EVALUATING THE EFFECTIVENESS OF CONSTRUCTED WETLAND IN POLISHING WASTEWATER EFFLUENT FROM GUSII TREATMENT PLANT IN KISII TOWN, KENYA

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DECLARATION

This thesis is my original work and has not been presented for award of a degree in any other university

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DEDICATION

Dedicated to my father Mr. Elias Otieno, mother Mrs. Grace Akinyi and brothers and sisters.

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LIST OF ABBREVIATIONS AND ACRONYMS

Abbreviations	**Description**
APHA	American Public Health Association
ASTM	American Standards for Testing and Materials
BOD	Biochemical Oxygen Demand
COD	Chemical Oxygen Demand
CRD	Complete Randomized Design
FWS	Free Water Surface System
HB	Hybrid System
HF	Horizontal Flow System
LVEMP	Lake Victoria Environmental Management Project
LVSWB	Lake Victoria South Water Services Board
MDG	Millennium Development Goals
NEMA	National Environmental Management Authority
OECD	Organization for Economic Co-operation and Development
SFS	Subsurface Flow Systems
TN	Total Nitrogen
TP	Total Phosphorous
TSS	Total Suspended Solids
UNEP	United Nations Environmental Program
UNESCO	United Nations Educational, Scientific and Cultural Organization
UNFPA	United Nations for Population Fund
USCS	Unified Soil Classification System
VF	Vertical Flow System
VSB	Vegetated Submerged Bed System
WHO	World Health Organization
WWC	World Water Council

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ABSTRACT

Constructed wetlands are widely recognized low cost wastewater treatment options, especially in developing countries where the conventional treatment systems are expensive to operate. To assess the potential in polishing of wastewater from Gusii municipal wastewater treatment plant, a horizontal, vertical and hybrid subsurface flows pilot scale constructed wetlands were designed, constructed, operated and the effluent analyzed for BOD_5, COD, TSS, TP and TN from systems either planted or not with Vetiver grass (*Chrysopogon zizanioides*). Vetiver grass tissues and roots were analyzed for Nitrogen and Phosphorous accumulation at the end of the experiment. Among the subsurface flow wetland systems planted with Vetiver grass, the hybrid system achieved significantly ($P \leq 0.05$) the highest removal of BOD_5 COD, TN, TP and TSS at 86.6, 82.4, 87.9, 65 and 94.6%, respectively compared to other wetland systems. The planted vertical system removed BOD_5 COD, TN and TP at 80.9, 72.9, 75.7, and 50.7%, respectively more efficiently ($P \leq 0.05$) than the horizontal system that achieved removal of BOD_5, COD, TN and TP at 75.8, 65.3, 70.0 and 43.8%, respectively. The planted horizontal system however showed better TSS removal at 89.9% compared to 83.2% achieved by vertical system. The unplanted subsurface flow wetland systems achieved significantly ($P \leq 0.05$) lower organics and nutrients removal efficiencies compared to the planted systems. The unplanted hybrid systems achieved the highest removal of BOD_5, COD, TN, TP and TSS at 73.8, 66.0, 61.4, 55.2 and 83.4%, respectively compared to other unplanted wetland systems. The unplanted vertical system removed BOD_5, COD, TN and TP at 63, 52.5, 51.7 and 35.9%, respectively more efficiently ($P \leq 0.05$) than horizontal system that achieved removal of BOD_5 COD, TN and TP at 56.9, 46.5, 33.3 and 32%, respectively. The unplanted horizontal system however showed better TSS removal at 79.4% compared to73.6% achieved by unplanted vertical system.

Vetiver grass accumulated 18,100 mg and 35.3 mg/kg Nitrogen and Phosphorous, respectively in the hybrid system as compared to 9,400 mg Nitrogen and 19 mg/kg Phosphorous, in the horizontal system and 10,400 Nitrogen and 18.3mg/kg Phosphorous in the vertical system. Accumulation of nitrogen and phosphorous by Vetiver grass in all the wetland systems were significantly different at 5% confidence level. There were also significant differences on N and P accumulation in the shoots and the roots with N accumulating more in the shoots while P in the roots.

CHAPTER ONE

1.0 GENERAL INTRODUCTION

1.1 Background Information

Poor wastewater management has contributed to challenges of water quality experienced globally. According to OECD (2012), proper wastewater management is important and contributes significantly in protecting water quality for domestic, industrial and agricultural uses. Proper treatment mechanism using environmentally friendly technologies is an approach that was mooted in the post-2015 Development Agenda to achieve water security (WHO, 2015).

The past focus by the Millennium Development Goals (MDG) on improving access to sanitation facilities without much consideration and emphasis on proper wastewater treatment resulted into the deteriorating water quality observed globally (WHO, 2015). Wastewater is the major contributor to the increasing anaerobic zones seen in the large water bodies (Diaz and Rosenberg, 2008). Lack of adequate wastewater management facilities in most cities (WWC, 2012), have escalated the problem of anaerobic conditions, leading to adverse effects on environment. The fourth World Water Development Report of 2012 indicated that only 20% of globally produced wastewater received proper treatment and the deteriorations resulting from eutrophication adversely affected biodiversity in water bodies (UNESCO, 2012).

The Lake Victoria Environmental Management Project reported a decline in water quality in the lake due to eutrophication from increased nutrient inflows (Juma et al., 2014). To cope up with the demands set by the Environmental Management and Coordination Act of 1999, adoption of low cost and effective wastewater treatment technologies such as constructed wetlands by industries in the Lake Victoria region could be viable options. According to Oketch (2006), adoption of constructed wetlands for wastewater management in Kenya is still low due to poor understanding of its potential. Lack of technical knowhow has hampered innovative approaches to wastewater management and hence the low adoption of this technology.

Wastewater treatment in constructed wetlands mimics the natural processes taking place in nature, but in a controlled environment (Tsang, 2015; Wu et al., 2015; Tournebize et al., 2016).

Zhang et al. (2016), defines constructed wetlands as artificially engineered ecosystems, designed and constructed to manipulate biological processes within a semi controlled natural environment.

Constructed wetlands and wastewater stabilization ponds combined may be important for polishing effluent from wastewater treatment systems leading to a more improved performance. Constructed wetland system is considered by Chaikumbung et al. (2016) to be relatively inexpensive and sustainable. The present study therefore evaluated the effectiveness of constructed wetland planted with Vetiver grass in addressing the challenges of water treatment works.

1.2 Statement of the problem

Gusii wastewater treatment plant receives wastewater from domestic, institutional and agricultural sources as well as urban runoff. This wastewater is subjected to conventional treatment by passing it through a series of waste stabilization processes namely anaerobic, facultative and maturation ponds and finally discharging it to river Riana, a source of domestic and agricultural water for residents living downstream. Wastewater analysis report of 2015 indicates a need to further polish the effluent before discharging it into the river. From the recent water analysis, the level of biochemical oxygen demand (BOD), chemical oxygen demand (COD) and total suspended solids (TSS) at the outlet of the maturation pond and discharge point into the river were above the maximum allowable limits (NEMA, 1999). The effluent BOD, COD and TSS levels were 61.53, 61.53 and 68.75%, respectively, above the NEMA maximum allowable effluent discharge levels. This study therefore aimed at designing and constructing a wetland to further polish the effluent from the maturation pond before discharging into Riana river.

1.3 Justification

Kenya a chronically water scarce country has one of the world’s lowest water per capita at 650 m^3 against the global recommendation of 1000 m^3 and this is expected to drop further to 250 m^3 by 2025 when the population is projected at 60 million persons (Mogaka et al., 2006; UNEP, 2006). To generate income and sustain household food requirements, the poor urban population continues to use untreated wastewater for irrigation purposes, which is a great threat to health. The use of cheap and environmentally friendly technologies such as constructed wetlands in wastewater treatment can contribute greatly in providing clean and safe water to these communities.

With devolution to county governments in Kenya, there is a likelihood of water quality crisis from urbanization, land use changes, industrialization, high living standards and with poor wastewater management. This therefore means that wastewater treatment plants such as Gusii are likely to receive more municipal waste as water demands rise and hence the need for better ways to manage and avail it for alternative uses.

1.4 Objectives

1.4.1 Overall objective

To evaluate the effectiveness of a model constructed wetland treatment system in polishing wastewater effluent from Gusii treatment plant.

1.4.2 Specific objectives

i. To characterize the hydraulic parameters of a model constructed wetland treatment system for polishing wastewater effluent from Gusii treatment plant.

ii. To compare effectiveness and recommend between the horizontal, vertical and hybrid subsurface constructed wetland systems in polishing wastewater effluent from Gusii treatment plant.

iii. To evaluate accumulation of nitrogen and phosphorous from the wastewater effluent by Vetiver grass (*Chrysopogon zizanioides)* planted in the model constructed wetland treatment system.

1.5 Hypotheses

i. Hydraulic parameters of a model constructed wetland system do not influence polishing efficiency for wastewater.

ii. Wastewater polishing efficiency is not influenced by the type of flow system in a constructed wetland.

iii. Planted Vetiver grass has no impact on the uptake of nitrogen and phosphorous from wastewater in a constructed wetland system hence does not polish the water effectively.

1.6 Outline of the thesis

This thesis is organized into seven chapters. Chapter one deals with background information, statement of the problem, justification, research objectives and hypotheses. Chapter two deals with general literature review while Chapter three details with the general materials and methods. Chapter four discusses the design and construction of subsurface flow wetland systems. Chapter five evaluates the performance of the horizontal, vertical and hybrid subsurface flow constructed wetland systems in polishing effluent from Gusii wastewater treatment plant while Chapter six evaluates the impact of planted Vetiver grass (***chrysopogon zizanioides*****)** on nitrogen and phosphorous uptake from the model constructed wetland treatment system. Chapter seven presents the general discussion, conclusions and recommendation of the study.

CHAPTER TWO

2.0 GENERAL LITERATURE REVIEW

2.1 Municipal wastewater and its characteristics

Wastewater can be defined as water that has been fouled by various uses. Corcoran et al. (2010) define wastewater as a combination of one or more of waste streams from domestic (black and grey water), commercial, institutional, industrial, urban run-off and agricultural sources either dissolved or as suspended matter.

Municipal wastewater contains nutrients and pathogens (Hanjra et al., 2012; Khatab et al., 2015; Elgallal et al., 2016) that lowers its quality for agricultural and livestock use resulting into high economic impacts when released into the environment untreated. According to Arend et al. (2011), eutrophication resulting from the wastewater inflows into water bodies alters species composition and dominance. Eutrophication has greatly been contributed by inadequately treated wastewater and agricultural run-off into water bodies (Hawkins et al., 2013; Kumar et al., 2016).

2.2 Wastewater quality analysis for the Gusii treatment plant

Kenya's National Environmental Management Authority (NEMA) and the Kenya Bureau of Standards (KEBS) have guidelines on quality requirement for effluent discharge into the environment in order to increase accountability for implementation of pollution control measures.

Table 2.1 shows the effluent quality of wastewater from the Gusii treatment plant against the expected standards for discharge into land or on water by NEMA.

Table 2.1: Quality of wastewater from Gusii treatment plant and water quality of recipient river (Sampling conducted during the rainy season) in 2015

Parameter	Units	NEMA Standards (Max)	Results Riana river upstream	Results Riana river downstream	Treatment plant Inlet	Treatment plant Outlet
pH	pH	6.5 – 8.5	6.8	7.4	7.08	7.65
TDS	Mg/l	1200	128.1	154	410	361
COD	Mg/l	50	40	96	814	130
BOD_5	Mg/l	30	35	48	364	78
TSS	Mg/l	30	60	78	424	96
Temperature	^oC	25-35	25.6	27.0	26.08	25.23
Total N	Mg/l	2	2.957	4.577	49.66	61.32
Total P	Mg/l	2	0.102	0.712	7.07	15.64

SOURCE: Gusii water and sanitation company (GWASCO, 2015)

TDS: Total Dissolved Solids, COD: Chemical Oxygen Demand, BOD: Biochemical Oxygen Demand, TSS: Total Suspended Solids.

The effluent biochemical oxygen demand (BOD), chemical oxygen demand(COD), total suspended solids (TSS), total nitrogen (TN) and total phosphorous (TP) levels were 78, 130 and 96, 61.32 and 15.64 mg/l respectively, in 2015. This was above the maximum allowable effluent discharge levels by the National Environment Management Authority of 30, 50, 30, 2 and 2mg/l for BOD_5, COD, TSS, TN and TP , respectively. This is an indication of inadequate polishing by the waste stabilization ponds. The up- and down-stream values are normally taken at 100m from the point of effluent discharge where it is assumed polished effluent will have mixed with flowing water in the receiving water body. Additionally, better quality water will have been realized from the self-purification ability of the river (Maina et al., 2010; Viswanathan et al., 2015; Bakar et al., 2016). However it was observed that the levels of BOD_5 and TN downstream were 37.5 and 56.3%, respectively above the allowable discharge levels. This poses health risks to residents and their livestock downstream, as well as bio-accumulation of heavy metals in crops which is hazardous to humans, livestock and even aquatic life such as fish that are consumed by humans.

2.3 Treatment processes at the Gusii municipal wastewater plant

Wastewater received by Gusii treatment plant is subjected to conventional treatment by passing it through bar screens, grit chamber and a series of wastewater stabilization ponds and finally released as effluent to river Riana. The bar screens remove large floating objects such as polythene and twigs while the grit chamber allows sand and grit particles to settle through sedimentation process (Goldman et al., 1986; Omelia, 1998). The waste stabilization ponds treat the wastewater entirely by natural process comprising of the anaerobic, facultative and maturation ponds (Rahmatiyar et al., 2015; Sabahet al., 2016). In the anaerobic pond, much of the organic loads settle at the bottom as sludge thus achieving high BOD reduction (Tchobanoglous et al., 2004; Pescod, 2016). The facultative ponds which are generally larger with longer retention period receives wastewater with low organic loads which is a favourable environment for algal proliferation that takes up the organics, nitrates and phosphates (Reinoso, 2011; Norvill et al., 2016). The maturation pond, the final treatment stage is designed for the removal of excreted pathogens through Ultra Violet (UV) disinfection as they are shallow and allow UV radiation penetration to the bottom of the pond (Reinoso, 2011; Verbyla et al., 2015; Vannoy, 2016).

2.4 Constructed wetland systems

Constructed wetlands and waste stabilization ponds when combined may be of importance for improved water cleaning systems (Chouinard et al., 2015; Banjoko et al., 2016). Constructed wetlands have been used for treating various types of wastewater in tropical and subtropical regions and thus considered as a sustainable wastewater management option for developing countries (Zhang et al., 2015). They are normally erected on a slope between 0.5 to 1 % and their shallowness permit better pollutants removal (Imfeld et al., 2009; Amacha et al., 2017).

2.4.1 Types of constructed wetlands

Two types of commonly constructed wetlands are free water surface systems (FWS) and subsurface flows systems (SBF) (Wu et al., 2015; Vymazal et al., 2015). In FWS system, water flows above the substrate and macrophytes are rooted below the water column where aerobic conditions prevail near the surface layer while anaerobic conditions dominate in the substrate (Maine et al., 2016; Zheng et al., 2016). The SBF systems are designed to maintain water level below the substrate upon which plants are established (Xu et al., 2015) and are suited to wastewater with low solid concentration to reduce clogging (Aiello et al., 2016; Miranda et al., 2016).

2.4.2 Types of flow directions

Three types of flow directions commonly used in constructed wetlands are namely, horizontal, vertical and hybrid systems (Cui et al., 2015; Dittrich et al.,2015). In horizontal flow constructed wetland, wastewater is fed continuously at the inlet, flows horizontally through the porous substrate until it reaches the outlet (Tsang, 2015; Zhang et al., 2016). Wastewater is cleansed through physical, biological and chemical processes as it passes through the substrate (Vymazal, 2010). This flow system can effectively remove organic pollutants from wastewater (Vymazal et al., 2015; Zhang et al., 2016), although nutrient removal especially nitrogen is low due to saturated conditions (Cooper et al., 1996; Coban et al., 2015).

In vertical flow constructed wetland, wastewater is fed intermittently from the top in large batches which then gradually percolates down through the bed under influence of gravity and is collected by a drainage network at the base (Vymazal et al., 2015; Fan et al., 2016). The intermittent feeding of wastewater allows the bed to be completely drained thus promoting nitrification and high organics removal (Xu et al., 2015; Jong et al., 2016). Hybrid system comprises of both the horizontal and vertical flow systems and the set up can either be horizontal flow followed by vertical flow wetland and vice versa thus achieving high treatment efficiency (Avila et al., 2015; Torrijos et al., 2016) especially with higher retention time.

2.5 Use of Vetiver grass for wastewater treatment

Vetiver grass (*Chrysopogon zizanioides*) belongs to the Graminae family and was first used for soil and water conservation in India in the 1980s by the World Bank (Truong and Loch, 2004). Since then, its role has been successfully extended to wastewater treatment (Soni et al., 2015; Mathew et al., 2016) works. Vetiver grass has proved to be an effective and low cost natural method of environmental protection (Adigun et al., 2015; Greenfield, 2002). According to Yeboah et al. (2015), vetiver grass has high nutrient removal from wastewater and thus can be used for pollution control. Due to its ability to thrive in unfavourable environments with high toxicities, vetiver grass has been considered suitable for wastewater treatment (Paz-Alberto and Sigua, 2013).

2.6 Pollutants removal mechanisms

Constructed wetlands mimic the natural chemical and biological processes occurring in wetlands in removing contaminants from the wastewater with the basic mechanisms being sedimentation, chemical precipitation, adsorption, microbial interactions and uptake by the vegetation (Fernandes et al., 2015; Tsang, 2015). Aerobic degradation of soluble organic matter is governed majorly by the aerobic heterotrophic bacteria due to their faster metabolic rate although ammonifying bacteria also degrade organic compounds containing nitrogen under aerobic conditions (Cooper et al., 1996). Settleable and suspended solids are effectively removed in the wetland by filtration and sedimentation (Jácome et al., 2016) and most of the removal occurs within a few meters beyond the inlet owing to the shallow depth of the liquid in the subsurface flow systems (Cooper et al., 1996).

Nitrogen is mainly removed by nitrification-denitrification (Fu et al., 2016; Paranychianakis et al., 2016) processes. Plant uptake also contributes to nitrogen removal in wetlands since they require nitrogen for growth (Billore et al., 2002). According to Hoffman et al. (2011), phosphorus removal can be achieved in constructed wetlands by adsorption and precipitation in the soil, and a small amount is also taken up by plants for growth. Pathogens are eliminated through the system mainly by sedimentation, filtration and adsorption by biomass and once entrapped within the system, their numbers decrease rapidly through predation and natural die-off (Cooper et al., 1996).

2.7 Abiotic factors and their influence on wetlands

Oxygen in wetland systems is utilized by heterotrophic bacterial for oxidation and growth (Giorgio et al., 1998), and biologically mediated processes such as nitrification and decomposition of organic matter (Nivala et al., 2013). The pH of wetland waters has an influence on the wetland performance since the biota of wetlands is impaired by sudden changes in pH (Batty et al., 2007; Iamchaturapatr et al., 2007). Temperature is also a widely fluctuating abiotic factor that strongly influences the rate of biological and chemical processes in wetlands (Kadlec and Reddy, 2001; Huang et al., 2013).

CHAPTER THREE

3.0 General Materials and Methods

3.1 Location of study

Gusii wastewater treatment plant is located in Kisii town; Suneka Division in Kisii County at latitude 0° 39' 30” S and longitude 34° 42' 30” E. Kisii town has a population of approximately 83,000 people within the municipal boundaries and about 200,000 people within the service area of the Kisii Water Supply System (UN-Habitat, 2008). Figure 3.1 shows the location of the study area.

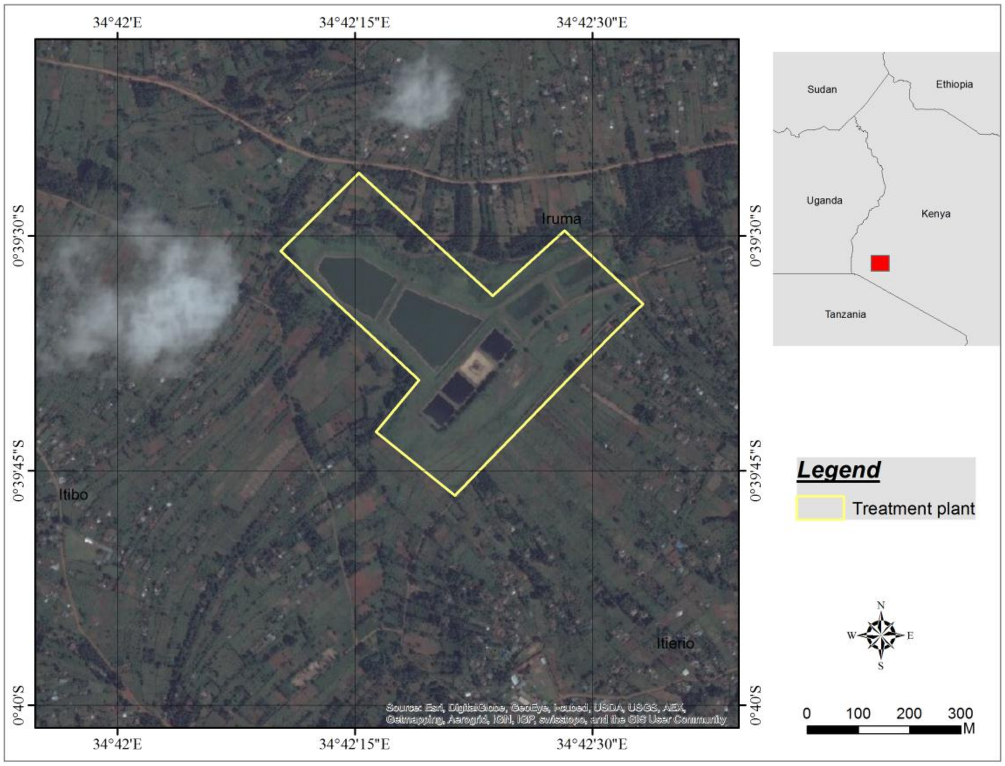

Figure 3.1: Layout of the study area

SOURCE: Topographical maps of Kenya (1990)

3.2 Topography

Kisii County is characterized by hilly topography and is endowed with several permanent rivers draining into Lake Victoria (Jaetzold et al., 2009; Wamalwa et al., 2016). Natural vegetation cover in the study area is low since 90% of the total area is under cultivation (GoK, 2009; Jaetzold et al., 2009).

3.3 Climate

The area has a highland equatorial climate resulting into bimodal rainfall pattern with the long rains occurring between February and June, and short rains between September and December. The area receives a mean annual rainfall of 1500mm (Wamalwa et al., 2016). The month of January and July are generally dry and the maximum temperatures range between 21 to 30°C, while the minimum are between 15 to 20°C (Jaetzold et al., 2009).

3.4 Soils

Seventy five percent of the county has deep red volcanic soils (Nitisols), rich in organic matter, while the remaining area comprises of clay, red loams, sandy soils, black cotton soils classified as Vertisols and organic peat soils classified as Planosols (Wamalwa et al., 2016; Wielemaker and Boxem, 1982) according to WRB (2006) classification.

3.5 Socio-economic aspects

Mixed farming is the main economic activity in the area and over 80% of the agricultural land is devoted to food and cash crops such as maize, finger millet, sorghum, beans, sweet potatoes, tea, coffee and sugarcane (Kisii Central District, 2008). According to Wamalwa et al. (2016), the high population density in the area has led to high food demand and reduction in farm sizes.

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CHAPTER FOUR

DESIGN, CONSTRUCTION AND TESTING OF HORIZONTAL, VERTICAL AND HYBRID SUBSURFACE FLOW CONSTRUCTED WETLAND SYSTEMS

Abstract

Constructed wetlands are eco-friendly alternatives for treating and reclaiming wastewater. Different types of constructed wetlands vary in their effectiveness to treat wastewater; however, they are practical low-cost alternatives to the conventional treatment systems. The study involved designing, constructing and evaluating the performance of horizontal, vertical and hybrid subsurface flow wetland systems. The wetland systems were designed using first order model developed by Kickuth (1977) that is based on BOD_5 removal. Among the subsurface flow wetlands planted with Vetiver grass, the hybrid system achieved significantly ($p \leq 0.05$) the highest BOD_5 removal at 86.6% followed by vertical system at 80.9%. The horizontal system achieved 75.8%. The unplanted systems exhibited a similar trend though with significantly ($p \leq 0.05$) lower BOD_5 removal compared to the planted systems. The unplanted hybrid system achieved BOD_5 removal at 73.8% followed by the vertical system at 63% and finally the horizontal system at 56.9%. Despite selecting coarse sand with higher porosity of 34.3% and lower silt content of 9.9% as the wetland substrate, clogging was experienced in the horizontal subsurface flow wetland planted with Vetiver grass mostly during rainy seasons. Restricting depth of wetland without considering rooting depth of Vetiver grass also contributed to clogging. Planting vegetation in rows resulted into wastewater flowing between the rows of Vetiver grass in the horizontal subsurface flow wetland when clogging occurred.

4.1 Introduction

Basic understanding of flow dynamics and environmental factors, and their interaction is important for the design and construction of a wetland. Moreover, when deciding on the materials and parameters to use for the construction of a wetland, certain considerations are critical. According to Metcalf (2003) and Patel et al. (2013), the principal data requirements for the design of a constructed wetland includes the expected influent volumes and concentrations (flow rate, BOD_5), the target effluent concentrations (hydraulic loading rates), climate (average daily precipitation and evapotranspiration) and substrate characteristics such as porosity and hydraulic conductivity. These factors greatly influence the hydraulic performance of any constructed wetland. In characterizing the hydraulic parameters of a wetland, the permeability of the substrate used is of great consideration in the design stage. Permeability, is the property that represents the ease with which water flows through a porous media (Salarashayeri and Siosemarde, 2012; Hunt and Manzoni, 2015). Grain size distribution of granular soils also affects permeability and it is characterized using the coefficient of uniformity (Cu) and coefficient of curvature (Cc) (Freeze and Cherry, 1979; Holtz et al., 2011; Wang et al., 2016).

Fluid viscosity is also an important parameter that determines internal resistance to flow. Read (2015) defines viscosity as a transport coefficient relating to transport of momentum. A higher viscosity corresponds to a thicker (more viscous) fluid, with the viscosities of semisolids and solids being the highest (Cooley and Gibson, 2016).Water having a low viscosity of 1.0 pascal seconds at 20 ^{o}C (Swindells et al., 1952; Cooley et al., 2016), flows easily through a porous substrate. Viscosity is highly dependent on temperature, with a higher temperature yielding more viscous gases and less viscous liquids (Elert, 2016; Malkovsky et al., 2016).

Vegetation in the wetland also affects the wastewater flow and hence plant density is of important consideration in the design of a wetland. They not only absorb the pollutants in wastewater but their roots also slow down water velocity thereby preventing water from taking preferential flow paths which can result into shorter retention time (Sehar et al., 2015; Sabokrouhiyeh et al., 2016).The flow rate assists in determining the size of wetland and the corresponding retention time. In the formula developed by Kickuth (1977), higher design flow results into larger surface area of the wetland bed. Likewise high water velocities in the wetland reduce the hydraulic residence time hence the longer water remains in the wetland, greater are the chances of higher sedimentation, adsorption, biotic processing and retention of nutrients (Wong et al., 2016; Tsang, 2016). As a management strategy, velocities can be kept low by regulating hydraulic loading rate, limiting the slope through the wetland, restricting outlet size and planting persistent emergent vegetation (Kadlec and Knight, 1996).

Constructed wetlands are broadly classified as free water surface wetlands and subsurface flow wetlands. The current study focused on the design and construction of horizontal, vertical and a hybrid subsurface flow constructed wetland systems with the purpose of polishing wastewater from the final maturation pond at Gusii wastewater treatment plant. The procedure used for the design was according to the UN-Habitat (2008) design manual.

4.2 Material and Methods

4.2.1 Study site

See chapter 3.0 section 3.1

4.2.2 Experimental Design and Treatments

The experimental design was a 3 by 2 factorial in a Completely Randomized Design (CRD) with six treatments replicated four times. The first factor are the three subsurface flow wetland systems (horizontal, vertical and hybrid system) and the second factor is whether the systems are planted with Vetiver grass or not resulting into the six treatments as follows:

i. Horizontal subsurface flow alone (HSSF)
ii. Vertical subsurface flow alone (VSSF)
iii. Hybrid subsurface flow system alone (HB)
iv. Horizontal subsurface flow + Vetiver grass (HSSF + VS)
v. Vertical subsurface flow + Vetiver grass (VSSF + VS)
vi. Hybrid subsurface flow system + Vetiver grass (HB + VS)

4.2.3 Layout for the experimental units

The experimental layout is summarized and presented in Table 4.1

Table 4.1: Layout of the experimental units

REP I	REP II	REP III	REP IV
VSSF	HSSF	HB	VSSF + VS
HB+VS	HB	HSSF+VS	HSSF
VSSF + VS	HSSF+VS	VSSF+VS	HB + VS
HSSF	HB + VS	HB + VS	HSSF + VS
HB	VSSF + VS	HSSF	VSSF
HSSF + VS	VSSF	VSSF	HB

REP: Replication, VSSF: Vertical subsurface flow wetland, HSSF: Horizontal subsurface flow wetland, HB: Hybrid system, VS: Vetiver grass

4.2.4 Sizing of the subsurface horizontal flow constructed wetland system

The bed surface area was obtained using Equation 4.1 as proposed by Kickuth (1977);

$$Ah = \frac{Qd(\ln Ci - \ln Ce)}{K_{BOD}} \qquad (4.1)$$

Where:

Ah = Surface area of bed (m^2)

Qd = average daily flow rate of wastewater (m^3/d)

Ci = influent BOD_5 concentration (mg/l)

Ce = effluent BOD_5 concentration (mg/l)

K_{BOD} = rate constant (m/d)

Recommended K_{BOD} value that was used was in the range of (0.067-0.1) as recommended by Cooper (1990) for better organics removal.

$$Average\ flowrate = \frac{(Inflowrate + Outflowrate)}{2} \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (4.2)$$

$$Outflowrate = Inflowrate + P - ET - I \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (4.3)$$

Where:

P= Precipitation (mm)

ETo= Reference Evapotranspiration (mm)

I = infiltration, which in this case was nil since the bed was lined with polythene of 0.3 mm thickness

The depth of 30cm was used for horizontal flow system. Horizontal subsurface flow wetland can have average depth of 27cm -50cm for more effective treatment according to Garcia et al. (2004) and 30-45cm according to (Steiner and Watson, 1993).

Length and width

To obtain length and width, an aspect ratio (L: W) of 4:1 was used. Aspect ratio of Length to Width of 4:1 is recommended for subsurface flow systems to avoid short-circuiting and to approach plug flow conditions (IWA, 2000). Length and Width was calculated as shown in Equation 4.4 and 4.5, respectively.

$$Length\ (L) = \frac{Bed\ surface\ area\ (Ah)}{Width\ (W)} \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (4.4)$$

$$Width(W) = \frac{Bed\ surface\ area\ (Ah)}{Length\ (L)} \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (4.5)$$

Hydraulic retention time was determined according to Equation 4.6

$$Hydraulic\ retention\ time(t) = \frac{nAd}{Q} \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (4.6)$$

Where:

n = porosity of the coarse sand (substrate) (%)

A = surface area of bed (m^2)

d = bed depth (m)

Q = the average daily flow rate (m^3/d)

Hydraulic loading rate was determined according to Equation 4.7

$$Hydraulic\ loading\ rate = \frac{average\ daily\ flow\ rate}{wetland\ surface\ area} \text{.......... ....}(4.7)$$

A slope of 0.5 to 1 % was used as is recommended for ease of construction and proper draining (UN-Habitat, 2008)

4.2.5 Sizing of the vertical flow constructed wetland

Bed surface area was determined from Equation 4.1 as proposed by Kickuth (1977), the hydraulic retention time, the hydraulic loading rate and slope was obtained using Equations 4.6 and 4.7 above, respectively.

Length and width was calculated as shown in Equation 4.4 and 4.5, respectively in section 4.2.4 in the sizing of horizontal flow constructed wetland.

Bed depth of 45cm was used for vertical flow wetland. Vertical subsurface flow wetland can have an average depth of 45cm-75cm for better nitrification (Philippi et al., 2006).

4.2.6 Sizing of the hybrid system

The hybrid system consisted of Vertical subsurface flow wetland (VSSF) linked to a horizontal subsurface flow wetland (HSSF). This is because in the VSSF, there is sufficient oxygen that nitrifies the ammonium in wastewater and in the horizontal system denitrification occurs since oxygen supply is limited. The size of each of the systems was as presented in the designs in sections 4.2.2 and 4.2.3 for the horizontal and vertical subsurface flow wetland systems, respectively.

4.2.7 Determination of the substrate hydraulic parameters

River sand from Kendu bay in Homa bay county and Sori in Migori county, Kenya was bought, transported, washed with clean water and sun dried. The dried sand was then used as substrate in the wetland systems. The hydraulic parameters that included soil porosity, hydraulic conductivity, coefficient of uniformity and specific gravity were determined before use. This processing was aimed at ensuring that the substrate selected met the required hydraulic conductivity that was free of clogging.

Hydraulic conductivity of the substrate sand was determined using the falling head permeability (permeametor) test according to BS1377 procedure for testing the permeability of granular soils (BSI, 2004). The coefficient of permeability was calculated from Equation 4.8 as:

$$K = \frac{2.3026\ a \times L}{A} \times \frac{Log_{10} H_1 - Log_{10} H_2}{t_2 - t_1} cm\sec^{-1} \text{.......... ....}(4.8)$$

Where:

K = Coefficient of permeability (cmsec^{-1})

a = Cross-sectional area of manometer tube (cm^2)

L = Length of sample under test (cm)

A= Cross sectional area of sample (cm^2)

H_1= initial height of water (cm)

H_2= head of water (in cm) indicated at the end of a particular period of time

t_2 = time corresponding to H_2 (sec)

t_1 = start time (sec)

2.3026 = conversion factor to log10

The soil porosity was determined according to BS1377 procedure (BSI, 2004) as shown in Equation 4.9

$$n = 1 - \frac{W_d}{VG\gamma_w} \text{.......... ....}(4.9)$$

Where:

n = soil porosity (%)

W_d = dry weight of soil sample (g)

V = volume of soil sampler (cm^3)

G = specific gravity of sand particles (dimensionless quantity)

Υ_w = unit weight of water (g/cm^3)

A gradation test was adapted for sieve analysis according to the procedure outlined by American Society for Testing and Materials (ASTM, 2006). The cumulative percent passing of the aggregate was determined and plotted against the sieve sizes. The graphs obtained were used in determining the coefficient of uniformity and coefficient of curvature of different sand aggregates.

The percent of particles retained in each sieve was then calculated using Equation 4.10

$$\%\,retained\ \ particles\,in\,each\,sieve\ \ = \frac{Wsieve}{Wtotal} \times 100\% \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots(4.10)$$

Where:

W_{sieve} = the weight of aggregate in the sieve (g)

W_{total} = the total weight of the aggregate (g)

The cumulative percent of aggregate retained in each sieve was obtained by adding up the total amount of aggregate that is retained in each sieve and the amount in the previous sieves.

The cumulative percent passing of the aggregate was found by subtracting the percent retained from 100% as shown in Equation 4.11

$$\%\,Cumulative\,aggregate\ passing = 100\% - \%\,Cumulative\,aggregate\,retained \ldots\ldots\ldots\ldots(4.11)$$

Coefficient of uniformity is a shape parameter and was calculated as given in Equation 4.12

$$Cu = \frac{D_{60}}{D_{10}} \qquad (4.12)$$

Where:

Cu = Coefficient of uniformity (dimensionless quantity)

D_{60} = Grain diameter at 60% passing (mm)

D_{10} = Grain diameter at 10% passing (mm)

Coefficient of curvature is a shape parameter and was calculated as given in Equation 4.13

$$Cc = \frac{(D_{30})^2}{D_{10} \times D_{60}} \qquad (4.13)$$

Where:

Cc = Coefficient of curvature /gradation (dimensionless quantity)

D_{30} = Grain diameter at 30% passing (mm)

D_{60} = Grain diameter at 60% passing (mm)

D_{10} = Grain diameter at 10% passing (mm)

Unified Soil Classification System modified from Airfield Classification system developed by Casagrande (Warren et al., 2015; Gambill et al., 2016) was used to grade the sand. The sand was classified as well graded when $C_u \geq 6$ & $1 < C_c < 3$. If both the criteria are not met then the sand was classified as poorly graded.

4.2.8 Water sampling and quality analysis

The water samples were collected at the inlet and outlets of the constructed wetland treatment systems planted with Vetiver grass and from the controls (unplanted) using one liter clean plastic bottles. Two sampling bottles were used for each treatment bi-weekly for duration of 6 weeks from 7th April to 19th May 2016. The samples were transported to the laboratory in cool boxes, filled with ice cubes to prevent deterioration and /or transformation of parameters. BOD_5 was determined using respirometric BOD OxiTop method that is based on pressure measurement in a closed system as described by Jouanneau et al. (2014).

4.2.9 Determination of BOD_5 Removal Efficiencies

Removal efficiencies of BOD_5 from the wetland systems were calculated as shown in Equation: 4.14

$$\mathrm{Re}\,moval Efficiency(\%) = \frac{Ci - Ce}{Ci} \times 100\% \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots(4.14)$$

Where:

Ci = Influent Concentration

Ce = Effluent Concentration

4.2.10 Statistical analysis

Data obtained for BOD_5, from the treatment systems were subjected to analysis of variance (ANOVA) at 5% level of significance using SPSS statistical software version 21.Means were separated using LSD test to determine if there were significant differences between treatment pairs.

4.3 Results and Discussion

4.3.1 Parameters of the horizontal, vertical and hybrid subsurface flow constructed wetland systems

Table 4.2 shows the design parameters of horizontal, vertical and hybrid subsurface flow wetland system that were used in the study.

Table 4.2: Design parameters of horizontal, vertical and hybrid subsurface flow wetland systems

Design Parameter	HSSF	VSSF	HYBRID(VSSF STAGE)	HYBRID(HSSF STAGE)
Wastewater type	Municipal wastewater	Municipal wastewater	Municipal wastewater	Municipal wastewater
Aspect ratio-length/width	4:1	4:1	4:1	4:1
Length	3.2	3.2	3.2	3.2
Width	0.8	0.8	0.8	0.8
Surface area(m^2)	2.845	2.845	2.845	2.845
Bed depth(m)	0.3	0.45	0.45	0.3
Wastewater flow rate(m^3d^{-1})	0.036(continuous flow)	0.018(two batches)	0.018(two batches)	-
Hydraulic retention time	8(days)	2 hours interval between batches	2 hours interval between batches	
Hydraulic loading rate(md^{-1})	0.013	0.0065	0.0065	
Vegetation Type	Vetiver grass	Vetiver grass	Vetiver grass	Vetiver grass

HSSF: Horizontal subsurface flow system, VSSF: Vertical subsurface flow system

The rooting depth of Vetiver grass was restricted to 0.3 m and 0.45 m in the horizontal and vertical subsurface flow system, respectively as proposed by Garcia et al. (2004) and Steiner et al. (1993) for effective treatment. However it was observed that clogging occurred in the horizontal subsurface flow wetland since wastewater had a tendency to follow rows between the Vetiver grasses. Limiting the depth of Vetiver grass to 30cm in the horizontal subsurface flow wetland could have caused the massive fibrous rooting system of Vetiver to interconnect with the adjacent roots of the Vetiver grasses. This could have trapped a lot of sediments in the subsurface which consequently could have reduced pore sizes that eventually caused the surfacing of wastewater between the rows of Vetiver grass. Yeboah et al. (2015) in a study on purification of industrial wastewater with Vetiver grass grown hydroponically on biogas wastewater observed that Vetiver root height reached 43cm within 90 days since planting. This observation is supported by Tanner et al. (1998) who observed that organic matter accumulation in the vegetated wetlands was 1300-3000 g/m^2/yr compared to the unvegetated wetlands that recorded 400-1600 g/m^2/yr. The authors concluded that the clogging that was experienced in the planted systems was as a result of the higher accumulation of organic matter in the subsurface.

However in the Vertical systems, the intermittent feeding could have caused turbulence that detached the trapped sediments which were then eliminated from the system.

The use of aspect ratio (4:1) to size the wetlands in this study instead of Darcy's Law was informed by the fact that Darcy's law is dependent on the reliability of the value of the hydraulic conductivity of the substrate. However according to Holtz et al. (2011), reliability of laboratory permeability test results depends on the quality of undisturbed soil samples collected in the field which is difficult to obtain for granular soils. According to Crites (1994), aspect ratio of 4:1 to 6:1 is suitable for design of wetlands to achieve high organic load reduction since sufficient surface area will be available for microorganisms to decompose the organic matter. This study achieved mean BOD_5 reduction of 75.8 and 80.9 % in the horizontal and vertical subsurface flow wetland planted with Vetiver grass. Lishenga et al. (2015) used an aspect ratio (3:1) to size horizontal subsurface flow wetland planted with Vetiver grass and achieved 75.12% BOD_5 reduction from domestic wastewater. Chen et al. (2006) used an aspect ratio (4:1) to size vertical subsurface flow wetland planted with *Phragmites communis* and achieved BOD_5 reduction of 89% from industrial wastewater. Klomjek et al. (2005) used aspect ratio (4:1) to size vertical subsurface flow planted with *Typha angustifolia* and achieved BOD_5 reduction of 74.3% from municipal wastewater. These results indicate that aspect ratio (4:1) is suitable for proper organics removal in a wetland.

4.3.2 Salient characteristics of river sand

Table 4.3 presents a summary of the characteristics of river sand from Kendu Bay and Sori.

Table 4.3: Characteristics of river sand from Kendu bay and Sori

Source	Porosity	Hydraulic Conductivity Cm/s	Specific Gravity	Coefficient Of uniformity	Coefficient of curvature	Silt content (%)
Kendu bay	0.343	2.766×10^{-3}	2.564	10.3	1.99	9.9
Sori	0.331	2.425×10^{-3}	2.551	48	2.82	16.2

The sand from Sori had higher uniformity coefficient (Cu) and coefficient of curvature (Cc) of 48 and 2.82, respectively compared to that from Kendu Bay that had a uniformity coefficient (Cu) and coefficient of curvature (Cc) of 10.3 and 1.99, respectively. The sand from both sources were classified as well graded according to unified soil classification systems (Warren et al., 2015; Gambill et al., 2016), since the test results fell within the criteria of $C_u \geq 6$ & $1 < C_c < 3$. Though sand from Sori was better graded, it had lower porosity and higher silt content of 33.1% and 16.2%, respectively compared to that from Kendu bay which had a higher porosity and lower silt content of 34.3% and 9.9%, respectively. These results indicate that the more the sand is well graded; the lower is its permeability which consequently lowers its suitability for use in a wetland. This could be attributed to the larger representation of fines in a well graded soil sample which consequently occupies available voids thus offers resistance to the easy flow of water through the soil. In a similar study, Onur (2014) observed that well graded soils have lower porosity since smaller grains tend to fill the voids between larger grains.

Clogging was experienced in the horizontal subsurface flow wetlands mostly during rainy seasons. This occurred despite selecting sand from Kendu bay to be used as the substrate in the wetland due its low silt content of 9.9% and high porosity of 34.3%. The massive fibrous rooting system of the Vetiver grass held the sand particles tightly together and this could have greatly reduced the porous nature. With pore size reduction, the flow of wastewater could have been restricted through the subsurface thereby causing surface overflow during rainy periods. Similar observation was reported by Aiello et al. (2016). The authors attributed it to development of biofilm and organic particle accumulation around the root zone which causes clogging in the subsurface flow wetland. These findings are further supported by studies conducted by George et al. (2000) who reported an estimated reduction of 2-8% in the void volume of coarse sand planted with vegetation which was much larger than reduction of 0.1-0.4% in the void volume in the unplanted substrate. Surfacing of wastewater was however not observed in the Vertical subsurface flow wetland planted with Vetiver grass. Intermittent flow in a vertical subsurface flow wetland, could have introduced turbulence thereby disturbing the sediments bound to the media in the constructed wetland.

In this study, sand from Kendu bay had a uniformity coefficient of 10.3. For the uniformity coefficient, the US EPA recommends a maximum Cu value of 4.0 (US EPA, 1993) for proper draining. According to Hwang et al. (2003), a larger uniformity coefficient implies that a wide range of particle sizes are well represented in a sand sample and hence the smaller particles fill in the voids consequently lowering the hydraulic conductivity. This could also explain the reason as to why water was observed on top of the substrate.

4.3.3 BOD_5 Removal

Table 4.4 presents the mean influent and effluent BOD_5 concentration for a period of 6 weeks from 7th April to 19th May 2016 when sampling and analysis was conducted.

Table 4.4: Mean influent and effluent BOD_5 concentration by various wetland units during the monitoring period

Treatment	Mean Influent BOD_5 Concentration (mg/L)	Mean Effluent BOD_5 concentration (mg/L)	Removal Efficiency (%)
HSSF + Vetiver grass	52.75	12.75^a	75.83
VSSF + Vetiver grass	52.75	10.03^b	80.99
Hybrid + Vetiver grass	52.75	7.07^c	86.60
HSSF (without grass)	52.75	22.75^d	56.87
VSSF(without grass)	52.75	19.50^e	63.03
Hybrid(without grass)	52.75	13.79^f	73.83

HSSF: Horizontal subsurface flow wetland, VSSF: Vertical subsurface flow wetland, HB: Hybrid subsurface flow wetland, Mean Effluent BOD_5 concentration with the same letter (a, b, c, d, e, f) in the same column are not significantly different at 5% confidence level.

Among the subsurface flow wetland systems planted with Vetiver grass, the hybrid system significantly ($p \leq 0.05$) achieved the lowest mean effluent BOD_5 of 7.07 mg/L followed by the vertical system at 10.03 mg/L .The horizontal system was at 12.75 mg/L. The more efficient polishing by the hybrid system could be attributed to the longer wastewater retention time in the coarse sand media at a length of 6.4 m compared to 3.2 m in the horizontal and vertical subsurface flow systems, thus allowing microorganisms more time to degrade organics. Five day biochemical oxygen demand (BOD_5) is the amount of oxygen required by microorganisms to degrade organic matter in wastewater within five days (APHA, 2005; Jouanneau et al., 2014). It therefore implies that if more organics are degraded in the wetland, the microorganisms in the effluent will demand less oxygen to decompose the remaining organic waste. Sirianuntapiboon et al. (2006) similarly observed BOD_5 reduction of 92±5% under longer hydraulic retention time of

3 days compared to 83±5% obtained at 0.75 day retention time. The authors attributed the better processes of organic solid biodegradation to the longer retention time in the wetland beds. Trang et al. (2010) also observed that at the highest hydraulic loading rate of 146mmday^{-1} which corresponded to 3 days retention time, the removal of BOD_5 was76±2% compared to 83±6% at 31mmday^{-1} corresponding to 13.9 days retention time. This occurred because as more wastewater was applied (hydraulic loading rate) the retention time decreased due to increasing water velocity thereby lowering contact time between microorganisms and wastewater.

However the significantly ($p \leq 0.05$) better polishing by the vertical compared to horizontal subsurface flow systems planted with Vetiver grass could be attributed to the intermittent feeding of wastewater in the vertical subsurface flow system. This could have created better aeration in the coarse sand media favorable for microbial decomposition of organics than in the horizontal subsurface flow system that was fed continuously with wastewater and hence saturated. The importance of oxygen level in the wetland is demonstrated by Boonsong and Chansiri (2008) who observed that the dissolved oxygen in the effluent from the system fed with highly concentrated wastewater (94.88 mg/l BOD_5) was lower at 0.96 mg/l compared to 1.45mg/l in the system fed with low concentrated wastewater (58.92 mg/L BOD_5). This was attributed to the consumption of more oxygen in aerobic decomposition of organic matter by microorganisms in the highly concentrated wastewater. Chandrakanth et al. (2016) observed similar results whereby 66.2 % BOD_5 removal was achieved in vertical subsurface flow wetland compared to 59.72% in horizontal subsurface flow wetland both at 5 hours retention time. The authors however attributed the better removal rates of BOD_5 in vertical subsurface flow wetland to the involvement of the total root zone as wastewater percolates downwards which increases the contact area of wastewater with the roots resulting into dominance of biological activity. Consequently as more organics are decomposed in the wetland the level for demand of oxygen by microorganisms in the effluent wastewater decreases.

On the side of the unplanted subsurface flow systems, the hybrid system again significantly ($p \leq 0.05$) achieved lowest mean effluent BOD_5 of 13.79 mg/L followed by vertical and horizontal system at 19.50 and 22.75 mg/L, respectively. The best performance of the hybrid system is as explained in the planted system above. Again the same argument provided in planted systems applies to better performance of vertical compared to horizontal sub-surface flow system.

The planted systems achieved significantly ($p \leq 0.05$) lower mean effluent BOD_5 compared to the unplanted systems with the planted hybrid system being the most efficient in BOD_5 removal. The planted hybrid system achieved lower mean effluent BOD_5 of 7.07 mg/L compared to the unplanted hybrid system at 13.79 mg/L and this could be attributed to the uptake of organic matter by Vetiver grass. When more organic matter is utilized the demand for oxygen by microorganisms to decompose the remaining organic matter in the effluent wastewater is reduced hence the low BOD_5 value in the planted system. The higher uptake of nutrients by Vetiver grass in wastewater is demonstrated by Mudhiriza et al. (2008) who observed that the average dry mass of Vetiver grass tillers significantly ($p \leq 0.05$) increased from 8.9g at the start of the experiment to 26.5g on the 21st day of effluent retention under Vetiver grass. Zhao et al. (2014) similarly observed that of all the wetland plants under study (Giant reed, Vetiver grass, Green umbrella plant, Alligator flag and Canna), Vetiver grass had significantly the highest leaf biomass of $1.57 kgm^{-2}$. The authors attributed the higher biomass of Vetiver grass to its herbal properties and longer duration of green leaves for photosynthesis as it utilized nutrients in wastewater.

The reduction of organic load in the unplanted hybrid system though lower than in the planted however shows that the coarse sand media in this study could have contributed to BOD_5 reduction by providing a good habitat for microorganisms to proliferate and degrade organic matter in wastewater. Calheiros et al. (2009) in a study on changes in the bacterial community structure in horizontal flow constructed wetland system planted with *Typha latifolia* and *Phragmites australis* for treating tannery wastewater observed that bacterial counts from roots and substrate (clay aggregates) samples of each unit were not significantly different. This is an indication that the substrate offers habitat for the microorganisms. Soric et al. (2011) further demonstrated the significance of wetland media. They observed that after two weeks, the effluent total organic carbon was lower at 57 ± 6 mg/l in the column filled with plastic beads as media compared to column filled with glass beads at76 ±8 mg/l. The authors attributed this observation to higher biofilm development in the plastic beads and thus concluded that metabolic pathways are influenced by the porous media dedicated to biofilm growth.

The planted vertical system also achieved lower mean effluent BOD_5 of 10.03 mg/L compared to the unplanted vertical system at 19.50 mg/L. The fibrous rooting system of the Vetiver grass in the planted vertical system could have reduced the flow rate of wastewater through the coarse sand media thereby giving micro-organisms ample time to degrade the organic matter in wastewater. With reduction in organic matter, less oxygen was thus required by microorganisms in the effluent wastewater to degrade the remaining organics hence the low BOD_5 value. The planted horizontal system also achieved lower mean effluent BOD_5 of 12.75 mg/L compared to the unplanted horizontal system at 22.75 mg/L. Since micro-organisms contribute to the degradation of organic matter in wastewater (Hijosa-Valsero et al., 2010; Wang et al., 2012), the massive fibrous rooting system of Vetiver grass could have increased the surface area for their attachment consequently improving the performance of the planted system. Li et al. (2010) observed that 78.9% of the clones affiliated with *Proteobacteria* which plays important roles in the metabolism of organic compounds were attached in the roots. This indicates that the roots provide significant support and shelter to the micro-biota involved in the transformation of organic pollutants. Gagnon et al. (2007) in a study on the influence of macrophyte species (*Phalaris arundinacea*, *Phragmites australis* and *Typha angustifolia*) on microbial density and activity in constructed wetlands made a similar observation. The authors observed that *Phalaris* which had significantly the highest root surface area had the greatest density of aerobic and facultative bacteria on the root surface suggesting root oxygen release required for metabolism.

4.4 Conclusions

The permeability of coarse sand with Vetiver grass as wetland plants was reduced thereby causing clogging in the horizontal subsurface flow wetland system. In selecting coarse sand to be used as the media in a subsurface flow wetland system, the uniformity coefficient which indicates the particle size distribution should be the most important parameter to be considered rather than relying on the porosity values. Being shallow water systems, the constructed wetlands are susceptible to pore filling in by incoming sediments consequently reducing the porosity. Using the first order model developed by Kickuth resulted into significantly higher BOD_5

removal of 75.83, 80.9 and 86.6% in the planted horizontal, vertical and hybrid subsurface flow wetland, respectively.

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CHAPTER FIVE

EFFECTIVENESS OF THE HORIZONTAL, VERTICAL AND HYBRID SUBSURFACE FLOW CONSTRUCTED WETLAND SYSTEMS IN POLISHING EFFLUENT IN THE GUSII TREATMENT WORKS

Abstract

Protection of fresh water resources against pollution from wastewater is important to achieve water security. This study aimed at comparing the performance of horizontal, vertical and hybrid subsurface flow system in polishing wastewater effluent from the maturation pond at Gusii wastewater treatment plant. The treatments were monitored for six weeks duration for chemical oxygen demand, total suspended solids, total nitrogen and total phosphorous against Kenya's National Environmental Management Authority standards for effluent discharge. Constructed systems planted with Vetiver grass performed significantly ($P \leq 0.05$) better compared to the others in pollutants removal. Among the systems planted with Vetiver grass, the hybrid subsurface flow system significantly removed the pollutants more efficiently than the single operated systems. The Vetiver planted hybrid subsurface flow wetland systems achieved the highest removal of COD, TN, TP and TSS at 82.4, 87.9, 65 and 94.6%, respectively compared to the others. The planted vertical subsurface flow removed COD, TN and TP at 72.9, 75.7, and 50.7%, respectively more efficiently than the horizontal subsurface flow system that achieved removal of COD, TN and TP at 65.3, 70.0 and 43.8%, respectively. The planted horizontal subsurface flow wetland however showed better TSS removal at 89.9% compared to 83.2% achieved by vertical subsurface flow system. The unplanted systems exhibited a similar trend whereby the hybrid subsurface flow systems achieved better performance than the single systems though with lower organics and nutrients removal efficiencies compared to planted systems. The unplanted hybrid subsurface flow wetland systems achieved the highest removal of COD, TN, TP and TSS at 66.0, 61.4, 55.2 and 83.4%, respectively as compared to other unplanted constructed wetland systems. The unplanted vertical subsurface flow removed COD, TN and TP at 52.5, 51.7 and 35.9%, respectively more efficiently than horizontal subsurface flow system that achieved removal of COD, TN and TP at 46.5, 33.3 and 32% , respectively. The unplanted horizontal subsurface flow wetland however showed better TSS removal at 79.4% compared to73.6% achieved by unplanted vertical subsurface flow system.

5.1 Introduction

Demand for fresh water resources is expected to rise with the growing global population yet this precious resource is under constant threat of pollution. Although there are natural causes, much of the eutrophication seen currently is a result of inadequately treated wastewater and agricultural run-off that end up in receiving water bodies (Cai et al., 2013). Adequate treatment of wastewater for reuse will therefore be a viable option in ameliorating the challenge of water scarcity and environmental degradation. Many industries in developing countries use conventional wastewater treatment systems to treat their wastewater before release into the environment (Konnerup et al., 2011; Zhang et al., 2014). However, these conventional treatment technologies have been found to be either ineffective, wasteful and/ or costly (Nhapi, 2004). In Kenya, the National Environment Management Authority (NEMA) has set guidelines on the permissible effluent discharge limits into the environment and these standards are rarely met by the conventional treatment methods used. Adoption of low cost and effective technologies such as phyto-remediation will therefore be a suitable option for many industries and households involved in wastewater treatment.

Constructed wetlands are considered to be the best choice to treat wastewater since they are economical and effective in pollutants removal (William, 1999; Mthembu et al., 2013). Vegetation plays a critical role in the performance of constructed wetlands and hence selection of the most efficient vegetation type is important. The vegetation not only absorb pollutants from wastewater but their roots prevent wastewater from taking preferential paths in the substrate that can result to hydraulic short circuiting which would consequently reduce the retention time in the wetlands (Stottmeisteret al., 2003; Sehar et al., 2015). The roots also provide a large surface area for attachment of micro-organisms that degrade the organics in the wastewater (Wu et al., 2014; Yuan et al., 2016). The use of aquatic plants is thus becoming increasingly common in wastewater management as it integrates treatment, recycling and re-use (Lishenga et al., 2015). Many studies have used different macrophytes such as, *Typha angustifolia* (Li et al., 2016), *Phragmites australis* (Bhatia et al., 2016), *Cyperus papyrus* (Kipasikaet et al., 2016), *Typha orientalis* (Wang et al., 2016), *Iris australis* (Lv et al., 2016), *Scirpus grossus* (Tangahu et al., 2016), *Canna iridiflora* (Weragoda et al., 2012) for industrial or domestic wastewater treatment with varying success.

Vetiver grass (*Chrysopogon zizanioides)* has gained wider acceptance in wastewater treatment due to its ability to thrive in unfavourable environments. Vetiver grass can tolerate a wide range of pH, salinity, sodicity, acidity and heavy metals (Chomchalow, 2000; Vimala and Kataria, 2005; Raude et al., 2009).

In many cases, Vetiver grass has been used to clean up many kinds of pollutants including metals, pesticides, oils and organic contaminants from wastewater (Minh et al., 2015;Kamtekar and Verma, 2016; Darajeh et al., 2016; Mathew et al., 2016). According to US EPA (2012), Vetiver grass eliminates several kinds of pollutants by completely destroying or converting them to carbon dioxide and water rather than simply immobilizing or storing them.

5.2 Materials and Methods

5.2.1 Study site

See chapter 3.0 section 3.1

5.2.2 Water sampling and quality analysis

The water samples were collected at the inlet and outlets of the constructed wetland treatment systems planted with Vetiver grass and from the controls (unplanted) using one liter clean plastic bottles. Bi-weekly sampling for each treatment for duration of 6 weeks from 7th April to 19th May 2016 was done in duplicates. The samples were transported to the laboratory in cool boxes filled with ice cubes to prevent deterioration and /or transformation of parameters.

Water quality parameters i.e. COD, TSS, TN and TP were determined according to the Standard Methods for the Examination of Water and Wastewater (APHA, 2005). Chemical oxygen demand (COD) was determined using the closed reflux titrimetric method as described by Ademoroti (1996) with potassium dichromate in sulphuric acid as oxidation reagent. Total suspended solids (TSS) was determined using the filtration method as described in ASTM (2007) procedure whereby filters (whatman glass fibre filter) of 1.58mm was used for filtration and then oven dried at 105^{0}C for 24 hours. The dry weight of the solids retained was divided by filtered volume to obtain mg/L of TSS.

The total nitrogen (TN) was determined using cadmium reduction method as described by Campbell et al. (2006). Total phosphorous (TP) was determined using Calorimetric Ascorbic acid method as described by Eaton et al. (2005). Other Parameters such as pH, temperature, electrical conductivity and total dissolved solids were determined in-situ using multimeter probe.

5.2.3 Determination of Pollutants Removal efficiencies

Removal efficiencies of pollutants from the wetland systems were calculated as shown in Equation: 5.1

$$\mathrm{Re}\,moval Efficiency(\%) = \frac{Ci - Ce}{Ci} \times 100\% \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots(5.1)$$

Where:

Ci = Influent Concentration

Ce = Effluent Concentration

5.2.4 Statistical analysis

Data obtained for BOD_5, COD, TSS, TN and TP from the constructed wetland treatment systems were subjected to analysis of variance (ANOVA) at 5% level of significance using SPSS statistical software version 21. Means were separated using LSD test to determine if there were significant differences between treatments.

5.3 Results and Discussions

5.3.1 Effluent and Influent Characterization

5.3.1.1 COD Removal

Effluent and influent COD concentrations for all the wetland units analyzed during the monitoring period are presented in Figure 5.1.

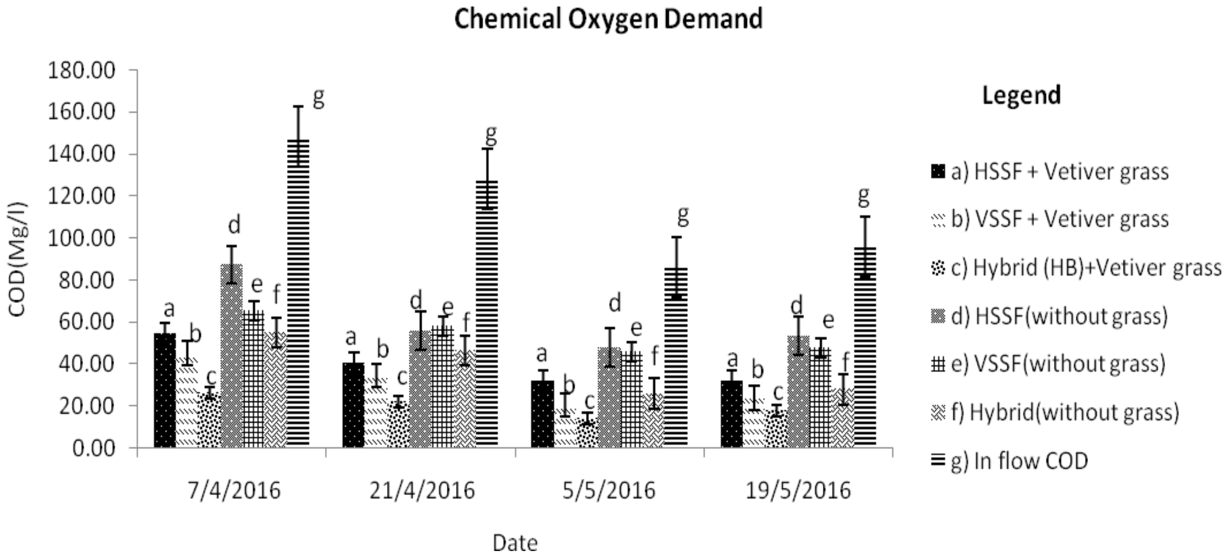

Figure 5.1: Effluent and influent COD for all the wetland units during the monitoring period

Among the subsurface flow wetland systems planted with Vetiver grass, the hybrid system achieved significantly ($p \leq 0.05$) the lowest mean effluent COD of 20.19 mg/L followed by the vertical system at 31.06 mg/L. The horizontal system was highest at 39.75 mg/L. The more efficient polishing by the hybrid system could be attributed to the longer wastewater retention in the coarse sand media at a length of 6.4 m compared to 3.2 m length in the horizontal and vertical subsurface flow systems, thus hybrid system allowed microorganism's ample time to degrade organics. Chemical oxygen demand is the amount of oxygen required to chemically oxidize the organic matter in wastewater (Ademoroti, 1996; APHA, 2005). It therefore means that if more organic matter is used or degraded in the wetland, there will be less oxygen requirement to chemically degrade the remaining organics in the effluent. Deblina and Brij (2010) similarly observed that the higher retention time of 4 days helped achieve maximum removal of COD at 85% compared to 45% at 1 day retention time. Ewemoje et al. (2015) also observed that removal efficiency of COD increased with retention time. The authors obtained 84, 92.4 and 95.3% COD removal at 3, 5 and 7 days retention time, respectively and attributed it to better contact time for microbial degradation of organic matter.

However the significantly ($p \leq 0.05$) better performance of the vertical than the horizontal subsurface flow systems planted with Vetiver grass could be due to the intermittent feeding of wastewater in the vertical system that created better aeration as opposed to the horizontal system that is fed continuously and hence always saturated. This increased the oxygen content in the wastewater required by microorganisms to degrade the organics thereby lowering the amount of oxygen required to chemically oxidize organic matter in the effluent. Pan et al. (2012) in a study on full-scale experiment on domestic wastewater treatment by vertical- and horizontal-flow constructed wetlands system observed effluent COD from vertical system was significantly lower at 30.9 mg/L compared to 33.2 mg/L in the horizontal system. This was associated with the initial increased oxygen level in the vertical system that promoted aerobic degradation of organic matter thereby decreasing the oxygen requirement in the effluent to chemically degrade the remaining portion. The significance of oxygen in wetland performance is further demonstrated by Ong et al. (2011) in a study on treatment of textile wastewater in aerated and non aerated wetland reactors where COD removal of 95 and 62%, respectively was observed. The authors noted that aerobic conditions facilitated the growth and proliferation of aerobic microbes which enhanced biodegradation of organic matters. Studies by Boonsong and Chansiri (2008) give further support as they observed dissolved oxygen in the effluent from the system fed with highly concentrated wastewater was lower at 0.96 mg/l compared to 1.45mg/l in the low concentrated wastewater. They attributed this to the consumption of oxygen in aerobic decomposition of organic matter by microorganisms.

On the other hand, of the unplanted subsurface flow systems, the hybrid system achieved significantly ($p \leq 0.05$) lowest mean effluent COD of 38.91 mg/L followed by vertical and horizontal system at 54.38 and 61.25 mg/L, respectively. The best performance of the hybrid system is on retention capacity as explained in the planted system above. Again the same argument provided in planted systems applies to better performance of vertical compared to horizontal sub-surface flow system.

The planted systems achieved significantly ($p \leq 0.05$) lower mean effluent COD compared to the unplanted systems with the planted hybrid system being the best in COD removal. The planted hybrid system achieved lower mean effluent COD of 20.19 mg/L compared to the unplanted hybrid system at 38.91 mg/L as Vetiver grass played a significant role in utilizing nutrients such N and P from wastewater. Lin et al. (2008) observed that the total biomass of Vetiver grass planted on gravel media significantly increased from 26±0 g at the start of the experiment to 352±33g after 35 days and attributed it to nutrient uptake for biomass yield. The good performance of the unplanted hybrid system however shows that the coarse sand media could have also contributed to COD reduction by providing good environmental conditions for microorganisms to proliferate and degrade organics in wastewater. The data obtained indicate that planted vertical system also achieved lower mean effluent COD of 31.06 mg/L compared to the unplanted vertical system at 54.38 mg/L. This could be attributed to the massive rooting system from the vetiver providing a larger surface area for microbial attachment, which consequently degraded the organic matter. Gagnon et al. (2007) observed a bacterial density ratio of 10.3 between planted and unplanted wetlands. They attributed this to micro aerobic environment in the rhizosphere of plants that is suitable for microbial species growth and diversity that digests organic matter. Njau and Mlay (2003) similarly observed that significant reduction of organic load was achieved in planted wetlands with Vetiver grass compared to the unplanted wetlands indicating that aquatic plants support the organic level reduction processes by availing atmospheric oxygen in their submerged stems, roots and tubers, which is then utilized by the microbial decomposers attached to them below the level of the water to digest the organic matter in wastewater

Similarly, the planted horizontal system achieved lower mean effluent COD of 39.75 mg/L compared to the unplanted horizontal system at 61.25 mg/L. The fibrous rooting system of the Vetiver grass could have reduced the flow rates of wastewater through the substrate thereby increasing the time for the microorganisms to degrade the organics in wastewater.

The significant ($p \leq 0.05$) variations observed in the influent COD concentrations throughout the monitoring period could be attributed to varying environmental factors in the waste stabilization ponds from which the wastewater originated. Alamgir et al. (2016) in a study on algal growth and waste stabilization ponds performance observed months with highest sunshine hours had higher amount of dissolved oxygen (DO) level in the effluent compared to those with shortest sunshine hours. The authors indicated that longer sunshine hours enhanced algal photosynthetic activities thus releasing oxygen required for organics decomposition by microbes in the ponds. Wallace et al. (2015) observed that high levels of floating green algae were present throughout the monitoring period in summer when temperatures were higher , with gradual die-off occurring as temperature decreased in spring. This may explain further the influence of varying temperature in the growth and consequently performance of algae in nutrients removal in waste stabilization ponds. Bartosh and Banks (2007) also observed the growth rates of both algae species (*C. vulgaris* and *S. subspicatus*) increased with increasing temperature and light intensity with growth ceasing at temperatures close to 0°C.

Table 5.1 presents the performance of the wetland systems in COD removal compared to the permissible limit according to Kenya's NEMA standards.

Table 5.1: Mean effluent COD concentration against NEMA standards

Treatment	Mean Influent COD Concentration (mg/L)	Mean Effluent COD concentration (mg/L)	Removal Efficiency (%)	NEMA Standards (mg/L) (Max)	Remarks Standards met (Yes or No)
HSSF + Vetiver grass	114.5	39.75	65.28	50	Yes
VSSF + Vetiver grass	114.5	31.06	72.87	50	Yes
Hybrid + Vetiver grass	114.5	20.19	82.37	50	Yes
HSSF (without grass)	114.5	61.25	46.51	50	No
VSSF(without grass)	114.5	54.38	52.51	50	No
Hybrid(without grass)	114.5	38.91	66.02	50	Yes

HSSF: Horizontal subsurface flow wetland, VSSF: Vertical subsurface flow wetland, HB: Hybrid subsurface flow wetland

The mean influent and effluent COD concentration presented are for a period of 6 weeks study from 7th April to 19th May 2016 when sampling and analysis was conducted. Levels of effluent COD achieved by all the constructed wetland systems planted with Vetiver grass in the study met the standards of maximum 30mg/L stipulated by the Kenya's National Environmental Management Authority (1999) for wastewater discharges into water or on land. However among the unplanted systems, only the hybrid system achieved the required effluent standards.

5.3.1.2 Nitrogen Removal

Figure 5.2 presents the total nitrogen (TN) concentrations in effluent and influent for all the units during the monitoring period.

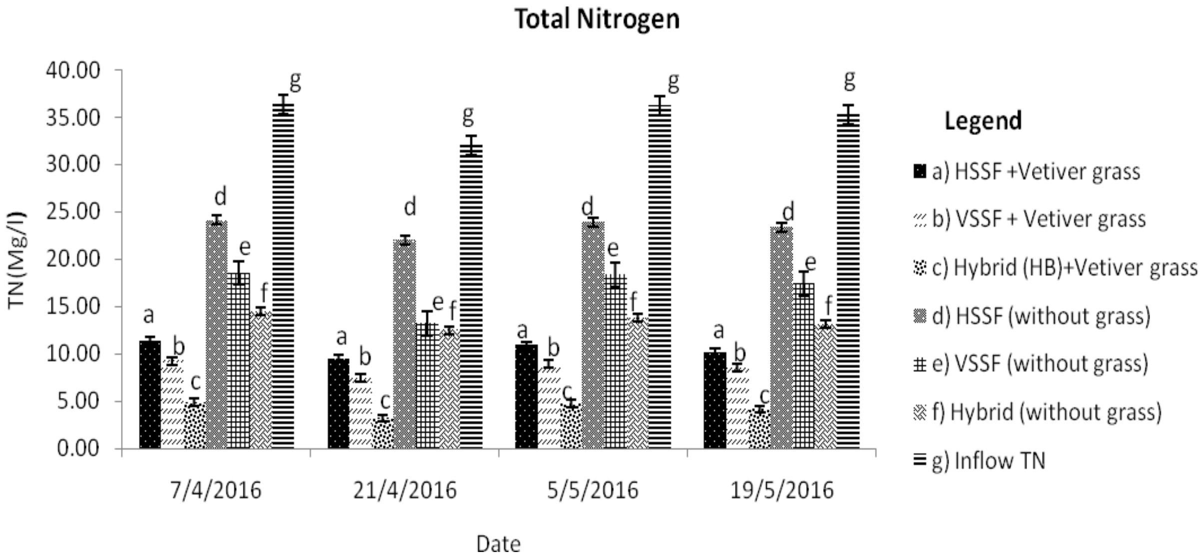

Figure 5.2: Effluent and influent Total Nitrogen for all the wetland units during the monitoring period

Among the subsurface flow wetland systems planted with Vetiver grass, the hybrid system achieved significantly ($p \leq 0.05$) the lowest mean effluent of TN at 4.23 mg/L followed by the vertical system at 8.51 mg/L. The TN in horizontal system was at 10.48 mg/L. The first stage of the hybrid system consists of the vertical subsurface flow system which is aerated due to intermittent feeding of wastewater. This promotes the conversion of ammonium in wastewater to nitrates by Nitosomonas bacteria (Fan et al., 2013; Yan et al., 2016) and the nitrates formed easily taken up by Vetiver grass (Billore et al., 2002; Njau and Mlay, 2003). As wastewater flowed to the next stage of the hybrid system which consist of horizontal subsurface flow system, anaerobic conditions dominates as it is always saturated with wastewater. This in turn promotes reduction of the nitrates by chemo-autrotrophic bacteria to gaseous forms of nitrogen (nitric oxide, nitrous oxide and dinitrogen) (Saeed and Sun, 2012; Vymazal, 2007) which greatly reduced the effluent TN levels. Zhang et al. (2015) observed that hybrid system achieved 75.4% TN removal compared to vertical and horizontal system at 53.35% and 50.3%, respectively. The authors attributed the better performance of the hybrid system to its ability to provide both aerobic and anaerobic conditions simultaneously for multipurpose microorganisms. Similarly Vymazal (2007) observed that hybrid constructed wetlands are primarily used for enhanced TN removal because the various types of wetland environments provide different redox conditions suitable for nitrification and denitrification processes.

The significantly ($p \leq 0.05$) better performance of the vertical subsurface flow system planted with Vetiver grass as compared to horizontal subsurface flow system planted with Vetiver grass could be due to their better aeration facilitated by the intermittent feeding of wastewater. This promoted conversion of ammonia to nitrates (Wu et al., 2016) that are easily taken up by plants. Plant uptake of N is one of the key processes of its removal from wetlands (Billore et al., 2002; Shivhare and Roy, 2013). Similar observations were noted by Wu et al. (2015) and Pan et al. (2012) that batch feeding greatly reduced TN in wastewater due to enhanced nitrification.
Of the unplanted subsurface flow systems, the hybrid system achieved significantly ($p \leq 0.05$) lowest mean effluent TN load of 13.48 mg/L followed by vertical and horizontal system at 16.87 and 23.31 mg/L, respectively. The efficiency of the hybrid system could be attributed to the longer wastewater retention in the coarse sand media at a length of 6.4 m compared to 3.2 m in the horizontal and vertical subsurface flow systems, allowing microorganisms time to degrade organics. Bioaloweic et al. (2011) observed 59.5% of N removal occurred through microbiological processes in the gravel used as substrate while volatilization and plant uptake accounted for only 13 and 15%, respectively. Coarse sand thus acts as a habitat for microorganism communities who assist in effectively removing nitrogen from contaminated water for their physiological need. Significance of substrate in nitrogen removal is further demonstrated in a study by Kantawanichkul et al. (2013) who noted that altering the media from sand to gravel decreased nitrogen removal efficiency from 65 to 46.8%. The authors indicated that sand particles have a larger surface area to support microorganisms and provide a longer retention time for biological processes such as nitrification and denitrification.

However the significantly ($p \leq 0.05$) better polishing efficiency by vertical than the horizontal unplanted subsurface flow systems could be due to the influence of better aeration on nitrogen removal as explained above in the planted systems. Cottingham et al. (1999) noted that prior to aeration, the NH_4^+-N removal rate was 18% but after aeration, the rate increased to 68% thus concluding that high removal was due to increased nitrification activity and the NO_3 subsequent utilization by the plants.

Jamieson et al. (2003) also observed that the introduction of aeration to a pilot scale constructed wetland model improved the mean ammonia-nitrogen removal efficiency from 50.5 to 93.3%, following a 2 week lag phase. Increased removal was primarily attributed to increased nitrification indicating continual aeration has great potential to enhance nitrification in constructed wetlands and NO_3 formed used as nutrients by the macrophytes thereby reducing TN level in wastewater.

The planted systems had significantly ($p \leq 0.05$) lower mean effluent TN compared to the unplanted systems with the planted hybrid system being the best in TN removal. The planted hybrid system achieved lower mean effluent TN of 4.23 mg/L compared to the unplanted hybrid system at 13.48 mg/L. Despite these two systems having similar dimensions as well as substrate and wastewater flow rates; the difference in performance is an indication that Vetiver grass played a significant role through nutrient uptake from wastewater. Mairi et al. (2012) observed performance efficiency of nitrogenous chemical removal was greatly increased by macrophytes absorption as percent NO_3 removal averaged 58.1% for planted cells and 21.6% for unplanted cells in his study. Similar observations were made by Bioaloweic at al. (2011) whereby plant uptake accounted for 15% nitrogen removal in a vertical flow wetland.

The planted vertical system mean effluent TN of 8.51 mg/L was significantly lower compared to 16.87 mg/L in the unplanted vertical system. Chang et al. (2013) observed that plants enhance nitrate removal by plant assimilation which accounted for 2-10% of removal efficiency. Tanner et al. (2012) noted that vegetation increases aeration within the constructed wetland system hence assisting nitrification process and the nitrates released taken up by the vegetation.

Vetiver planted horizontal system achieved significantly lower mean effluent TN of 10.48 mg/L compared to the unplanted horizontal system at 23.31 mg/L. This could also be attributed to the uptake of nutrients from wastewater as explained in the case of the planted hybrid and vertical system above. According to Njau and Mlay (2003), aquatic plants support the organic level reduction processes by availing atmospheric oxygen in their submerged stems, roots and tubers, which is then utilized by the microbial decomposers attached to them below the level of water to digest organic matter in wastewater which possibly contain nitrogenous compound. Additionaly the availed oxygen could have favoured the conversion of ammonium to nitrates which is then utilized by Vetiver grass as explained in the case of planted vertical and hybrid system.

The influent TN concentration was observed to significantly ($p \leq 0.05$) vary throughout the monitoring period and was attributed to the varying environmental factors such as light intensity and temperature. Rockne and Brezonik (2006) observed significantly low ammonium concentration in the effluent during warmer periods compared to colder periods. They attributed this to rapid uptake of ammonium by the growing algae coupled with volatilization of any residual ammonia at higher temperatures. Similarly, Maynard et al. (1999) observed that at higher temperatures and higher pH (>10), ammonia volatilization was the main nitrogen removal mechanisms.

Table 5.2 shows the mean effluent TN concentration values from the constructed wetlands compared to the permissible limit according to Kenya's NEMA.

Table 5.2: Mean effluent TN concentration against NEMA standards

Treatment	Mean Influent TN Concentration (mg/L)	Mean Effluent TN concentration (mg/L)	Removal Efficiency (%)	NEMA standards (mg/L) (Max)	Remarks Standards met (Yes or No)
HSSF + Vetiver grass	34.95	10.48	70.01	2	No
VSSF + Vetiver grass	34.95	8.51	75.65	2	No
Hybrid + Vetiver grass	34.95	4.23	87.89	2	No
HSSF (without grass)	34.95	23.31	33.3	2	No
VSSF(without grass)	34.95	16.87	51.73	2	No
Hybrid(without grass)	34.95	13.48	61.43	2	No

HSSF= Horizontal subsurface flow wetland, VSSF= Vertical subsurface flow wetland, HB: Hybrid subsurface flow wetland, TN= Total Nitrogen

The mean influent and effluent TN concentration presented are for a period of 6 weeks from 7th April to 19th May 2016 when sampling and analysis was conducted. Levels of effluent TN achieved by all the constructed wetland systems in this research did not meet the standards of maximum 2mg/L stipulated by the Kenya's NEMA (1999) for wastewater discharges into water or on land.

5.3.1.3 Total Phosphorous Removal

Figure 5.3 presents the total phosphorous (TP) concentrations in effluent and influent analyzed in the wetland units during the monitoring period.

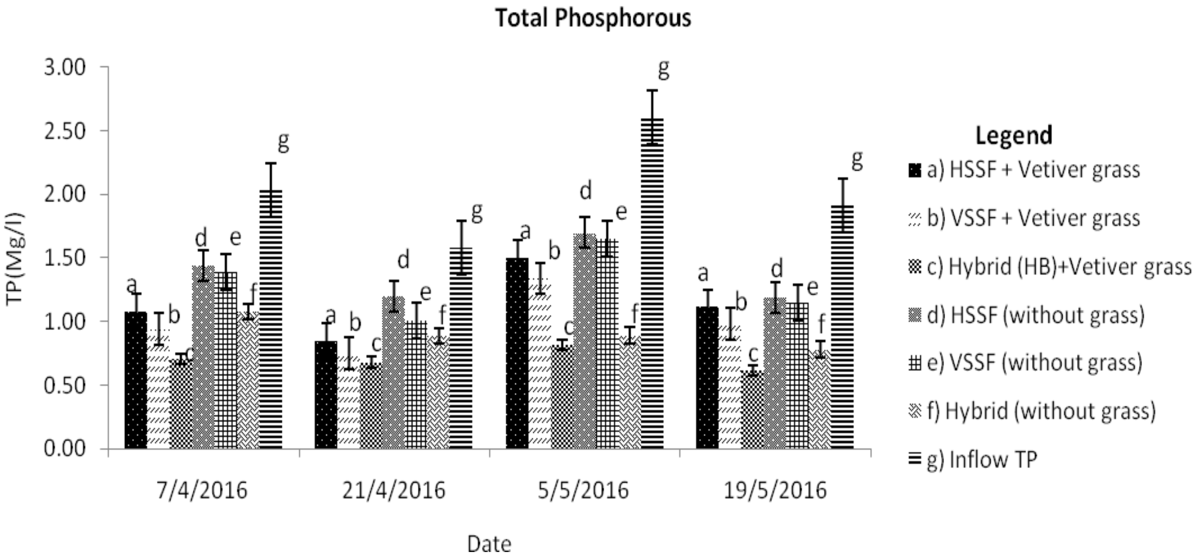

Figure 5.3: Total Phosphorous content in effluent and influent in the wetland units during the monitoring period

Among the subsurface flow wetland systems planted with Vetiver grass, the hybrid system achieved significantly ($p \leq 0.05$) the lowest mean TP of 0.71 mg/L in the effluent followed by the vertical at 1.00 mg/L and finally at 1.14 mg/L in the horizontal system. The best performance of the hybrid system could be attributed to the longer wastewater retention in the substrate compared to the horizontal and vertical subsurface flow systems, which allowed the coarse sand media to adsorb more phosphorous. Adsorption of phosphorous has been reported to be the major mechanism of its removal from wetlands. Njau et al. (2003) using pumice as a substrate to adsorb P from wastewater observed that 39% of all dissolved P was removed via sorption to the pumice soil substrate. Ayoub et al. (2001) using sand coated in iron aluminium hydroxide observed that 70% of P was adsorbed to the coarse sand.

The significant ($p \leq 0.05$) better performance of the planted vertical subsurface flow system compared to planted horizontal system could be due to the saturated conditions in the horizontal system that inhibits microbial decomposition of organic matter containing P as opposed to well aerated conditions in the vertical system.

Forbes et al. (2009) observed the wetland that was intermittently fed with wastewater achieved the highest removal rate of P compared to those operated on a continuous flow. This suggests that the intermittent dosing improved dissolved P removal, perhaps by higher iron-P precipitation rates occurring under oxidized conditions. According to Tang et al. (2009) during the loading period in a vertical subsurface flow wetland, air is forced out of the soil and during the percolation phase, the surface soil dries out drawing air back into the soil pore spaces consequently providing alternating oxidizing/reducing conditions in the soil thus promoting P adsorption.

Of the unplanted subsurface flow systems, the hybrid significantly ($p \leq 0.05$) achieved lowest mean effluent TP of 0.9 mg/L followed by vertical and horizontal system at 1.30 and 1.38 mg/L, respectively. The best performance of the hybrid system is as explained in the planted system above. The same argument provided in planted systems applies to better efficiency of vertical compared to horizontal sub-surface flow system.

The planted systems achieved significantly ($p \leq 0.05$) lower mean effluent TP compared to the unplanted systems with the planted hybrid system being the best in TP removal. The planted hybrid system achieved lower mean effluent TP of 0.71 mg/L compared to the unplanted hybrid system at 0.9 mg/L and was attributed to the utilization of nutrients from wastewater by Vetiver grass in the wetland. Lishenga et al. (2015) observed that soil based vetiver system achieved 32.9% TP removal efficiency compared to the unplanted system at 14.85%. This was because Vetiver grass absorb phosphate-P and their roots slow down water velocity thereby increasing TP removal through sedimentation as organic phosphorous. Mng'anya et al. (2000) observed that Vetiver grass contributed about 3% in phosphorous removal from the wetland through uptake.

The planted vertical system also achieved lower mean effluent TP of 1.00 mg/L compared to the unplanted vertical system at 1.30 mg/L. Yeboah et al. (2015) in a study on purification of industrial wastewater with Vetiver grass grown hydroponically on palm oil mill effluent observed that phosphate level was reduced from 10.5mg/l to 1.62mg/l corresponding to 84.57% reduction. This was attributed to Vetiver's high affinity for phosphate for its root development.

According to Hoffman et al. (2011), phosphorus removal can be achieved in constructed wetlands by adsorption and precipitation, and a small amount is also taken up by plant growth. Similarly, the planted horizontal system achieved lower mean effluent TP of 1.14 mg/L compared to the unplanted horizontal system at 1.38 mg/L. This is further attributed to uptake of phosphates by Vetiver grass as explained in the planted hybrid and vertical system.

The influent TP concentration was observed to significantly ($p \leq 0.05$) vary throughout the monitoring period due to the varying environmental factors that influence treatment performance of waste stabilization ponds. Richmond (2004) reported that phosphate content of algal dry biomass grown in wastewater could reach up to 3.3%. However the rate of utilization of nutrients by algae during photosynthesis is a function of light intensity and temperature which varies both diurnally and seasonally (Bartosh and Banks, 2007; Alamgir et al., 2016). Similarly, Powell et al. (2008) observed that phosphate content of algae varied between 0.41 and 3.16% depending on the conditions they were exposed to in the waste stabilization ponds. The authors conclude that accumulation of phosphate is a function of light intensity and temperature with higher temperatures and light intensity resulting into higher accumulation.

Table 5.3 shows the mean TP concentration values in effluent from the constructed wetlands compared to the permissible limit according to Kenya's NEMA.

Table 5.3: Mean TP concentration in effluent against NEMA standards

Treatment	Mean Influent TP Concentration (mg/L)	Mean Effluent TP concentration (mg/L)	Removal Efficiency (%)	NEMA standards (mg/L) (Max)	Remarks Standards met (Yes or No)
HSSF + Vetiver grass	2.03	1.14	43.84	2	Yes
VSSF + Vetiver grass	2.03	1.00	50.73	2	Yes
Hybrid + Vetiver grass	2.03	0.71	65.02	2	Yes
HSSF (without grass)	2.03	1.38	32.02	2	Yes
VSSF(without grass)	2.03	1.30	35.96	2	Yes
Hybrid(without grass)	2.03	0.91	55.17	2	Yes

HSSF= Horizontal subsurface flow wetland, VSSF= Vertical subsurface flow wetland, HB= Hybrid subsurface flow wetland, TP= Total Phosphorous

The mean influent and effluent TP concentration presented are for a period of 6 weeks from 7th April to 19th May 2016 when sampling and analysis was conducted. Levels of TP content in effluent by all constructed wetland systems met the standards of a maximum 2mg/l as stipulated by the Kenya's NEMA (1999) for wastewater discharges into water or on land. The constructed wetlands met the objective as in P removal.

5.3.1.4 Total Suspended Solids Removal

Figure 5.4 presents the total suspended solids (TSS) concentrations in effluent and influent analyzed for wetland units during the monitoring period.

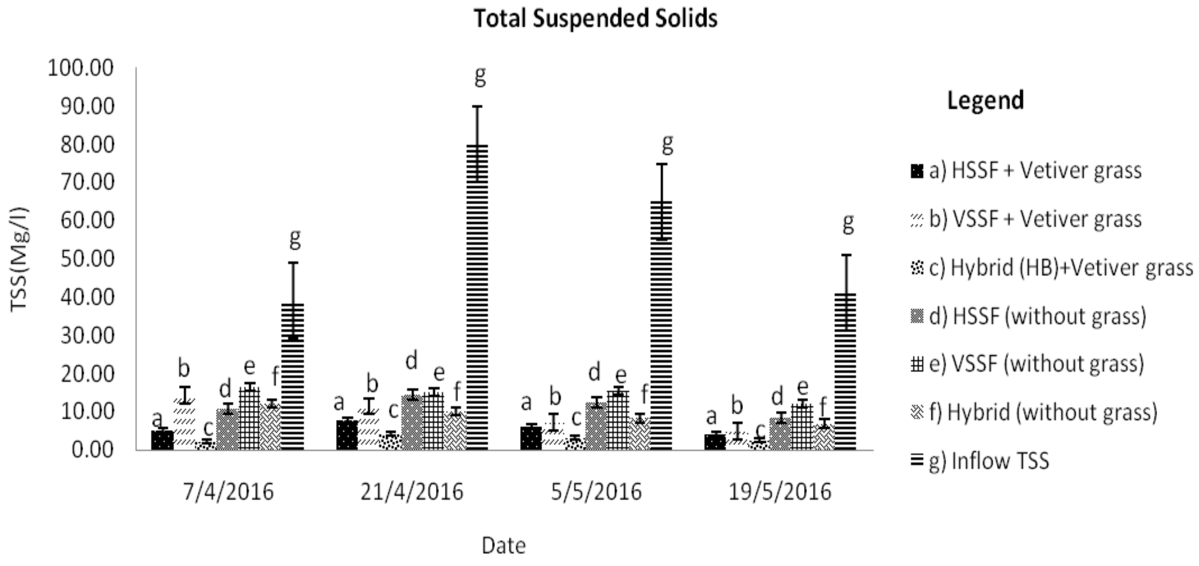

Figure 5.4: Total Suspended Solids in effluent and influent during the monitoring period

Among the subsurface flow wetland systems planted with Vetiver grass, hybrid system achieved significantly ($p \leq 0.05$) lowest mean TSS of 3.04 mg/L in effluent followed by the horizontal at 5.71 mg/L and finally 9.44 mg/L for the vertical system. The best performance of the hybrid system could be attributed to the longer wastewater retention in the substrate at a length of 6.4 m compared to 3.2 m in the horizontal and vertical subsurface flow systems, which allowed the substrate to filter much of the suspended solids from the wastewater. Shruthi and Lokeshappa (2015) using Vetiver grass observed better removal efficiencies of TSS at 60 and 66% were achieved at 4 and 6 days retention time, respectively compared to 58% achieved at 2 days . Ewemoje et al. (2015) in a study on the effect of hydraulic retention time on pollutant removal in a wetland planted with *Coix lacryma jobi,* observed TSS removal was 26.1, 41.9 and 47.8% at 3, 5 and 7 days retention time, respectively.

Of the vetiver grassed plots, significantly ($p \leq 0.05$) better performance of the horizontal subsurface flow system compared to vertical could be attributed to the uniform flow in the horizontal system due to consistent and continuous feeding of wastewater. This could have reduced disturbance on the particles that have been trapped in the system. However in the vertical system, the batch feeding led to turbulence as wastewater flow downwards under gravity thereby disturbing the sediments bound to the media in the constructed wetland. The particles are consequently dislodged from the wetland and contribute to the high TSS level in the effluent.

Of the unplanted systems, the hybrid again achieved significantly ($p \leq 0.05$) lowest mean TSS in effluent of 9.36 mg/L followed by horizontal and vertical system at 11.57 and 14.83 mg/L, respectively. The best performance of the hybrid system is as explained in the planted system above. Again the same argument provided in planted systems applies to better performance of horizontal compared to vertical sub-surface flow system.

The planted systems achieved significantly ($p \leq 0.05$) lower mean effluent TSS compared to the unplanted systems with the planted hybrid system being the best in TSS removal. The planted hybrid system achieved lower mean effluent TSS of 3.04 mg/L compared to the unplanted hybrid system at 9.36 mg/L. The difference in performance could be attributed to the trapping of the solids by the fibrous rooting system of the Vetiver grass. Barakati et al. (2011) in a study on use of Vetiver grass instead of reed in municipal wastewater treatment reported 82% and 96.5% TSS removal for reed and Vetiver grass, respectively. They attributed it to long, branched and bulky rooting system of Vetiver grass that like a powerful filter traps the coarse sediments in wastewater. Abolfazl et al. (2014) also in a study on treatment of hospital wastewater by Vetiver and typical reed plants in a horizontal flow wetland observed that the removal value for TSS for Vetiver grass had a better increasing trend than reed during a period of 3 months. However, no meaningful difference was observed based ($p \geq 0.05$). The authors attributed it to the massive and bulky rooting system of Vetiver grass that traps sediments effectively.

The planted vertical system also achieved lower mean effluent TSS of 9.44 mg/L compared to the unplanted vertical system at 14.83 mg/L. This could also be attributed to trapping of solids by the fibrous rooting system of the Vetiver grass. Mburu et al. (2013) observed that vegetated cells with *cyperus papyrus* achieved 50% TSS removal compared to 18.4% in the unplanted cells. The authors explained that at constant hydraulic loads, roots and rhizome contribute to stabilise the wetland beds and increase the interception and sedimentation. In a study regarding the extent of the trapping of TSS by plant roots, Smith and Kalin (2000) measured the mass of solids trapped amongst roots of a two year old floating *Typha* vegetation mat on an acid mine drainage pond and estimated that a mature system would capture at the least, approximately 2.2 kg of solids per m^2 of floating vegetation. This could also explain the reason as to why the units containing Vetiver grass, known to be deep-rooted, posted higher TSS removal efficiency than the unplanted.

The planted horizontal system achieved lower mean effluent TSS of 5.71 mg/L compared to the unplanted horizontal system at 11.57 mg/L. The roots of Vetiver grass are likely to have slowed down the velocity of wastewater through the coarse sand media thereby increasing the retention time which consequently improved filtration level. Karathanasis et al. (2003) observed that the vegetated systems with cattails (*Typha latifolia*) showed significantly greater ($p \leq 0.05$) removal efficiencies for TSS at 88% compared to the unplanted systems at 46%. They attributed it to rooting biomass of the vegetated systems which provides more effective filtration of the TSS load as well as contributing complimentary treatment of the organic portion of the TSS load through microbial decomposition by offering extensive surface area for microbial attachment.

The influent TSS concentration were observed to be significantly ($p \leq 0.05$) varying throughout the monitoring period which could be attributed to influence of environmental factors on TSS removal in the waste stabilization ponds. Wind velocity is an environmental factor that can influence performance of the waste stabilization ponds since during strong winds; turbulence tends to occur in the maturation and facultative ponds. This in turn can interfere with the settling of the suspended particles at the bottom of the ponds and which consequently get out of the system resulting into high TSS level. Therefore variations in wind velocity could cause the varying influent TSS level in the wetlands.

Table 5.4 shows the performance of all the wetland systems in TSS removal compared to the permissible limit according to Kenya's NEMA.

Table 5.4: Mean effluent TSS concentration against NEMA standards

Treatment	Mean Influent TSS Concentration (mg/L)	Mean Effluent TSS concentration (mg/L)	Removal Efficiency (%)	NEMA standards (mg/L) (Max)	Remarks Standards met (Yes or No)
HSSF + Vetiver grass	56.25	5.71	89.85	30	Yes
VSSF + Vetiver grass	56.25	9.44	83.22	30	Yes
Hybrid + Vetiver grass	56.25	3.04	94.6	30	Yes
HSSF (without grass)	56.25	11.57	79.43	30	Yes
VSSF(without grass)	56.25	14.83	73.64	30	Yes
Hybrid(without grass)	56.25	9.36	83.36	30	Yes

HSSF= Horizontal subsurface flow wetland, VSSF= Vertical subsurface flow wetland, HB= Hybrid subsurface flow wetland, TSS= Total Suspended Solids

The mean influent and effluent TSS concentration presented are for a period of 6 weeks from 7th April to 19th May 2016 when sampling and analysis was conducted. Levels of effluent TSS achieved by all the constructed wetland systems in this research met the standards of maximum 30mg/l stipulated by the Kenya's NEMA (1999) for wastewater discharges into water or on land. The constructed wetlands met the objective as in TSS removal.

5.4 Conclusions

Constructed wetlands are effective in pollutants removal from municipal wastewater. Among the subsurface flow wetland systems planted with Vetiver grass, the Hybrid systems achieved significantly ($p \leq 0.05$) the highest pollutants (COD TSS, TN and TP) removal, compared to the horizontal and vertical subsurface flow systems. The vertical subsurface flow system also performed significantly ($p \leq 0.05$) better in COD, TN and TP removal compared to horizontal system except in TSS removal. Similar trend was exhibited in the unplanted systems. Overally, the planted systems performed significantly ($p \leq 0.05$) better than the unplanted systems in pollutants removal with the hybrid system planted with Vetiver grass being the best in polishing municipal wastewater.

5.5 References

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CHAPTER SIX

ACCUMULATION OF NITROGEN AND PHOSPHOROUS BY VETIVER GRASS (*CHRYSOPOGONZIZANIOIDES*) IN THE MODEL CONSTRUCTED WETLAND TREATMENT SYSTEM

Abstract

Kenya is classified as water scarce country yet the existing fresh water resources are under constant threat of pollution resulting from wastewater inflows. Wastewater contains nitrates and phosphates that stimulate excessive plant growth when released into water bodies thus deteriorating their quality. The purpose of the study was to evaluate the performance of Vetiver grass in the uptake of Nitrogen and Phosphorous from the three (horizontal, vertical and hybrid subsurface flow wetland systems) model constructed wetland units for treating municipal wastewater. Nitrogen and phosphorous accumulation in the roots and shoots of the Vetiver grass was determined and the data subjected to ANOVA at 5% confidence level. Vetiver grass accumulated 18,100 mg and 35.3 mg/kg Nitrogen and Phosphorous, respectively in the hybrid system compared to 9,400 mg Nitrogen and 19 mg/kg Phosphorous, in the horizontal subsurface flow system and 10,400 Nitrogen and 18.3mg/kg Phosphorous in the vertical subsurface flow system. Accumulation of nitrogen and phosphorous by Vetiver grass in all the wetland systems were significantly different ($P \leq 0.05$). There was also significant ($P \leq 0.05$) difference of N and P accumulation in the shoots and the roots with N accumulating more in the shoots while P in the roots.

6.1 Introduction

Fresh water has increasingly become one of the rare valuable resources under the constant threat of pollution. The rapid build-up of toxic pollutants in soil and water bodies not only affects natural resources, but also causes major strains on ecosystems (Arias-Estévez et al., 2008; Paz-Alberto & Sigua, 2013) thereby affecting their functions. The deteriorations in water quality resulting from eutrophication are estimated to reduce biodiversity in water bodies and wetlands by a third globally (UNESCO, 2012). Nutrients discharged into water bodies stimulate excess plant growth resulting in decreased water quality (Arend et al., 2011; Herfindahl et al., 2015;

Yan et al., 2016). Inadequately treated wastewater and agricultural run-off into water bodies has contributed significantly to most of the eutrophication seen today (Cai et al., 2013). The use of conventional wastewater treatment system has proved costly and ineffective (Kumar et al., 2016; Chirisa et al., 2017) and this necessitates the need to develop low energy, effective and low cost technologies in developing countries such as Kenya for efficient treatment.

Phyto-remediation as a green technology is one of the main environmentally friendly technologies that are gaining wider use for wastewater treatment (Mojiri et al., 2016; Vymazal and Březinová, 2016). Diamond (2016), defines phyto-remediation, as the use of plants and their associated microorganisms to stabilize or remove contamination in water. Plant roots exude a wide variety of organic compounds which support the microbial community and can facilitate absorption of some heavy metals (Zang et al., 2013) that are hazardous to both human and livestock.

The use of Vetiver grass for phyto-remediation has gained wider use in the recent years as it has proved to be very effective, low cost natural methods of environmental protection (Greenfield, 2002; Raharjo et al., 2015; Darajeh et al., 2016). Vetiver grass (*Chrysopogon zizanioides*) belongs to the graminae family and was first used for soil and water conservation in India in the 1980s by the World Bank (Truong and Loch, 2004). Since then, its role has been successfully extended to wastewater treatment (Soni et al., 2015; Shahsavari et al., 2016) works. In the process of wastewater treatment, the Vetiver grass absorbs essential plant nutrients such as nitrogen (N) and phosphorus (P) and stores them for other physiological uses (Dhir, 2013; Islands, 2016). The objective of this study was therefore to evaluate the effectiveness of Vetiver grass in the uptake and accumulation of N and P from municipal wastewater passing through a horizontal, vertical and hybrid subsurface flow constructed wetland treatment systems in Gusii wastewater treatment plant.

6.2 Materials and Methods

6.2.1 Study site

See chapter 3.0 section 3.1

6.2.2 Planting and establishment of Vetiver grass

The Vetiver grass slips of 300 mm height were obtained from Kenya Agricultural and Livestock Research Organization (KALRO) in Kisii and planted at spacing 100 mm within and 150 mm between rows in the substrate of the Horizontal, Vertical and Hybrid subsurface flow wetland systems. Diammonium Phosphate (DAP) fertilizer was used at planting to enable root establishment since the substrate had low N and P content of 1200 and 19 mg/kg, respectively. For a period of one month since planting, they were watered with fresh water and subsequently in the 2nd and 3rd month with wastewater from the maturation pond. The Vetiver grass in all the planted wetland units began to continuously receive wastewater based on the experimental flow rate of $0.036 m^3 d^{-1}$ at the beginning of the fourth month for a period of 8 weeks into the Horizontal subsurface flow system. In the planted Vertical subsurface systems, it was intermittently fed with two batches daily of wastewater with each batch having $0.018\ m^3$.

6.2.3 Harvesting of Vetiver grass and Data collection

Five stems of Vetiver grass were randomly harvested from each wastewater polishing unit at the 138th day after planting. The shoots and roots from each wetland unit was air dried, weighed and analyzed for total N and P concentration using atomic absorption spectroscopy (Thomas et al., 1967; Parkinson and Allen, 1975).The data obtained was subjected to a two way ANOVA at 5% level of significance. Means were separated using LSD test to determine if there were significant differences between treatment pairs.

6.3 Results and Discussion

6.3.1 Establishment of Vetiver grass

Figure 6.1 shows the variation of Vetiver grass shoot height with time in the constructed wetland systems during the monitoring period at the 96th, 110th, 124th and 138th day after planting.

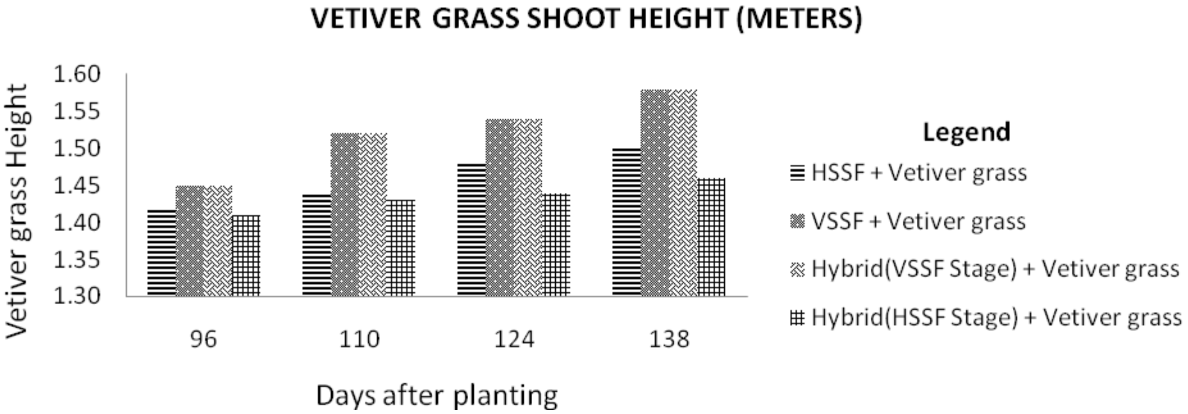

Figure 6.1: Variations of Vetiver Grass shoot height during the monitoring period

Vetiver grass achieved significantly ($p \leq 0.05$) higher mean height of 1.52m in the vertical subsurface flow system, compared to the horizontal flow system at 1.46m as from the 96th upto 138th day after planting. Continuous water flow in the horizontal system could have occupied the voids thereby creating waterlogged conditions thus inhibiting Vetiver grass uptake of nutrients and thereby lowering its growth. Parent et al. (2008) observed that as water saturates the soil pores, gases are displaced and reduction in gas diffusion occurs which reduces photosynthesis and translocation of photoassimilates. Similar observation was noted by Steffens et al. (2005) in a study to investigate the effect of water logging on growth and plant nutrient concentrations where water logging resulted in a significant decrease of shoot dry weight production. The authors explained this observation that due to oxygen deficiency in the root medium of waterlogged soils, synthesis of ATP may be inhibited thus lowering energy status of the plant which consequently leads to a decrease in nutrient uptake.

Despite the saturated conditions in the horizontal subsurface flow wetland system, the progressive growth observed indicates that Vetiver grass has strong adaptation to excess moist conditions. Boonsong and Chansiri (2008) using Vetiver grass cultivated with floating platform technique demonstrated it’s ability to thrive in waterlogged conditions. They observed that after eight weeks, in both experimental set up with highly concentrated wastewater and low concentrated wastewater, the survival percentages of Vetiver grass were ranging from 75-100%. Yeboah et al. (2015) in a study on purification of industrial wastewater with vetiver grasses grown hydroponically, also observed shoot height at the start of the hydroponic treatment in the biogas effluent was 20cm which progressed to 30cm, 45cm , 90cm and 122cm after 30, 60, 90 and 120 days, respectively. In the same study the authors also observed at the start of hydroponic treatment in food and beverage wastewater, the Vetiver shoot height was 20cm which then progressed to 22cm, 23cm, 25cm and 28cm at 30, 60, 90 and 120 days, respectively thus demonstrating it could thrive optimally in water logged conditions.

In the hybrid set up, growth of Vetiver grass in the first stage (Vertical subsurface flow) and in the second stage (Horizontal subsurface flow) varied significantly ($p \leq 0.05$). The grass in the first stage (Vertical subsurface flow) grew taller to a mean height of 1.52m compared to the height in the second stage (Horizontal subsurface flow) of 1.44m as from the 96th upto 138th day since planting. This could have been attributed to the better uptake of nutrients by Vetiver grass in the first stage (vertical subsurface flow) which is well aerated and thus wastewater that flowed to the second stage (horizontal subsurface flow) had lower nutrient content.

In all the wetland systems however, there was progressive increase in Vetiver grass height during the monitoring period which could be attributed to the increase in the uptake of nutrients with physiological age of Vetiver grass. Similar observation was noted by Xia et al. (2003), that the purifying capacity of Vetiver grass in the vertical subsurface flow wetland treating oil refined wastewater gradually increased with the gradual growth and development resulting in gradual increase of biomass. This is further supported by studies by Dhanya and Jaya (2013) who reported that domestic wastewater were rich in nutrients like N, P and K consequently resulting into faster growth of Vetiver grass in the constructed wetland.

6.3.2 Nitrogen Accumulation in the Roots and Shoots of Vetiver Grass in the various treatments during the monitoring period

Table 6.1 shows N accumulation in the roots and shoots of Vetiver grass in the Horizontal, Vertical and Hybrid subsurface flow constructed wetland systems during the monitoring period.

Table 6.1: Nitrogen accumulation in the roots and shoots of Vetiver Grass

Wetland System	N-root (mg/kg)	N-shoot (mg/kg)	Total Accumulation (N-root + N-shoot) (mg/kg)
HSSF + VS	4200^a	5200^a	9400^a
VSSF + VS	4500^b	5900^b	10400^b
HB(VSSF stage + VS)	4500^b	5900^b	10400^b
HB(HSSF stage + VS)	3200^c	4500^c	7700^c

HSSF=Horizontal subsurface flow system, VSSF=Vertical subsurface flow system, HB= Hybrid subsurface flow system, VS= Vetiver Grass, N-root= Nitrogen accumulation in root, N-shoot= Nitrogen accumulation in shoot, Total accumulation with the same letter (a,b,c) in the same column are not significantly different at 5% confidence level.

Nitrogen accumulation in the roots and shoots of Vetiver grass in the horizontal subsurface flow system was 4200 mg and 5200 mg/kg, respectively as at 138th day after planting. Accumulation of N in the Vertical subsurface flow constructed wetland system in the roots and shoots of Vetiver grass was 4500mg and 5900mg/kg, respectively as at 138th day after planting. In the hybrid system, accumulation of N in the roots and shoots was 7700mg and 10400mg/kg, respectively as at 138th day after planting. Nitrogen accumulated was significantly ($p \leq 0.05$) more in the shoots than in the roots of Vetiver grass in all the systems at the end of monitoring period which corresponded to 138th day after planting. This could be an indication that Vetiver grass has higher translocation rate of N from roots to shoots to meet its high nitrogen requirement for stem and leaf growth. Gerrard (2008) observed that when Vetiver grass was grown hydroponically in raw sewage, the accumulation of N was significantly higher in the shoot at 2.37% compared to 1.54% in the roots. Akbarzadeh et al. (2014) observed that Vetiver grass grown hydroponically on domestic wastewater had significantly ($p \leq 0.05$) higher total nitrogen accumulation in the shoots than in the roots. The authors attributed their observations to rapid growth rate and high biomass yield in the grass.

In total, Vetiver grass accumulated significantly ($p \leq 0.05$) the highest N content of 18, 100 mg/kg in the hybrid system, followed by vertical system at 10, 400 mg/kg and finally in the horizontal system at 9,400 mg/kg as at 138th day after planting. This could be attributed to the N uptake by Vetiver grass over a length of 6.4m in the hybrid system compared to 3.2m in both the horizontal and vertical system. However, the significantly ($p \leq 0.05$) higher accumulation of N in the vertical system than in the horizontal system could be due to the better aeration in the vertical subsurface flow system that favours oxidation of ammonia in wastewater to nitrate (NO_3^-) and ammonium (NH^{4+}) that is easily taken up by Vetiver grass (Billore et al., 2002; Njau and Mlay, 2003). This observation is supported by Reddy (1982) in a study on N cycling in a flooded soil ecosystem planted to rice who noted that in aerobic soils where nitrification can occur, nitrate is usually the predominant form of available nitrogen that is absorbed as opposed to water logged conditions that inhibit the biological oxidation of ammonia. Mengel and Kirkby (1987) in a study on plant nutrition, noted that ammonium accumulates in the soil when N conversion is limited or completely stopped if water logged soil conditions persists further supports the findings of this study.

6.3.3 Phosphorous Accumulation in the Roots and Shoots of Vetiver Grass in the various treatments

Table 6.2 shows P accumulation in the roots and shoots of Vetiver grass in the horizontal, vertical and hybrid subsurface flow constructed wetland systems.

Table 6.2: Phosphorous accumulation in the roots and shoots of Vetiver Grass

Wetland System	P-root (mg/kg)	P-shoot (mg/kg)	Total Accumulation (P-root + P-shoot) (mg/kg)
HSSF + VS	10.50^{a}	8.50^{a}	19.00^{a}
VSSF + VS	9.80^{b}	8.50^{a}	18.30^{b}
HB(VSSF stage + VS)	9.00^{c}	8.00^{b}	17.00^{c}
HB(HSSF stage + VS)	9.50^{d}	8.80^{c}	18.30^{b}

HSSF=Horizontal subsurface flow system, VSSF=Vertical subsurface flow system, HB= Hybrid subsurface flow system, VS= Vetiver Grass, P-root= Phosphorous accumulation in root, P-shoot= Phosphorous accumulation in shoot, Total accumulation with the same letter (a,b,c,d) in the same column are not significantly different at 5% confidence level

Phosphorous accumulation in the roots and shoots of Vetiver grass in the Horizontal subsurface flow system was 10.5 and 8.5 mg/kg, respectively compared to Vertical subsurface flow system at 9.8 and 8.5 mg/kg, respectively as at 138th day after planting. In the hybrid system, accumulation of phosphorous in the roots and shoots was 18.5 and 16.8 mg/kg, respectively as at 138th day after planting. Phosphorous accumulated significantly ($p \leq 0.05$) more in the roots than in the shoots of Vetiver grass in all the wetland systems. This could indicate that Vetiver grass utilizes more P for root development. Gerrard (2008) observed that when Vetiver grass was grown hydroponically in raw sewage, the accumulation of P was significantly higher in the root at 0.41% compared to 0.29% in the shoots. Boonsong and Chansiri (2008), it was observed that P accumulation in the shoots of Vetiver grass grown in the highly concentrated wastewater was significantly lower compared to the accumulation in the roots. The authors explained that phosphorous was the macronutrient required in high amounts for root development.

In total, Vetiver grass accumulated significantly ($p \leq 0.05$) the highest amount of P at 35.3 mg/kg in the hybrid system, followed by horizontal system at 19 mg/kg and finally in the Vertical system at 18.3 mg/kg as at 138th day after planting. This could be attributed to the uptake of phosphates by Vetiver grass over a length of 6.4m in the hybrid system compared to 3.2m in both the horizontal and vertical system. However, the significantly ($p \leq 0.05$) higher accumulation of P in the horizontal system than in the vertical system could be due to longer contact time between Vetiver grass roots and wastewater as opposed to vertical system whereby wastewater is uniformly spread over the whole surface area and flows downwards under gravitational influence. This influence of gravity could cause wastewater to drain out faster thereby shortening contact time with Vetiver grass roots.

It was also observed in this study that P accumulation in Vetiver grass in all the wetland systems were significantly ($p \leq 0.05$) lower compared to N. For instance, in the hybrid system, Vetiver grass accumulated 35.3 mg/kg P compared to 19,100 mg/kg N a fact attributed to adsorption of P by the sandy substrate making it unavailable for Vetiver grass uptake. Similar observation was noted by Holford (1997) where, more than 80% of the phosphorous in soil become immobile and unavailable for plant uptake due to adsorption, precipitation or conversion to the organic form. According to Hoffman et al. (2011), phosphorous removal can be achieved in constructed wetland by adsorption and precipitation and only a small amount is taken up by plant growth. Wagner et al. (2003) observed that Vetiver requirement for P was not as high as for N and no growth response occurred at rates higher than 250kg/ha/year under P supply while for N supply, the growth increased significantly upto an application rate of 6000kg/ha/year.

6.4 Conclusions

Vetiver grass accumulated 18,100 mg/kg and 35.3 mg/kg N and P, respectively in the hybrid system as compared to 9,400 N and 19 mg/kg P, in the horizontal subsurface flow system and 10,400 N and 18.3mg/kg P in the vertical subsurface flow system. Hence it can be concluded that Vetiver grass accumulates more N and P in the hybrid systems than in single systems (horizontal and vertical system) and it up takes more N from wastewater in well aerated soils in vertical subsurface flow systems than under waterlogged conditions in the horizontal subsurface flow systems. P uptake is generally low compared to N and it is independent of substrate aeration but on the contact time between wastewater, substrate and Vetiver grass. Purifying ability of Vetiver grass also increases with time.

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CHAPTER SEVEN

7.0 General Discussion, Conclusions and Recommendations

7.1 General Discussion

This study emphasized on the need for proper wastewater treatment before release into water or land to protect the existing fresh water bodies from pollution. According to WHO (2015), the past focus by the Millennium Development Goals on increasing access to improved sanitary facilities with little emphasis on wastewater management have resulted into the deteriorating water quality globally. Adoption of low cost technologies like constructed wetlands for wastewater treatment is an idea that should be implemented in the third world countries where conventional treatment systems are deemed to be expensive and ineffective (Mthembu et al., 2013). Recognizing the challenge of water pollution in water scarce countries like Kenya (Mogaka et al., 2006), there is urgent need for emphasis to be put on proper wastewater treatment by industries before release into water bodies that are depended on by downstream users for domestic and livestock use. Reuse of treated wastewater for purposes that doesn't require high quality water should also be adopted to ease the stress on the existing fresh water reserves.

7.2 Conclusion

The study made the following conclusions:

- Well graded sand was found to have low porosity and low hydraulic conductivity and hence not suitable for use in constructed wetland.
- In selecting coarse sand as a media in subsurface flow wetland, the particle size distribution (uniformity coefficient) should be an important consideration rather than relying on porosity values.
- The size of wetland cells is also highly dependent on the design flow rate and the BOD rate constant if first order model proposed by Kickuth is used but the actual operational flow rate determines the retention time in the wetland.

- Hybrid constructed wetlands exhibited better pollutants removal than single operated constructed systems (horizontal and vertical systems).

- BOD_5, COD and TSS removal was high in all types of constructed wetlands planted with Vetiver grass. However the nutrient removal especially phosphorous was low in the single systems (vertical and horizontal subsurface wetlands).

- Vetiver grass can thrive in waterlogged conditions in the horizontal subsurface flow wetlands though this made the grass not to grow vigorously.

- Vetiver grass accumulated more N in the shoots than in the roots but it accumulates more P in the roots than in the shoots.

- Accumulation of N in Vetiver grass was higher in well aerated soils than in anaerobic conditions.

- Phosphorous removal in constructed wetlands is more dependent on the contact time between wastewater and the substrate rather on plant uptake.

7.3 Recommendations

The study made the following recommendations:

- Constructed wetlands is a suitable technology that should be adopted to ameliorate the low availability of irrigation water and to protect the existing fresh water bodies against pollution in water scarce countries like Kenya.

- Constructed wetland treatment system should be combined with the conventional wastewater treatment plant in Gusii so as to further polish the effluent to meet the expected standards of discharge of wastewater into the receiving river.

- Constructed wetlands should involve the use of a substrate with high adsorption capacity of phosphorous to improve Total Phosphorus reduction.
- Vetiver grass should be planted randomly in a horizontal subsurface flow wetland to prevent wastewater from taking preferential paths.

- Variation of experimental flow rates should be carried out during the monitoring period to determine its effect on the treatment performance.
- Longer periods of monitoring are recommended.

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APPENDICES

APPENDIX I: SIEVE ANALYSIS RESULTS FOR RIVER SAND OBTAINED FROM SORI

Pan mass=100gm

Initial dry sample mass + pan=1188gm

Initial dry sample mass =1088 gm

Washed dry sample mass + pan=1012gm

Washed dry mass=912 gm

Fine mass =176gm

Table 1: Sieve analysis results of river sand from Sori

Sieve size (mm)	Retained mass (gm)	% retained	Cumulative passed percentage(%)	Remarks
14	0	0.0	100.0	
10	0	0.0	100.0	
4.76	30	2.8	97.2	
2.36	184	16.9	80.3	
1.18	280	25.7	54.6	
0.6	148	13.6	41.0	
0.3	154	14.2	26.8	
0.15	73	6.7	20.1	
0.075	43	4.0	16.2	Clay/Silt content=16.2%
Pan	176	16.2		
Total	1088			

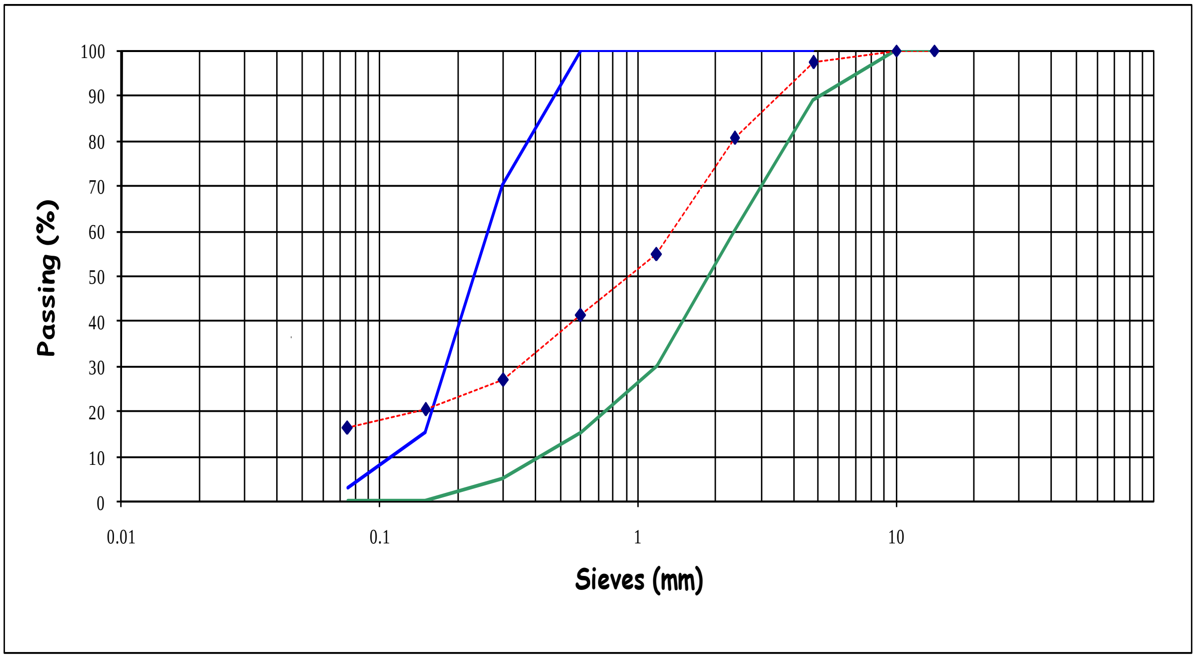

Figure 8.1: Grading curve for river sand from Sori

From the grading curve the coefficient of uniformity and coefficient of curvature can be obtained as in equation 1.1 and 1.2 respectively

$$Cu = \frac{D60}{D10} = \frac{1.45}{0.03} = 48 \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (1.1)$$

Coefficient of curvature Cc

$$Cc = \frac{(D30)^2}{D10 \times D60} = \frac{0.35 \times 0.35}{0.03 \times 1.45}. = 2.82 \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (1.2)$$

APPENDIX II: SIEVE ANALYSIS RESULTS FOR RIVER SAND OBTAINED FROM KENDU BAY

Pan mass=100gm

Initial dry sample mass + pan=986gm

Initial dry sample mass =886 gm

Washed dry sample mass + pan=898gm

Washed dry mass=798 gm

Fine mass =88gm

Fine percent=9.9

Table 2: Sieve analysis results of river sand from Kendu bay

Sieve size (mm)	Retained mass (gm)	% retained	Cumulative passed percentage(%)	Remarks
14	0	0.0	100.0	
10	0	0.0	100.0	
4.76	8	0.9	99.1	
2.36	38	4.3	94.8	
1.18	208	23.5	71.3	
0.6	199	22.5	48.9	
0.3	207	23.4	25.5	
0.15	71	8.0	17.5	
0.075	67	7.6	9.9	Clay/Silt content=9.9%
Pan	88	9.9		
Total	886			

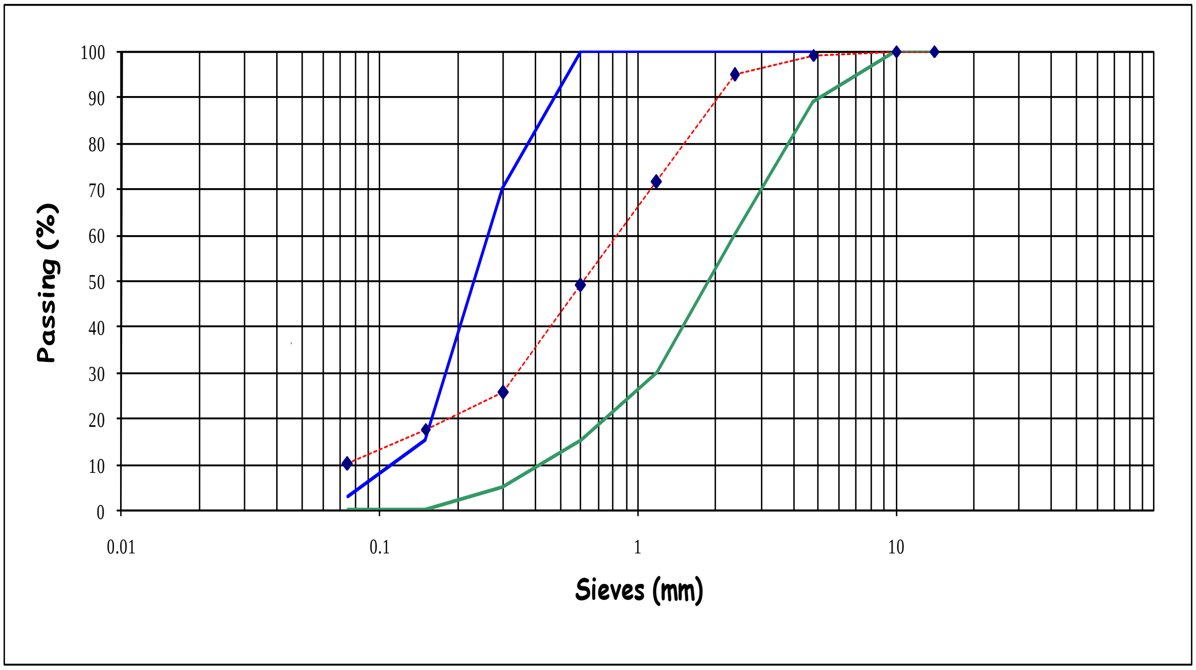

Figure 8.2: Grading curve for river sand from Kendu bay

From the grading curve the coefficient of uniformity and coefficient of curvature can be obtained as in equation 2.1 and 2.2 respectively

$$Cu = \frac{D60}{D10} = \frac{0.82}{0.075} = 10.93\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots(2.1)$$

Coefficient of curvature Cc

$$Cc = \frac{(D30)^2}{D10 \times D60} = \frac{0.35 \times 0.35}{0.075 \times 0.82}. = 1.99 \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (2.2)$$

APPENDIX III: DETERMINATION OF SPECIFIC GRAVIY OF RIVER SAND FROM KENDU BAY AND SORI

Table 3: Specific gravity of river sand from Kendu bay and Sori

Sample	Kendu bay sand	Sori
Mass of empty bottle(W1)	59.6	50.5
Mass of bottle + Soil (W2)	69.6	57.9
Mass of bottle + Soil + Water (W3)	170.3	152.6
Mass of bottle full of water(W4)	164.2	148.1
Massof water used (W3-W2)	100.7	94.7
Mass of Soil used (W2-W1)	10	7.4
Volume of soil(W4-W1) - (W3-W2)	3.9	2.9
Specific gravity of Soil: $Gs = \frac{(W2 - W1)}{(W4 - W1) - (W3 - W2)}$	2.564	2.551

APPENDIX IV: POROSITY TEST RESULTS FOR RIVER SAND FROM KENDU BAY

Weight of empty can=975.2g

Can + sample=1752.8g

Initial height of relative density can=15.7 cm

Diameter of relative density can= 6.97cm

Volume of relative density can= 600cm3

Height displaced =3.6 cm

Difference in height: 15.7-3.6=12.1 cm

Volume after fall(shaking)= $V2 = \frac{\pi \times D^2 h}{4}. = .\frac{3.142 \times 6.97 \times 6.97 \times 12.1}{4}. = 461.7cm^3$

Porosity= $n = 1 - \frac{(1752.8 - 975.2)}{(461.7 \times 2.564 \times 1)} = 0.343$

APPENDIX V: POROSITY TEST RESULTS FOR RIVER SAND FROM SORI

Weight of empty can=975.2g

Can + sample=1716.6g

Initial height of relative density can=15.7 cm

Diameter of relative density can= 6.97cm

Volume of relative density can= 600cm^3

Height displaced =4.3cm

Difference in height: 15.7-4.3=11.4 cm

Volume after fall (shaking)= $V2 = \dfrac{\pi \times D^2 h}{4}. = .\dfrac{3.142 \times 6.97 \times 6.97 \times 11.4}{4}. = 434.97\,cm^3$

Porosity= $n = 1 - \dfrac{(1716.6 - 975.2)}{(434.97 \times 2.564 \times 1)} = 0.331$

The soil from Kendu bay has higher porosity and hence better hydraulic conductivity than soil from Sori.

APPENDIX VI: DETERMINATION OF PERMEABILITY OF RIVER SAND FROM KENDUBAY USING FALLING HEAD PERMEABILITY TEST

The coefficient of permeability using falling head method is obtained from the formula:-

$$K = \frac{2.3026\,a \times L}{A} \times \frac{Log_{10} H_1 - Log_{10} H_2}{t_2 - t_1}\, cm/\sec$$

Where:

K= coefficient of permeability (cm/sec)

a= crossectional area of manometer tube (cm^2)

L= length of sample under test (cm)

A= cross sectional area of sample(cm^2)

H_1= initial height of water (cm)

H_2= head of water in cm indicated at the end of a particular period of time

t_2= time corresponding to H2 (sec)

t_2= start time (sec)

Results of river sand from Kendu bay

Trial 1:

Table 4: Trial 1 of permeability test for river sand from Kendu bay

Time (sec)	Height of water (cm)
0	96.5
5	86.4
10	76.5
15	66.5

Trial 2:

Table 5: Trial 2 of permeability test for river sand from Kendu bay

Time (sec)	Height of water (cm)
0	96.5
5	86.2
10	75.9
15	65.5

The permeability of Kendu bay sand was calculated to be: 2.766×10^{-3}cm/s

APPENDIX VII: DETERMINATION OF PERMEABILITY OF RIVER SAND FROM SORI USING FALLING HEAD PERMEABILITY TEST

Results of river sand from Sori

Trial 1:

Table 6: Trial 1 of permeability test for river sand from Sori

Time (sec)	Height of water (cm)
0	96.5
5	87.4
10	78.27
15	69.1

Trial 2:

Table 7: Trial 2 of permeability test for river sand from Sori

Time (sec)	Height of water (cm)
0	96.5
5	87.3
10	78.1
15	68.9

The permeability of Sori sand was calculated to be: 2.425×10^{-3} cm/s

APPENDIX VIII: PLANTING OF VETIVER, SAMPLING AND ANALYSIS OF WASTEWATER

Plate 1: Planting of Vetiver grass slips

Plate 2: Vetiver grass at three months since planting

Plate 3: Sampling of effluent wastewater from the wetlands

Plate 4: Wastewater analysis in the laboratory

APPENDIX IX: STATISTICAL ANALYSIS USING SPSS

Table 8: Chemical Oxygen Demand ANOVA Results

Measure	Constructed wetland units	Constructed wetland units	Significance
LSD	HSSF + VETIVER	VSSF + VETIVER	0.000
		HB + VETIVER	0.000
		HSSF(CONTROL)	0.000
		VSSF(CONTROL)	0.000
		HB(CONTROL)	0.242

HSSF: Horizontal subsurface flow wetland system, VSSF: Vertical subsurface flow wetland system, HB= Hybrid subsurface flow wetland system, CONTROL: Unplanted systems

Table 9: Biochemical Oxygen Demand ANOVA Results

Measure	Constructed wetland units	Constructed wetland units	Significance
LSD	HSSF + VETIVER	VSSF + VETIVER	0.012
		HB + VETIVER	0.000
		HSSF(CONTROL)	0.000
		VSSF(CONTROL)	0.000
		HB(CONTROL)	0.303

HSSF: Horizontal subsurface flow wetland system, VSSF: Vertical subsurface flow wetland system, HB= Hybrid subsurface flow wetland system, CONTROL: Unplanted systems

Table 10: Total Nitrogen ANOVA results

Measure	Constructed wetland units	Constructed wetland units	Significance
LSD	HSSF + VETIVER	VSSF + VETIVER	0.000
		HB + VETIVER	0.000
		HSSF(CONTROL)	0.000
		VSSF(CONTROL)	0.000
		HB(CONTROL)	0.000

HSSF: Horizontal subsurface flow wetland system, VSSF: Vertical subsurface flow wetland system, HB= Hybrid subsurface flow wetland system, CONTROL: Unplanted systems

Table 11: Total Phosphorous ANOVA Results

Measure	Constructed wetland units	Constructed wetland units	Siginificance
LSD	HSSF + VETIVER	VSSF + VETIVER	0.000
		HB + VETIVER	0.000
		HSSF(CONTROL)	0.000
		VSSF(CONTROL)	0.000
		HB(CONTROL)	0.000

HSSF: Horizontal subsurface flow wetland system, VSSF: Vertical subsurface flow wetland system, HB= Hybrid subsurface flow wetland system, CONTROL: Unplanted systems

Table 12: Total Suspended Solids ANOVA Results

Measure	Constructed wetland units	Constructed wetland units	Significance
LSD	HSSF + VETIVER	VSSF + VETIVER	0.000
		HB + VETIVER	0.000
		HSSF(CONTROL)	0.000
		VSSF(CONTROL)	0.000
		HB(CONTROL)	0.000

HSSF: Horizontal subsurface flow wetland system, VSSF: Vertical subsurface flow wetland system, HB= Hybrid subsurface flow wetland system, CONTROL: Unplanted systems