nitrogen forms include ammonium, nitrite and nitrate.

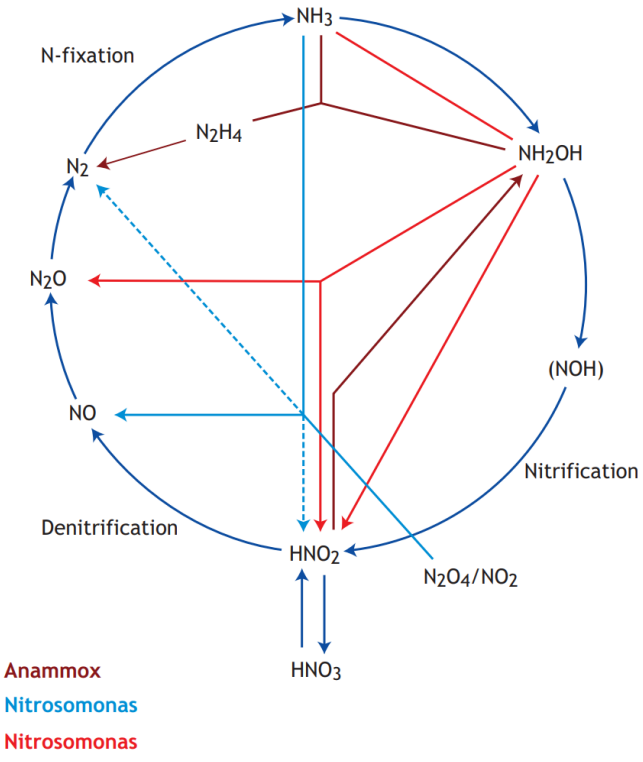

Figure 9: Nitrogen cycle and associated pathways for transformation between different forms

There are many conversion and removal mechanisms of nitrogen in TWs which have been studied extensively. These include biological mechanisms such as ammonification, nitrification, denitrification, plant uptake, biomass assimilation and annamox (anaerobic ammonium oxidation) awhile adsorption on media surfaces occurs as a physiochemical mechanism.

Plants can directly absorb and use nutrients (in this case nitrogen) from wastewater to support growth. They are able to consume two nitrogen forms, nitrate and ammonia, with ammonia being the more favourable nutrient for most plants used in treatment wetlands (Vymazal, 2010). This uptake results in the growth of the plants, which slows down during colder seasons. However, for net nitrogen removal the plants must be harvested. If the plants are not harvested and given a chance to decompose nitrogen in the plant matter will be released again (Dotro, 2017). Harvesting of plants is task that contributes extra costs and maintenance labour, undesirable for this scenario. This nitrogen removal mechanism compared with the nitrification and subsequent denitrification process contributes little in overall nitrogen removal. Also, as this occurs in the FDZ of PSVF wetland units it is of little concern for the excess nitrate concentrations passing into the SZ.

Biomass assimilation occurs to meet nutrient requirements for microbial activity that forms biofilm upon plant roots and media (Saeed & Sun, 2012). With microbial activity requiring such small amounts of nutrients for growth this mechanism is responsible for negligible nutrient (and subsequently nitrogen) removal.

Ammonium adsorption occurs within the FDZ of a PSVF wetland. In this case ammonium adsorbs to zeolite during loading and as a result during non-loading periods where oxygen mass transfer occurs allows for nitrification of the adsorbed ammonium. Upon the next loading cycle nitrate is then released into flowing wastewater into the SZ, which is a contributor to the issue of high nitrate concentrations experienced.

Anammox, a relatively new nitrogen removal mechanism, refers to the oxidation of ammonium under anaerobic conditions where nitrate is used as an electron acceptor to form di-nitrogen (N_2) gas. This process has a few advantages over the classical nitrification and denitrification process in requiring no carbon source, lower oxygen demand and low energy consumption. However, the bacteria responsible for this process

is slow growing resulting in long start up periods for bioreactors relying on this mechanism. These bacteria are also inhibited by excess presence of various substrates such as sulphide, ammonium and nitrate which are present in wastewater. Therefore, this mechanism would not be a major contributor in nitrogen removal in TWs treating wastewater.

Ammonification (mineralisation) is a biological process where organic nitrogen is converted to ammonia (NH_3) and inorganic ammonium (NH_4^+) as the end product. The process is performed by heterotrophic organisms (Kadlec & Wallace, 2008) which can occur in septic tanks. Urea and amino acids are the most prevalent examples of organic nitrogen in wastewater and are broken down as follows (1-1), (1-2);

- Urea

$$(NH_2)_2CO + H_2O \rightarrow 2NH_3 + CO_2 \quad (1\text{-}1)$$

- Amino acid

$$RCH(NH_2)COOH + H_2O \rightarrow NH_3 + CO_2 \quad (1\text{-}2)$$

Complete ammonification, the first step in breaking down organic nitrogen into ammonia, can occur in a well-designed septic tank which is achievable by updating septic tanks on-site at marae. This step achieves no nitrogen removal as such, but ammonia can be processed via nitrification and denitrification for nitrogen removal.

Conventional nitrification is a microbiological process that occurs in aerobic conditions and converts ammonia (NH_4^+) into nitrite (NO_2^-) and nitrite into nitrate (NO_3^-) (Henze et al., 1997). This occurs in the FDZ in PSVF wetlands where aerobic conditions are achieved between loadings causing oxygen mass transfer to ensue. Nitrification is facilitated by specific chemolithoautotrophic organisms which use ammonia (NH_4^+) or nitrate as an energy source, molecular oxygen as an electron acceptor and dissolved carbon dioxide as their carbon source (Ahn, 2006). Bacteria from the *Nitrosomonas* bacterial genus responsible for the reduction of ammonium to nitrite and bacteria from the *Nitrobacter* bacterial genus are responsible for the oxidisation of nitrite to nitrate. This results in two stoichiometric reduction-oxidisation reactions occurring (1-3), (1-4) (Saeed & Sun, 2012)

$$NH_4^+ + \frac{3}{2}\,O_2 \rightarrow NO_2^- + H_2O + 2H^+ \quad (1\text{-}3)$$

$$NO_2^- + \frac{1}{2}\,O_2 \rightarrow NO_3^- \quad (1\text{-}4)$$

Combining the two reactions above the overall stoichiometric reaction is given (1-5)

$$NH_4^+ + 2O_2 \rightarrow NO_3^- + H_2O + 2H^+ \quad (1\text{-}5)$$

Similar to the ammonification process, denitrification process only provides conversion of nitrogen from ammonia into nitrate and no actual total nitrogen removal. Therefore, this process needs to be coupled with denitrification to achieve total nitrogen removal from wastewater.

Denitrification is the major mechanism where total nitrogen removal occurs in treatment wetlands (Saeed & Sun, 2012). This mechanism is the final step of the conventional nitrogen removal route where nitrogen oxides, such as nitrite and nitrate produced in the nitrification process, are reduced to di-nitrogen (N_2). Heterotrophic bacteria responsible for this reduction include *Pseudomonas*, *Enterbacter*, *Bacillus*, *Micrococcus* and *Spirillium* (Knight et al., 2000) which under anoxic (oxygen deficient) conditions use the nitrogen oxides as primary electron acceptors and organic carbon compounds serve as electron donors to cause reduction. However in the presence of oxygen, oxygen is the preferred electron acceptor for these bacteria (Saeed & Sun, 2012). These conditions are met within the PSVF design previously mentioned where the SZ provides the anoxic conditions for denitrification.

Generally, denitrification occurs in the sequence below, with intermediate products of the process being toxic and objectionable (1-6) (Henze et al., 1997)

$$NO_3^- \rightarrow NO_2^- \rightarrow NO \rightarrow N_2O \rightarrow N_2 \quad (1\text{-}6)$$

Henze et al., 1997 states the denitrification process is affected by temperature slowing considerably when temperature is below 5°C, with optimum temperature being between 20°C and 25°C. The capabilities of the process are also affected by pH with the optimum pH being between 6 and 8 pH, with the process generating alkalinity pH is parameter that needs to be monitored for these systems.

Since these bacteria requiring organic carbon to serve as electron donors, availability of organic carbon is another restricting parameter to avoid restrictions on denitrification (Henze et al., 1997) suggests the C:N (carbon to nitrogen) ratio should at least be 5:1. Despite organic carbon being present in wastewater there usually is not enough available to be above this limiting ratio. Typically PSVFs are designed with gravel or sand as media and therefore do not provide any organic carbon to promote denitrification. Previously in horizontal flow wetlands where excess nitrate has been observed the systems are dosed with external organic carbon sources such as methanol, ethanol and glucose among just a few to aid in denitrification. However, these sources for organic carbon can become expensive, require a constant supply to be accessible and is not environmentally friendly, not suitable for the criteria desired by marae.

2.6 Materials to Improve Denitrification

The result of previous research leads to the problem this research is directed at, finding a suitable source of organic carbon to improve denitrification. The idea of using organic materials as media to serve as a solid carbon source for denitrification is not foreign however there is little research into their use in conjunction with a PSVF where nitrification is not the issue. There are multiple sources of organic carbon to consider which can serve as media in the SZ however not all these sources meet the criteria set out by marae in designing/operating a wastewater treatment system. Furthermore, for implementation in the SZ of PSVF units some other factors need to be considered;

- Longevity, as materials that are quickly consumed will need regular replacement
- Disposal of material post use, materials that require excessive processes in disposal add to the cost of implementation
- Hydraulic conductivity, media used in the saturated zone must be able to allow wastewater to pass through effectively

The use of media in horizontal flow subsurface treatment wetlands and other laboratory scale experiments with saturated media to enhance denitrification provides some idea of performance for this situation.

One example of media previously examined to support denitrification is shredded tyre chips, although in this example iron ions (Fe^{2+}) are the electron donors for autotrophic denitrification bacteria rather than organic carbon for heterotrophic denitrification

bacteria. Chyan et al., 2013 observed an average nitrate removal rate of 62.2% in horizontal flow wetlands (a good estimate for performance in SZ of a PSVF wetland unit). While the idea of reusing a waste material is ideal, procuring this material in constructing a PSVF unit for marae could be costly or inaccessible when more environmentally materials are available for use. Also the idea of excess heavy metals leeching into soil upon land application is not desirable.

Cotton is a material that has previously been tested to enhance denitrification. Si et al., 2018 reported 71% nitrate removal (at roughly room temperature) for vertical column filled with processed cotton and dosed with a synthetic wastewater. However in another experiment where low-quality unprocessed short fibre cotton was used denitrification was very promising but the entirety of the 380 g of cotton was consumed within 7 months (Volokita et al., 1996). This poses a potential problem for large scale applications at a marae where the constant replacement of cotton would become an issue. Furthermore, in New Zealand cotton is not abundantly produced which would cause issues in procurement of material and induce a large cost.

Coffee grounds are an excellent example of a by-product that could be used to promote denitrification. Keilhauer et al., 2019 observed up to 96.1% nitrogen removal in mesocosm scale floating treatment wetlands (a version of surface flow wetlands), however attributed a lot of nitrogen removal to plant growth and uptake. While the nitrogen removal is promising there will not be plant growth occurring in the SZ of a PSVF to remove nitrogen. Another study observed that coffee grounds struggled with denitrification as nitrate load increased (Jimenez et al., 1990). This could present issues with performance when loading rates vary constantly due to the nature of marae wastewater. Along with concerns over the hydraulic conductivity of the material would make it an inappropriate material for implementation as a carbon source in PSVF units.

Cork is another example of by-product being used as a solid carbon source for wetlands. Lorena Aguilara et al., 2019 used cork in hybrid TWs (vertical flow cell followed by horizontal flow subsurface flow cell) achieving up to 99% nitrate removal. This would be a great potential media given the high nitrate removal observed, however due to the lack of this by-product in New Zealand industry this material would be difficult to obtain at a low cost.

Newspaper is a solid carbon source available in many parts of the world. One study showed up to 98.87% nitrate removal suggesting that newspaper being a cellulose rich material provided ample carbon for denitrification (Si et al., 2018). Another study by Volokita et al., 1996 similarly observed complete denitrification using newspaper as carbon source in vertically packed columns. However, the study also observed the decrease in denitrification over time with mass loss of newspaper, issues with bacteria attaching to printed sections of newspaper and potential for severe clogging when nitrogen gas is trapped within the media matrix. These issues along with the move away from printed media suggests that newspaper would not be a suitable carbon source in this case.

(Martínez et al., 2018) conducted a study comparing nitrogen removal in PSVFs with and without an internal solid organic carbon source in the form of corncobs. This one of the few examples of using an organic carbon media in PSVFs. Figure 10 (Martínez et al., 2018) gives a graphical overview of the two systems compared.

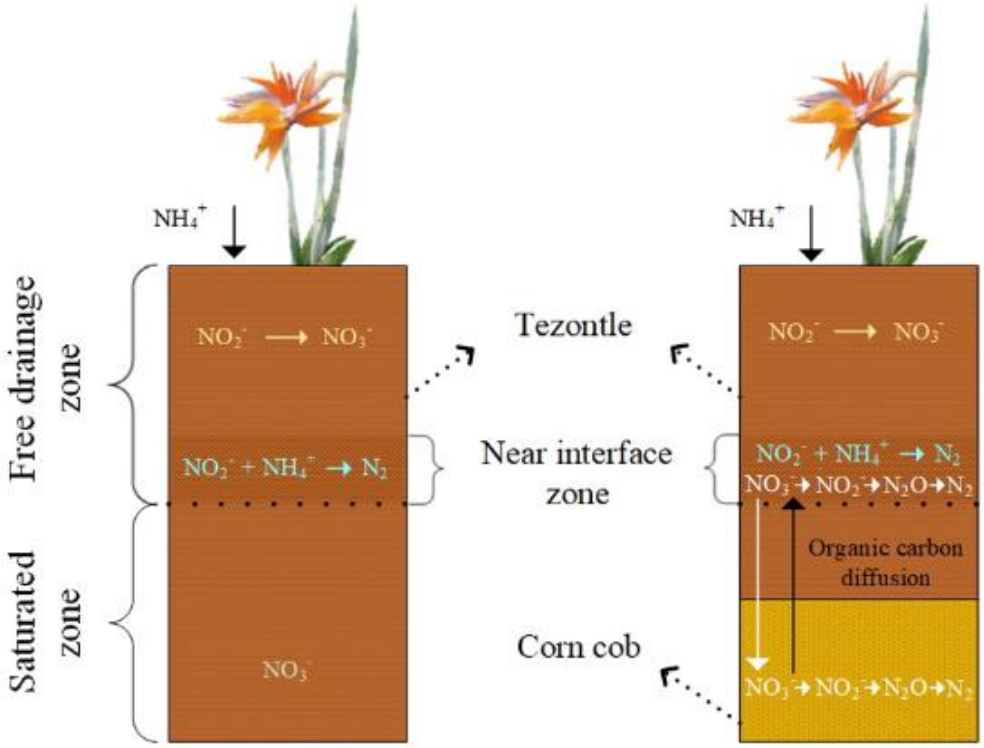

Figure 10: Graphical representation of pilot scale PSVFs used to compare the effect of an internal organic carbon source on nitrogen removal

The results showed that PSVFs with an internal organic carbon source of corn cob improved TN removal compared to the traditional PSVF without. The highest average

TN removal achieved using corn cob as media in the saturated zone was 73.2% but performance suffered as the biodegradable organics supplied by the corncob media was removed (i.e. mass loss). This is a strong reference point going forward to measure performance of PSVFs for TN removal for various organic media in the SZ.

There are vastly more materials that could be used as media in the SZ of a PSVF unit such as plant matter ranging from straw to coconut husks or even agave husks resulting from tequila production. However, with the criteria set by marae and finding obtainable materials within New Zealand seems to be the main factor, organic media options seem limited. One option is woodchips, in New Zealand there is a large amount of fauna therefore woodchips from various species of trees are available readily as waste from the arborist profession making them a good option to pursue. As for options provided by industry in the form of by-products there is brewers spent grain (BSG) from the brewing industry and slum gum from bee-wax industry are readily available options. Finding a use for by-products is an environmentally friendly approach and align well with values held by tangata whenua.

Woodchips have previously examined to determine the effect of certain parameters on denitrification in treatment wetlands, such as temperature, flow rate and hydraulic retention time. Tanner et al., 2012 monitored a treatment system made up of a gravel vertical flow wetland followed by two bioreactors in series, the first containing a woodchip mix of *Pinus raidata* and *Pseudotusa menziesii*, followed by one filled with coconut husks. The system had been running for two years suggesting woodchips have good longevity in this kind of deployment. The mesocosm scale PSVF scale systems were run using Pine woodchips without noticeable degradation, although there was noticeable colour present in effluent. As a material woodchips have proven to be robust (good longevity), have good hydraulic conductivity and provide a good environment (i.e. porous) for bacterial biofilm adhesion/growth making it suitable for use in this context. Thus the exploration of the large number of species of trees and how they perform in terms of denitrification going forward is the focus of this research.

Brewers spent grain has previously used as biofilter media to treat nitrate-rich synthetic groundwater at a lab scale (Benyoucef et al., 2013). The study used a packed up-flow column operating at pH 7-7.5, hydraulic retention time of 100 minutes and a nitrate load

of 199 mg/L resulted in complete (upwards of 99%) nitrate removal. A promising result for the media to be used in this case.

Slum Gum has not been used for denitrification as such previously but it is known to contain a large amount of organic matter (Morales-Corts et al., 2010) and is primarily used as nutrients to enhance plant growth agriculturally.

It is important to note that previous research has determined that a myriad of parameters play a factor in denitrification performance of configurations of wetlands, including temperature, particle size and system sizing which should be taken into account case by case. For by-products there is also the concern of pollution swapping with other minerals and chemicals being present as a result of industry, which should be closely monitored and checked before implementing on any large scale.

Zhou et al., 2022 performed a study on the effects of dosing sucrose, a high organic carbon content liquid, into microcosm scale vertical flow constructed wetlands filled. The results showing the sucrose does systems showed a TN removal efficiency of 93.5 ± 1.8% compared to 57.3 ± 3.3% of control systems filled with gravel. More importantly, nitrate removal efficiency of the sucrose dosed systems was 99.8% compared to the control systems of 52.5 ± 1.5%. While the cost of regularly dosing with an organic carbon rich source like sucrose for TWs when treating marae wastewater is not practical, this experiments validates the idea of using liquids rich in organic carbon to enhance not only nitrate removal (i.e. denitrification) but also total nitrogen removal.

2.7 Conclusion

In conclusion, the rural location of marae, values held by Māori and the criteria set out by marae proves challenging in finding a suitable solution to improve on-site wastewater treatment. Intensified PSVF wetland units are a promising potential solution in meeting those requirements. However, despite the many criteria this configuration of TWs are meeting, the poor denitrification performance of pilot scale versions of these are proving to be an issue yet to be solved. The research gap identified is surrounding the vast variety of organic materials that could be suitable for enhancing denitrification and even producing a leachate suitable for dosing PSVF wetland units. While woodchips have been previously examined for denitrification/nitrate removal capacity, the large variety of

fauna present in New Zealand offers many potential species which can be evaluated. Finding a purpose for industry by-products is supported by the environment friendly ideology of New Zealand, however with limited industrial processes producing high organic carbon content materials many of the options explored in other areas of the world do not apply in this situation. Therefore investigation into the materials available in New Zealand to local marae for the purpose of denitrification/nitrate removal is needed and is the goal of this research.

Chapter 3

Methods

This section covers the experimental setups to achieve each objective followed by how parameters are monitored and how data is analysed.

3.1 Experimental Setup

3.1.1 Phase 1

The objective of phase 1 is to monitor the ability of the selected materials to release carbon. Five litres of material are allowed to soak in five litres of tap water for 30 days. The material and water reside in 20 litre plastic buckets fitted with taps (and sealed) (**Figure** 11) to allow for easy extraction of leachate without disturbing media.

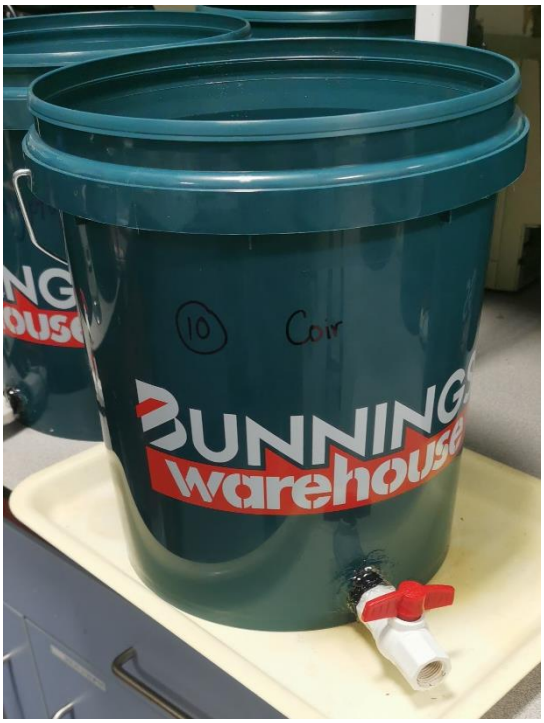

Figure 11: Example of bucket setup for phase 1 of experiments

Materials are dried prior to measurement until consistent weight in a 60°C oven. They are measured using a five litre measuring jug, filled half way then tapped 50 times, filled to the five litre mark and tapped 50 times again and then topped up to the 5 litre mark again. Mass of the materials are taken upon prior to soaking and again at the end of the 30 day experimental period after drying to consistent weight.

The large quantity of water used is to ensure media is completely submerged to ensure maximum contact areas between water and media allowing for maximum carbon release. It also allows for the extraction of samples without causing material to be unsaturated (above water). The long period of soaking is to ensure leachate and media stabilise to give a measure of how much carbon can be released into leachate without time restrictions.

A small sample (around 150 mL) is extracted via the tap every 2 days and tested for pH, conductivity and carbon oxygen demand (COD). Temperature is also taken for each time parameters are taken. Excess sample is returned to the bucket.

The results should give an indication of carbon release rates and maximum potential carbon available in leachate produced for each material. The carbon available can be used to evaluate the suitability of leachate produced by each material to be used for dosing a saturated zone in CWs to contribute extra organic carbon for denitrification. This should also give an idea on the ability of each material to serve as media in a saturated zone and their capacity for denitrification.

3.1.2 Phase 2

Using the buckets established in phase 1, the same material used in phase 1 is soaked in five litres of tap water. The leachate is replaced with fresh water every ten days and allowed to soak for three cycles.

The purpose of implementing repeating ten day cycles is to observe the effect on carbon release when the same media is reused with fresh water being introduced. This should give an indication if media is suitable for producing leachate, how many cycles are required for systems to reach steady-state and capacity for carbon release upon reaching steady state (i.e. operational CWs).

Mass measurements are taken of dried material (dried until consistent weight in a 60°C oven) prior to soaking and after the three cycles have been allowed to run. Temperature, COD, pH and conductivity is taken every 2 days during soaking. Small samples (around 150 mL) are drawn from the installed tap, with excess being returned to the bucket.

3.1.3 Phase 3

Figure 12: Plastic buckets used for systems in phase 3

The experiment is scaled down to one litre of material and one litre of nitrate solution soaked in a two litre BPA plastic bucket (Figure 12) with two replicates for each material due to work area restrictions. To emulate hydraulic residence time (HRT) experienced in the mesocosm systems setup at the Pukete site the nitrate solution is refreshed every 24 hours. However, the systems are allowed to soak for weekends without refreshing solution, resulting in longer HRT of 3 days. The selection of materials is narrowed down after analysis of results from phase 1 and phase 2 for this phase.

For phase 3, each replicate is dosed with 25 mg/L NO_3^- (nitrate) solution refreshed every day. One litre of concentrated solution (5 g/L $NO_3^{-)}$ is made up by dissolving 36.091 g KNO_3 (dried to consistent weight) in one litre of distilled water. The 25 mg/L NO_3^- solution is the result of diluting 5 mL of the concentrated solution with distilled water to a one litre volume.

Upon one day of HRT water/effluent is emptied via decantation and a small handheld screen (Figure 13) before refreshing with fresh nitrate solution. This allows the material to stay saturated while loading fresh nitrate into the systems. Solids caught on the screen are washed back into the system with new nitrate solution to minimise solid loss. This method was preferable over installing taps due to the small size of the system and to emulate similar conditions in a CW system where some fines will be lost to effluent as a

fine mesh/filter would cause severe clogging to occur and cause issues with system operation.

Figure 13: Handheld sieve used for draining leachate/effluent

Effluent for each replicate was monitored for temperature, pH, conductivity and nitrate concentration once per day prior to refreshing the nitrate solution. With COD taken at the end of each week in an effort to produce results as similar to an operational system as possible since the system has had 5 days of solution being refreshed each day. Results obtained from the two replicates are averaged to obtain a set of data to be analysed for each material, with maximum coefficient of variation calculated for all nitrate concentrations (being the parameter with the largest potential for variability)for each material to ensure validity of the results.

3.2 Monitoring Parameters

3.2.1 Mass and Volume

Figure 14: Mettler Toledo precision scale used for precision mass measurements

Figure 15: AND EK-15KL scale used for larger mass measurements

For precise mass measurements, such as preparing standards and nitrate solution, the Mettler Toledo precision scale in Figure 14 was used. While for larger mass measurements an AND EK-15KL scale was used (Figure 15).

Auto pipettes are used for small regular volume (1 mL–10 mL) measurements such as making nitrate solution in phase 3 and preparing COD samples. These are checked using distilled water upon setting volume and adjustments made to ensure accuracy.

Remaining volume measurements of large capacity are taken with appropriate sized laboratory standard measuring cylinders and jugs. Volume measurements for woodchips and other media are also taken using these with application of human judgement which will cause some inaccuracy due to trying to measure volume for solids. However, this will also occur in filling CWs experiencing similar variability.

3.2.2 pH, Temperature and Conductivity

pH, temperature and conductivity are monitored using handheld measurement devices. These devices are maintained to laboratory regulations with regular calibration and checked against standards (with adjustments to results made as required) prior to use.

Figure 16: Eutech pH 150 pH meter

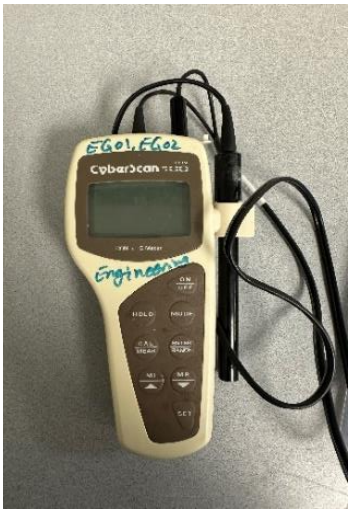

Figure 17: Cyberscan CON 100 conductivity meter

Figure 18: Center 372 RTD thermometer

The handheld devices used to monitor pH (Figure 16), conductivity (Figure 17) and temperature (Figure 18) are shown above. Buffer solutions of 4 and 7 pH were used to check accuracy prior to use. A conductivity standard solution of 1413 µS/cm was used to check for variation prior to use.

3.2.3 Chemical Oxygen Demand

Chemical oxygen demand (COD) was measured using the HACH high range COD vials and following method 8000 detailed by HACH (DOC316.53.01099). A HACH DRB200 reactor was used for digestion and HACH 2800 DR spectrophotometer (regularly calibrated and maintained) was used to obtain COD readings. A blank is produced using deionised water for each phase. Samples are diluted to fit within the range of 3 – 150 mg/L COD.

A brief overview of the method is as follows:

1. 100 mL of sample is blended for 30 seconds or until homogenisation
2. The homogenised sample is gently stirred with a magnetic stir plate in a beaker
3. Preheat the DRB200 reactor to 150°C
4. Remove the cap from the vial. Hold at an angle of 45 degrees and 2 mL of sample is pipetted into the vial (a new pipette tip being used for each sample)
5. Close vials, clean with water and paper towels. Invert the vial gently several times to mix
6. Place vials in the reactor and heat for 2 hours
7. Allow vials to cool to approximately 120°C or less (indicated by reactor)

8. Invert each vial several times while warm and place vials in a tube rack to cool to room temperature
9. Insert light shield and select program 430 COD LR on the HACH 2800 DR spectrophotometer
10. Clean the blank sample and insert the blank into the cell holder
11. Zero the spectrophotometer
12. Clean the prepared sample cell and insert into the cell holder
13. Read the result and multiple by dilution factor as required

3.2.4 Nitrate concentration

Nitrate concentration leachate samples in phase 3 was tested using method 4500-NO_3^- B. Ultraviolet Spectrophotometric Screening Method detailed in Bird et al., 2017 due to the nitrate range (0.2 – 11 mg NO_3^-- N/L) that needs to be monitored, the simplicity and speed of the test and availability of equipment.

The basis of the method is that nitrate (NO_3^-) calibration curve follows Beer's law up to 11 mg N/L. Measuring UV absorption at 220 nm allows a rapid determination of nitrate, however dissolved organic matter can also absorb at 220 nm but not at 275 nm, therefore the second measurement at 275 nm can be used to correct the nitrate value from the 220 nm reading if required.

The method from Bird et al., 2017 is suggested to be used for water with low organic matter and due to previous results should be true with denitrification consuming most organic carbon however, despite filtering, significant interference readings at 275nm were recorded for the majority of values obtained. The method quantifies significant interference if two times the 275 nm reading being more than 10% of 220 nm reading. In this case we ignore the adjustment of subtracting two times the absorbance reading at 275nm from the reading at 220nm as it would give negative absorbance readings and therefore negative nitrate concentration. As a result the nitrate concentrations obtained can only be used as rough estimates.

In using this method multiple nitrate standards must be created to fit a linear regression to calculate nitrate concentration. Details are as follows:

1. Prepare stock nitrate solution
    a. Dry potassium nitrate (KNO_3) in an oven at 103-105°C for 24 hours

b. Dissolve 0.7281 g ± 0.0005 g potassium nitrate in deionised water and dilute to 1000 mL
c. Preserve with 2 mL chloroform ($CHCl_3$)/L. Resulting in a solution stable for 6 months
d. Here 1 mL of solution contains 100 µg NO_3^-N

2. Prepare intermediate nitrate solution
    a. Dilute 100 mL of the previous stock nitrate solution to 1000 mL with deionised water
    b. Preserve with 2 mL $CHCl_3$/L
    c. Resulting in 1 mL of solution containing 10 µg NO_3^-N
3. Prepare calibration standards
    a. Create standards in the range 0 – 7 mg NO_3^-/L by diluting 0, 5.0, 10.0, 15.0, 20.0, 25.0, 30.0 and 35.0 mL of the intermediate solution to 50 mL using deionised water
    b. Read absorbance (after zeroing with deionised water) at 220 and 275 nm wavelengths
    c. Adjust the 220 nm absorbance with 275 nm absorbance if required (220 nm reading minus two times the 275 nm reading)
    d. Plot the results (Figure 19) and fit linear regression, obtaining intercept and slope of the regression (slope being 0.2313 and intercept 0.008)

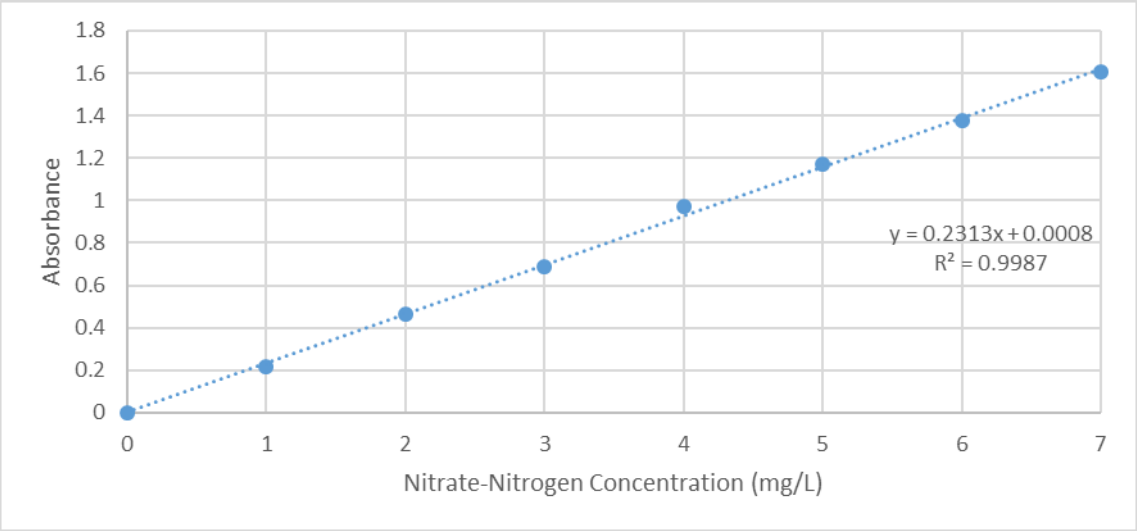

Figure 19: Nitrate-nitrogen standards plotted and fitted with linear regression

Roughly 100 mL (after mixing) of sample is passed through a GA-55 grade glass fibre filter (0.6 micron particle retention), changed for each sample, via a vacuum pump and flask. To 50 mL of the sample 1 mL of 1 M hydrochloric acid (HCl) solution is added to prevent interference from hydroxide or carbonate concentrations.

Samples are checked for absorption in a silica cell of 1 cm light path with a Shimadzu UV-1900 spectrophotometer (Figure 20). The silica cell is washed with deionised water then new sample between samples to ensure no cross contamination.

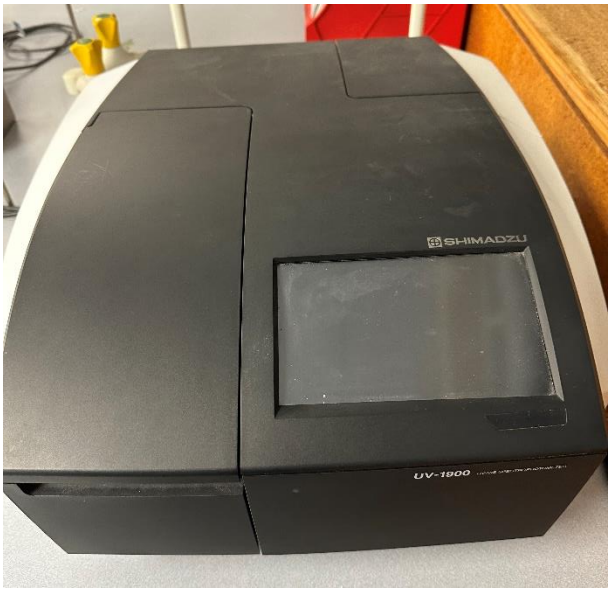

Figure 20: Shimadzu UV-100 spectrophotometer used for obtaining nitrate concentration

Samples and standards are then calculated using (1-7)

$$C = \frac{A - I}{S} \tag{1-7}$$

Where:

C = nitrate concentration (mg/L),

A = absorbance (samples without correction),

I = intercept of the regression line, and

S = slope of the regression line

3.2.5 Dissolved Oxygen

At the end of phase 3 dissolved oxygen (DO) was obtained using a Cyberscan DO 300 dissolved (Figure 21) oxygen meter to check whether systems are achieving anaerobic/anoxic conditions with some caveats explained in section 4.3.9.

Figure 21: Cyberscan DO 300 dissolved oxygen meter from Eutech instruments used to monitor DO

DO is measured by inserting the probe into sample water, continuously stirring releasing any air bubbles and providing fresh sample to the sensor until DO reading stabilises.

3.3 Analysis Methods

3.3.1 Mass Balance

In checking for mass before and after each phase of experiments mass balances can be performed for each system during each phase. Mass balances for the systems in combination COD and denitrification gives the ability to ascertain carbon release and nitrate removal capabilities of media on a weight basis. This information will also allow calculations regarding longevity of each material in phase 3 where operating conditions for larger systems are emulated. This also gives information on bulk density of the materials allowing sizing of CW systems based on carbon demand for denitrification.

3.3.2 pH

Collecting pH data for the laboratory scale systems is important in checking if systems are operating within the range for optimal denitrification (6-8 pH), a system high in

acidity or alkalinity can reduce denitrification capabilities. Therefore the pH of a system is an important consideration when selecting media to implement into CWs in future.

In phase 1 plotting pH to check for trends along with average pH over the 30 days will be used to estimate the pH of an operating system or leachate produced, checking if pH is within the optimal range for denitrification. In phase 2 average pH of the 10 day cycles for each material system allows for observation in trends occurring over multiple cycles. While average pH of the final cycle gives the best indication of pH when materials are used in-situ in a CW. The last average pH (between the two replicates) measured for each material in phase 3 gives the best estimate for systems operating at steady state.

3.3.3 Conductivity

Measuring conductivity for leachate gives an indication of the amount of impurities in the water. In phase 1 plotting conductivity over the 30 day experimental period gives an idea when impurities are being released and therefore how long it takes each material/leachate to achieve stability. Averaging conductivity for each of the cycles in phase 2 shows how the systems behave over multiple cycles and if/when the systems reach steady-state. Conductivity is less important and not analysed in phase 3 with the large number of cycles and differing HRT, however during the experimental period conductivity is used to ensure there are not any anomalies occurring.

3.3.4 Chemical Oxygen Demand

Chemical oxygen demand is the key parameter in monitoring carbon release from media into water/leachate. Using (1-8) from Dubber & Gray, 2010 COD can be converted to total organic carbon (TOC).

$$\mathrm{COD} = 49.2 + (3 \times TOC) \qquad (1\text{-}8)$$

In phase 1 plotting COD results and fitting with linear regression to the appropriate data points will give a carbon release rate. These plots (along with pH and conductivity data) will give an idea on the length of time required for the systems reach steady-state. Plotting, averaging, obtaining standard deviation and coefficient of variation of COD results for each 10 day cycle in phase 2 gives an idea on if/when systems reach steady-state

In conjunction with COD results and mass balances in phase 1 and 2 allows for sizing/viability of theoretical CWs and viability of dosing with leachate based upon carbon requirements for optimal denitrification. While COD results for phase 3 are used to monitor if the materials would contribute to COD in effluent from a constructed wetland.

3.3.5 Nitrate Concentration

Nitrate concentration and interference values in phase 3 is plotted to observe how systems behave on startup and therefore when/if systems reach steady state. As an estimate the lowest average nitrate concentration (of the two replicates) for each material is used to give an estimate on nitrate removal percentage if the materials were to be implemented into CWs operating at steady-state.

Chapter 4

Results and Discussion

4.1 Phase 1

In phase 1 the ability of the media to release carbon is examined when allowed to soak for 30 days, thus retaining all carbon released in that time. COD is used as an indicator for the released carbon in present in the leachate and an indication on the carbon release rate into water from each media source. pH is also monitored to observe potential alkalinity problems and conductivity as an indicator for the amount of impurities are present in the leachate. The result should an idea on how much carbon is available for each media, a release rate and the initial indicator for the suitability of each media for producing leachate for dosing CW systems.

4.1.1 Eastern Cottonwood Woodchips

Eastern Cottonwood or *Populus deltoides* is a low cost woodchip that is appropriate for use in the specified requirements of iwi and is international available. It has multiple uses in New Zealand and could be sourced as a waste product. As a woodchip material it can be used as woodchip on land as a means of disposal among other options. It is considered a hardwood and is light in appearance with past testing showing a similar denitrification capability as some other woodchips. Thus giving it unique properties in these tests as a light coloured hardwood which is readily available and at low cost.

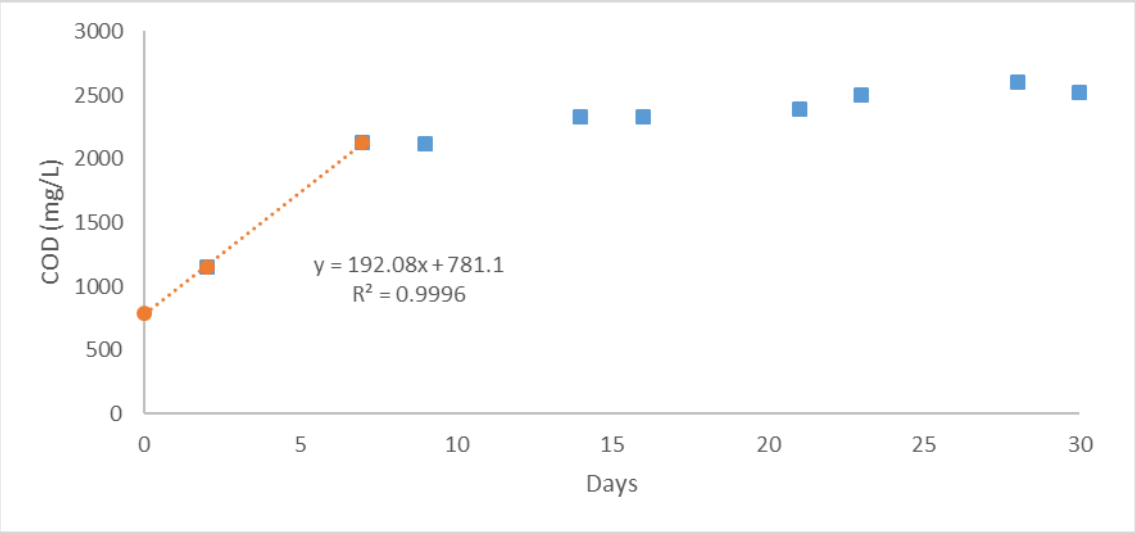

Figure 22: COD (mg/L) of Eastern Cottonwood woodchip leachate over 30 days with linear regression fitted for initial release and logarithmic regression for overall release

Figure 22 shows the Eastern Cottonwood leachate had a high rate of release for carbon into the water in the first 7 days before slowing down to stabilise at roughly 2500 COD mg/L. Examining the initial release rate of carbon over the first 7 days, where the majority of release occurs, to be at a rate of 192.1 COD mg/L.day over the first 7 days.

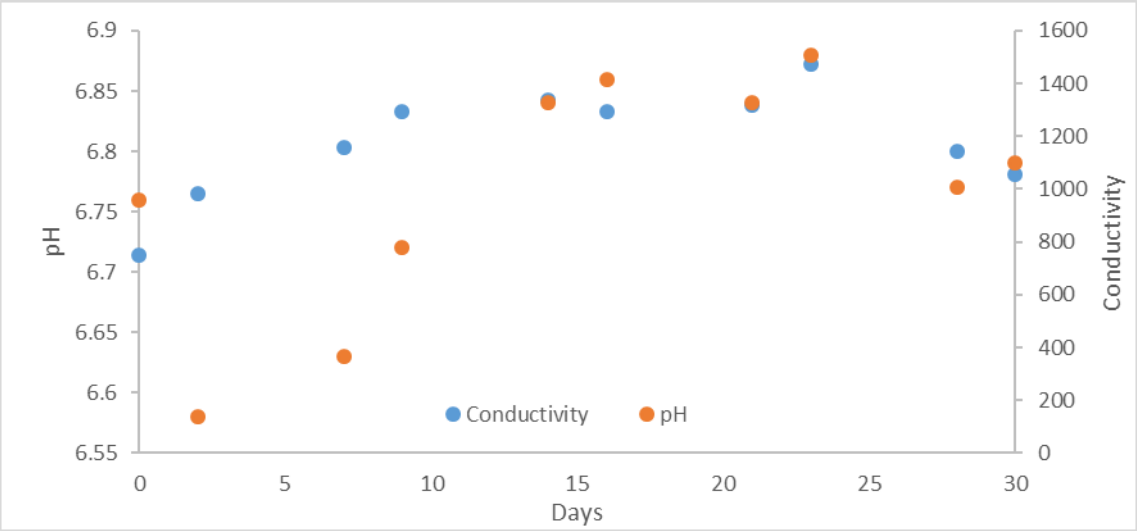

Figure 23: pH and Conductivity of Eastern Cottonwood woodchip leachate over 30 days

Figure 23 shows a steady increase in pH of the leachate over the first 14-15 days before stabilising. The system achieved an average pH of 6.8 with a relatively low range of only 0.3, leaving in it the optimum range (6-8 pH) for denitrification to occur. Conductivity started stabilising around day 9 suggesting that is when the media stopped releasing impurities into the leachate, while the variability could be potential microbial or fungal activity being present.

Figure 24: Eastern Cottonwood woodchips dry prior to soaking

Figure 25: Eastern Cottonwood woodchips and leachate at 22 days

The appearance of the material prior to soaking, shown in Figure 24, is relatively light in colour (apart from the bark present) with small to medium sized chips and reasonably

uniform without a large amount of fines present. There was minor fungal/ microbial growth visible as shown in Figure 25 and the leachate produced showed relatively light colour in line with the colour of the woodchips.

4.1.2 Mahogany Woodchips

Mahogany is available as a low cost woodchip as it can be a result of waste from uses in making furniture. Being a woodchip source it meets the criteria for use in the intended systems. It is a reddish-brown darker appearing hardwood which is relatively free of voids. The density and colouring of this hardwood make it unique with a relatively available source not only in New Zealand but also internationally.

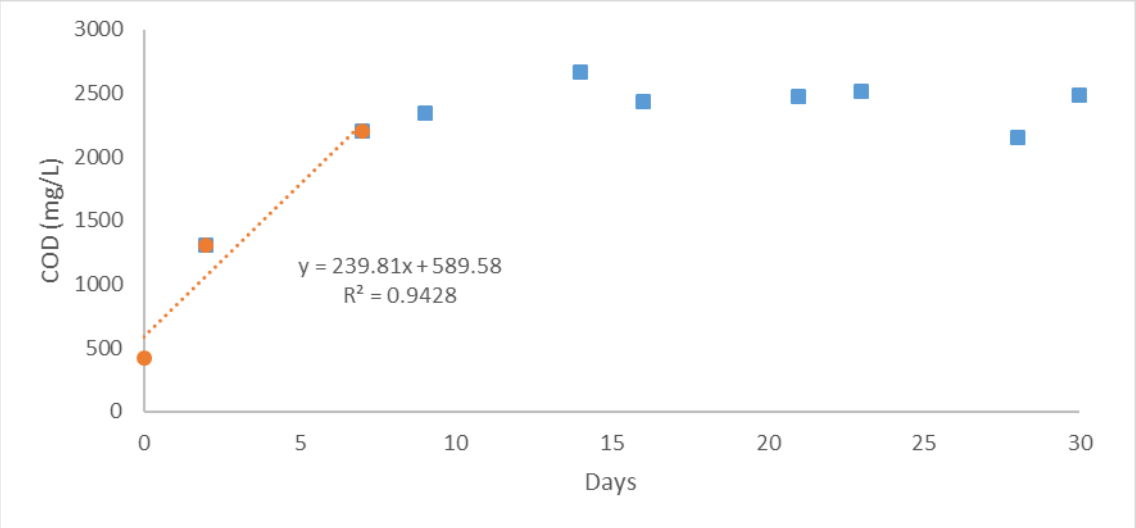

Figure 26: COD (mg/L) of Mahogany woodchip leachate over 30 days with linear regression fitted for initial release and logarithmic regression for overall release

COD of the Mahogany leachate (Figure 26) behaved similarly to the Eastern Cottonwood leachate (Figure 22) where the uptake carbon release is high in the first 7 days before stabilising. Similarly COD of the leachate seemed to stabilise around 2500 mg/L and stabilisation to occur around roughly 7 days in the system. However Mahogany had a faster initial release rate of 239.8 COD mg/L day compared to the Eastern Cottonwood material.

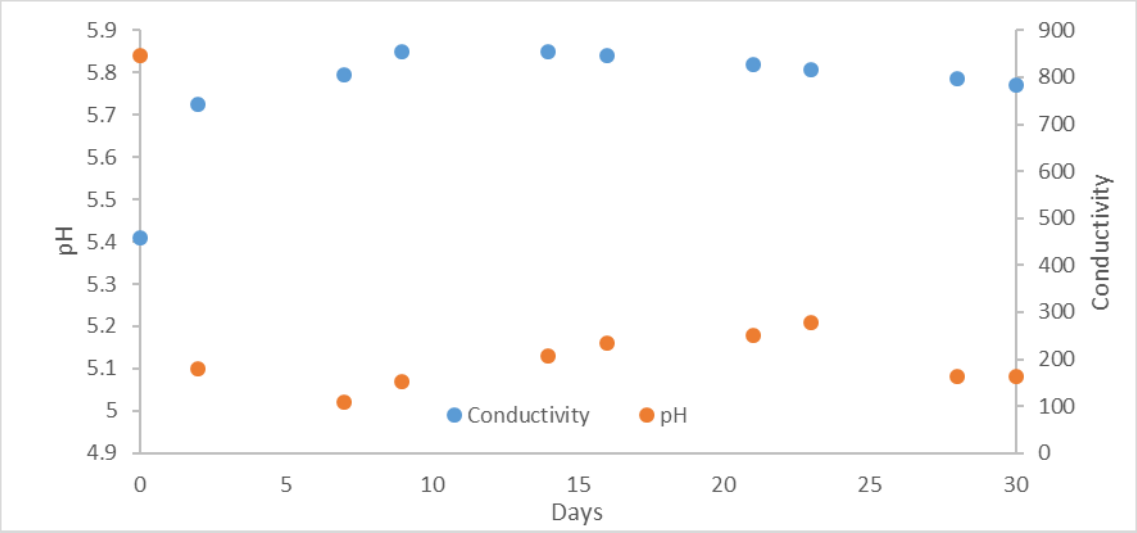

Figure 27: pH and Conductivity of Mahogany leachate over 30 days

pH and Conductivity (Figure 27) of the leachate followed the same trend established by COD in the leachate, stabilising after 7 days. The slow decrease in conductivity suggests impurities in the system started to decrease, perhaps suggesting fungal or microbial activity. Average pH achieved by the system was 5.2 with a range of 0.8 leaving it slightly outside the desired levels for optimum denitrification to occur.

Figure 28: Mahogany woodchips dry prior to soaking

Figure 29: Mahogany woodchips and leachate at 22 days

Mahogany woodchips are noted to be quite a darker wood and this sample containing lots of fine with a reasonably significant variability in the size of the chips (Figure 28). The leachate produced from this material thus also had a relatively darker colour to it in accordance with the colour of the material. Fungal/ microbial growth was not clearly visible in Figure 29 but could be present and would explain the variability in the previous conditions monitored.

4.1.3 Manuka Woodchips

Manuka is a tree native to New Zealand and south east Australia, therefore more out of reach for international applications. The material is relatively low cost material which can be sourced as a waste product from many uses including furniture and uses as cooking material. Therefore meeting criteria set previously. As a native hardwood with a light brownish colour it presents distinguishing characteristics for testing. The material used is a commercial version (still relatively cheap) intended for use in cooking applications.

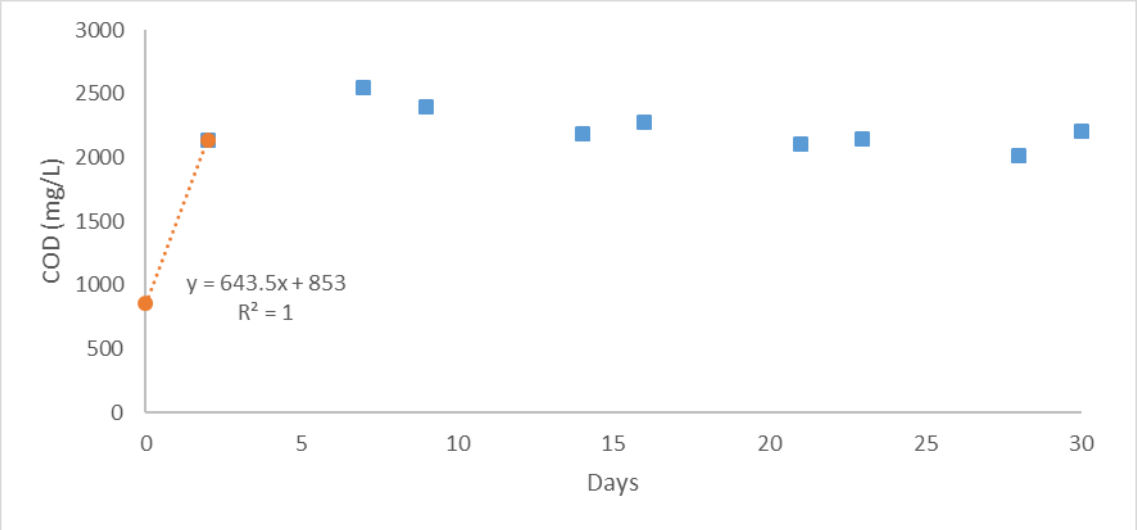

Figure 30: COD (mg/L) of Manuka woodchip leachate over 30 days with linear regression fitted for initial release and logarithmic regression for overall release

Figure 30 shows the Manuka leachate had a quicker uptake than the previous two systems with majority of carbon uptake occurring in the first 2 days at an estimated rate of 643.5 COD mg/L.day. However the COD in the leachate starts to trend downwards after 7 days, suggesting carbon in the leachate being used elsewhere.

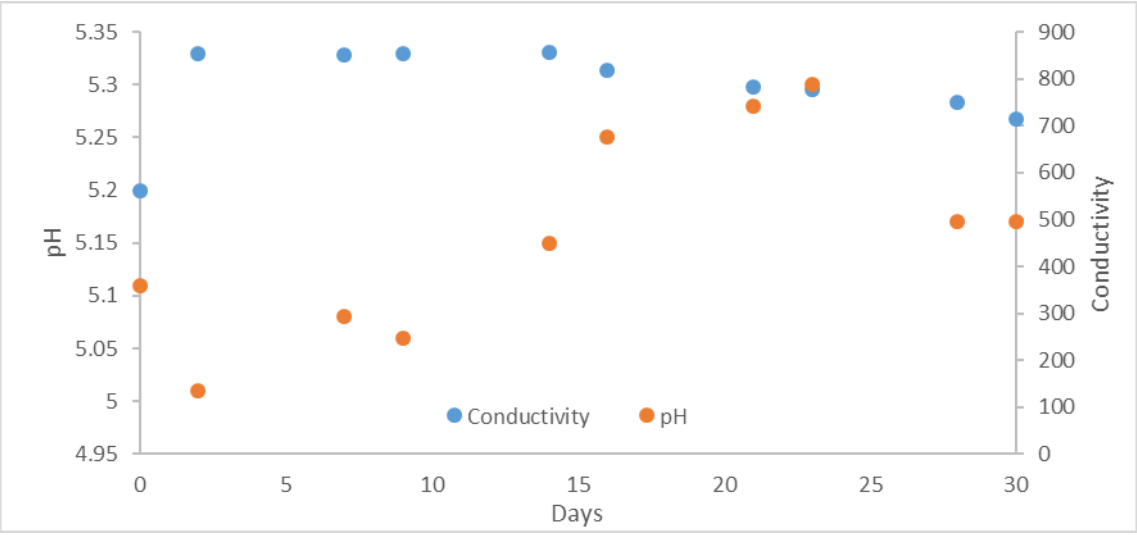

Figure 31: pH and Conductivity of Manuka leachate over 30 days

Figure 31 shows pH for the Manuka leachate does not show a steady trend over the 30 days, however with a low range over the 30 days of 0.3 and an average of 5.2. Thus this system sits outside the optimum range for denitrification but is rather stable in pH. Figure 31 also shows conductivity peaks at day 3 suggesting fungal or microbial activity starting and potentially removing some impurities in the leachate.

Figure 32: Manuka woodchips prior to soaking

Figure 33: Manuka woodchips and leachate at 22 days

Being a commercial product intended for cooking use the dry material is clean and did not have many fines with chips varying in size as can be seen in Figure 32. At day 22 (Figure 33) there is clearly visible fungal/microbial growth explaining the decrease in COD (Figure 30) and conductivity (Figure 31). Leachate took a little of the darker/red colour of the material but remained relatively clear.

4.1.4 Eucalyptus Woodchips

Eucalyptus is native to Australia that can be found all over the world. As a woodchip material it meets criteria required for a real-world system with low cost as it could be sourced as a waste product with multiple different uses. The material is also known for anti-bacterial properties which could also have implications in treating wastewater.

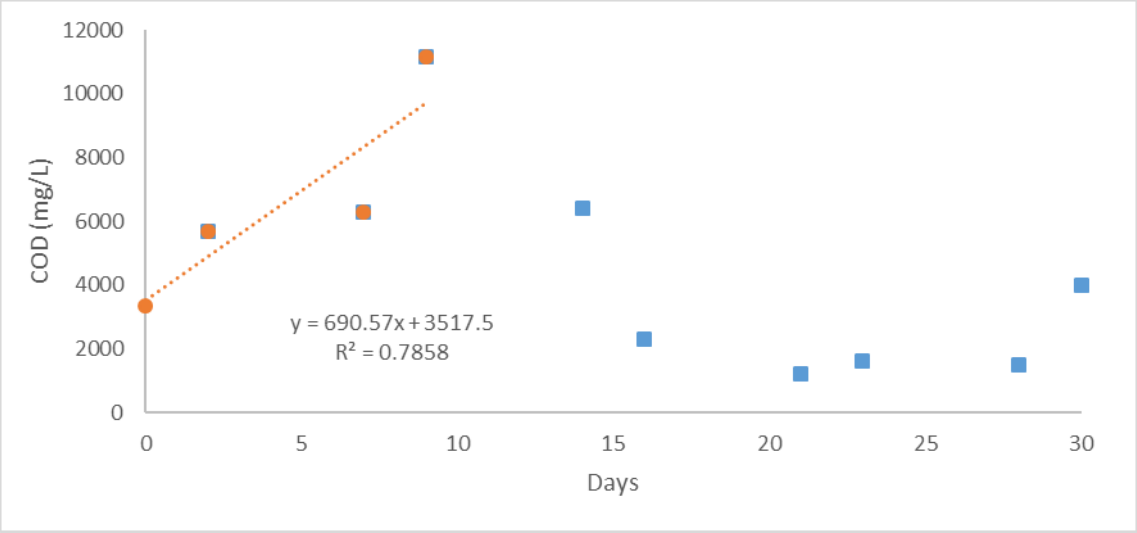

Figure 34: COD (mg/L) of Eucalyptus woodchip leachate over 30 days

Eucalyptus leachate produced the most variable trend of all woodchips tested in phase 1. Figure 34 shows a high peak of COD in the leachate at day 9, however the system drops drastically in the following days to around 2000 COD mg/L. This result makes the system somewhat unpredictable however with the peak being so high the material has good potential for carbon release. For the first 9 days the system achieved the highest initial release rate of 690.6 COD mg/L.day out of all the woodchip systems. However COD in leachate for this material rapidly declines after this peak suggesting carbon in the system is being consumed via fungal/microbial growth.

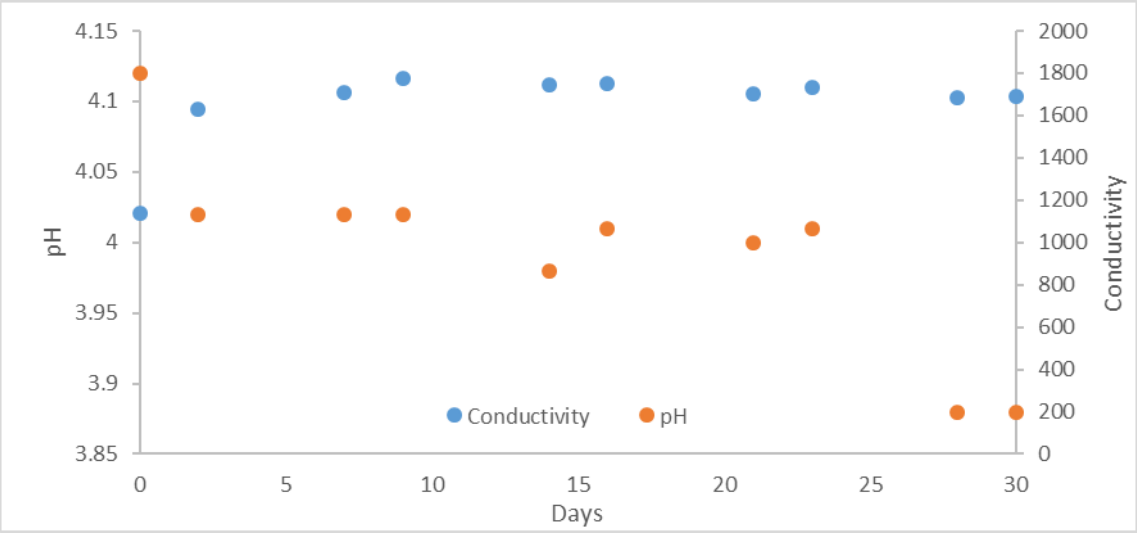

Figure 35: pH and Conductivity of Eucalyptus leachate over 30 days

Figure 35 shows pH of the Eucalyptus leachate started stabilising after day 2 however towards the end of the 30 day period there is a slight drop. The system averaged 4 with a small range of 0.2 placing it firmly outside the optimum range for denitrification. Conductivity also started stabilising after day 2 between 1600-1800µs/cm suggesting majority carbon uptake had already occurred in concurrence with pH stabilising.

Figure 36: Eucalyptus woodchips prior to soaking

Figure 37: Eucalyptus woodchips and leachate at 22 days

Chip size seemed to be relatively small on average and contained a lot of fines however this sample also had large pieces present shown in Figure 36. Figure 37 shows there was significant fungal/microbial growth present potentially explaining some of the variance in the trend for COD in the leachate, as the carbon could have been used as a food source for the growth. The Eucalyptus leachate had the darkest colour of all the woodchips possibly an indication of the high COD in the leachate.

4.1.5 Crack Willow Woodchips

Crack willow or *Salix x fragilis* is a relatively soft wood with multiple uses from cultivating honey to being grown for firewood. This is classed as a pest plant in New Zealand and is fast growing. Thus making this a relatively low cost material/ waste material which also meets requirements for real-world implementation.

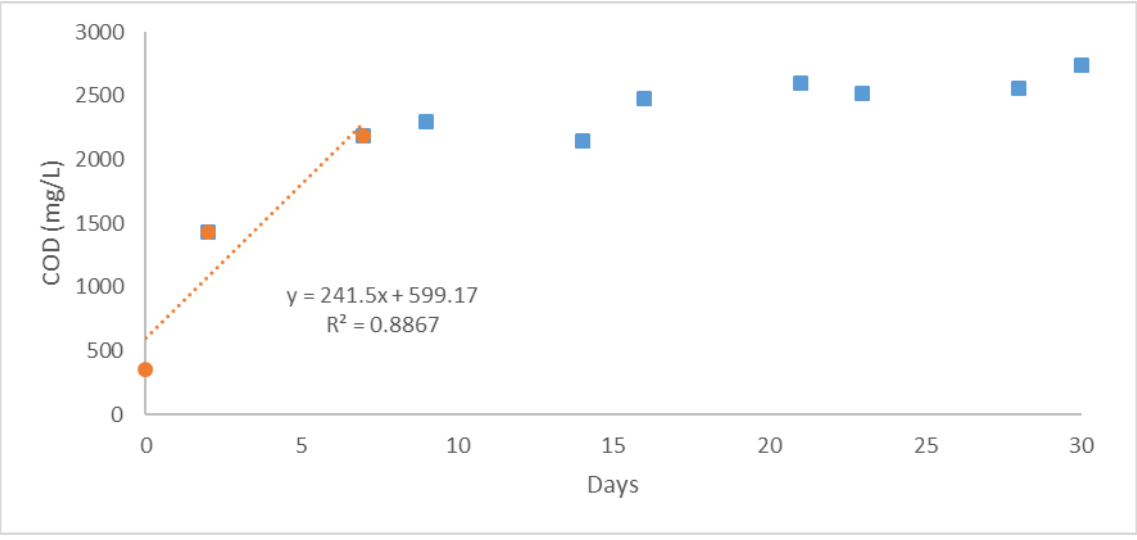

Figure 38: COD (mg/L) of Crack Willow woodchip leachate over 30 days

Figure 38 shows the leachate had an initial release rate of 241.5 COD mg/L.day the first 7 days before starting to stabilise. This initial release period and rate is consistent with some of the other woodchip material (Eastern Cottonwood and Mahogany) observed in this phase. However COD in the leachate appears to still be steadily rising towards the end of the 30 days of monitoring, albeit slowly, suggesting the carbon release is still occurring.

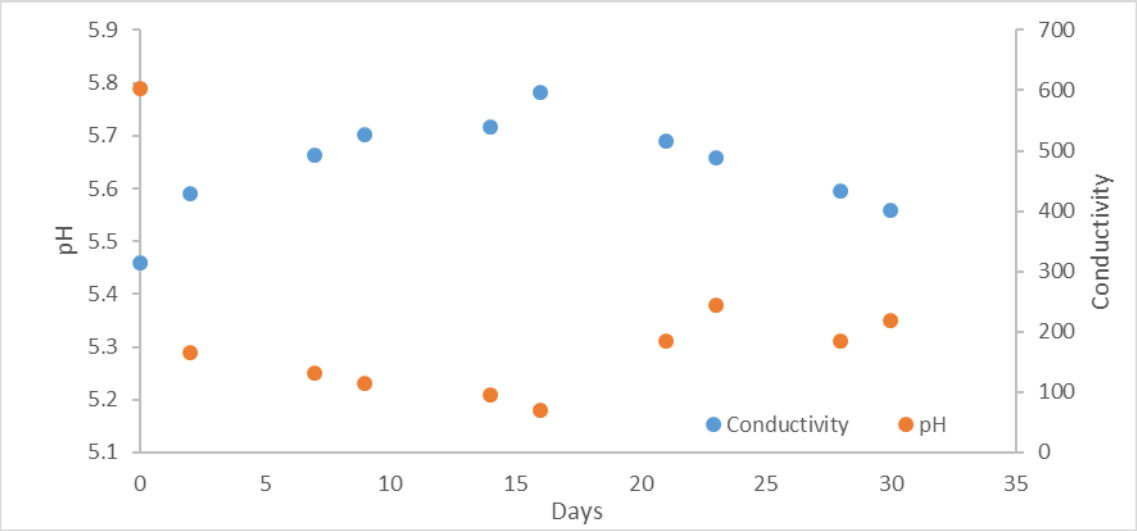

Figure 39: pH and Conductivity of Crack Willow leachate over 30 days

pH of the system quickly dropped in the first 2 days and then varies between 5.2 and 5.4 suggesting there is some acidity present in the woodchips which transfers to the leachate, as shown in Figure 39. Average pH of the system was 5.3 with a range of 0.6, which places it outside of the ideal range for denitrification but is a reasonably stable system. Conductivity peaks at day 16 and there is a steady rise prior and decrease after the peak, later than the previous woodchips examined. This longer period before the peak along with the trend shown by COD in the leachate (Figure 38) suggests this material would take longer than some other woodchips tested.

Figure 40: Crack Willow woodchips prior to soaking

Figure 41: Crack Willow woodchips and leachate at 22 days

Figure 40 shows the large variability in the sizing of the woodchips with more large pieces than majority of the other woodchips used in testing. The presence of these larger pieces could explain the slower uptake of carbon in the leachate. There was not visible fungal/ microbial growth at 22 days (Figure 41) which could also be an indicator of the slower release of carbon of this system

4.1.6 Princess Tree Woodchips

Princess tree or *Paulownia tomentosa* is a hardwood native to central and western China and is extremely fast growing and tolerant of many conditions. Being an ornamental tree woodchips could be a result of trimming or culling making it a low cost material but could be harder to obtain but meets conditions for potential real-world implementation. The Princess tree has also been suggested for use in carbon capture projects (Thunder Said Energy, 2020), giving this material an interesting set of properties.

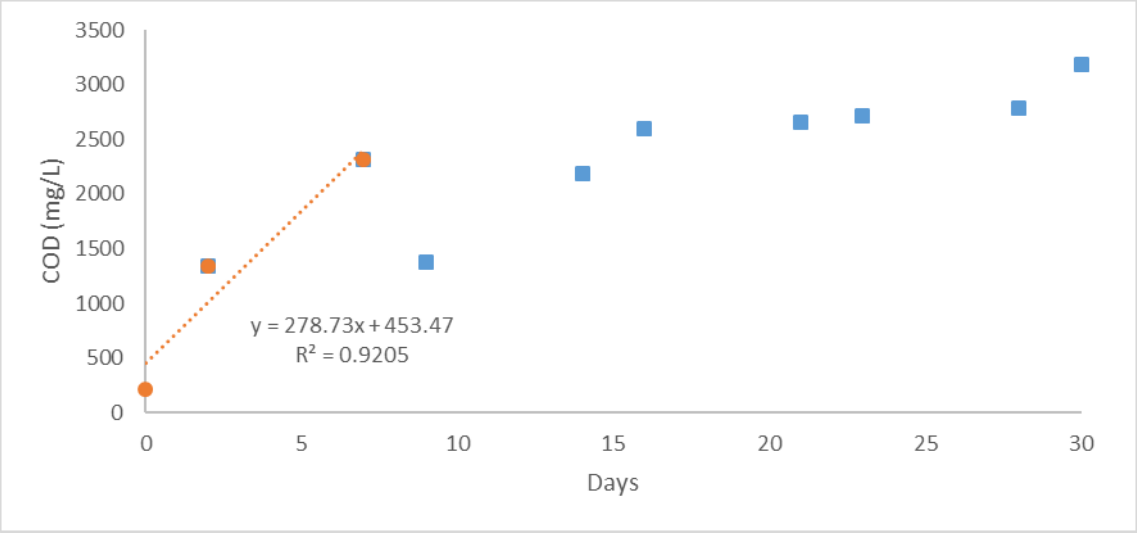

Figure 42: COD (mg/L) of Princess tree woodchip leachate over 30 days

Figure 42 shows COD in the leachate quickly rises over the first 7 days at a rate of 278.7 COD mg/L.day, before a decrease in COD and then rising again. The initial release period is the concurrent with those observed in Eastern Cottonwood, Mahogany and Crack willow systems but had the highest release rate. The slight decrease in COD could be the start of fungal/microbial growth and carbon being used as a food source. However despite this drop in COD the system indicates a quick uptake of carbon in the first 7 days and then uptake rate slows down but still steadily trends upwards, even peaking on day 30.

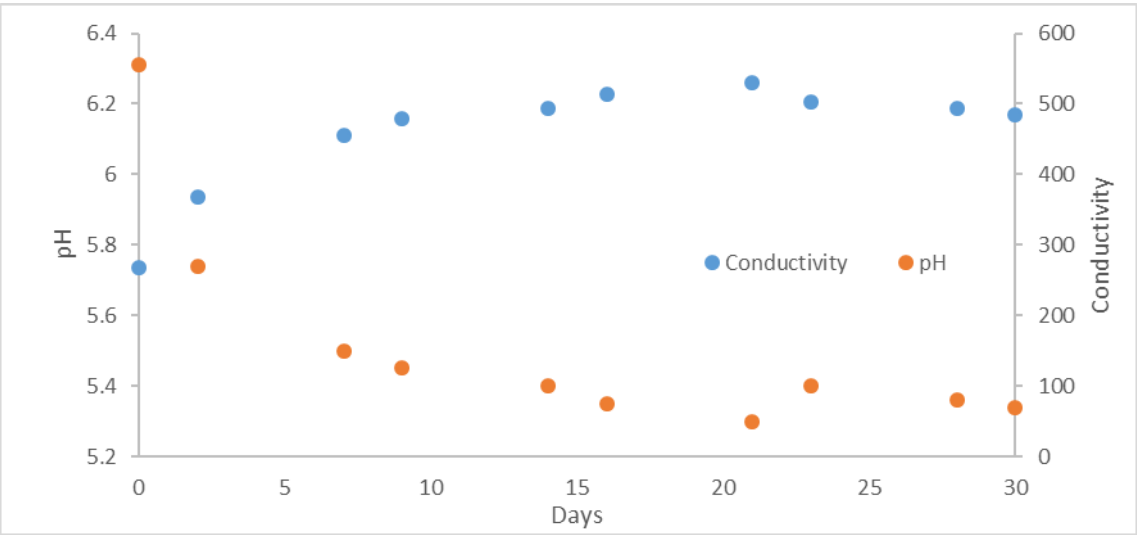

Figure 43: pH and Conductivity of Princess tree leachate over 30 days

Figure 43 shows a steady decrease in pH over the first 7 days before slowing the rate of decrease. As with the other woodchip leachates the pH drops however the Princess tree leachate does this over a longer period of time before stabilizing. Average pH of the system is 5.5 with a range of 1, however it is important to note the system started at 6.31 on initiation. Therefore the system has the potential with a lower HRT to operate within the optimum pH range for denitrification. Conductivity quickly increases over the first 7

days then stabilizes. This is in concurrence to the trends shown by pH and COD content of the leachate where the system starts to stabilise around day 7.

Figure 44: Princess tree woodchips prior to soaking

Figure 45:Princess tree woodchips and leachate at 22 days

Princess tree woodchips had a light appearance and few fines, as shown in Figure 44, with chip sizes being reasonably small. A small amount of fungal/microbial growth is visible which can be seen in Figure 45 providing some context to the variability in monitored for this phase.

4.1.7 Pine Woodchips

Pine is one of the more extensively used types of lumber, therefore should be readily available as offcuts or waste being produced as a result of industry. Pine is available internationally and its properties has been examined prior therefore can set a benchmark for comparison. These factors and being a woodchip material, pine is well suited candidate for our real-world application. The woodchips used here are of the *Pinus subg. Pinus* variant, a hard wood with a yellowish appearance.

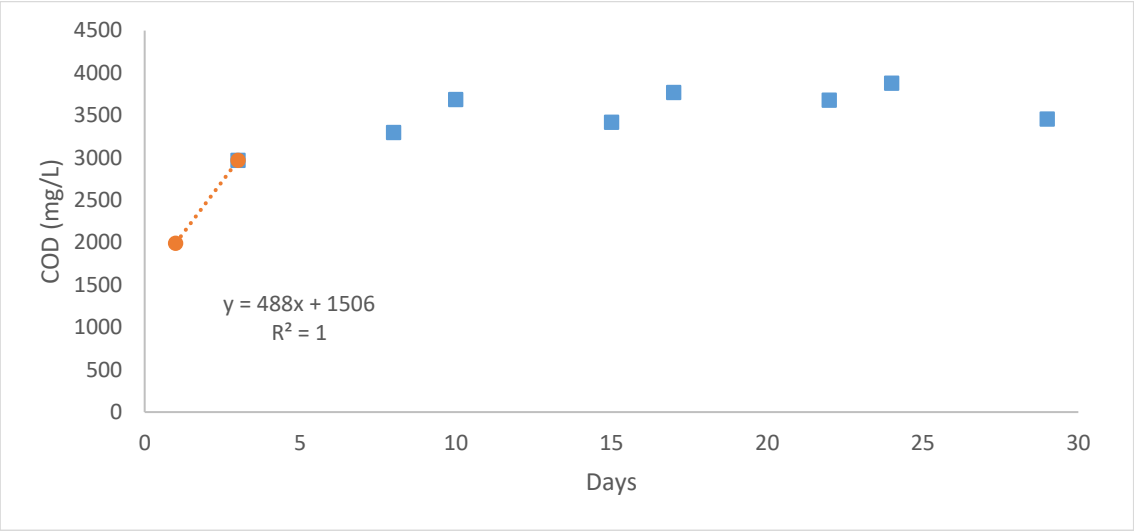

Figure 46: COD (mg/L) of Pine woodchip leachate over 31 days

Figure 46 shows an initial release carbon in the leachate in the first 3 days at a rate of 488 COD mg/L.day comparable to the Manuka woodchip system but with a higher rate of release. The system starts to stabilise above 3500 mg/L of COD after this initial release period. While this does not peak as high as the Eucalyptus leachate it stabilises at a higher point than the other woodchip materials examined. This suggests that carbon available in the leachate would also be higher than the other woodchip materials. However, it can be seen that COD follows an upwards trend followed by a downwards trend after stabilising suggesting there is some carbon being used, perhaps due to fungal/microbial growth or other processes in play.

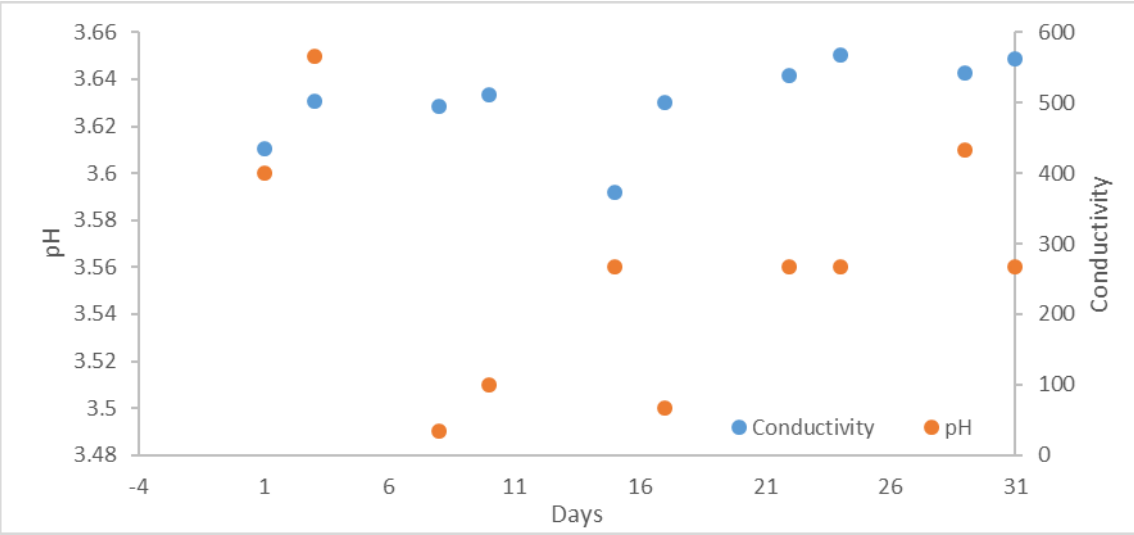

Figure 47: pH and Conductivity of pine leachate over 31 days

Figure 47 shows that pH of the system increases in the first 3 days before following a similar trend to COD with no steady trend established, however this is within a small range of 0.16 which could be due to inaccuracies of the pH meter. With an average pH of 3.6 and a low range the leachate produced is outside the optimal range for denitrification.

Conductivity over the 31 days also behaved similarly with variability being shown but does seem to slowly increase over the experiment period. The low range of results for pH, conductivity and COD suggests Pine woodchips achieve a reasonably stable leachate relatively quick.

Figure 48: Pine woodchips on initiation of phase 1

Figure 49: Pine woodchips and leachate at 31 days

Figure 48 shows the Pine woodchips were relatively small in size but included some medium sized chips and fines. The appearance is slightly yellowish and caused the leachate to be quite yellow in appearance. Figure 49 shows there was no significant fungal/microbial growth at 31 days of soaking. This could be due to the acidic nature of the leachate.

4.1.8 Brewers Spent Grain

Brewers spent grain (BSG) is a by-product of the brewing industry (originally barley) thus is readily available and should be reasonably cheap being a waste material. BSG is a material composed of cellulose, hemicelluloses, lignin and protein, which is similar to the woodchips previously examined. The waste material has a multitude of uses including animal feed and fertilizer making disposal low impact and easy. With these factors and being natural material, BSG meets requirements for real-world implementation.

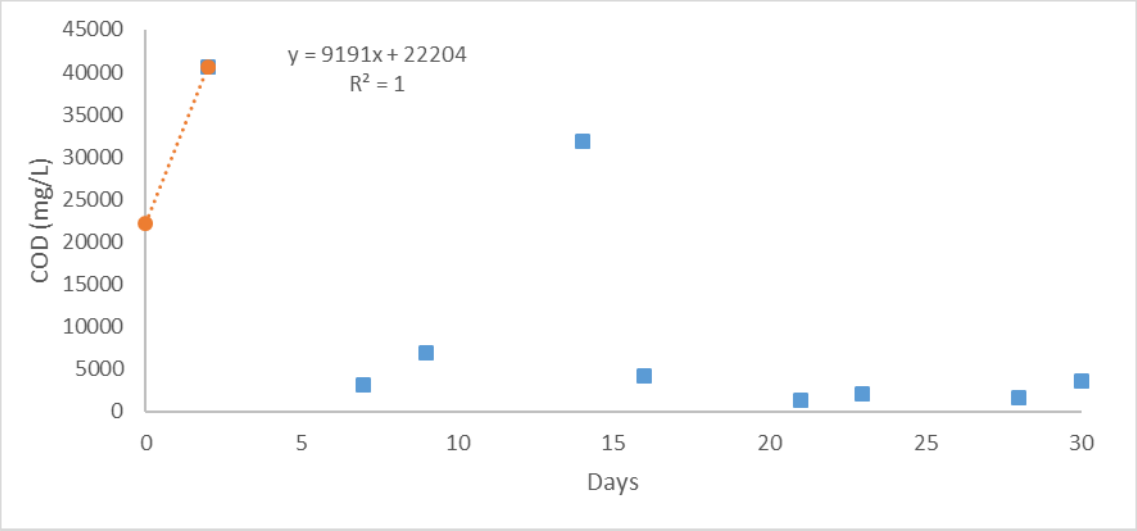

Figure 50: COD (mg/L) of Brewers Spent Grain leachate over 30 days

Figure 50 shows significantly higher peaks of COD in the leachate produced compared to other systems on day two and day 14. The initial carbon release rate of 9191 COD mg/L.day is the highest observed of all the materials used. The system seems to start stabilising around day 16 around 2000-3000 COD mg/L, lower than some of the woodchip materials examined. Despite where the system stabilizes, the peaks and trends shown suggest that the material has great potential for a large of amount of carbon release to the leachate within a reasonably short amount of time. The extremely high peaks and quick uptake means there is potential to produce a leachate from this material.

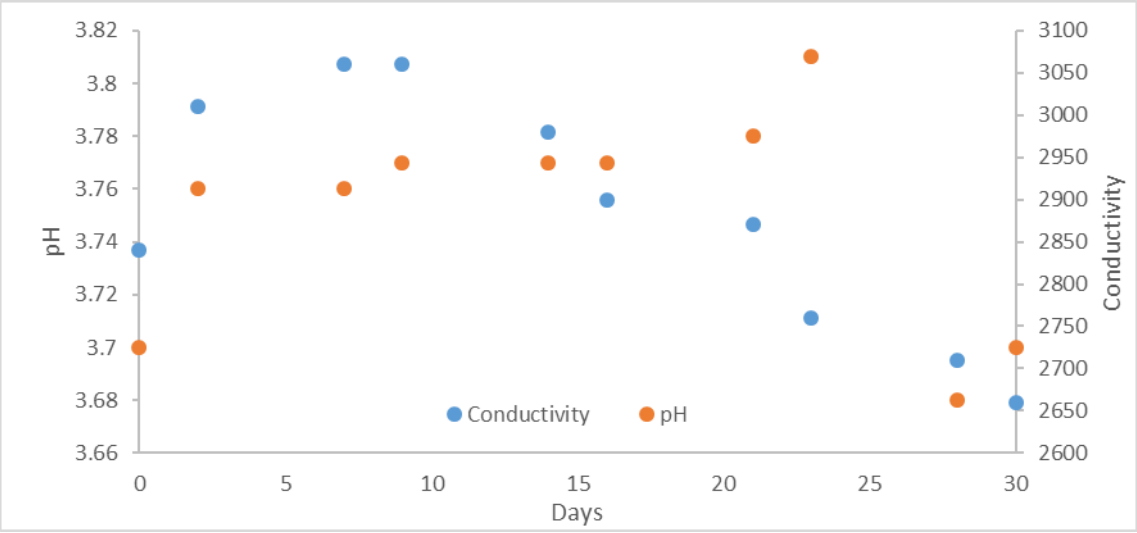

Figure 51: pH and Conductivity of BSG leachate over 30 days

The pH of the leachate, shown in Figure 51, quickly rises in the first two days and begins to stabilise, however it dips quite drastically for the final two days. This also supports the COD trend that majority of uptake occurs in the first two days. However, an average pH of 3.8 with a range of 0.1 was achieved which places it outside of the optimum range for denitrification. The conductivity shows a similar trend where impurities rapidly increase

in the first two days. However, a steady decline in conductivity after can be seen after day nine, meaning the impurities in the system are being removed.

Figure 52: Brewers Spent grain prior to soaking

Figure 53: Brewers Spent Grain and leachate after 22 days

Figure 52 shows that the grains from BSG are relatively small giving a large surface area in contact with the water for carbon uptake. Figure 53 shows how prevalent fungal/microbial growth is, more than other materials examined, which could be explained by the proteins present in the material. This extreme amount of growth could explain the decrease in carbon content and conductivity of the leachate. This also resulted in a fermenting smell which could cause a problem in a real-world implementation.

4.1.9 Slum Gum

Slum gum is a waste by-product from the beeswax rendering process, therefore would be relatively low cost and readily available material (even internationally). There were some issues with drainage suggesting there could be issues with blocking and hydraulic conductivity. However due to the nature of the material it still meets requirements for real-world implementation.

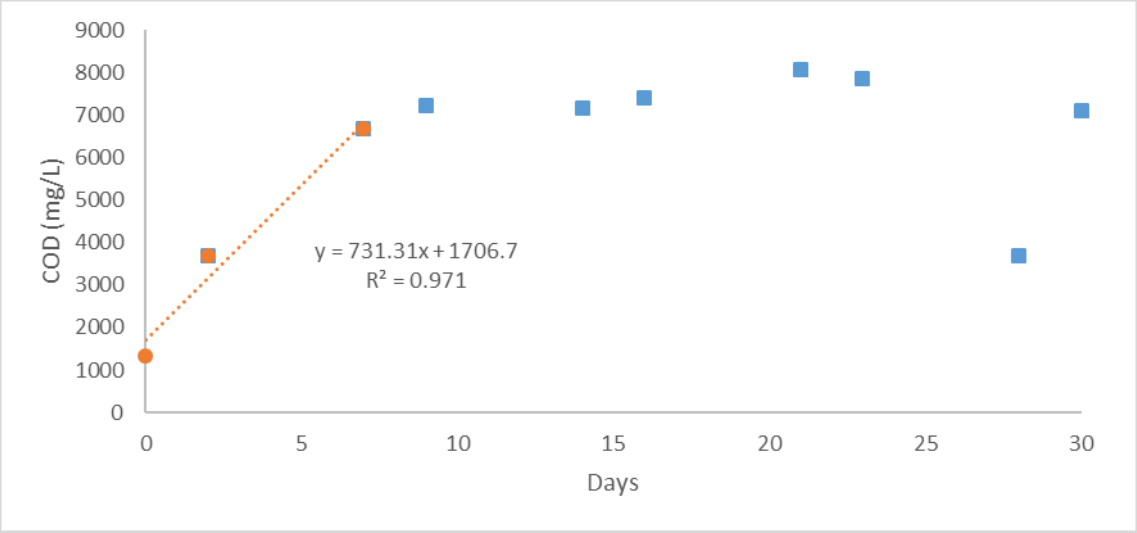

Figure 54: COD (mg/L) of Slum Gum leachate over 30 days

Figure 54 shows there is a quick initial release of carbon over the first seven days, at a rate of 731 COD mg/L.day and then the carbon release slows before dropping on day 23. This drop in COD content of leachate could be due to the presence of fungal/microbial activity as shown in other materials monitored during this phase. Carbon uptake slows after day 7 but steadily increases between 7000-8000mg/L, significantly higher than the woodchip material examined. This is promising for denitrification despite the drainage issues and could potentially be a candidate for producing leachate.

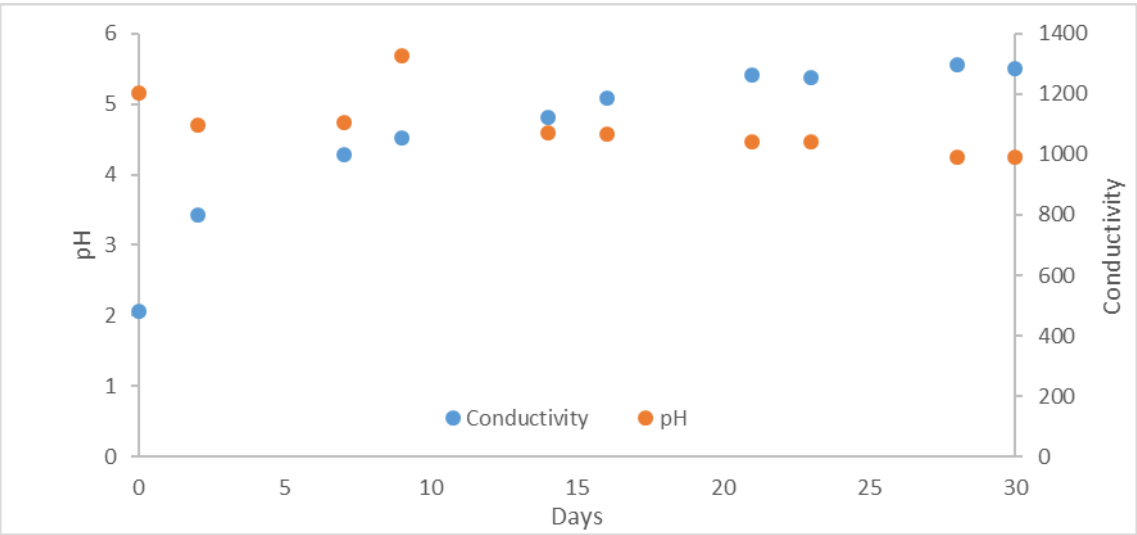

Figure 55: pH and Conductivity of Slum Gum leachate over 30 days

Figure 55 shows a steady decrease in pH over 30 days except one outlier suggesting the system is yet to reach stability. This occurs with conductivity also, with a noticeable increase over the 30 days suggesting uptake is still occurring. Average pH of the system was 4.7 with a range of 1.5, a noticeably larger range than previous woodchip materials. This is outside the optimum range for denitrification, however due to higher carbon content in the leachate the material could still prove useful for denitrification.

Figure 56: Slum Gum prior to soaking

Figure 57: Slum Gum and leachate at 22 days

Slum gum occurs as small grains however as seen in Figure 56 these small grains end up clumping even if dried. The size of these grains could explain the issues with drainage. Figure 57 clearly shows fungal/microbial growth occurring which explains some of the outliers in the monitored parameters.

4.1.10 Coir

Coir is a natural fibre derived from the outer husk of coconut and has multiple uses. Here a commercially available coir briquette intended for potting mix is used. Being a natural product which is relatively cheap and accessible, coir meets criteria set for real-world implementation. During this experiment a significant issue with drainage is noticed due to how small the fibres were. Future research could be directed at different forms of coconut husks with larger or consolidated fibres to develop a more useful material.

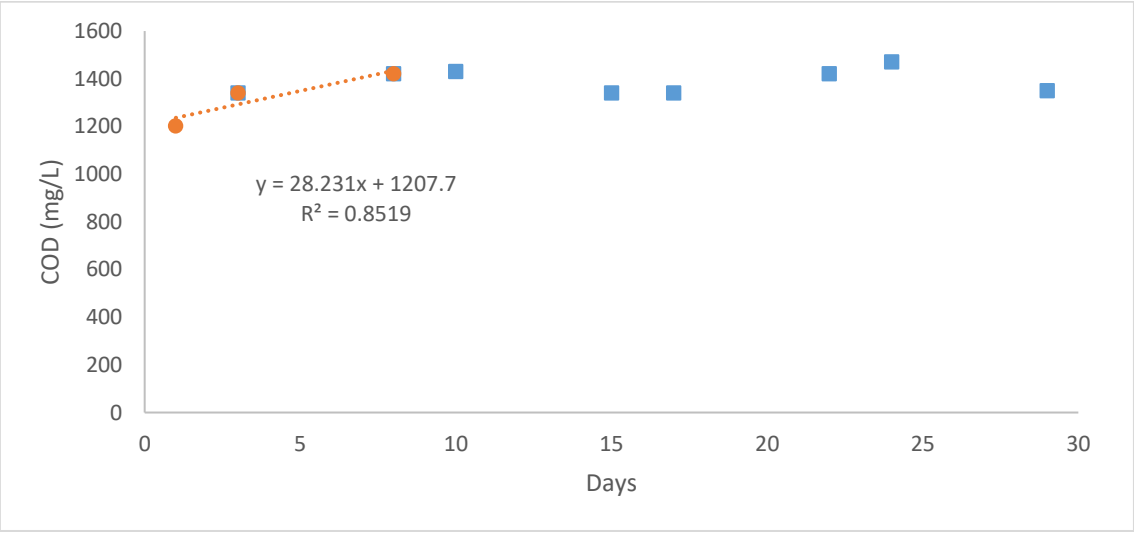

Figure 58: COD (mg/L) of Coir leachate over 31 days

Figure 58 shows that the leachate produced from the coir material stays relative stable between 1200-1400 COD mg/L over the 30 days. This could be due to the small sized fibres offering a large surface area exposed to the water for uptake. This means uptake is extremely quick occurring almost instantly. However, with COD stabilising at such a low reading suggests there is not much carbon available for denitrification in the leachate produced. Upon modelling the first eight days of carbon release as initial release the Coir system had a low rate of 28.2 COD mg/L.day.

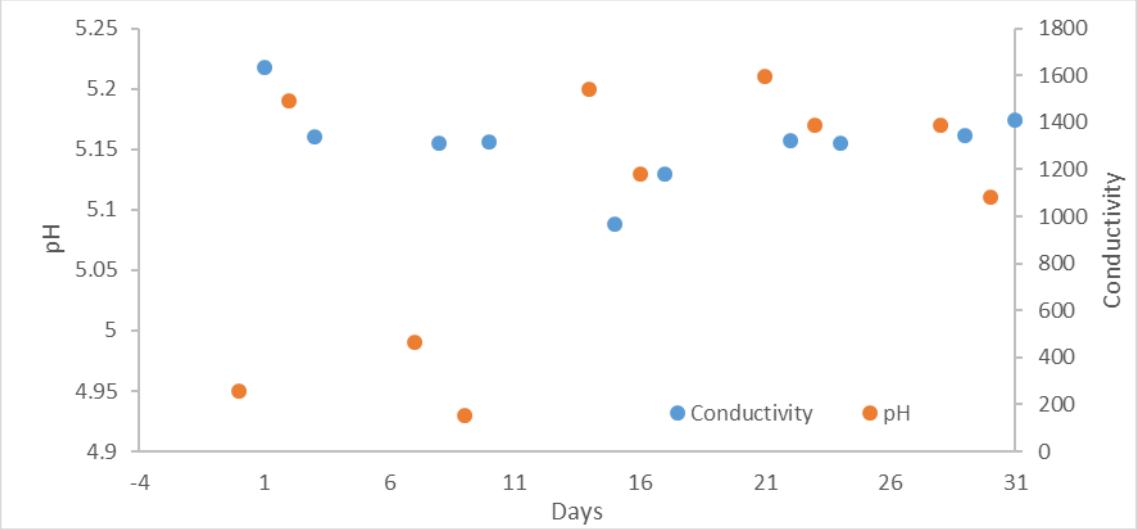

Figure 59: pH and Conductivity of Coir leachate over 31 days

No significant can be seen trend in pH or conductivity shown in the Figure 59 above however both seem to be stable with relatively small ranges. Average pH of the system was 5.1 with a range of 0.3 placing it outside the range for optimum denitrification, however this is less alkaline than some of the woodchip systems.

Figure 60: Coir on initiation of phase 1

Figure 61: Coir and leachate at 31 days

The brick form factor of the material used is shown in Figure 60, the brick could only be sized roughly for a 5L system. The brick quickly solubilised into the water and resulted

in what the form in Figure 61, the small the fibres are visible. There was no fungal/microbial growth to note, an indication of how little carbon was available in the system.

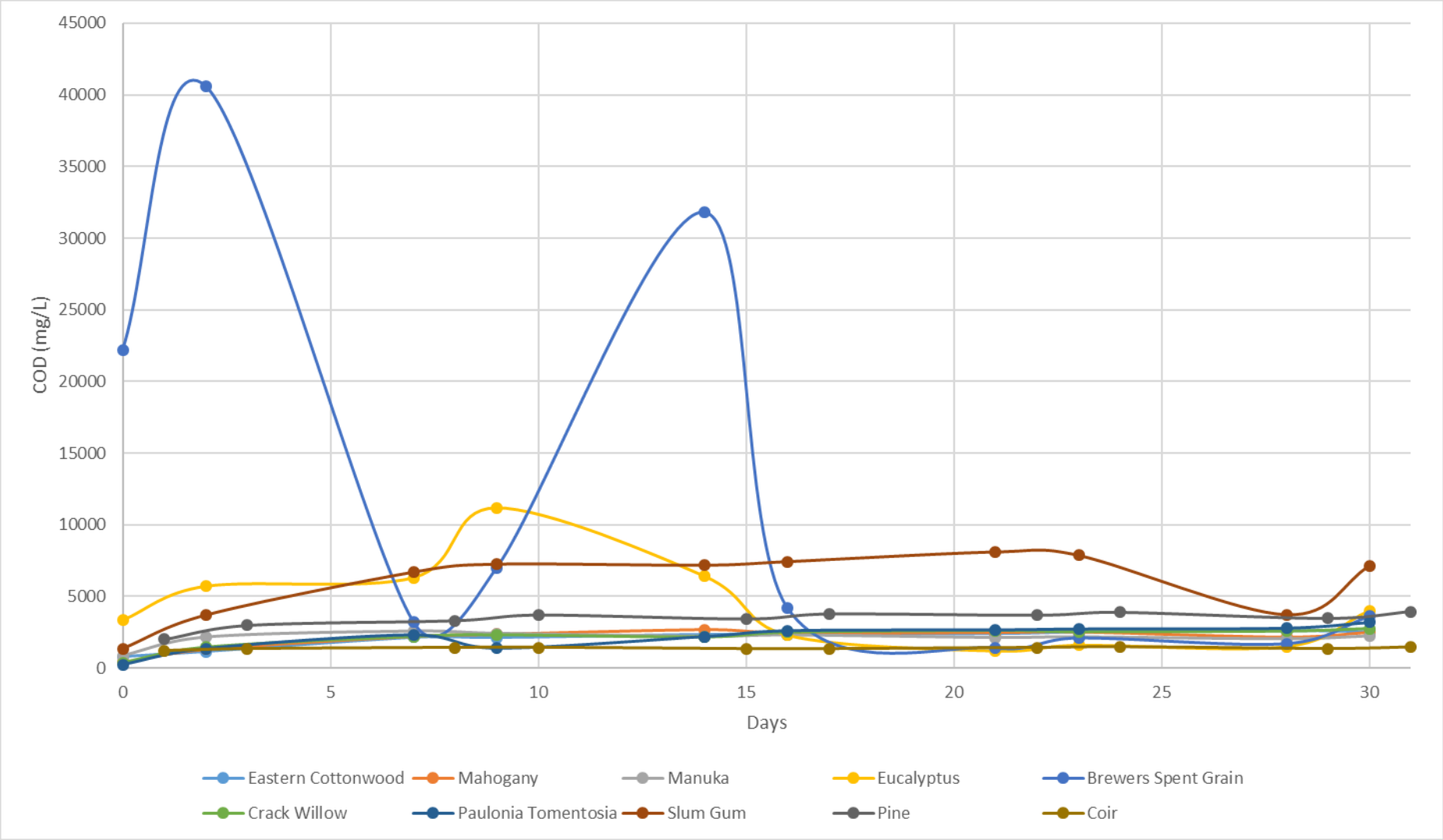

Figure 62: Comparison of COD in leachate of all materials monitored in phase 1

4.1.11 COD in Leachate

Figure 62 shows COD content in the leachate of each material plotted together. Most of the woodchip materials examined produced similar results except Eucalyptus. The waste by-products (Slum gum and BSG) seems to produce much higher COD content in the leachates, therefore suggesting more carbon available for denitrification.

Table 2: Comparison of COD present in leachate for each material

Material	Average COD (mg/L)	Std. Dev COD (mg/L)	Min. COD (mg/L)	Max. COD (mg/L)
Eastern Cottonwood	2086	582	792	2600
Mahogany	2097	663	419	2660
Manuka	2090	437	853	2550
Eucalyptus	4350	2950	1200	11150
Crack Willow	2132	689	350	2740
Princess Tree	2137	853	217	3180
Pine	3410	547	1994	3940
BSG	11779	13650	1400	40586
Slum Gum	6023	2159	1342	8080
Coir	1376	75	1202	1470

Error! Reference source not found. supports the results previously mentioned with higher average COD in leachate present for BSG and Slum gum compared to the woodchip systems. However variability in the BSG system is significantly larger compared to the other systems, but this could be due to the excessive fungal/microbial growth observed. Slum gum on the other hand also had a noticeably larger variability compared to the other woodchips (except Eucalyptus) but also had noticeable fungal/microbial growth, while Coir had the lowest carbon uptake in leachate with the lowest variability.

Table 2 also shows Eucalyptus woodchips produced the largest carbon uptake in leachate of all woodchips but had a large variability compared to the other woodchips. This could be explained by the fungal/microbial growth seen during the experiment. The Pine woodchips also showed a higher carbon uptake than majority of other woodchip systems but was more stable, along with previous research this is another reason to use this as a benchmark material. Crack Willow and the Princess tree woodchip systems were similar however the Princess tree woodchip leachate had a higher peak COD comparatively but also more variability. The other three woodchip systems (Eastern Cottonwood, Mahogany and Manuka) showed similar results but each material has their own distinguishing characteristics.

4.1.12 Mass Balance

Table 3: Mass balance for each material for phase 1

Material	Initial (g)	Final (g)	Difference (g)	Mass Loss %	Density (g/L)
Brewers Spent Grain	859.6	510.8	348.8	40.6%	171.9
Pine	944.3	652.1	292.2	30.9%	188.9
Easter Cottonwood	931.1	785.4	145.7	15.6%	186.2
Coir	292.2	250.1	42.1	14.4%	58.4
Princess Tree	560.5	483.4	77.0	13.7%	112.1
Mahogany	988.1	855.7	132.4	13.4%	197.6
Crack Willow	787.0	694.5	92.5	11.7%	157.4
Manuka	1367.6	1228.1	139.6	10.2%	273.5
Slum Gum	1182.7	1122.8	59.8	5.1%	236.5
Eucalyptus	1394.7	1413.6	-18.9	-1.4%	278.9

Upon examining mass loss for each material, shown in Table 3, significant mass loss observed in the BSG system which would explain the high carbon uptake into leachate. Interestingly Slum Gum had very little mass loss in comparison despite releasing a large amount of carbon, perhaps due to high sugar content present in the material.

For the woodchip material systems, roughly a 10-15% mass loss is observed for each system. The Pine woodchip system lost significantly more mass over the experiment period at over 30% perhaps due to the amount of fines present in the system, potentially causing issues with solids in effluent. Interestingly Eucalyptus gained mass which could be due to the fungal/microbial growth observed. Coir had mass loss in line with other woodchip material.

The density of each material for each system gives us an idea on cost for each material when implementing in a constructed wetland system. More material (by weight) is required for Manuka and Eucalyptus compared to other woodchip systems. While Crack Willow and Princess tree woodchips require the least amount of material by weight. Examining the by-product materials, BSG requires slightly more material compared to the two previously mentioned woodchip systems, however Slum Gum requires significantly more.

$$\text{COD } per\ kg\ of\ material\ per\ L\ of\ water = \frac{\frac{Average\ COD\ (mg/L)}{Initial\ Mass\ (kg)}}{Water\ Volume\ (L)} \quad (1\text{-}9)$$

Comparing COD mg/L per kg of material per L of water in producing leachate calculated via (1-9) for each material gives us a better idea of the capabilities of each material to release carbon.

Table 4: Average COD (mg/L) content per kg of material per L of water/leachate

Material	COD $mg/L^2.kg$
Easter Cottonwood	2241
Mahogany	2122
Manuka	1528
Eucalyptus	3119
Crack Willow	2709
Princess Tree	3813
Pine	3612
Brewers Spent Grain	13703
Slum Gum	5093
Coir	4710

Error! Reference source not found. shows that when examined in this format that the by-product materials, BSG and Slum Gum outperformed the woodchip materials. However, Princess Tree, Pine and Eucalyptus woodchip systems were the 3 highest performing woodchip material based systems. Manuka examined via this format had the lowest capability to release carbon out of all the systems examined. Interestingly Coir is third when compared in this format, however when examining these materials for implementation in the proposed CW systems other factors need to be considered.

4.1.13 Initial Carbon/COD Release Rate

Table 5: Summary of carbon release rates measured in leachate from fitted linear regression sorted in ascending order

Material	Release Rate COD (mg/L)/day
Coir	28.3
Eastern Cottonwood	192.1
Mahogany	239.8
Crack Willow	241.5
Princess Tree	278.7
Pine	488.0
Manuka	643.5
Eucalyptus	690.6
Slum Gum	731.3
Brewers Spent Grain	9191.0

Table 5 indicates BSG had the highest release rate observed followed by the other by-product material Slum gum. The woodchip materials showed varying release rates with Eucalyptus, Manuka and Pine being the highest. These release rates give an indication of how quickly carbon can be consistently released into a system with low hydraulic retention time as would be the case in real-world implementation. However, results should be considered as very rough estimates due to the lack of resolution in data over the initial days.

4.1.14 Implications for Wetland Filters

From the materials examined using the leachates produced to dose an existing system to assist with available carbon does not seem possible due the concentrations of carbon produced. Henze et al., 1997 suggests a C: N ratio of at least 5:1 to avoid carbon restrictions for denitrification. From data collected at the Phuket site there is an estimated 25 mg/L of Nitrogen entering the saturated zone, thus requiring 125 mg/L of organic carbon.

Using (1-8) (Dubber & Gray, 2010) to convert TOC to COD results in a concentration of 424.2 COD mg/L required to avoid these restrictions when applying to a system with no organic carbon available in the saturated zone. The primary issue is adding additional volume to the influent, theoretically then 8484 COD mg/L concentration is required to dose 50 mL per L of influent. Therefore, Table 2 shows BSG leachate would be the only option. However, this material was rather unstable and showed the most fungal/microbial growth which would present odour and visual issues in producing and storing the material/leachate. The data presented here does not represent how each system would behave over several cycles of producing leachate, in the next phase average COD of the leachate is investigated to show a trend over multiple cycles. Therefore, it would be impractical to pursue the idea of using leachate to dose wetland systems proposed unless a material with a significantly higher carbon release that remains stable over several cycles can be found.

As an estimate, the parameters of the Pukete site wetland systems implemented estimate that there is a 100 L saturated zone with a flow rate of 100 L/day meaning 2500 mg/L nitrogen is being loaded, therefore requiring 37549.2 COD mg/L per day for complete denitrification. Using these parameters in combination with average COD per system and density of the material the weight of material required for complete denitrification can be determined.

Table 6: Amount of material required for optimal denitrification compared to how much material can fit in the saturated zone using parameters of Pukete wetland filter systems sorted by material in 100 L/material required % for phase 1

Material	Average COD mg/L.day.kg material	Material Required (kg)	Material in 100 L (kg)	Material in 100 L/ Material Required %
Brewers Spent Grain	2284	16.44	17.19	105%
Slum Gum	849	44.24	23.65	53%
Eucalyptus	520	72.23	27.89	39%
Pine	583	64.46	18.89	29%
Princess Tree	635	59.09	11.21	19%
Crack Willow	452	83.16	15.74	19%
Mahogany	354	106.17	19.76	19%
Manuka	255	147.40	27.35	19%
Easter Cottonwood	373	100.55	18.62	19%
Coir	760	49.43	5.84	12%

Table 6 shows only BSG would have enough material to meet optimum denitrification conditions when implemented. However this experiment was only with loosely filled material for a 5 L volume whereas in constructing a wetland filter the saturated zone is tightly packed meaning more material (by weight) would be present. Therefore, suggesting a BSG filled saturated zone wetland filter would actually contain more carbon than required potentially leading to COD swapping in effluent. Slum Gum followed by Eucalyptus and Pine woodchips would be the most suitable with Coir being the worst candidate. The remaining woodchips all showed similar results using these parameters. Ideally results from this phase of experiments show a correlation with denitrification ability of organic materials and could be shortened and replicated to evaluate future materials for suitability in wetland filters.

4.1.15 Limitations

These experiments were conducted in buckets open to air in a lab with temperature of the leachate measured anywhere from 18.5°C to 25°C which in previous research proved to alter denitrification performance. However, in the proposed constructed wetland implementation for these materials they will also be exposed to changing temperatures with great variability. Regrettably the materials/leachate were exposed to air giving a chance for fungal/microbial activity to occur. However in producing leachate on a large scale the conditions would be similar but as the material would be in a saturated zone under constructed wetland conditions, it would be more anaerobic.

Another factor with potential impact on carbon uptake is the chip size/ surface area exposed to water. The materials obtained were collected from various sources as waste or by-product (i.e. tree culling) with the exception of Manuka woodchips and Coir. Ideally woodchips would be graded and surface area exposed/chip size controlled. However, in implementing these materials in constructed wetland filters for Maraes, woodchips and industry by-product would be used in the form they are obtained to keep costs low.
These materials should also be monitored for potential pollution swapping for example an excess of carbon would result in COD contaminants being present in the outlet for a CW filter. This is especially important when using industry by-products where other pollutants are present.

Ideally the release rate experiments would be run with replicates to account for variations in the systems however with the size of the systems and the limited space available this was not able to be implemented.

4.2 Phase 2 Results and Discussion

In phase 2 the ability of the materials to release carbon over multiple cycles is examined. These results should give a better idea of what the consistent carbon release capacity of each material giving an indication of what would occur in producing batches of leachate using the same material. It should also give an idea of how much carbon is available when using the materials in-situ in a wetland filter once the system reaches an operational steady state.

4.2.1 Eastern Cottonwood Woodchips

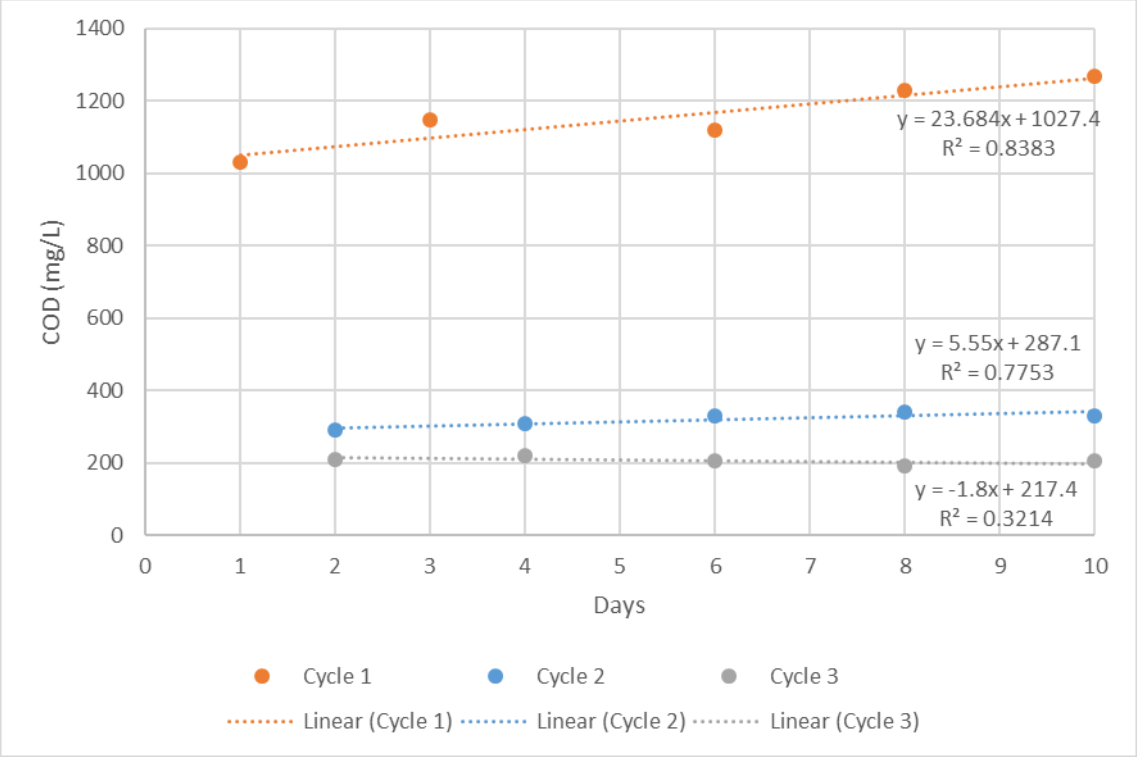

Figure 63: COD mg/L of Eastern Cottonwood woodchip leachate over three cycles with fitted linear regression

Table 7: Statistics for pH, Conductivity and COD for Eastern Cottonwood woodchip leachate over three cycles

Cycle	Average pH	Average Conductivity (μs^{-1})	Average COD (mg/L)	Std. Dev COD (mg/L)	Coefficient of Variation
1	6.76	614.2	1160	84.38	7.3%
2	6.76	374.2	320.4	17.83	5.6%
3	6.43	312.2	206.6	8.98	4.4%

Figure 63 shows carbon release rate declining over each cycle and suggests the system is beginning to near equilibrium on the third cycle as can be seen by the similarities of cycle 2 and 3. The negative rate of release over the third cycle suggests that fungal/microbial activity is consuming carbon in the system despite no visible signs observed. Table 7 supports this verdict with changes in average COD and average conductivity showing similar behaviour to the changes in release rate. Average pH for all cycles are within the optimal range for denitrification to occur so alkalinity should not be an issue when implementing this material in wetlands.

4.2.2 Mahogany Woodchips

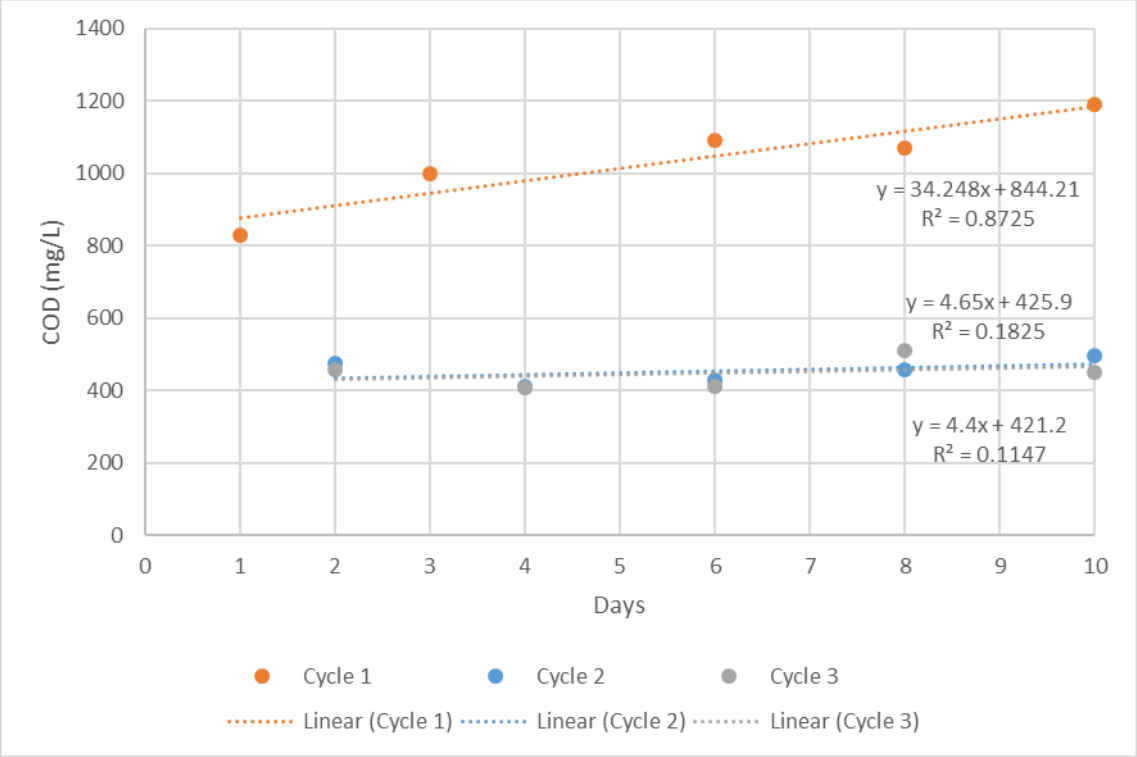

Figure 64: COD mg/L of Mahogany woodchip leachate over three cycles with fitted linear regression

Table 8: Statistics for pH, Conductivity and COD for Mahogany woodchip leachate over three cycles

Cycle	Average pH	Average Conductivity (μs^{-1})	Average COD (mg/L)	Std. Dev COD (mg/L)	Coefficient of Variation
1	5.57	434	1036	119.60	11.5%
2	5.74	250	453.8	30.79	6.8%
3	6.01	268.8	447.6	36.75	8.2%

The system showed the same decrease in carbon release rate over the three cycles as with the previously examined materials. The similar behaviour of cycle 2 and 3 shown in Figure 64 suggests this system has reached equilibrium by the third cycle, this is supported by the comparable results for average COD (Table 8) for the two cycles. The average pH rises to within the optimal pH range for denitrification for the third cycle, suggesting some of the material responsible for the acidity of the system are being removed with each cycle. Average conductivity also supports the indication that the system has reached equilibrium with results of cycle 2 and 3 being comparable.

4.2.3 Manuka Woodchips

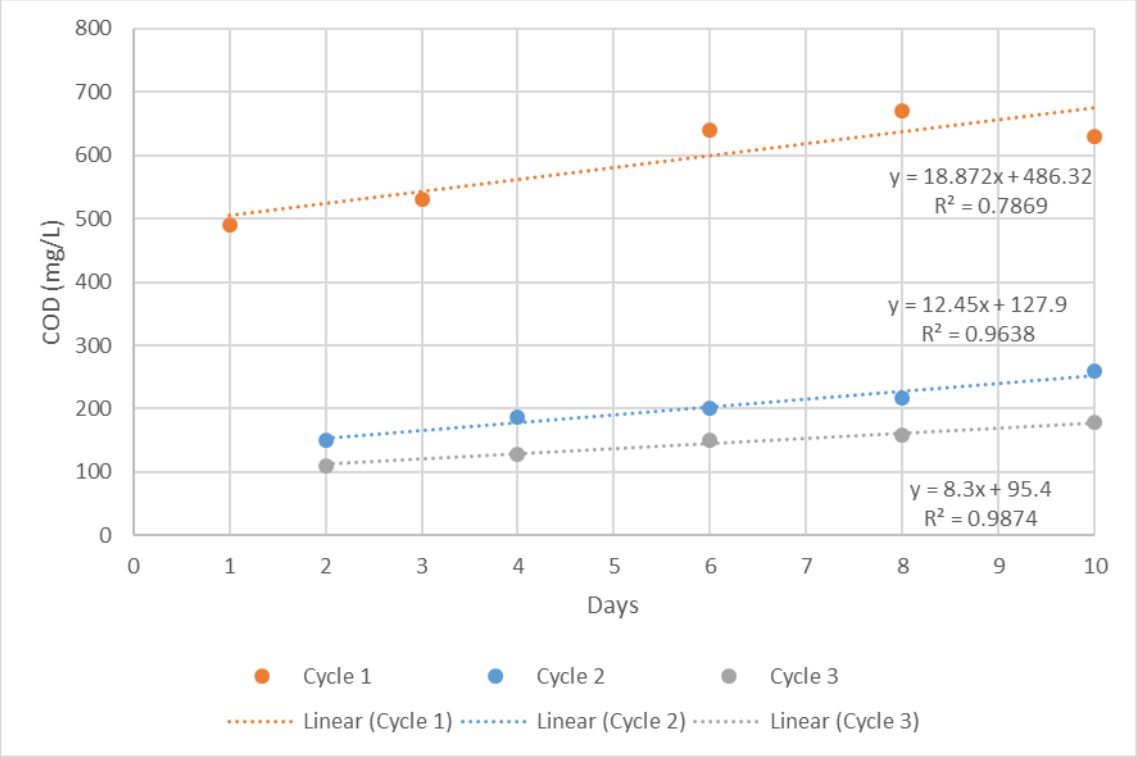

Figure 65: COD mg/L of Manuka woodchip leachate over three cycles with fitted linear regression

Table 9: Statistics for pH, Conductivity and COD for Manuka woodchip leachate over three cycles

Cycle	Average pH	Average Conductivity (μs^{-1})	Average COD (mg/L)	Std. Dev COD (mg/L)	Coefficient of Variation
1	5.57	330.4	592	69.40	11.7%
2	5.86	174.84	202.6	35.87	17.7%
3	5.91	150.8	145.2	23.63	16.3%

Figure 65 shows a decrease in carbon release rate over each cycle and a similar behaviour for cycle 2 and 3 suggesting the system is near equilibrium. This supported by the marginal change in average COD (Table 9) between the second and third cycle compared to the first and second cycle. Over a longer period carbon release rate should be slightly lower than observed in the third cycle. Average pH increases over the three cycles with a final average pH of 5.9, just outside the optimum range for denitrification, however at given enough time to reach steady state this may fall within the aforementioned range. Average conductivity also follows these trends of suggesting impurities in the leachate produced are close to reaching equilibrium.

4.2.4 Eucalyptus Woodchips

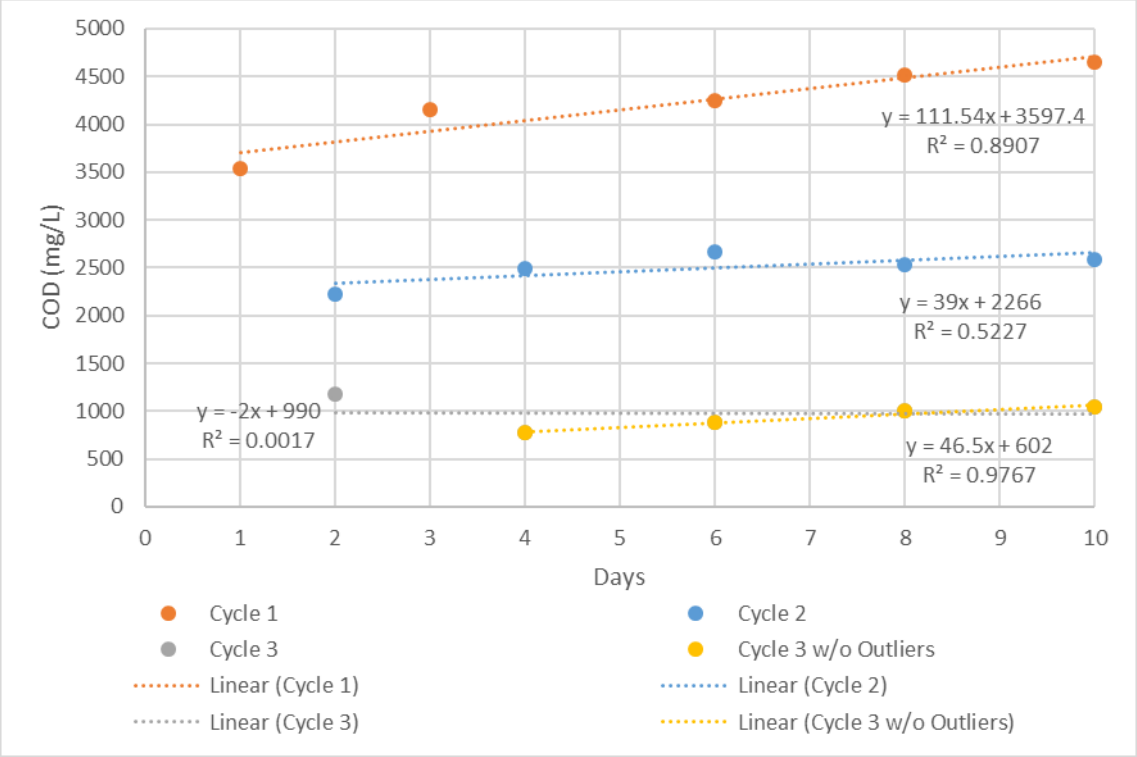

Figure 66: COD mg/L of Eucalyptus woodchip leachate over three cycles and removed outlier with fitted linear regression

Table 10: Statistics for pH, Conductivity and COD for Eucalyptus woodchip leachate over three cycles

Cycle	Average pH	Average Conductivity (μs^{-1})	Average COD (mg/L)	Std. Dev COD (mg/L)	Coefficient of Variation
1	4.20	779.4	4222	385.51	9.13%
2	4.45	457.4	2500	152.58	6.10%
3	5.19	274.8	978	138.04	14.11%

Figure 66 shows the Eucalyptus system follows the trend of COD increasing over each cycle except cycle 3 but a steady decrease in carbon release rate over each cycle. The third cycle resulted in a negative carbon release rate which could be due to the substantial fungal/microbial activity observed or the outlier observed on day two of the third cycle. Removing this outlier gives a more accurate estimate of rate of release (46.5 COD mg/L.day) for the final cycle. Table 10 suggests that the system has not reached equilibrium where average COD are significantly lower with each subsequent cycle. **Error! Reference source not found.** also shows average pH rising over each cycle. An average pH of 5.19 during cycle 3 this is still outside the optimum range for denitrification. However, the rising pH trend observed could put pH of the system at equilibrium to be within this range. The trend for average conductivity further supports the system has not reached equilibrium. The removal of the outlier for the third cycle

previously mentioned results a standard deviation of 105.2 COD mg/L and a coefficient variation of 11.34%.

4.2.5 Crack Willow Woodchips

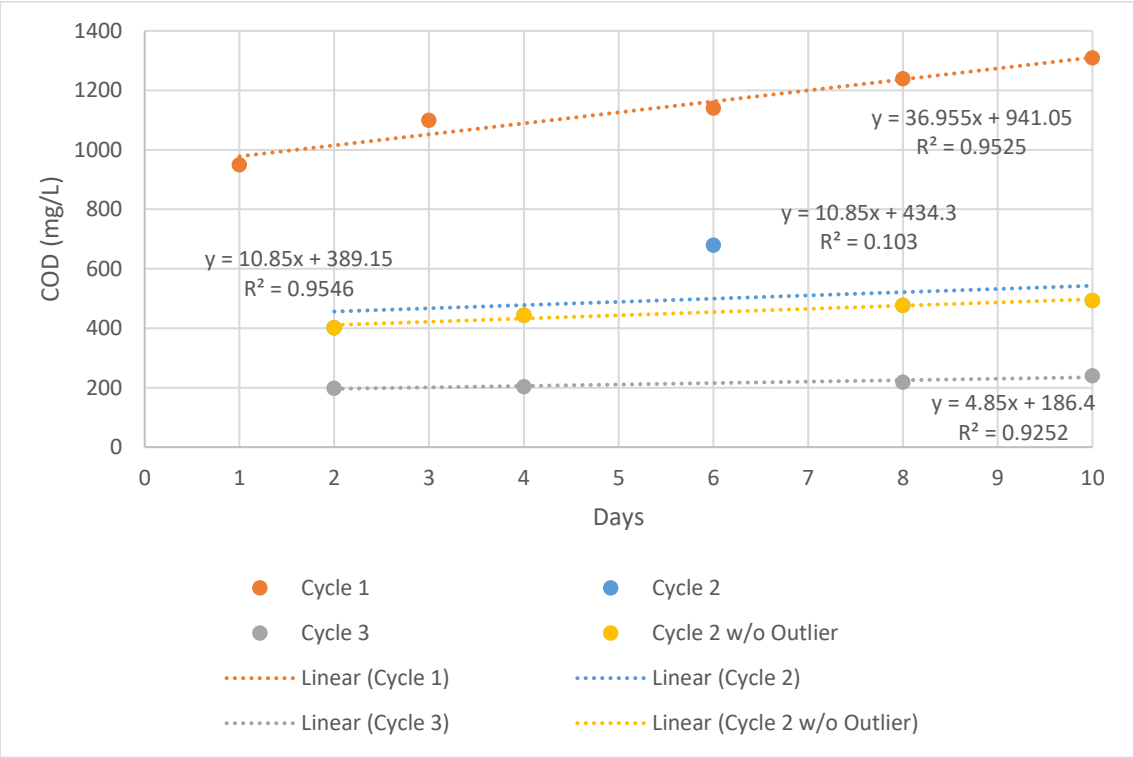

Figure 67: COD mg/L of Crack Willow woodchip leachate over three cycles and removed outliers with fitted linear regression

Table 11: Statistics for pH, Conductivity and COD for Crack Willow woodchip leachate over three cycles

Cycle	Average pH	Average Conductivity (μs^{-1})	Average COD (mg/L)	Std. Dev COD (mg/L)	Coefficient of Variation
1	5.61	350.6	1148	123.52	10.76%
2	5.90	216	499.4	95.61	19.14%
3	6.05	184.94	226.4	26.05	11.51%

The Crack Willow woodchip system showed a decreasing trend for carbon release rate over each cycle shown in Figure 67. The similarities between cycle 2 and 3 shows that the system is close to equilibrium. Table 11 supports this with changes in average COD and average conductivity over each cycle being smaller. Average pH of the third cycle is already within the range for optimal denitrification to occur the trend suggesting this would be even higher than the 6.05 average observed in the third cycle. There was one outlier result observed in the second cycle on day six with a spike in COD of the leachate, omitting this result gives an insignificantly lower average COD of 454 mg/L and gives the same rate of release for the second cycle with more accuracy. This also reduces the

standard deviation of the second cycle to 35 COD mg/L and a coefficient of variation of 7.73%.

4.2.6 Princess Tree Woodchips

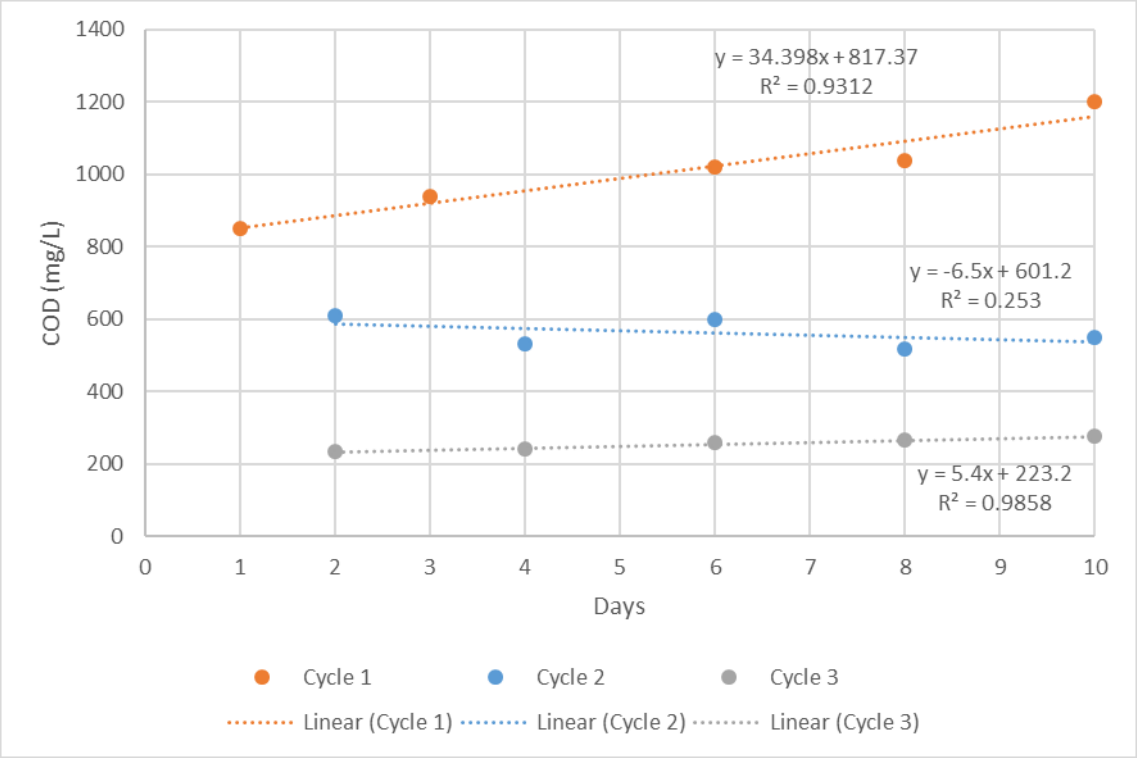

Figure 68: COD mg/L of Princess tree woodchip leachate over 3 cycles with fitted linear regression

Table 12: Statistics for pH, Conductivity and COD for Princess tree woodchip leachate over three cycles

Cycle	Average pH	Average Conductivity (μs^{-1})	Average COD (mg/L)	Std. Dev COD (mg/L)	Coefficient of Variation
1	5.88	283.8	1010	116.28	11.51%
2	5.97	219.8	562.2	36.55	6.50%
3	6.10	173.56	255.6	15.38	6.02%

Figure 68 shows the Princess tree system showed the same trend of decreasing rate of carbon release decreasing over each cycle, however cycle 2 showed a negative rate of release perhaps due some fungal/microbial activity being present. Changes in average COD and conductivity (Table 12) over the three cycles suggests the system is beginning to stabilise by the third cycle. Average pH of the system rises with each cycle, by cycle 3 the system is within the optimal denitrification pH range, as the system is yet to reach equilibrium pH of a functioning system should be slightly higher placing it well within the optimal denitrification range.

4.2.7 Pine Woodchips

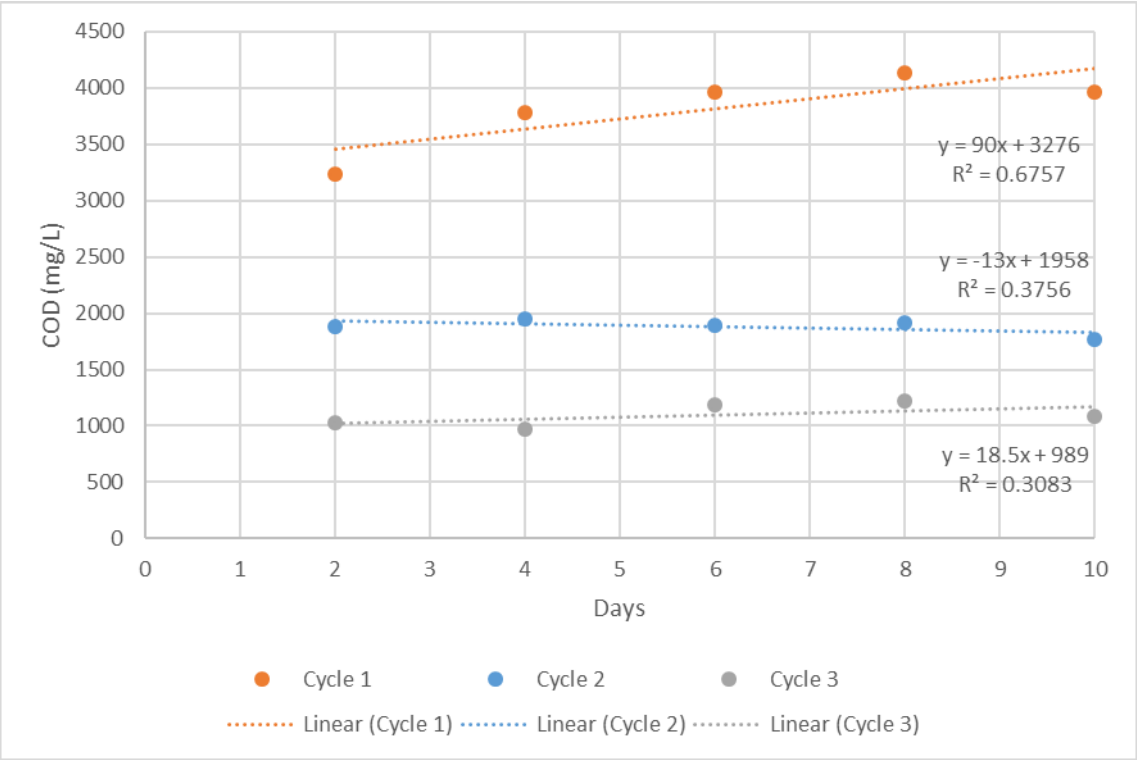

Figure 69: COD mg/L of Pine woodchip leachate over three cycles with fitted linear regression

Table 13: Statistics for pH, Conductivity and COD for Pine woodchip leachate over three cycles

Cycle	Average pH	Average Conductivity (μs^{-1})	Average COD (mg/L)	Std. Dev COD (mg/L)	Coefficient of Variation
1	3.62	543.6	3816	309.68	8.12%
2	3.82	307.2	1880	60.00	3.19%
3	3.91	221.18	1100	94.23	8.57%

Figure 69 shows the same decrease in carbon release rate over each cycle except cycle 2 showing a negative release rate which could be due to fungal/microbial activity beginning. This system appears to start stabilising by the third cycle supported by changes in average conductivity and COD (Table 13) starting to reduce between the second and third cycle. The average pH for this system never managed to reach the optimum pH range for denitrification, despite the pH rising over each cycle it is unlikely for the system at equilibrium to reach a pH of higher than 6.

4.2.8 Brewers Spent Grain

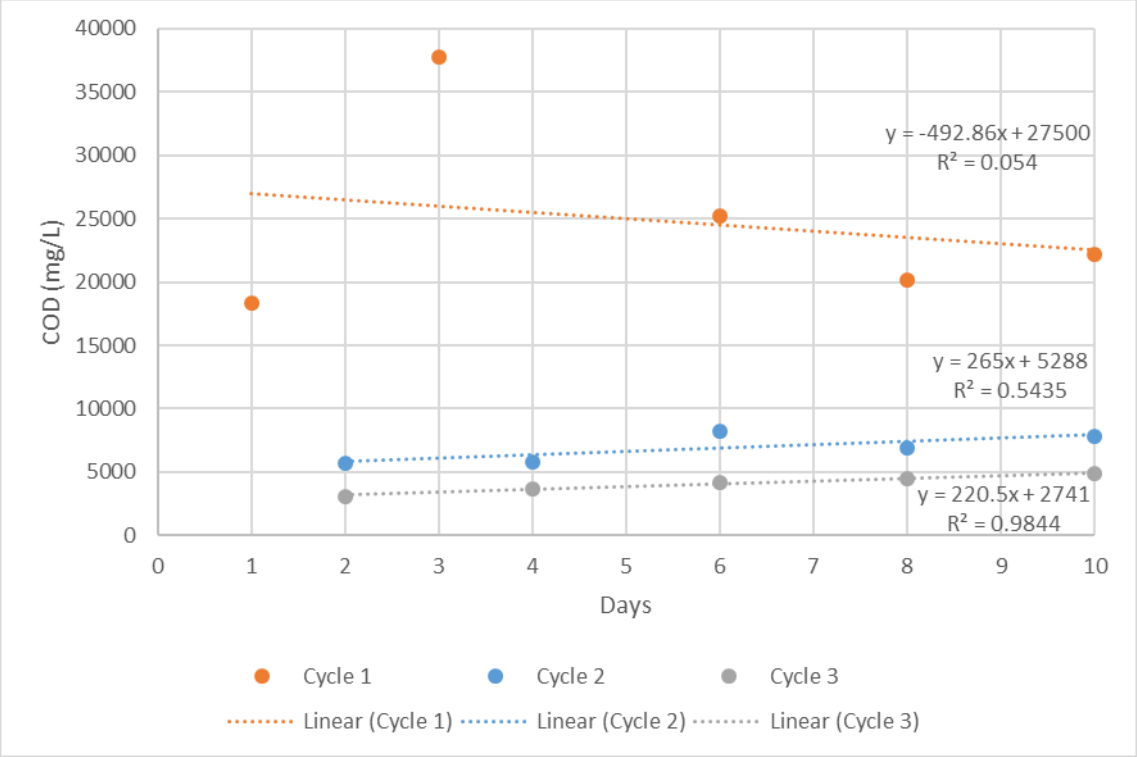

Figure 70: COD mg/L of Brewers Spent Grain leachate over three cycles with fitted linear regression

Table 14: Statistics for pH, Conductivity and COD for Brewers Spent Grain leachate over three cycles

Cycle	Average pH	Average Conductivity (μs^{-1})	Average COD (mg/L)	Std. Dev COD (mg/L)	Coefficient of Variation
1	4.31	2412	24740	6918.27	27.96%
2	4.45	1149.8	6878	1016.67	14.78%
3	5.29	1016.2	4064	628.60	15.47%

The first cycle of the Brewers spent grain system showed a similar trend (Figure 70) to the results from phase in the first cycle (Figure 50) with carbon quickly releasing however with quick fungal/microbial action results in large variation in carbon release rate. Fungal/microbial growth was observed over this cycle explaining the large amount of variation. The second and third cycles showed a more predictable release rate similar to the other materials. Comparing the final two cycles suggests the system is close to reaching steady state by the third cycle. This is supported by the smaller change in average COD and conductivity between the cycles (Table 14). The larger variation in average COD over these cycles that is not clearly visible with the scaling of Figure 70 but due to the larger quantities of carbon in the leachate. Average pH trended upwards over each cycle with the value for cycle 3 being just out from the optimum pH range for denitrification, however this could potentially fall within the range given more cycles.

4.2.9 Slum Gum

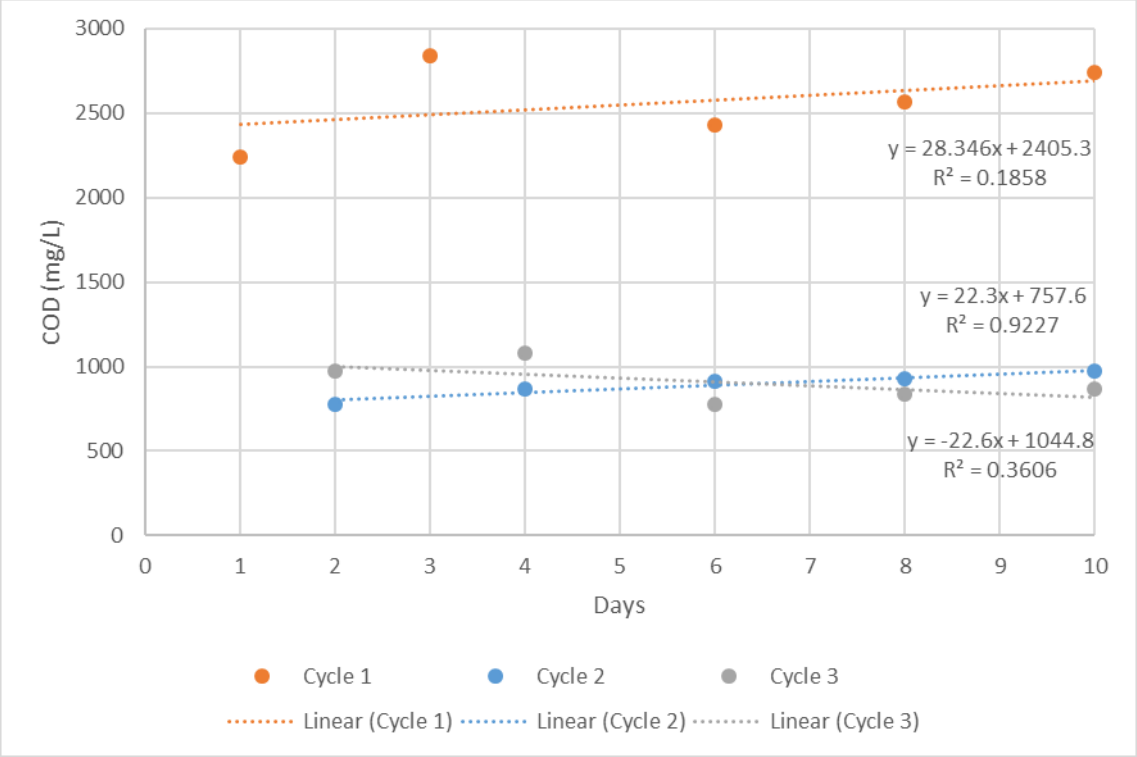

Figure 71: COD mg/L of Slum Gum leachate over three cycles with fitted linear regression

Table 15: Statistics for pH, Conductivity and COD for Slum Gum leachate over three cycles

Cycle	Average pH	Average Conductivity (μs^{-1})	Average COD (mg/L)	Std. Dev COD (mg/L)	Coefficient of Variation
1	5.00	509.8	2564	214.53	8.37%
2	5.33	286.6	891.4	65.66	7.37%
3	5.04	228.4	909.2	106.45	11.71%

Figure 71 suggests the system reached equilibrium within the 3 cycles observed. We see a steady increase in COD observed over each cycle except cycle 3 suggesting that's when the observed fungal/microbial growth occurred. The result is a negative release rate for carbon over this final cycle. Therefore, results obtained in cycle 2 for this material should be a reasonable representation for the system at steady state. These trends are mirrored by the average COD (Table 15) and the changes observed in average conductivity also suggests the system is near steady state. Average pH showed some variation but no general upward trend and therefore it is estimated a steady state would be outside the optimal pH range for denitrification.

4.2.10 Coir

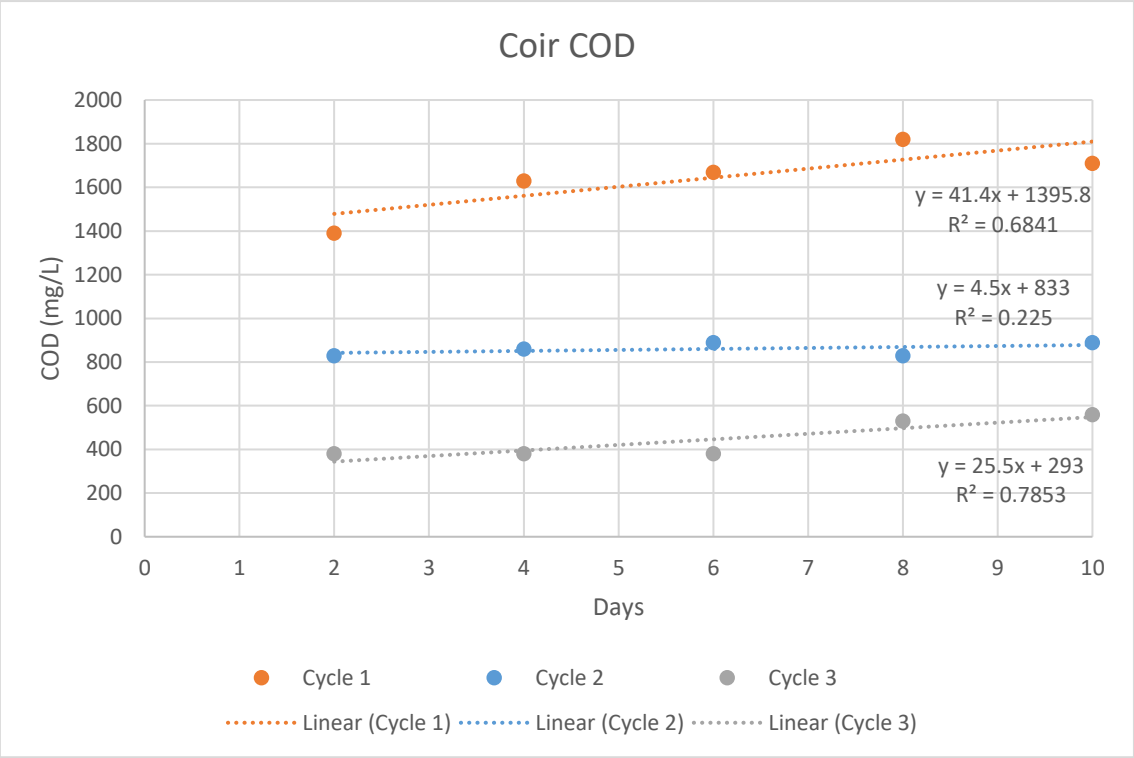

Figure 72: COD mg/L of Coir leachate over three cycles with fitted linear regression

Table 16: Statistics for pH, Conductivity and COD for Coir leachate over three cycles

Cycle	Average pH	Average Conductivity (μs^{-1})	Average COD (mg/L)	Range COD (mg/L)	Std. Dev COD (mg/L)	Coefficient of Variation
1	4.96	890.2	1644.2	429	141.57	8.61%
2	5.05	450.4	860	60	26.83	3.12%
3	5.21	270	446	180	81.39	18.25%

Figure 72 showed carbon release rates showed no general trend over the 3 cycles showing a decrease in the second cycle before an increase observed in third. The trends shown by average COD and conductivity (Table 16) showed the system has not reached equilibrium but is beginning to stabilise. Average pH rises over the 3 cycles however the small changes observed suggests given time to reach steady state system could potentially be within the optimum pH range for denitrification.

4.2.11 Mass Balance

Comparing Table 17 and Table 3 (mass balance from phase 1) the mass loss % is relatively lower for the woodchip systems in phase 2, apart from the Eucalyptus system which could be due to the large amount of fines present in the material being washed out with each cycle. While the by-product material systems showed an increase in mass loss

percentage over phase 2 also due to finer particles being washed out with each cycle. These results give an idea on how quickly each material would degrade when implemented in a wetland filter system for example Brewers Spent Grain had the highest capacity for carbon release but will degrade at a much faster rate.

Table 17: Mass balance of systems for phase 2

Material	Initial Weight (g)	Final Weight (g)	Difference (g)	Mass Loss %	Density (g/L)
Eastern Cottonwood	785.37	729.3	56.07	7.1%	157.1
Mahogany	855.69	783.1	72.59	8.5%	171.1
Manuka	1228.05	1178.8	49.25	4.0%	245.6
Eucalyptus	1413.6	1174.4	239.2	16.9%	282.7
Crack Willow	694.52	661.5	33.02	4.8%	138.9
Princess Tree	483.44	429.4	54.04	11.2%	96.7
Pine	652.1	623.3	28.8	4.4%	130.4
Brewers Spent Grain	510.76	283.3	227.46	44.5%	102.2
Slum Gum	1122.84	1010	112.84	10.0%	224.6
Coir	250.1	214.1	36	14.4%	50.0

4.2.12 Implications for Leachate production

This phase of experiments examined the ability of each material to maintain carbon release over several cycles with the final cycle serving as an estimate for average COD (mg/L) in leachate produced. The release rates obtained are not as significant as they are relatively low (apart from the BSG system) despite COD measured generally rising over a cycle, the trends show COD does not change overly much (per cycle).

Table 18: Comparison of average COD (mg/L) and average pH of each system sorted by largest average COD

Material	Average COD (mg/L)	Average pH
Brewers Spent Grain	4064	4.45
Pine	1100	3.91
Eucalyptus	927.5	5.19
Slum Gum	909.2	5.04
Mahogany	447.6	6.01
Coir	446	5.21
Princess Tree	255.6	6.10
Crack Willow	226.4	6.05
Eastern Cottonwood	206.6	6.43
Manuka	145.2	5.91

Average COD and average pH results shown in Table 18 are pulled from the third cycle for each system. Despite most systems not quite reaching steady state, this data is the best

estimate obtainable for a system operating at steady state. From the results we can see Brewers Spent Grain had the highest average results expected by the results from phase 1 (Table 2) with carbon release much higher than the other materials. The unexpected result is the Pine and Eucalyptus systems producing more average COD than the Slum Gum system. Referring to results from Table 2, it is expected the Slum Gum system to have a higher average COD in leachate and the Coir system to have a lower value. The results following are as expected with Princess Tree and Crack Willow systems performing similarly followed by the remaining materials. Average pH for the high carbon release systems fell below the optimum range for denitrification however were trending upwards over each cycle.

The previous leachate dosing example required 8484 COD mg/L concentration to avoid carbon restrictions for denitrification. Comparing with results obtained in this phase, none of the systems were able to produce a leachate that met more than half of the carbon requirements for a proposed leachate dosing wetland filter. As a result, materials would have to be replaced after producing one batch of leachate, along with the odour and visual concerns shown by high carbon release materials in phase makes the proposition of dosing impractical.

4.2.13 Implications for Wetland Filters

The release rates obtained for each material were omitting the value for the start of each cycle, seeing the carbon release rates being reasonably accurate and not trending towards 0 COD mg/L on initiation of each cycle suggests most of the carbon release occurs in each system in within the first or second day. This quick initial release of carbon is important as the mesocosm scale systems at the Pukete site have a hydraulic retention time of only 24 hours (similar to those proposed for real-world systems), therefore a material that takes a longer time to release carbon would restrict the systems denitrification ability. The trends observed means we treat average COD (after converting to TOC) of the leachates produced as the carbon available for denitrification in the saturated zone.

Table 19: Amount of material required for optimal denitrification compared to how much material can fit in the saturated zone using parameters of Pukete wetland filter systems. Sorted by material in 100 L/material required % for phase 2.

Material	Average COD mg/L.day.kg material	Material Required (kg)	Material in 100 L (kg)	Material in 100 L/ Material Required %
Brewers Spent Grain	3978.4	9.4	10.2	108%
Pine	843.4	44.5	13.0	29%
Eucalyptus	328.1	114.5	28.3	25%
Slum Gum	404.9	92.7	22.5	24%
Mahogany	261.5	143.6	17.1	12%
Coir	891.6	42.1	5.0	12%
Princess Tree	264.4	142.0	9.7	7%
Crack Willow	163.0	230.4	13.9	6%
Easter Cottonwood	131.5	285.5	15.7	6%
Manuka	59.1	635.2	24.6	4%

Table 19 uses the same parameters specified in phase 1 for the Pukete wetland to determine whether a material (by weight) will release enough carbon to not limit denitrification once operating at (estimated) steady state. The Brewers Spent Grain system is the only system that meets and exceeds this criteria.

Concerns of the laboratory scale experiment being loosely packed (and therefore have a lower density) while a saturated zone would be compacted still apply, as in phase 1 results. Therefore, the BSG system may have excess carbon present in effluent while the materials that did not manage to meet the carbon requirements, according to Table 19, could fit more material and be closer to meeting those requirements.

Being limited in space moving forward to phase 3 Coir is eliminated as a potential material due to drainage issues observed. Crack Willow woodchip system performed similarly but slightly worse than the Princess tree woodchip system and therefore will also be removed from further testing as the results should be similar. The lower carbon releasing woodchip materials will be kept as a reference to see if the experiments performed for here and for phase 1 can be used to indicate denitrification performance of a material.

4.2.14 Limitations

The same limitations apply as those specified in phase 1 concerning temperature, chip size/surface area, pollution swapping monitoring and the lack of replicates. Temperature

varying between 16°C - 21°C as this phase was conducted during the colder winter months. Fungal/microbial activity was still present in this phase however replacing the leachate with fresh water every 10 days and perhaps lower temperatures seemed to reduce growth, resulting in more predictable results in the second and third cycles.

Most systems reached steady state or were starting to stabilise with the second and third cycles returning similar results however the values obtained in the third cycle could only be an estimate for an operational wetland.

4.3 Phase 3 Results

In phase 3, the selected media is observed in two replicate systems for denitrification ability of a 25 mg/L nitrogen-nitrate solution over 60 days with one day hydraulic residence time (HRT). Although the solution does not imitate the nutrients and organic carbon that are present in influent, the presence of nitrate and organic carbon from the materials should allow for the growth of heterotrophic microorganisms (along with other pathways) for denitrification.

Due to the adjustments made (section 3.2.4) to the original method from Bird et al., 2017, obtained data should be considered just a rough estimate and indicates the presence of organic matter for effluent from these systems, especially upon establishing the systems. Some systems managed to achieve accurate values towards the end of the experiment, which are highlighted on the plots for nitrate concentration for each material.

With two replicates for each material, the examined data below represents an average of both replicates for each material. Due to the systems not being operated over weekends the systems have 3 days (instead of one day) HRT when sampling on the first day of the week which should be taken into consideration when examining data obtained on these days.

4.3.1 Eastern Cottonwood Woodchips

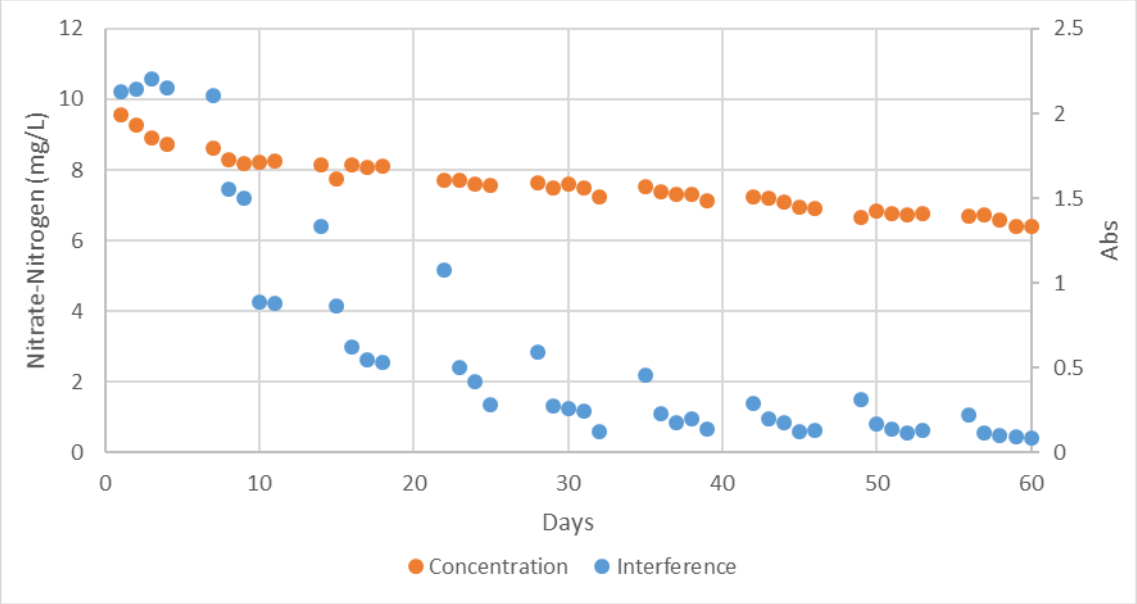

Figure 73: Average nitrate-nitrogen concentration (mg/L) and interference reading at 275nm (abs) for effluent from Eastern Cottonwood woodchip replicates.

Figure 73 shows a trend of decreasing NO_3^-N concentration in effluent (increasing nitrate reduction %) over the 60 days monitored for this system. The system does not appear to be at steady-state by day 60 suggesting allowed to run longer nitrate reduction would be even higher. As an estimate, the lowest (average of the two replicates) nitrate-nitrogen concentration observed was 6.39 mg/L giving a nitrate reduction of 74.4% for this material. Interference readings observed for this system decreased over the time monitored, given enough time results obtained using this method could be reasonably accurate. The effect of the increased HRT over weekends can be seen by the increased interference on the first day of each week monitored. Maximum coefficient of variation observed for nitrate concentration between the two replicates was 1.42%, more than acceptable in validating results obtained.

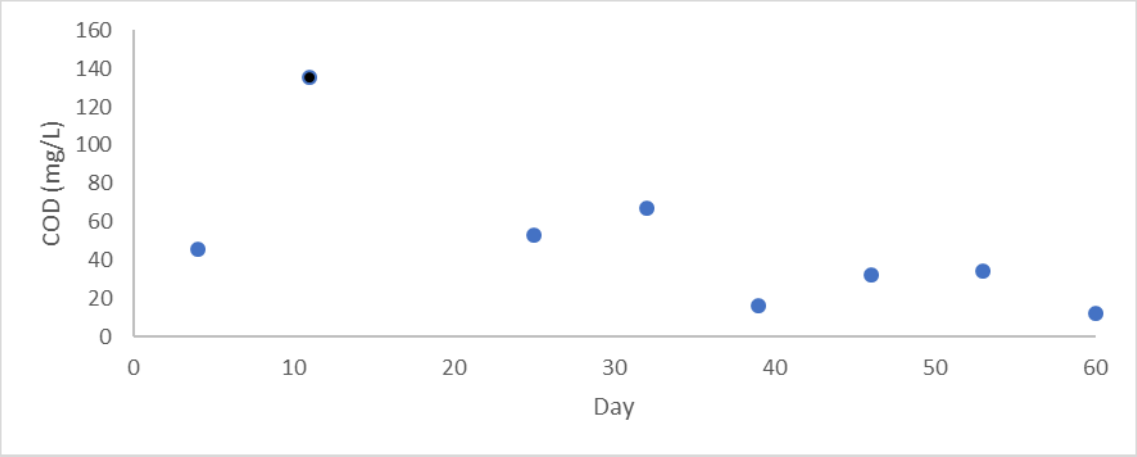

Figure 74: Average COD of effluent of the two replicates for Eastern Cottonwood woodchip replicates over 60 days. Outliers marked in black.

Figure 74 suggests COD in effluent for these replicates were varying at a low range of 13-67 COD mg/L upon excluding outliers, although the lower values obtained towards the final 4 weeks of the experiment are a reflection of the larger amount of nitrate removed during that period of time. As a result it can be determined that there is not a large amount of excess carbon in the effluent from this material when used for denitrification. Equipment malfunctions in controlling temperature during COD digestion resulted in the outlier on day 11 (highlighted in black) and damaged test on day 18 (not shown).

4.3.2 Mahogany Woodchips

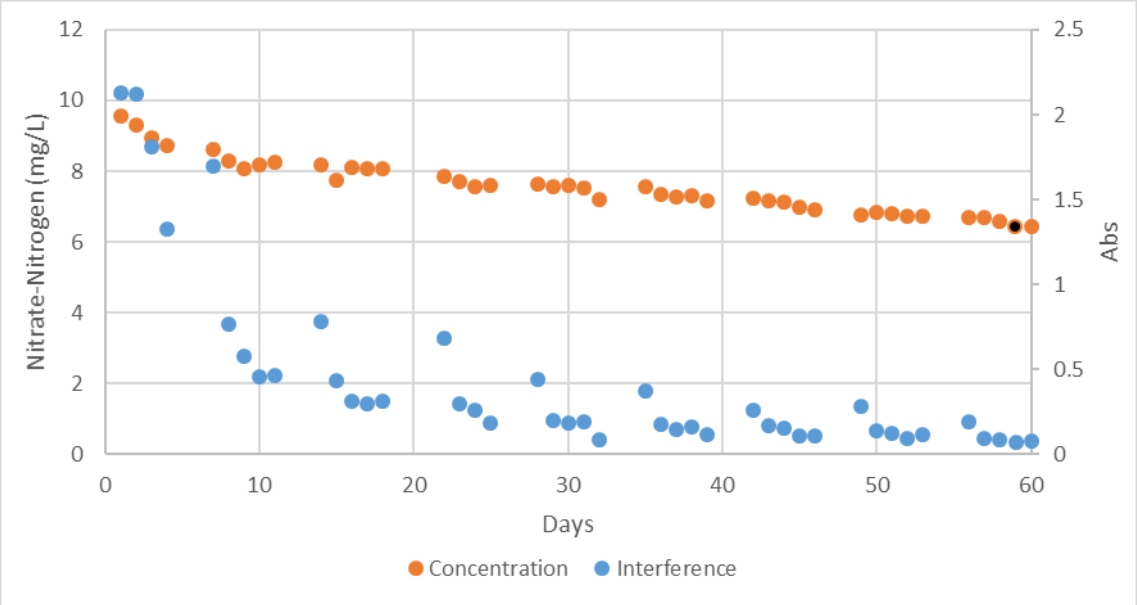

Figure 75: Average nitrate-nitrogen concentration (mg/L) and interference reading at 275nm (abs) for effluent from Mahogany woodchip replicates. Accurate readings are highlighted in black.

Figure 75 shows a trend of decreasing nitrate-nitrogen concentration (increasing nitrate reduction) in effluent over the 60 days monitored, suggesting if allowed to run for longer nitrate concentration in effluent would be even lower. As an estimate, the lowest nitrate-nitrogen concentration observed was 6.41 mg NO_3^-N/L resulting in a nitrate reduction of 74.4% similar to that observed in the Eastern Cottonwood woodchip systems. The data point highlighted in black meets the criteria from the method for an accurate reading and is the lowest concentration observed. The trend shown by interference readings for these systems suggests the system is close to steady state with the majority of reduction in organic matter in effluent occurring within the first 30 days. The two replicates achieved a maximum coefficient of variation for nitrate concentration observed was 0.89% acceptable for validating results obtained.

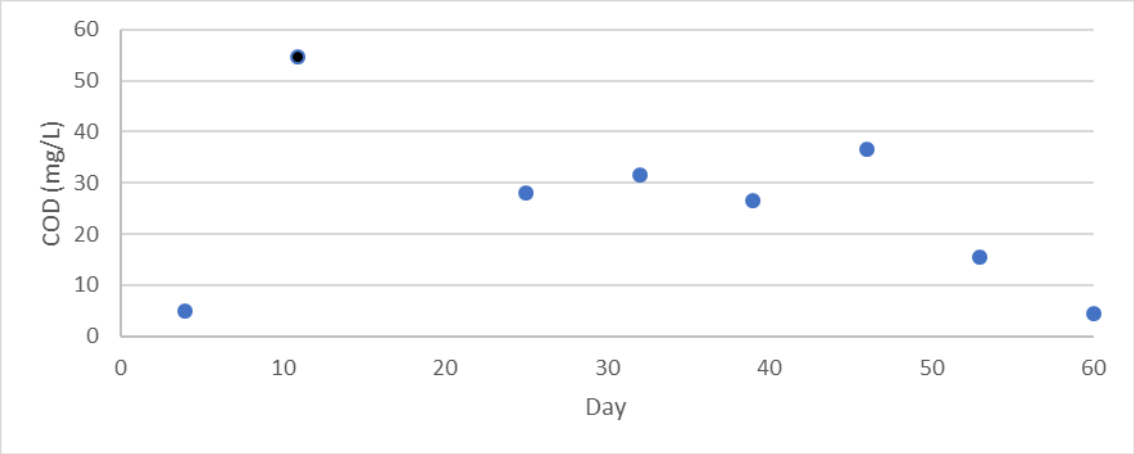

Figure 76: Average COD of effluent of the two replicates for Mahogany woodchip replicates over 60 days. Outliers marked in black.

Figure 76 showed that COD in effluent of the Mahogany woodchip replicates were reasonably stable and varied within a low range (5-37 COD mg/L) when excluding outliers for the same reason as stated for Eastern Cottonwood woodchip results. However lower values were observed towards the end of this phase coinciding with the higher nitrate-nitrogen removal observed. The results suggests this material would not contribute significant excess carbon to the effluent from the denitrification process.

4.3.3 Manuka Woodchips

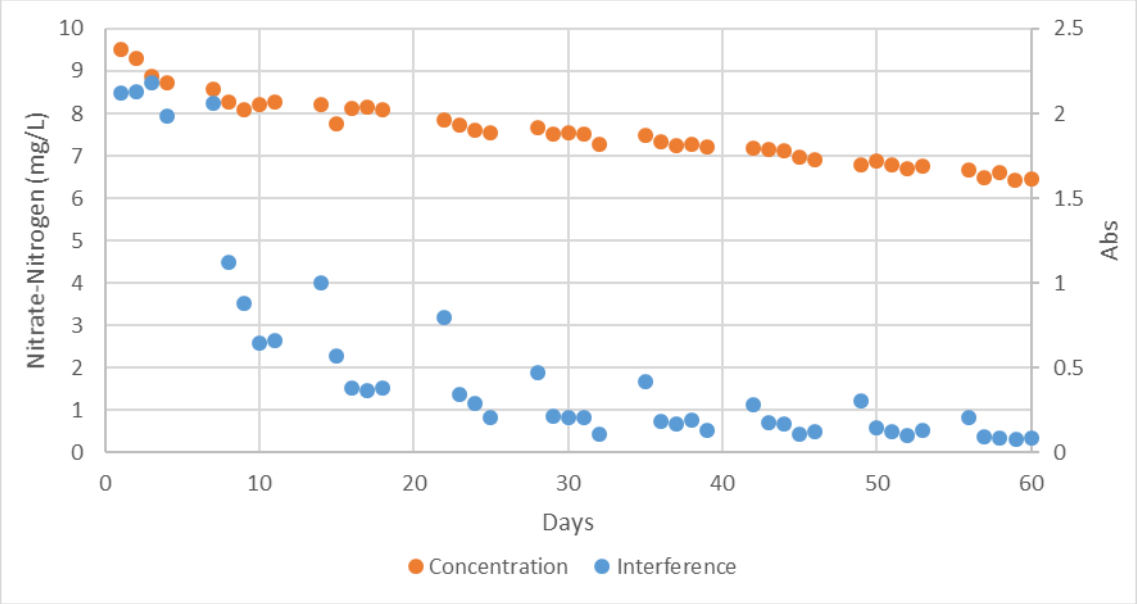

Figure 77: Average nitrate-nitrogen concentration (mg/L) and interference reading at 275nm (abs) for effluent from Manuka woodchip replicates.

Figure 77 shows a decreasing trend in nitrate (increasing nitrate removal) observed in effluent over the 60 days monitored, however without reaching steady-state suggests nitrate concentration upon achieving steady-state would be even lower. As an estimate the lowest nitrate-nitrogen concentration observed 6.41 mg NO_3^-N/L resulting in a nitrate reduction of 74.4% similar to the previously examined systems. The majority of organic matter causing interference in the readings were not present in effluent by day 30 (as with the previous two woodchip materials). Although no data obtained met the criteria for interference to not be considered significant, two times the 275 nm values were below 12% of the respective 225 nm values suggesting the results are a reasonable reflection of true nitrate concentration in effluent. Maximum coefficient of variation between the two replicates for nitrate concentration was 0.99% acceptable for validating results obtained.

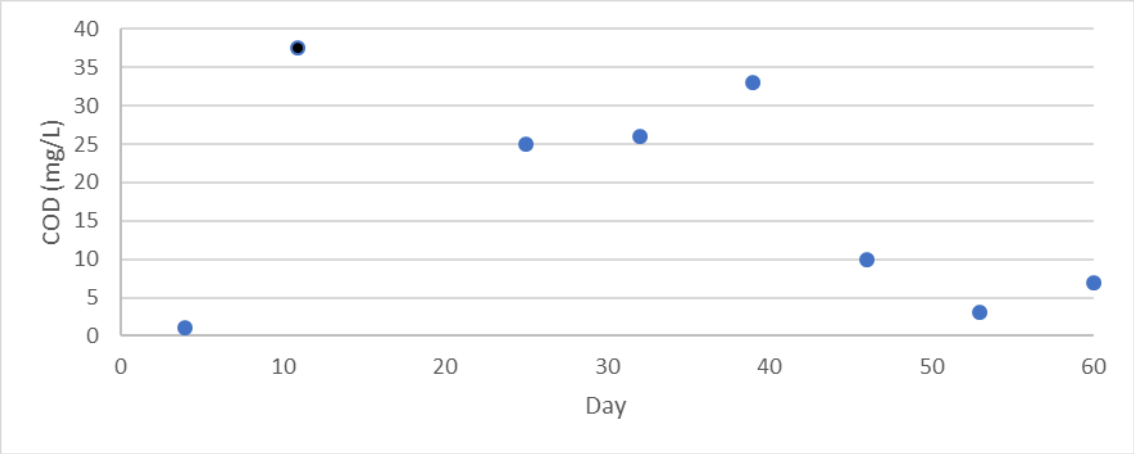

Figure 78: Average COD of effluent of the two replicates for Manuka woodchip replicates. Outliers marked in black.

Results for COD (Figure 78) in effluent for Manuka woodchips are similar to the two previously examined woodchip materials varying within a low range (1-33 COD mg/L), with an outlier observed for the reason previously mentioned. The same lower values were observed towards the end of the experiment period where higher nitrate removal was observed, suggesting Manuka woodchips would not contribute significant excess carbon to the effluent as a result of the denitrification process.

4.3.4 Eucalyptus Woodchips

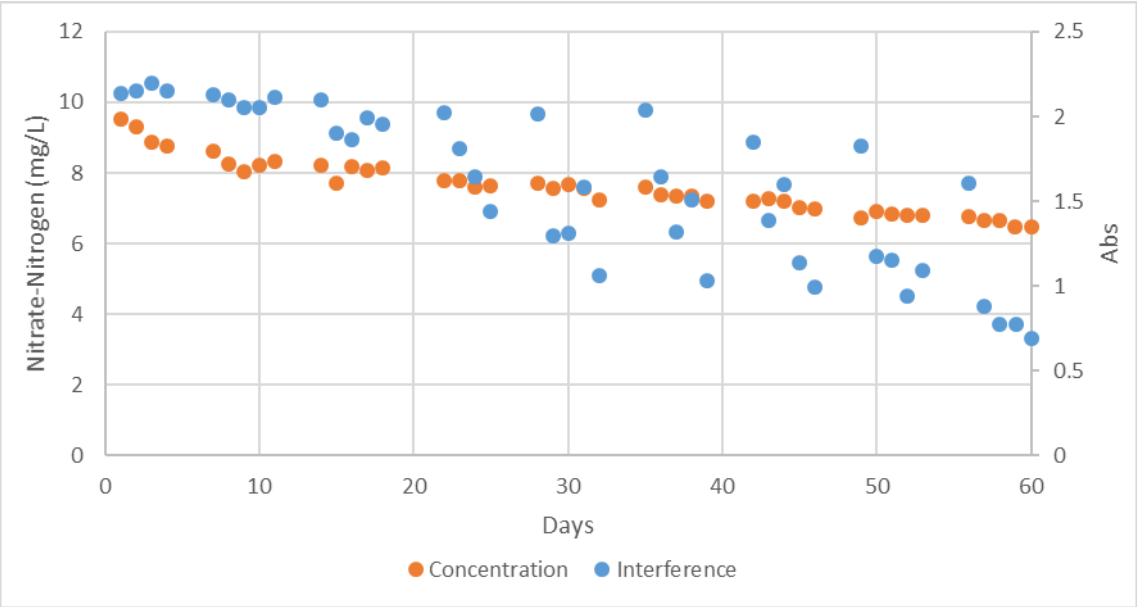

Figure 79: Average nitrate-nitrogen concentration (mg/L) and interference reading at 275nm (abs) for effluent from Eucalyptus woodchip systems.

Figure 79 showed that the Eucalyptus replicates displayed a similar decreasing trend in nitrate-nitrogen concentration (increasing nitrate reduction) in effluent with the systems yet to reach steady-state which would result in a lower nitrate concentration. As an

estimate the lowest achieved nitrate-nitrogen concentration was 6.45 mg NO_3^-N/L resulting in a nitrate reduction of 74.2%. However the replicates showed much higher interference values, as a result the results obtained should only be taken as an estimate without further more accurate testing. The higher interference values only showed some noticeable reduction in the final two weeks observed, which could be due to the fines and nature of the material which caused significant fungal/microbial activity noted in the first phase of experiments causing significant organic matter to be present in the effluent. The Eucalyptus replicates also showed a much darker visible colour serving as an indicator of the presence of organic matter. The maximum coefficient of variation for nitrate concentration for the two replicates for this material was 1.61%, acceptable for validating the results obtained.

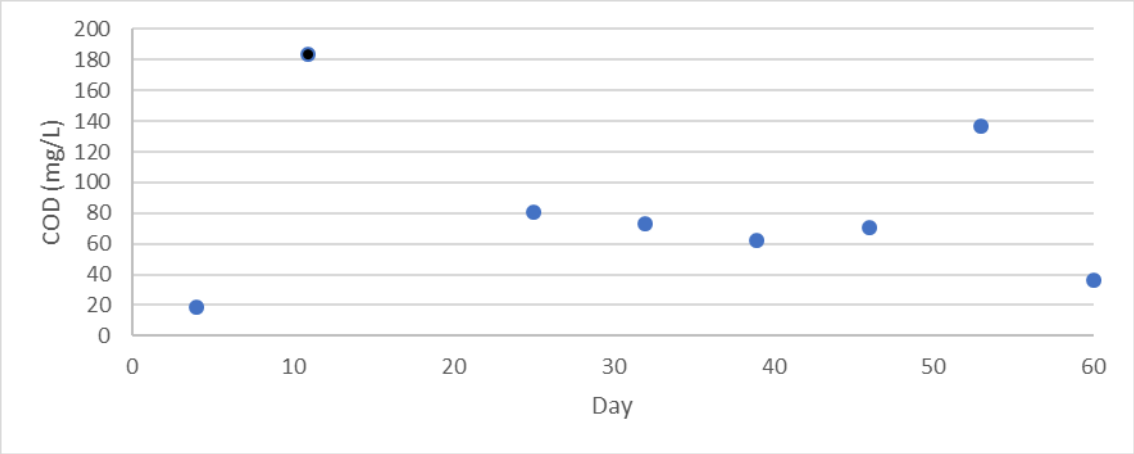

Figure 80: Average COD of effluent of the two replicates for Eucalyptus woodchip replicates. Outliers marked in black.

Figure 80 has the same limitations in regards to outliers as the previous systems. COD observed for the two replicates over the 60 days varied within a slightly higher range (19-137 COD mg/L) than the previous woodchip materials examined, which is expected due to the higher carbon release observed for Eucalyptus woodchips from the previous phases. The low COD observed in the final week of the experiment suggests that this material behaves similarly to the other woodchip materials with COD in effluent decreasing as nitrate reduction increases, however would require more than one data point to validate this claim. The higher range observed could result in some noticeable contribution to COD in effluent.

4.3.5 Princess Tree Woodchips

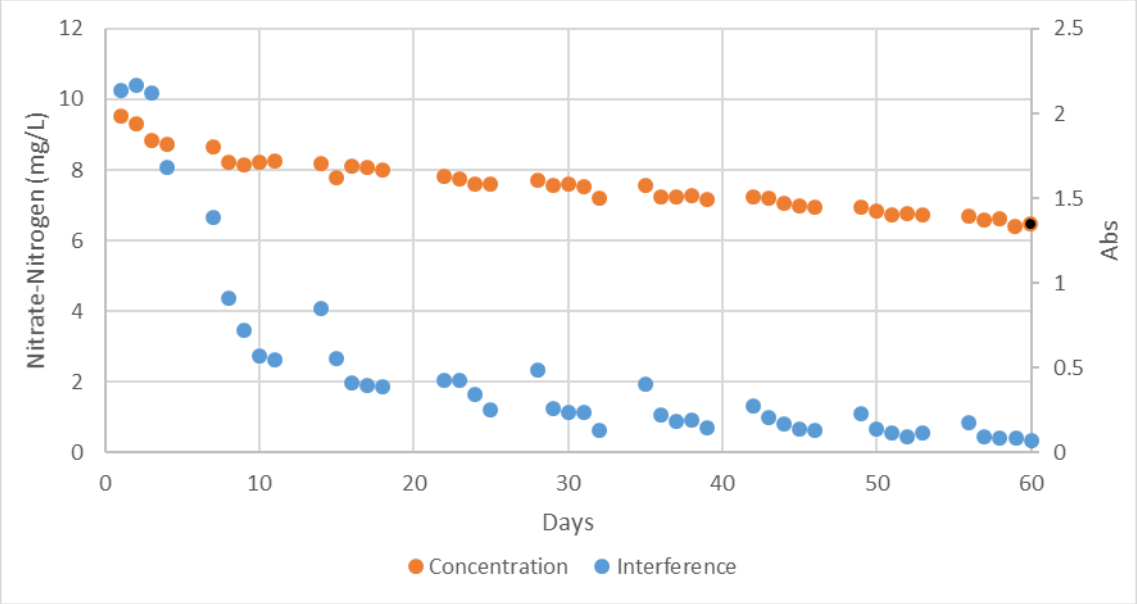

Figure 81: Average nitrate-nitrogen concentration (mg/L) and interference reading at 275nm (abs) for effluent from Princess Tree woodchip replicates. Accurate result highlighted in black.

Figure 81 shows the decreasing nitrate-nitrogen concentration (increasing nitrate reduction) in effluent observed over the experimental period and the replicates were yet to reach steady-state which should result in lower nitrate concentration. As an estimate the lowest nitrate concentration observed was 6.37 mg NO_3^-N/L resulting in a 74.5% reduction in nitrate being achieved. Interference readings were noticeably reduced by day 30 with an 'accurate' nitrate concentration obtained on the final day of monitoring. The effluent produced by these replicates showed the least amount of visible colour as an indicator for lower organic matter present. Maximum coefficient of variation for nitrate concentrations between the two replicates was 0.96% validating results obtained.

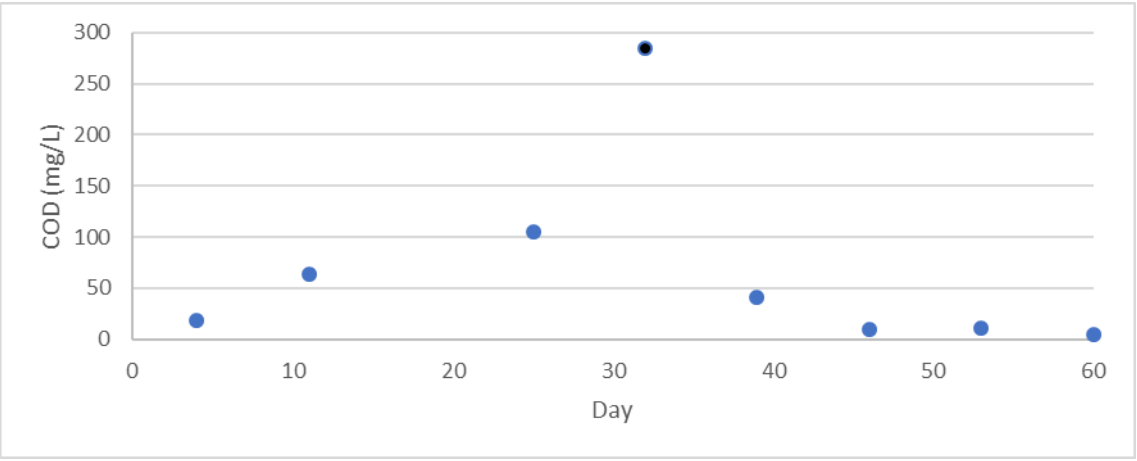

Figure 82: Average COD of effluent of the two replicates for Princess tree woodchip replicates. Outliers marked in black.

Figure 82 shows the stable values for COD in effluent varying in a low range (5-104 COD mg/L) with a notable outlier on day 32 due to issues with temperature control during digestion. The lower COD values obtained during the final 3 weeks of the experiment concur with the previous materials results in suggesting higher nitrate reduction reduces COD present in effluent. The low values obtained suggest Princess tree woodchip would not contribute noticeable COD to effluent during the denitrification process.

4.3.6 Pine Woodchips

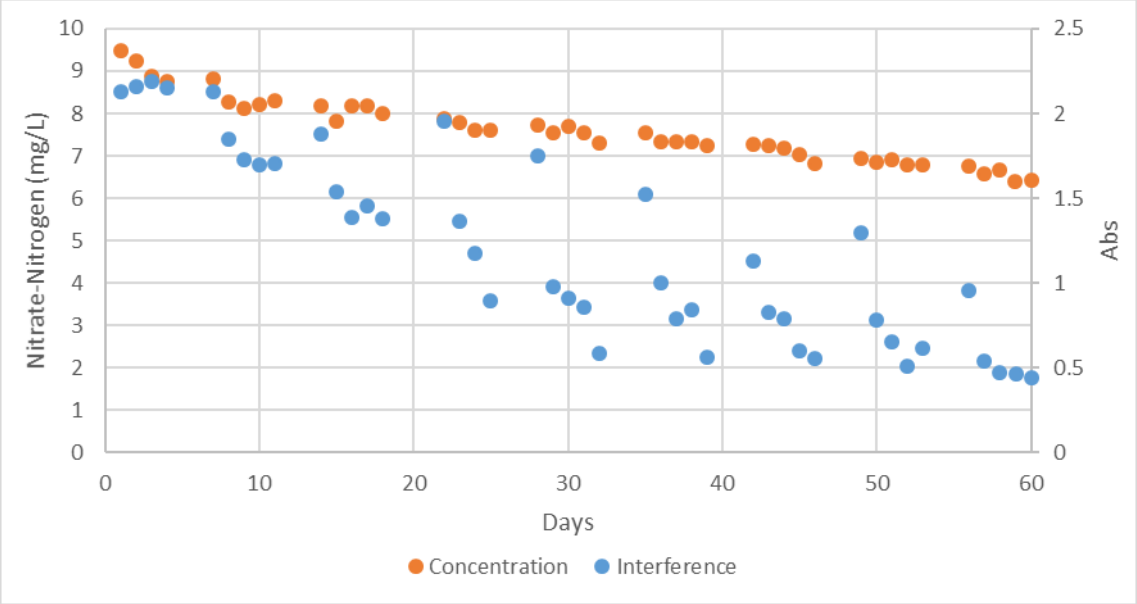

Figure 83: Average nitrate-nitrogen concentration (mg/L) and interference reading at 275nm (abs) for effluent from Pine woodchip replicates.

Figure 83 shows the decreasing trend of nitrate-nitrogen concentration in effluent, but the Pine woodchip replicates had not yet reached steady-state which would result in even lower nitrate concentration than those observed. Therefore as an estimate, the lowest nitrate concentration observed was 6.39 mg NO_3^-N/L resulting in a 74.4% nitrate reduction achieved. Interference values observed showed a decreasing trend further supporting that the replicates had not achieved steady-state. The values observed were also higher than most woodchip systems displayed (apart from Eucalyptus woodchip systems) suggesting higher organic matter present in effluent. This is in concurrence with results in phase 1 where the material had more fines present and showed some fungal/microbial activity. Maximum coefficient of variation for nitrate-nitrogen concentration between the two replicates was 2.08%, although slightly higher than other woodchip systems, is low enough to validate results.

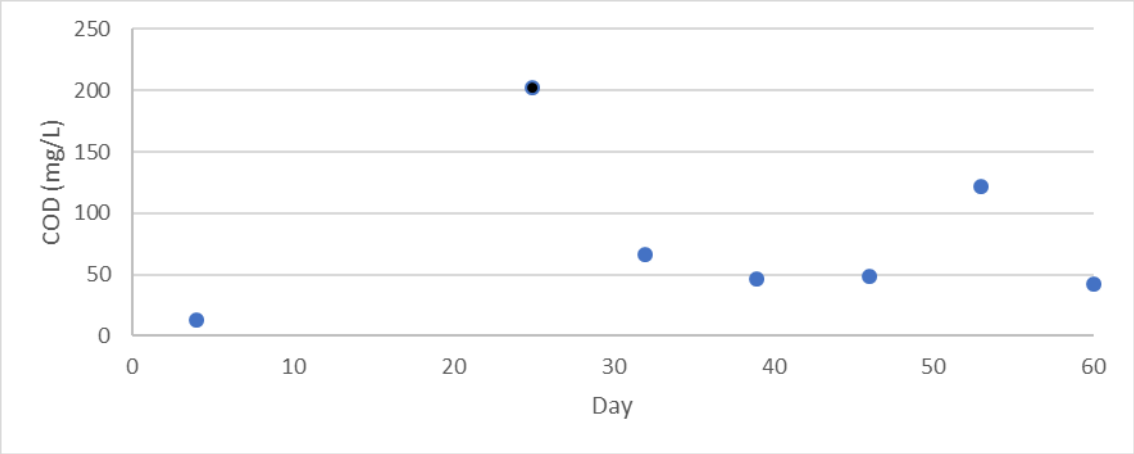

Figure 84: Average COD of effluent of the two replicates for Pine woodchip replicates. Outliers marked in black.

Average COD for the Pine woodchip replicates in effluent (Figure 84) were stable and varied at a low range (13-122 COD mg/L) suggesting there would no significant COD contribution to effluent from this process. Issues with temperature control (tested in different batches than other materials due to digester capacity) during digestion caused the tests for day 11 to be damaged and results on day 25 to be an outlier. Pine replicates did not show the same trend with lower COD in effluent as nitrate reduction increased.

4.3.7 Brewers Spent Grain

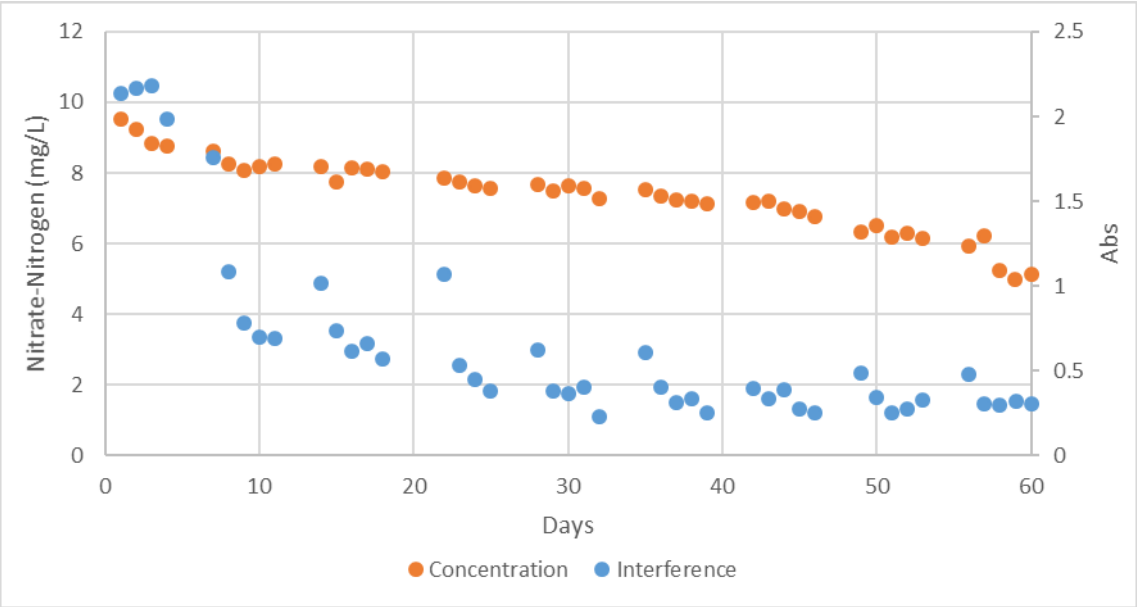

Figure 85: Average nitrate-nitrogen concentration (mg/L) and interference reading at 275nm (abs) for effluent from Brewers Spent Grain replicates.

Figure 85 shows a decreasing trend for nitrate-nitrogen concentration in effluent for Brewers Spent Grain replicates, with the replicates not reaching steady-state nitrate-nitrogen concentration can be expected to be even lower in effluent during operation at

steady-state. As an estimate the lowest nitrate-nitrogen concentration achieved was 4.98 mg NO_3^-N/L translating to an 80.1% nitrate reduction, noticeably higher than those achieved by the woodchip systems. The interference readings observed followed a similar trend to those shown by the majority of woodchip systems, decreasing rapidly within the first week and becoming more stable by day 30 and onwards. Maximum coefficient of variation for nitrate-nitrogen concentration between the two replicates was 16.44%, although high this is still within the scope to validate results. One replicate performed noticeably better for nitrate removal than the other especially towards the end of the experiment period, suggesting there is some variation in start-up time (or time to reach steady-state performance) when using this material for denitrification.

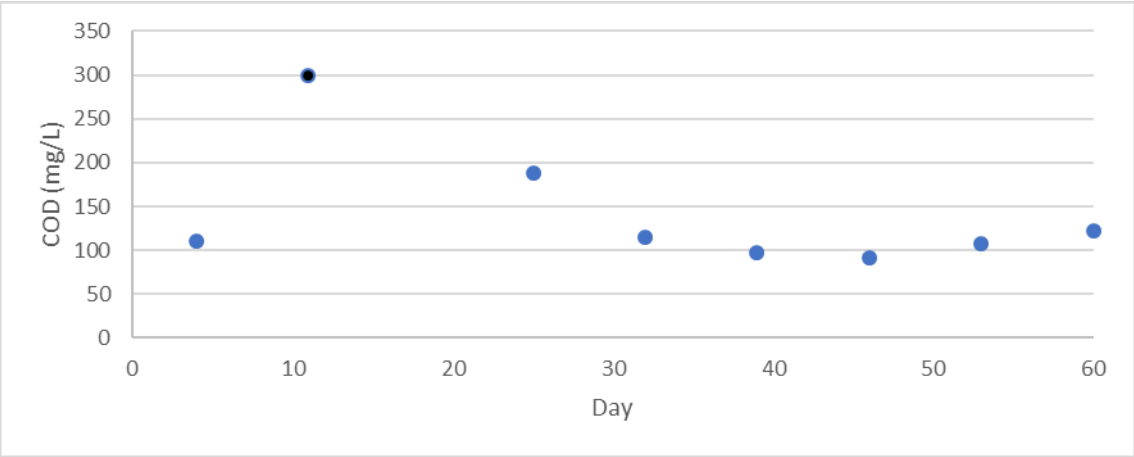

Figure 86: Average COD of effluent of the two replicates for Brewers Spent Grain replicates. Outliers marked in black.

Figure 86 shows the average COD in effluent achieved by the two replicates to be stable outside of the outlier identified on day 11 due to temperature control issues. However the range is significantly higher than the woodchip materials being 83–188 COD mg/L, which could cause issues contributing additional COD to effluent despite higher nitrate reduction observed.

4.3.8 Slum Gum

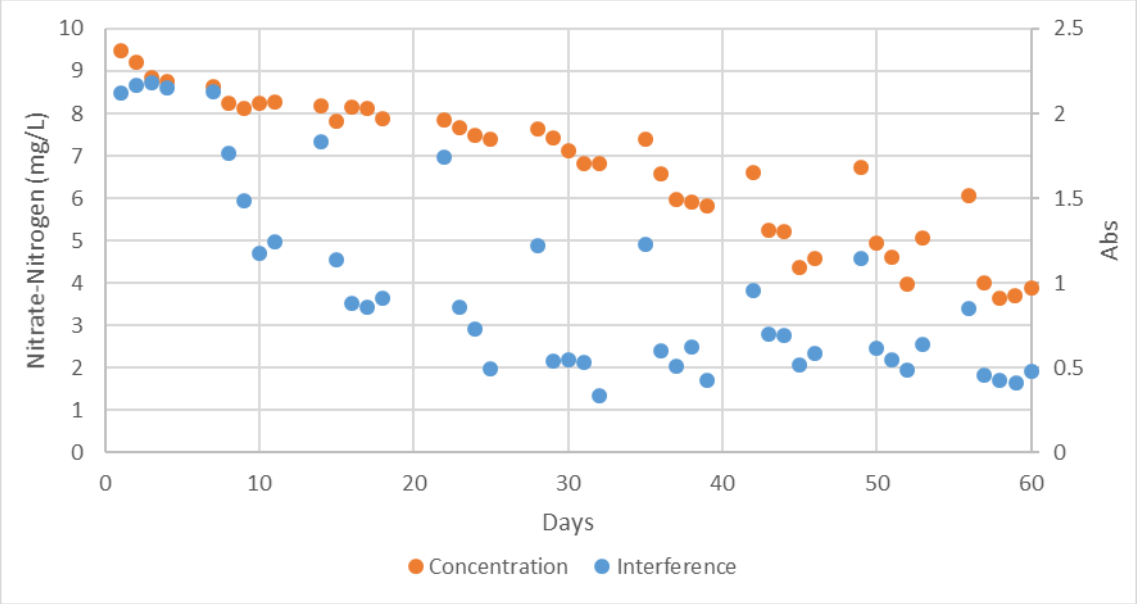

Figure 87: Average nitrate-nitrogen concentration (mg/L) and interference reading at 275nm (abs) for effluent from Slum Gum replicates.

Figure 87 shows a decrease in nitrate-nitrogen concentration over the 60 days the replicates were monitored with steady-state not being achieved the trend suggests lower nitrate-nitrogen concentrations once stable. The impact of longer HRT during the weekends resulted in higher nitrate-nitrogen concentrations in the last 4 weeks of operation that was not observed with the other materials. As an estimate the lowest nitrate-nitrogen concentration obtained was 3.64 mg NO_3^-N/L resulting in a nitrate reduction of 85.5%, the highest nitrate reduction observed out of all materials monitored. Maximum coefficient of variation between the two replicates for nitrate concentration was 10.03%, higher than observed in the woodchip materials but within acceptable limits to validate results. The longer HRT over weekends also caused higher interference values, however excluding these interference values behaved similarly to the other systems stabilising after 30 days operation.

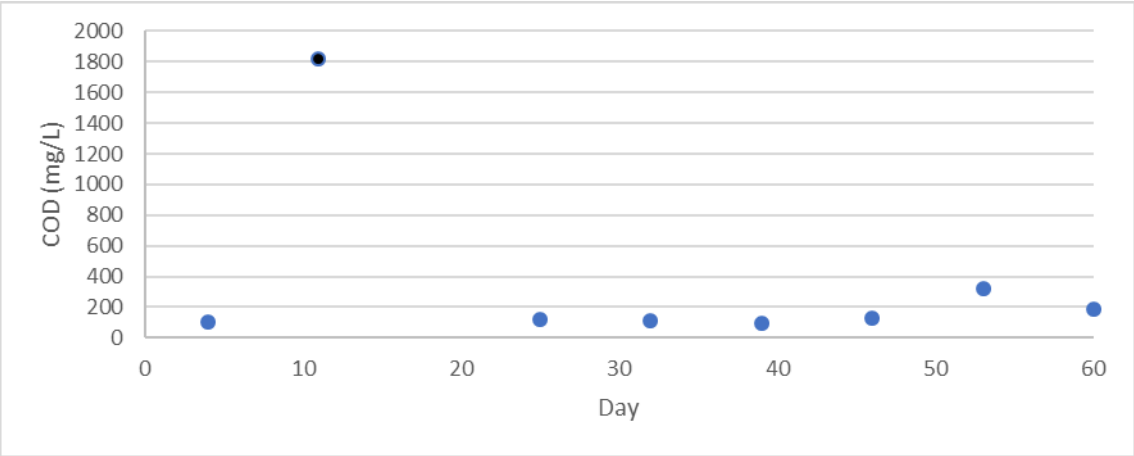

Figure 88: Average COD of effluent of the two replicates for Slum gum replicates. Outliers marked in black

Figure 88 shows a stable average COD varying between 109 and 322 COD mg/L over the 60 days monitored upon exclusion outliers. The outlier on day 11 occurred due to temperature control issues during incubation which also caused the COD vial to be damaged for the test on day 18. As with the Brewers Spent Grain replicates the Slum gum replicates showed an increase in COD in effluent despite having higher nitrate removal. With the higher range observed the material could also cause issues contributing excess COD to effluent when used in a wetland filter.

4.3.9 Dissolved Oxygen in Replicates

Table 20: Average dissolved oxygen present in replicates for each material sorted from lowest to highest and DI water used in dosing

Material	Average DO (mg/L)
Brewers Spent Grain	5.17
Slum Gum	4.57
Mahogany	5.17
Manuka	5.55
Pine	4.73
Eucalyptus	3.51
Eastern Cottonwood	5.73
Princess Tree	4.70
DI Water	9.97

Table 20 shows that no systems achieved anoxic conditions (DO < 0.5 mg/L) for denitrification to occur via anaerobic pathways. However, DO was measured at the end of the experiment after most of the soluble COD has been removed or used in denitrification resulting in less oxygen demand within the systems. Also due to the small

volumes of the systems there would be sufficient mass transfer of oxygen to maintain a higher DO experienced.

The results suggest the nitrate reductions observed do not include anaerobic pathways for nitrate removal (i.e. denitrification) however this could be occurring in anoxic environments within the pores of the materials. As a result when implemented in wetland filters that can achieve anaerobic conditions in the saturated zone denitrification capabilities could be even higher.

4.3.10 pH in Replicates

pH observed on day 60 for the systems would be the best estimate for operational steady-state CWs, which are summarised below in Table 21

Table 21: pH of systems on day 60 of experimental period

Material	pH (day 60)
Pine	4.6
Eucalyptus	5.15
Manuka	5.35
Princess Tree	5.5
Mahogany	5.87
Brewers Spent Grain	6.11
Eastern Cottonwood	6.12
Slum Gum	6.85

The results show that only the BSG, Eastern Cottonwood and Slum Gum systems had a pH within the optimum range for denitrification. Interestingly Pine woodchips produced a significantly acidic leachate which could cause issues as effluent, while the other woodchips were all within 5-6 pH. These results should be taken into account when selecting a material for implementation into CWs.

4.3.11 Mass Balance

Table 22 indicates mass gain for Manuka, Mahogany and Princess Tree woodchip replicates. This could be a result of adsorption of nitrate loaded into the systems or fungal/microbial growth that was not clearly visible causing net mass gain over the experimental period, however practically these materials should be losing mass over an extended period. Pine, Eastern Cottonwood and Eucalyptus woodchip replicates had minimal mass loss over the experimental period. Taking 2% mass loss over 60 days as an

estimate for woodchip materials, woodchip materials used in an operating wetland filter would experience complete mass loss in roughly 3000 days (just over 8 years).

Table 22: Average mass loss between both replicates for each material and percentage mass loss over the 60 days experimental period sorted from lowest to highest.

Material	Average (g)	Mass Loss %
Manuka	-15.1	-5.2%
Mahogany	-10.1	-5.3%
Princess Tree	-2.35	-2.2%
Eastern Cottonwood	0	0.0%
Pine	2.95	2.0%
Eucalyptus	6.5	2.2%
Slum Gum	40.3	14.7%
Brewers Spent Grain	87.8	52.0%

The two waste/by-product materials Slum gum and Brewers Spent Grain experienced significantly more mass loss compared to the woodchip materials. Brewers Spent Grain replicates losing more than half of their initial weight in 60 days, suggesting the material would be completely consumed within around 142 days. While Slum gum would last around 408 days before complete consumption. Despite the significant mass loss no decrease in denitrification performance was observed in these waste/by-product replicates.

In practical implementation replacement would occur before complete consumption of the saturated zone material would occur as theoretically denitrification performance would decrease as significant mass loss would limited carbon available. Brewers Spent Grain showed 52% mass loss without reducing nitrate removal capacity therefore as a safe estimate 50% complete mass loss can be used to determine operational life span for the materials. This results in woodchip materials requiring replacement around every four years, Slum gum requiring replacement every six months and Brewers Spent Grain to be replaced every two months. The short operational lifespan of Brewers Spent Grain systems would be impractical in this scenario requiring much more maintenance than available whereas Slum gum systems would be border line. A more practical solution would be a hybrid mix of woodchip and by-product materials, however further testing for denitrification performance and longevity of said hybrid mixes would be required before recommending this as a solution.

4.3.12 Implications for Wetland Filters

Table 23 summarises percentage of nitrate removal achieved for each material mentioned previously, showing woodchip materials perform similarly while the waste/by-product materials achieved higher nitrate removal capabilities. These results suggest Slum gum and BSG to be good materials for nitrate removal, however other factors such as longevity, odour and appearance issues and cost need to be taken into account when implementing into real-world applications. Further research could be directed at a mix of woodchip materials (for longevity) and waste/by-product materials (for higher carbon release) to meet criteria required for wetland filter systems.

Table 23: Nitrate removal % for each material and sorted in ascending order

Material	Nitrate removal %
Eucalyptus	74.2%
Eastern Cottonwood	74.4%
Mahogany	74.4%
Manuka	74.4%
Pine	74.4%
Princess Tree	74.5%
Brewers Spent Grain	80.1%
Slum Gum	85.5%

Comparing with results from phase 1 (Table 2), a correlation between organic carbon release capacities for materials and denitrification performance is not clear. Suggesting organic carbon release is not the only key parameter restricting denitrification performance. However, by-product materials serving as media for denitrification did outperform woodchips concurring with results from phase 1 where carbon release for by-products was significantly higher than woodchips. Therefore, a simple carbon release test (i.e. phase 1 of the experiment) could be an early screening tool for materials to evaluate suitability as media to be organic carbon sources rather than a direct indicator of denitrification performance for materials.

Chapter 5
Conclusions

5.1 Phase 1

The objective of phase 1 of this research examines the ability of materials (Eastern Cottonwood, Mahogany, Manuka, Eucalyptus, Crack Willow, Princess Tree and Pine woodchips, Brewers Spent Grain, Slum Gum and Coir) to release carbon when allowed to soak for 30 days, therefore giving an indication on the amount of organic carbon available for the denitrification process and viability of creating a leachate appropriate for supplementing organic carbon sources in the saturated zone of PSVF wetland units.

Results showed that BSG, Slum Gum, Eucalyptus and Pine woodchips achieved the highest average COD and therefore highest concentration of organic carbon present in leachate over the 30 day experimental (11779 COD mg/L, 6023 COD mg/L and 4350 COD mg/L respectively). These three materials also exhibited the highest initial release rates, 9191 COD mg/L.day, 731.3 COD mg/L.day and 690.6 COD mg/L. respectively. As a result, these three materials seem to be the most appealing candidates in enhancing denitrification in the saturated zone of PSVF wetland units. However, other factors experienced by these systems need to be considered in implementation for marae grounds; the BSG and slum gum systems experienced significant mass loss (41% for BSG and 31% for slum gum) over the experimental period and had a pungent fermentation/degradation smell. The significant mass loss indicates the materials would require replacement or topping up more often and low odour is part of the criteria for maraes in wastewater systems. While the eucalyptus (and pine) woodchip systems produced an effluent/leachate with a dark yellow colour representative of the media colour, undesirable in effluent.

The remaining materials ranked by organic carbon present in leachate (average COD) as follows; Pine, Princess Tree, Crack Willow, Mahogany, Manuka and Eastern Cottonwood woodchips followed by the Coir briquette. Woodchip systems showed similar organic carbon release (albeit Pine woodchips with slightly higher average COD observed in leachate), while Coir had the least organic carbon present in leachate.

Using parameters obtained from the Pukete mesocosm scale trials to estimate organic carbon requirements (where C:N ratio is above 5:1) for not restricting denitrification, only BSG proved to be a viable material to serve as media (in the SZ) and for dosing at a rate of 50 mL leachate per litre of influent. However, these results are from a single cycle with fresh material which is not how TWs operate and therefore only serve as an indicator.

5.2 Phase 2

Phase 2 examined the selected materials ability to release carbon over three 10 day cycles, giving a more accurate indication of organic carbon, and therefore denitrification performance (with organic carbon being a limiting factor in denitrification) as well as viability of materials to produce leachate for dosing over more than one cycle.

COD concentrations over the 10 day cycles suggested that carbon release occurs mostly within 1-2 days. The average COD of the final cycle of each material/system is used to estimate organic carbon available when the materials are used in-situ and for constant production of leachate. The results showed that BSG, Pine and Eucalyptus woodchips, and Slum Gum were the best performing resulting in average COD (of the final 10 day cycle) of 4064 COD mg/L, 1100 COD mg/L, 927.5 COD mg/L and 909.2 COD mg/L respectively. The remaining materials ranked as follows for average COD; Mahogany woodchips, Coir, Princess Tree woodchips, Crack willow woodchips, Eastern Cottonwood woodchips and Manuka woodchips.

Using the same parameters (from Pukete trials) as in phase 1 only BSG met the organic carbon requirements for not restricting denitrification performance. However, no materials met the available organic carbon requirements for dosing at 50 mL leachate per litre of influent. Although due to the packing used in experimental systems there are some concerns of excess organic carbon passing through to effluent, which is undesirable. As a result, leachate production would require fresh material every batch making it impractical for the intended use at marae.

5.3 Phase 3

Upon analysing results from phase 1 and phase 2, Coir and Crack Willow woodchips from the pool of selected materials for testing in phase 3. The remaining materials were

dosed with 25 mg/L nitrate with a single day HRT (except weekends) over 60 days to give an indication of denitrification performance.

Monitoring of pH during this phase indicated that only Eastern Cottonwood woodchips, BSG and Slum gum systems fell within the pH range for optimum denitrification (6-8 pH), suggesting the use of other materials would require pH adjustment if implemented as media.

Mass loss of materials during this phase gives the most accurate indication of longevity of materials. BSG experienced a mass loss of 52% over the 60 day experimental period, followed by Slum gum experiencing a mass loss of 14.7%. As a result, the estimated lifetime of BSG serving as media in the SZ of a PSVF unit is roughly 142 days before complete consumption and Slum gum a lifetime of 408 days before complete consumption. Woodchip materials on the other hand experienced negligible mass loss indicating a long lifespan requiring less maintenance in the form of replacement or addition.

The two by-products, BSG and Slum gum, showed the best nitrate removal at 80.1% and 85.5% removal respectively. The remaining woodchip materials all showed similar denitrification performance ranging between 74.2-74.5% nitrate removal, which are reasonably comparable to that achieved by the by-product materials. These results in combination with results from phase 1 suggest that organic carbon release directly correlated to denitrification and is not the only key factor in denitrification performance for these materials. However, the stand out performance of by-product materials in phase 1 is similar to those experienced in phase 3. Therefore suggesting that the simple test administered in phase 1 could be used in mass screening of materials rather than a direct indicator of denitrification capability in future applications.

While the denitrification performance of the by-product materials is higher than woodchip materials, the lower lifespan of these materials is a concern. As a recommendation, for materials to serve as media contributing organic carbon in the SZ of PSVFs investigation could be directed at a mix of woodchip and by-product materials or other by-product materials with longer lifespans or even other species of woodchip.

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