MACROPHYTE BASED INDEX OF BIOTIC INTEGRITY FOR MONITORING ECOLOGICAL INTEGRITY OF CHEPKOILEL RIVER SWAMP, KENYA

BY

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DECLARATION

Declaration by the candidate

I duly declare that this thesis is my original work and has not been presented to any other University for the award of a degree. This work should not be reproduced without the express authority of the author and/or University of Eldoret.

Kipkorir Regina Jeruiyot **Signature……………** **Date…………….**
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Declaration by the supervisor

This thesis has been submitted with my approval as University supervisor.

Dr. Josephine Mulei **Signature…………….** **Date……………**

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DEDICATION

This thesis is dedicated to my beloved parents Mr. and Mrs. Samson Cheronoh, my husband Daniel Kiprop and siblings: Irene, Gideon, Raymond and Bravin.

ABSTRACT

The study set out to investigate the uses of macrophytes within Chepkoilel river swamp, the impact of those uses on macrophyte community structure and to develop an index for monitoring the ecological integrity of the swamp. Macrophytes were sampled at seven stations along Chepkoilel river swamp one of which became the reference station. Stations were chosen to correspond to different human activities and intensity of human presence Sampling was done at each station for six months. Selected physico-chemical parameters of water were measured at each sampling site. Sampled macrophytes were identified, and taxon diversity, abundance, evenness and non-taxonomic attributes estimated for each station. Composition and distribution results were used to develop a Macrophyte Index of Biotic Integrity that was used for bioassessment of ecological integrity of the swamp. Ten metrics were tested using box-and-whisker plots to determine their variability across a gradient of human disturbance. The riparian community was found to have diverse uses for macrophytes from this swamp with animal grazing, fuelwood collection, wild fruits, and medicinal uses showing high respondent proportion on frequency of use and use preference. Results showed significant ($p<0.05$) spatial variation in macrophyte community attributes and water quality parameters. The macrophyte attributes further showed significant relationships with water quality parameters. Stations with high human disturbance recorded low abundance and diversity compared to those with low human disturbance. Nine metrics met the test criteria and were used to develop Index of Biotic Integrity development along the swamp. These were total abundance, diversity index, evenness index, vascular plant diversity, non-vascular plant diversity, grasses, sedges, true aquatic plants, and plants with persistent litter. Station 1 obtained a total macrophyte index of Biotic Integrity score of 37 while station 3 obtained a total score of 11. This result indicated that more disturbed stations got low IBI scores compared to the less disturbed ones. Conclusions from the study showed that human disturbance influenced macrophyte composition and abundance along Chepkoilel river swamp consequently influencing the ecological integrity of the swamp. The macrophytes in the swamp can be considered to be reliable indicators of disturbance and the IBI developed should therefore be used to constantly monitor the ecological integrity of the swamp.

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LIST OF ACRONYMS AND ABBREVIATIONS

ANOVA	Analysis of variance
BOD	Biological oxygen demand
DO	Dissolved Oxygen
GEF	Global Environmental Facility
pH	Potential hydrogen
RBP	Rapid Biomonitoring Program
SEM	Standard error of the mean
TSS	Total Suspended Solids

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CHAPTER ONE

INTRODUCTION

1.1 Background Information

Wetlands play an ecological role and have the potential of great economic, cultural and scientific value (Ministry of Environment and Mineral Resources (MEMR), 2010). They provide habitats for a wide range of flora and fauna and are important sources of water for human consumption. Further, wetlands also provide good soils for agriculture, livestock grazing, hydrological function of recharge and discharge of water, water purification, flood control, and stratum of carbon dioxide while the economic benefits include fisheries, recreation, and also serve as spawning grounds for fish and resting ground for birds (Ambasa, 2005).

In Kenya, wetlands face rapid degradation as a result of both anthropogenic disturbance and natural causes like urbanization, climate change, overexploitation, inadequate awareness on conservation of wetlands and unsustainable management together with inadequate legislative framework (MEMR, 2010).

Macrophytes are aquatic plants, growing in water and are usually emergent, submergent or floating (Achieng, 2011). They include macro-algae, mosses and liverworts (bryophytes), fern (Pteridophytes) and Tracheophytes (Sosiak, 2002). The life forms of macrophytes which include emergent, floating-leafed and submerged together with zonation and community patterns of these aquatic plants are also useful in describing the plants structures as well as the form and condition of the environment (Sosiak, 2002).

Macrophytes have for a long time been used as bioindicators of aquatic health for example Charophytes have since 1930 been used as indicators of good ecological state of water (Pelechaty *et al*., 2004) and eelgrass is regarded as a useful indicator of water quality since water clarity influences its distribution within a specific habitat (Krause-Jensen *et al*., 2005).

1.2 Statement of the Problem and Justification

Chepkoilel River originates from Kaptagat forest and along its course; various human activities are known to take place. These activities include crop cultivation, animal grazing and brick making activities that often lead to wetland degradation (Ambasa, 2005). The swamp supports a big flower farm, a university fish farm and the university sewage treatment ponds also drain into it. Despite the presence of these activities, little effort has been done to characterize this swamp in terms of its ecological integrity. Most of the studies done in this swamp have largely been on water quality status using physico-chemical parameters and its fauna such as macroinvertebrates and fish, with few studies working on macrophyte diversity (Mulei, 2011). None of the studies previously done have strived to develop a biomonitoring index using macrophytes which is equally reliable as fish and macro-invertebrates.

Plants are now being used for bio-assessment of ecological integrity is therefore currently the focus as bio-indicators (Penniung, 2008). However much of the evidence to support their positive response and reliability is only documented in developed countries where intensive critical examination is employed to develop universal index that can be used for measuring aquatic ecological integrity (Goethals *et al*., 2001; Hrivnak *et al*., 2007 and Penniung, 2008).

Plants in Chepkoilel river swamp are directly affected by human disturbance and changes in water chemistry. Their utilization for bio-assessment will therefore be important since they are organisms found either seasonally or permanently within the area and can provide reliable information. Macrophytes are sedentary, sensitive to environmental variations, convenient for sampling (Mason, 1981; Zhou *et al*., 2008), and react more rapidly to the presence of pollutants than higher organisms (Ferrat *et al*, 2003). They are important in nutrient cycling, control of water quality, sediment stabilization and shelter for aquatic organisms and wildlife (Janauer, 2001; Demireze and Aksoy, 2006), bioaccumulations (Camargo and Alonso, 2006) and good indicators of the changes occurring in water bodies (Melzer, 1999). Many community-habitat relationships have not only regional but also general characteristics (Balazs *et al*., 2009). Macrophytes respond easily to hydro morphological degradation with multiple stressors (Mason, 1981). Perennial macrophytes are good indicators of long term habitat changes; an integral of temporal effect of disturbance (Hering *et al*., 2006). Despite these facts, plant use as biomonitors of Ecotoxicological studies in wetlands is scanty (Rader *et al*., 2001). This study therefore used macrophytes to assess the ecological integrity of Chepkoilel river swamp along different disturbance gradients.

1.3 Objectives

The overall objective of this study was to assess the variation in macrophyte community within different disturbance gradients along Chepkoilel river swamp with an aim of developing a bio monitoring tool for assessing ecological integrity of the swamp.

Specific objectives

i. To document the various uses and use preference of macrophytes along of Chepkoilel river swamp by the riparian community.
ii. To determine the diversity and abundance of macrophytes within Chepkoilel river swamp
iii. To determine the relationship between water quality parameters and macrophyte attributes like diversity, abundance, evenness, grasses, sedges, plants with persistent litter, vascular and non-vascular plants
iv. To develop a macrophyte based index of biotic integrity for monitoring the ecological integrity of the swamp

1.4 Hypothesis

H_1: The communities along Chepkoilel river swamp have various uses and use preference for the various macrophytes occurring in the swamp

H_2: There is variation in diversity and abundance of macrophytes along Chepkoilel river swamp.

H_3: Water quality parameters along Chepkoilel river swamp has an influence on macrophyte community attributes like diversity, abundance, evenness, grasses, sedges, plants with persistent litter, vascular and non-vascular plants.

H_4: The response of macrophytes in Chepkoilel river swamp to changes in water quality is sufficient enough to be used to monitor changes in ecological integrity.

CHAPTER TWO

LITERATURE REVIEW

2.1 Use of Macrophyte

Macrophytes colonize many types of aquatic ecosystems, such as lakes, reservoirs, wetlands, streams, rivers, marine environments and even rapids and falls (Geothals *et al.*, 2001). They are important components of aquatic systems, influencing ecological processes like nutrient cycling and attributes of other aquatic attached assemblages such as species diversity. Macrophytes worldwide have several human and hydrological uses. They are harvested for the purpose of mat making, making fishing traps and crop mulching (Orwa *et al*., 2012). The macrophytes are also commonly harvested for medicinal value in most rural areas of Kenya (Obiero *et al*., 2012). The rural communities during the low rainfall period not only clear wetlands for crop cultivation but also collect fuel wood and graze livestock within wetlands, activities that have direct impact on macrophyte structure (Masese *et al*., 2009).

Animal grazing reduces the abundance and diversity of macrophytes in addition to increasing nutrient concentrations through urine and dung deposition (Aura *et al*., 2010, Orwa *et al*., 2012). Reduction in diversity and abundance of grasses in a wetland is an indicator of reduced habitat quality (Balazs, 2009). Collection of macrophytes for whichever purpose has been found to reduce the integrity of wetlands. Macrophyte harvesting reduces plant cover thereby exposing the fauna within the system to harsh conditions like direct sunlight which increases temperature and increase nutrient concentration as a result of reduced absorption capacity (Balazs *et al*., 2009 and Hering *et al*., 2006). Macrophytes offer shoreline protection and trap sediments before entering the water column. This protection capacity is greatly reduced when the plants are removed (Ntiba *et al.*, 2001).

A wetland where the riparian community greatly depends on its macrophytes for livelihood is likely to be of poor ecological integrity. (Geothal *et al.*, 2001. The situation is worsened when the use is extractive that is removal of plants since the buffering capacity is reduced. It is thus imperative to investigate the uses under which the swamp is put for the purpose of regulation. Although the buffering capacity of Chepkoilel river swamp has been studied before (Orwa, 2009); this could have changed with plant use changes.

2.2 Macrophyte Diversity and Abundance

Macrophyte species and distributions are diverse worldwide (Willby *et al.*, 2000). Over the last decades, various biological traits have been used to group plant species into coherent groups which have provided a valuable alternative approach for studying the ecology of a wide range of vegetation types (Willby *et al.*, 2000). The impact of habitat perturbation can also be predicted with broad sensitivity rather than being dependant on the presence of an individual species which may merely reflect chance dispersal and seasonal colonization events (Rolon *et al.*, 2008). The study of variation in macrophytes community attributes at different disturbance gradients is therefore essential for biomonitoring (Hrivnak, 2005). Chepkoilel river wetland though has numerous benefits for the riparian community has not received sufficient attention regarding its conservation in terms of macrophyte structure at various disturbance gradients.

The richness of plant community in aquatic and wetland habitats is relatively low compared to terrestrial communities (Rader *et al*., 2001). Adaptation and specialization to the aquatic habitat has been achieved by only a few angiosperms and pteridophytes which are though a few species float freely in water (Hrivnak, 2005). Wetland vegetations are also very sensitive to changes in environmental conditions in terms of spatial and temporal scales (Hrinvak, 2005). When considering plant sampling techniques for biomonitoring, it is important to consider their morphological adaptation (Hrinvak, 2005). Lack of macrophytes in a system where they are expected to occur suggests a reduced population of fish and waterfowl (Ervin and Wetzel, 2002).

2.3 Water Quality Parameters

The physical and chemical characteristics of aquatic environments are influenced by a number of processes. The character of rivers often reflects an integration of physical and biological processes occurring within the catchments. Some of the landscape properties that contribute directly to the structure and function of aquatic systems include prevailing climate, riparian land-use or cover patterns, channel slope, aspect, and bedrock geology, and hydrography (Richards *et al*., 1997). The physical factors that affect aquatic environments are temperature, discharge, turbidity, PH and conductivity (Boney, 1989).

In aquatic ecosystems pH and dissolved oxygen play an important role in determining the biotic community structure as it regulates metabolic processes (Busulwa and Bailey, 2004). In streams and the rivers amount of dissolved oxygen in water is controlled by a number of factors. High dissolved oxygen values can be maintained in upland streams as a result of their characteristic cascade, rapids and riffles as water moves from higher altitudes (Busulwa and Bailey, 2004). Temperature also affects the amount of dissolved oxygen in water (Kalff, 2002). Increase in temperature lowers its solubility resulting in low values.

The amounts of nutrients in the water play a significant role in influencing the chemistry of aquatic ecosystems particularly those that are often in short supply and those that limit primary productivity, like phosphorus, nitrogen or both (Kalff, 2002). Moreover, nutrient limitation is mostly reported in lakes but not in rivers and this is attributed to the enriching effect of water velocity and turbulence. Another reason is that streams are open systems with a large capacity to retain nutrients (Kalff, 2002).

Most of the nutrients in streams and rivers are supplied from the catchment. Land-use activities like agriculture serve as possible sources of nutrients. Problems of nutrient enrichment that cause eutrophication have, thus, been reported in most riverine systems of Lake Victoria basin (Osano *et al*., 2003; Raburu, 2003) and the lake itself (Okungu and Opango, 2005).

2.4 Macrophyte Index of Biotic Integrity

Ecological Indicators are measurable entities that are used to assess the status and trend of key ecological attributes or other factors (Wray and Baylay, 2006). A good indicator meets the criteria of being measurable, precise, consistent, relevant and sensitive (Groom *et al*., 2005). Potential indicators of wetland health may be physical, chemical or biological in nature (Wray and Baylay, 2006).

The physical changes to wetland hydrology may occur slowly over time and can be difficult to monitor since wetland functions such as flood control, ground water storage and peat accumulation may be disrupted by physical disturbance which might be difficult to quantify and more when natural physical conditions and hydrology are variable(Wray and Baylay, 2006). Reflection or impact of physical disturbance may be more easily quantified by the chemical and biological parameters of the wetland system for instance; change in water table can lead to change in plant species (Wray and Baylay, 2006).

Water chemistry is a very useful indicator for overall wetland health, however difficult, time consuming and expensive to measure the numerous chemicals parameters related to water chemistry (Njiru *et al*., 2008). Chemical interactions are furthermore complex and may be difficult to measure in terms of their degradation and transformation (Wray and Baylay, 2006). It is also not cost effective to frequently measure chemical parameters over a long period of time.

Biological assessments of water quality have proved very successful and easy to do as compared to routine chemical sampling (Mason, 1981). The biological indicators most useful and in consideration for assessment of wetland health condition include microbes, vascular and non-vascular plants, invertebrates and birds (Wray and Baylay, 2006). Wetland macrophytes are directly influenced by water quality and therefore, any impairment in wetland quality should be reflected by taxonomic composition of aquatic plant community (Croft and Chow-Fraser, 2007). Since plants function as an energy source for higher organism, they can indicate trophic status of a wetland and therefore, nutrient loading (Wray and Baylay, 2006). Change in community composition, biomass and change in plant health are therefore very useful as indicators.

CHAPTER THREE

MATERIALS AND METHODS

3.1 Study area

Chepkoilel river swamp (Figure 1) lies between latitude 0^0 40' N and 0 35' S and longitude 0^0 37' E and 0^0 50' E at an altitude of 2180 m. The swamp occurs on fertile volcanic soils in a gentle sloping terrain bordered by undulating plains. The soils are rich in montmorillonites and clays thereby encouraging extensive cracking during dry periods and water logging during wet seasons (Odongo, 1996). The swamp covers aproximately 5.6 km^2. Wetland vegetation is dominated by a central band of dense *Cyperus papyrus* flanked by shorter emergent vegetation dominated by other *Cyperus spp*. including *C. rotundus*, *C. triandra* and *C. laevigatus*.

The wetland has a catchment of 210 km^2 and is supplied with water by Chepkoilel River originating from Kaptagat forest where it is referred to as Misikuri River (Odongo, 1996). As the river moves downstream it enters the gently undulating plains where its flow and velocity reduce resulting in deposition and widening of the channel. The rainfall distribution is bimodal with an annual mean of 986 mm in two distinct seasons. The daily mean maximum and minimum temperature recorded in the area is 17.6 ºC and 10 ºC respectively (Jaetzold and Schmidt, 1983). The main human activities in the surrounding area include wheat and maize farming, horticulture, floriculture, poultry, fish farming, animal husbandry and brick making. Shallow and seasonally flooded sections of the wetland have been encroached by agricultural activities where the cultivation of *Solanum lycopersicum* L. (tomatoes) and *Brassica* L. (kales and cabbages) dominate. In these areas ditches have been dug either to drain water away or supply water for irrigation. There is also settlement on the riparian areas. The swamp receives wastewater from the University of Eldoret sewerage treatment ponds.

The wetland is owned by the local community who hold title deeds for parcels of land that project into it. Because of increasing human population and diminishing land resources in the adjacent area, there is a lot of encroachment into the wetland for agricultural activities. Land in the surrounding area is also being increasingly subdivided for sale and inheritance increasing the intensity of human activities (Regina, Pers.com).

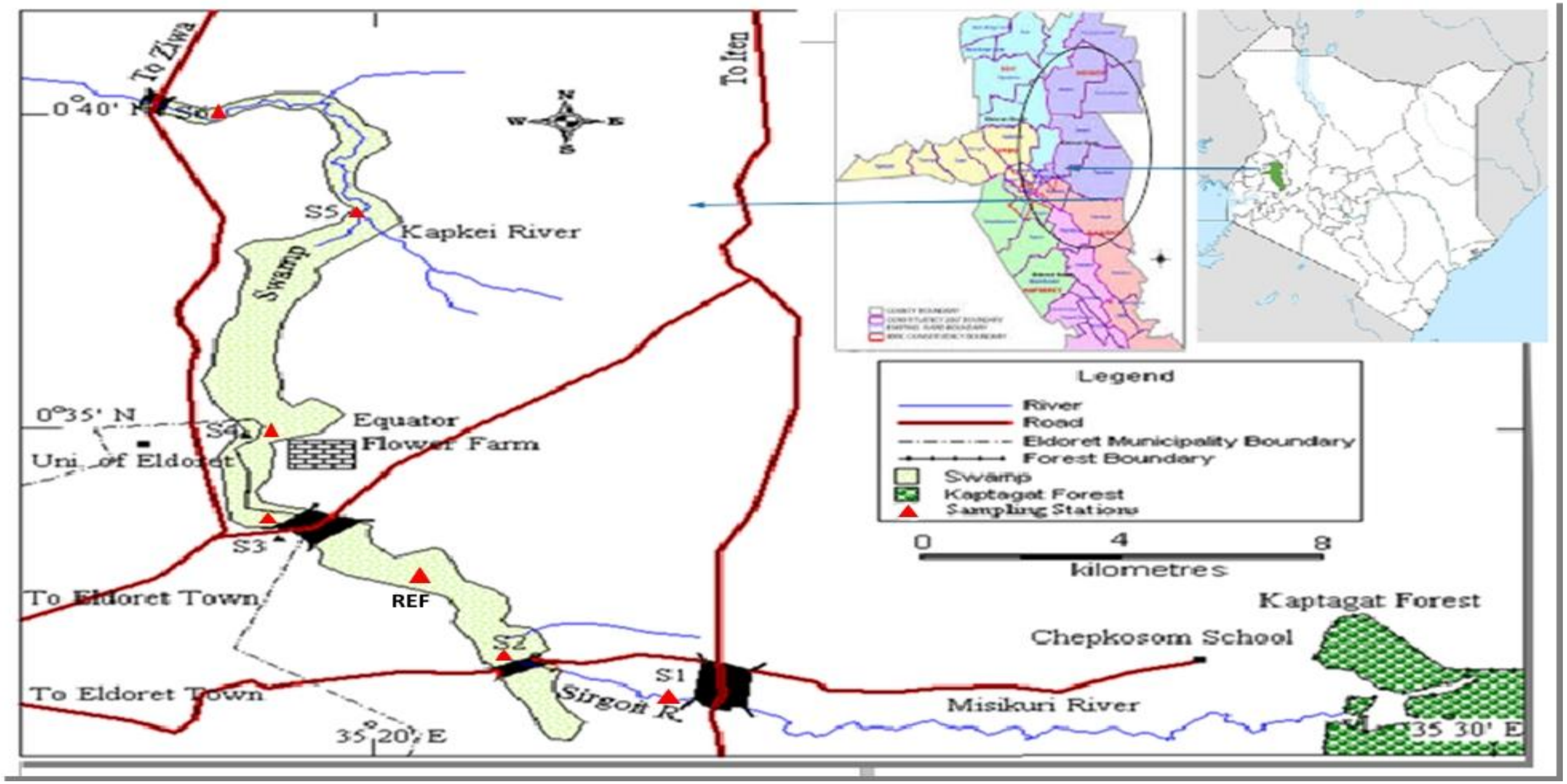

Figure 1: Chepkoilel River Swamp showing the Sampling Stations. Inset: Map of Kenya and Uasin Gishu County (Source: Author, 2015)

3.2 Study Sites

The study area was stratified into six stations on the basis of the types and intensity of human activities as follows.

Station 1 (S1)

The station (Figure 2) was located at Koilel Bridge. This is the point where the swamp begins and *Cyperus sp* dominated except *Cyperus papyrus*. The catchment of this area is dominated by large wheat farms with a clear buffer zone.

Figure 2: Station 1: Showing (a) Point where the river enters the swamp (b) Vegetation (Source: Author, 2015)

Station 2 (S2)

This is the point where *C. papyrus* begins to appear along the river. At this station there is human settlement, crop cultivation (maize and kales), animal grazing and plantations of eucalyptus trees.

Station 3 (S3)

This station was located at Marura Bridge along Eldoret - Iten road (Figure 3). There is a shopping centre at this station resulting to high human presence. Various substances originating from the shopping centre such as wastewaters, and packing papers are deposited into the swamp.

Figure 3- Showing, (a) Car wash and (b) Cattle grazing at Marura Bridge (Station 3) (Source: Author, 2015)

Station 4 (S4)

Station 4 (Figure 4) was placed at the University of Eldoret fish farm. At this station the wetland was protected or buffered from external disturbance due to restriction of access.

Figure 4: Showing, (a) fish ponds, (b) foot path leading to the swamp at Station 4 (Source: Author, 2015)

Station 5 (S5)

The station was located to the left of Limnyomoi Primary School about 600 m after the discharge point of the University of Eldoret's sewage treatment ponds. (Figure 5).

Figure 5: Showing (a) Maize farms and (b) Irrigated vegetable farms adjacent to the swamp at station 5 (Source: Author, 2015)

Station 6 (S6)

This station was placed on the right side of Kaprobu Bridge along Eldoret – Ziwa road (Figure 6). Beyond this point the swamp disappears and the river is again large as it is at station S1. There are large farms of wheat and maize at the catchment.

Figure 6: Showing (a) Footpath leading to the swamp, (b) River channel and (c) Emergent vegetation along the channel at Station 6. (Source: Author, 2015)

3.3 Sampling

3.3.1 Macrophyte Uses

Data on macrophyte uses was collected for three months using various participatory techniques, including in-depth interviews of Chepkoilel river swamp community, key Informants, and direct observations. Similar methods were used by Obiero *et al.,* (2012) to study community perception on lake recession in the Nyando wetlands. A total of 300 respondents, 50 key informants and 250 others were selected among the riparian community.

From each category respondents were picked on the basis of availability and accessibility (Convenience sampling). The key informants in this study were herbalists, area Chiefs, shopkeepers, and macrophyte harvesters.

The other respondents were grouped into high income earners, middle income earners and low income earners in the ratio 1:2:2. The high income earners were characterized by possession of permanent houses, assets such as personal vehicles and farm machinery and large tracts of land. Those with grass thatched houses; small pieces of land and no machinery of any kind were grouped as low income earners. Those respondents with semi-permanent houses and fewer machinery than high income earners were grouped under the middle income earners. This categorization was done in consultation with the area Chiefs.

Two different questionnaires (Appendix 2) were designed and administered for this research, one for the key informants and the other for the other respondents. In each questionnaire type there were both closed and open-ended questions. The questionnaires were self-administered to avoid ambiguity and no answer responses.

Questions administered to the respondents mainly touched on age, gender, occupation, use of macrophytes, frequency of use, use preference and reason for the preference. The herbalists were specifically interviewed on macrophyte usage in curing diseases.

3.3.2 Composition, Diversity and Abundance of macrophytes

In order to capture spatial variation in macrophyte community structure, data was collected for six months at the sites corresponding to the different human disturbance along the wetland.

Triplicate quadrats of $5m^2$ were marked at each site for plant community attribute assessment. From each quadrat, three sub-quadrats of $1m^2$ were further marked where macrophytes were identified to species level and the count of each species recorded.

Voucher specimens of each species were deposited at the University of Eldoret herbarium for verification and future reference. Identification of macrophytes was done according to keys developed by Agnew and Agnew (1994); Beentje (1994); Ibrahim and Kabuye (1987); Haines and Lye (1983); Clayton (1970, 1974, 1982). The plants were further categorized using non-taxonomic groupings. The groupings included categories such as herbs, grasses, sedges, shrubs, creepers, trees, climbers, and algae. The other grouping used was macrophyte habitat where the macrophytes were either classified as terrestrial, semi-aquatic and aquatic plants. Aquatic plants are those that occurred in permanently wet habitats whereas terrestrial plants were those located on dry land adjacent to the wet habitats.

The Shannon-Wiener diversity index was used to evaluate macrophyte diversity in all the sampling stations along Chepkoilel river swamp. The index measures the average degree of uncertainty of predicting the species of a given individual picked at random from a community. It is calculated using the standard equation (Magguran, 1988)

$$H' = -\sum ((n/N) \times \ln (n/N)) \quad \text{.................................... (Equation 1)}$$

Where n = number of macrophytes of a species

N = total number of macrophytes in a station.

ln = the natural logarithm

The evenness index (e) is a measure of how evenly the numbers of species are distributed in a station (Magguran, 1988) Theoretically, in an ideal stream maximum evenness is attainable when (e) is equal to 1.0, meaning that each taxa is equally represented in the population.

The index was calculated as:

$$e = H' / \ln N \quad \text{………………………………………………… (Equation 2)}$$

Where H', and N are as used in the Shannon-Weiner Index equation above, and 'ln' is the natural logarithm.

Relative abundance (R.A) is a composition measure that provides information on the make-up and the relative contribution of the populations of macrophytes to the total composition (Barbour *et al*., 1995). This was calculated using the formula:

$$RA = \frac{\text{Number of individuals of one grouping}}{\text{Total number of individuals in a station}} \times 100 \quad \text{……… (Equation 3)}$$

3.3.3 Physico-chemical parameters and Nutrients

Physico-chemical parameters

Physical and chemical parameters were measured in triplicates at each station. Conductivity was measured *in situ* using conductivity meter (OAKTON^{R}, Model WD-35607-10, Singapore), whereas the temperature and pH were measured *in situ* by a combined pH-and-temperature-meter, (OAKTON^{R}, Model pH/Mv/°C METER, Singapore).

The Winkler titration method (APHA, 1998) was used to determine dissolved oxygen (DO) and biological oxygen demand (BOD). Two sets of triplicate water samples were collected in glass stoppered bottles at each sampling station. The first set used to determine DO was fixed using 2 ml Manganous Sulphate followed by 2 ml of Winkler's reagent.

Dissolved oxygen was determined by titrating 150 ml of sample with a standardized 0.025M Sodium thiosulphate solution. The amount of dissolved oxygen was calculated using the formula:

$$\text{DO (mg/L)} = \frac{C_v C_b \times 8000}{S_v \times \frac{S_b - (a+b)}{S_b}} \quad \text{…………..……. (Equation 4)}$$

where : C_v = the volume of thiosulphate used

C_b = concentration of thiosulphate

S_v = volume of sample used

S_b = volume of manganese+ Winkler's reagent added and

8000 = a constant

Water samples for BOD were wrapped using aluminium foil immediately after sampling, stored in a dark box and transferred to a dark cabinet in the laboratory. On the fifth day the amount of DO was calculated as explained above. BOD was derived using the formula below;

$$BOD = DO_1 - DO_2 \quad \text{…………………………………. (Equation 5)}$$

Where DO_1 is the DO at the day of sampling while DO_2 is the DO after the fifth day.

Nutrients

Water samples for total phosphorus and total nitrogen determination were collected in triplicates during each sampling occasion using 250 ml bottles, fixed immediately using 1ml concentrated Sulphuric acid and transported to the laboratory where they were analyzed according to standard methods (APHA, 1998).

Total Nitrogen and Phosphorous was determined according to persulfate digestion method. The temperature DRB200 Reactor was set to 105 °C then three vials were cleaned and dried for use in this experiment. Using a funnel the contents of total Nitrogen persulfate Reagent Powder Pillow and total Phosphorous persulfate Reagent Powder Pillow was added to two HR Digestion Reagent vials. To one of the vials 0.5 mL of sample of total nitrogen persulfate Reagent Powder Pillow was added and to the other, 0.5 mL of deionized water was added to a prepared blank and the same was done for phosphorous. To the third vial only the sample was put into it. All the vials were capped and shaken vigorously for at least 30 seconds to mix. The vials were put in the reactor and left for exactly 30 minutes. The contents blank vial was put into the spectrophotometer and reading taken. This was followed by the sample vial with reagent and finally the one without reagent.

Total nitrogen (TN) and Total Phosphorous was (TP) was calculated using the formulae:

TN in mg/L = F (E_1 sample – (E_0 + E_{B1});

$$F = \frac{\text{Sample concentration in mg/l}}{E1 - EB1}$$ …………………………………… (Equation 6)

E_0 = absorbance of sample without reductant

E_{B1} = absorbance of distilled water + reagent

E_1 = absorbance of sample with reagent

TP in mg/L = F (E_1 sample – (E_0 + E_{B1})

$$F = \frac{\text{Sample concentration in mg/l}}{E1 - EB1}$$ ………………… (Equation 7)

E_0 = absorbance of sample without reductant

E_1 = absorbance of sample with reductant

E_{B1} = absorbance of distilled water + reagent

3.4 Index of Biotic Integrity (IBI) Development

For IBI development, metrics for the wetland health assessment were selected and tested using Box and Whisker plots (Aura *et al.*, 2009). All the metrics that met the test criteria of non-intersection of all or some 50% of the whiskers were used to develop the index. The metrics selected (Table 1) include relative number of vascular plants, number of non-vascular plants, number of grass and grass-like plants, number of sedges, presence or absence of bladderwort, the relative abundance of aquatic plant species, diversity of plants with persistent litter, diversity index, evenness index, macrophyte abundance, and ratio of trees to other macrophytes (Achieng, 2011 and Aura *et al.*, 2009).

Table 1- Description of some metrics used for IBI Development (Adapted from Melzer, 1999)

Metric	**Description and methodology**	**Expected response to disturbance**
Vascular plant diversity	Based on ecological principle that integrated and stable natural communities have greater richness. The total number of genera at each site plus diversity index was used.	Decrease
Non-vascular plant diversity	Non-vascular plants like mosses, liverwort, lichens and macroscopic algae depend on a healthy aquatic environment for reproduction hence sensitive to changes in environment	Decrease
Grasses and grass-like plants	A healthy wetland supports several grasses and grass-like plants	Decrease
Percent sedge cover	Sedges are especially sensitive to changes in wetland hydrology	Decrease
Presence of bladderworts	Bladderworts are carnivorous plants that feed on microscopic invertebrates. Their presence suggests good health	Decrease
Relative Abundance of true aquatic plants	Many of these plants float or are just below the water surface. They are sensitive to the quality of the aquatic environment	Decrease

Macrophytes with persistent litter	A higher cover of these plants means slower nutrient cycling and lower biodiversity. A low abundance of these plants suggests rapid nutrient and mineral cycling.	Increase
Shannon Weiner diversity Index	A higher value for this index shows good health.	Decrease
Evenness Index	A non- disturbed station is perceived to be evenly distributed thus a higher value depicts little or no disturbance	Decrease
Total Plant abundance	This measures the total count of the plants present at a given station. Due to extractive and destructive nature of man low abundance is an indication of human disturbance	Decrease
Tree: Macrophyte ratio	This is a ratio of tree species to other macrophytes within the swamp.	Increase

The 1, 3, 5 scoring system as used in developing IBIs (Karr, 1981; Kerans and Karr, 1994; Barbour *et al.,* 1999; Raburu *et al.,* 2009) was used depending on whether the value at a station slightly deviates from, or deviates largely from the reference values (Karr and Chu, 1997; Barbour *et al.,* 1999). For those metrics that decrease with disturbance; a score of 5 was made for values above 75% of the reference site value, a score of 3 for values between 50% and 75% and a score of 1 for values below 50%.

For the case of metrics that increase with disturbance, a score of 5 was made for values below the 25% of the reference value, a score of 3 for those between 25% and 50% of the reference site value and a score of 1 for those above the 50 percentile value of the reference site value. The values were then summed up to get the final

IBI score per site and further categorized into either excellent, moderate or poor (Table 2) depending on the total IBI value score

The percentage of IBI score was calculated as follows:

% Score = $\left(\frac{Total\ score\ per\ site}{Maximum\ possible\ score}\right) \times 100$ ………………….. (Equation 8)

Table 2: The scoring and Categorization scale (Adapted from Achieng', 2011)

IBI Score	Wetland Health Assessment
Above 75%	Excellent
60% - 75%	Good
50% - 59%	Fair
30% - 49%	Poor
Below 30%	Very Poor

3.5 Data analysis

Statistical analyses were performed using SPSS, version 20 for Windows. Descriptive statistics was used to analyze the data generated from the questionnaires. One-way analysis of variance (ANOVA), (Zar, 2001) was used to test for differences between stations for macrophyte abundance and water quality parameters at 95% confidence levels. The data on abundance was transformed, $\log_{10}$ (X+1), prior to doing ANOVA test to meet the statistical criteria for normality. Multiple comparisons of means were done using Duncan's Multiple Range Test (DMRT) to distinguish the specific stations that differed significantly from one another (Sokal and Rohlf, 1981).

CHAPTER FOUR

RESULTS

4.1 Respondents Characteristics

The respondents for this study were drawn from both genders with 60% males and 40% females. The respondents further represented several age groups with those between 31 years and 40 years dominating (32.0%). Those above 50 years were only 4.0% (Figure 7).

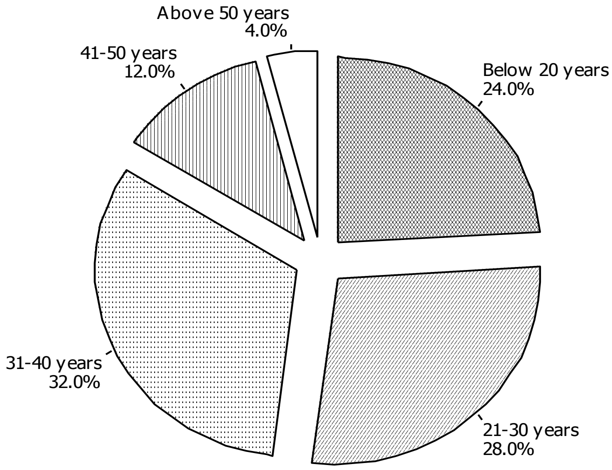

Figure 7: Age categories of respondents interviewed

With respect to the respondents education level 32.7% of the respondents were secondary school leavers and a small proportion (5.7%) holders of a postgraduate degree. (Figure 8)

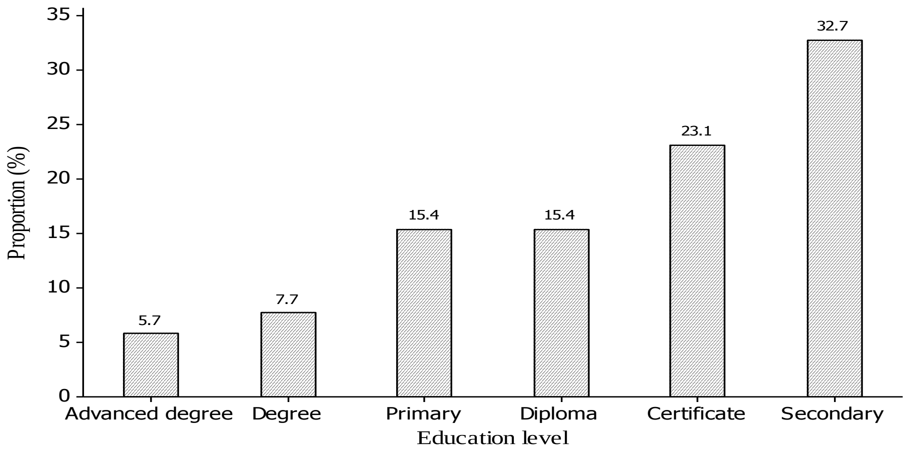

Figure 8: Proportion of respondents with different levels of education

4.2 Macrophyte Uses

Twenty uses were identified from the study. Most respondents use the swamp as a source of fuelwood (62.5%), wild fruits (41.7%), and for animal grazing (35.6%). Thirty three point three percent use it for medicinal purposes. Fencing and building small shops are also common uses by over 20% of the respondent proportion (Figure 9).

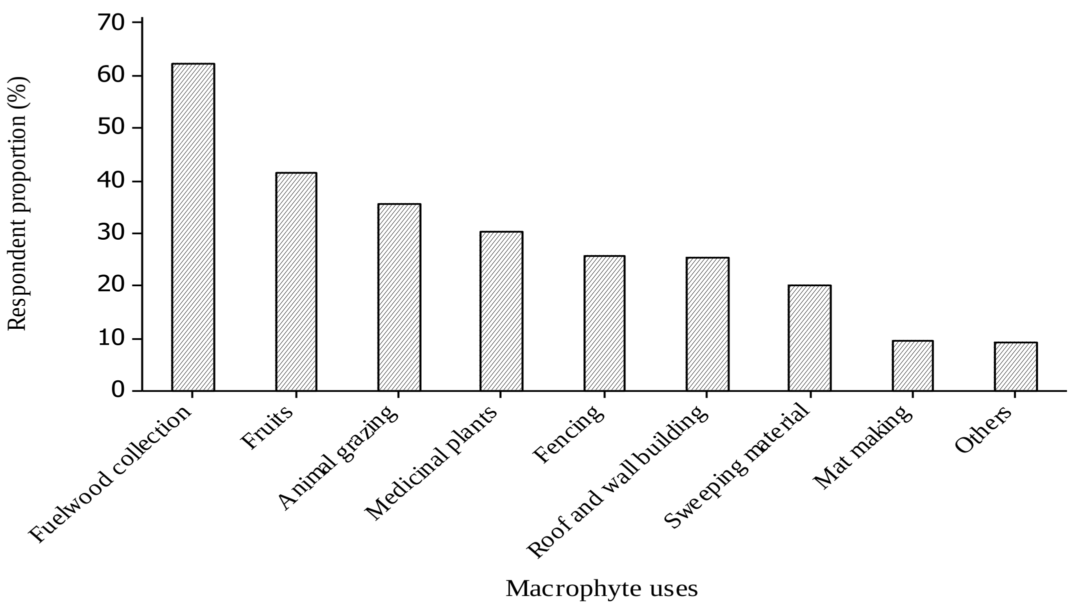

Figure 9: Macrophyte uses by the respondents

The main source of energy for the people was fuel wood, which was used by 62.5% of respondents. Majority of those who collect fuel wood from the swamp (83.7%) use it for domestic purposes while the remaining 16.3% use it for commercial purposes, either selling it directly or use it in their small food kiosks to supplement charcoal which is becoming expensive day by day.

Some respondents used fuel wood though they did participate in its actual collection. They instead bought it from fuel wood vendors who probably got it from the swamp or other forests supporting the swamp like Kaptagat forest. About two-thirds (70%) of fuel wood collectors collected fuelwood once or twice a week, while the rest did so more often.

Almost all (97.5%) of the respondents who grazed their livestock in the swamp did it on a daily basis except a few who occasionally practiced zero grazing. The animals commonly grazed in the swamp were sheep and cattle. Generally, men of youthful age take part in animal grazing or tethering. Over 30% of the respondents used various plants from the swamp for medicinal purposes to treat both human and animal diseases. Some of the diseases treated included stomach disorders, skin problems, headache, heart burn, tooth ache, chest pain, malaria and de-worming. (Table 3). Over 60% of those who used medicine obtained medicinal plants from the swamp plants obtained it from herbalists but did not know the exact plants used. Twenty five percent of the respondents knew some of the plants used but not the dosage and frequency of use and hence consulted the herbalists for assistance.

Table 3: Medicinal plants found within and used by the riparian community along Chepkoilel River swamp

	Common Name	**Scientific name**	**Local name**	**Part used**	**Diseases used to treat**
1	Sodoms apple	*Solanum incanum* L.	Lobotik	Fruit	Mouth ulcers
2	Castor plant	*Ricinus communis* L.	Maniyek	Seeds, leaves	Rushes, colon cleaning
3	Commelina	*Commelina africana* L.	Chepseper	Stem	Wounds
4	Aloe vera	*Aloe barbadensis* L.	Tengeretwet	Stem	Pimples,
5	Black Jack	*Bidens pilosa* L.	Kipkotiwet	Leaves	Wounds
6	True indigo	*Indigofera hombei* L.	Parkelat	Roots	Tooth ache
7	Round-leaved vine	*Cissus rotundifolia* L.	Cherorowet	Roots	Amoebiasis, typhoid, and female infertility.
8	Coffee senna	*Senna occidentalis L.*	Chema	Leaves	Malaria

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Table 4: Use preferences and reasons for the preference

Use	Reasons
Animal Grazing	• Lack of alternative grazing field due to extensive crop cultivation • Availability all year round • Variety of grasses that is good for animal health
Fuel wood collection	• Cheaper source of Energy • Easy to use since it is the traditional source of energy for cooking • Readily available • Rising economic value and demand from food kiosk operators
Medicinal Plants	• High cost of medical care in public and private hospitals • Little trust in public health care (negligence and insufficient drugs) • Ability to cure ailments brought about as a result of taboos and traditions that cannot be handled in hospitals.
Papyrus Harvesting	• Economic and aesthetic value of the products (Mats, roofing restaurants and food kiosks) • High cost of conventional and modern fencing materials • Ability to regenerate soon after harvesting

4.2.1 Relationships between Economic category and macrophyte uses

The high income earners used macrophytes for feeding their animals more than any other use whereas the middle income earners mainly used the macrophytes for animal grazing, as fuel wood and for fencing (Figure 10). The low income earners had several uses for the macrophytes. A big proportion of them used them for fuel wood, mat making and for medicinal purposes.

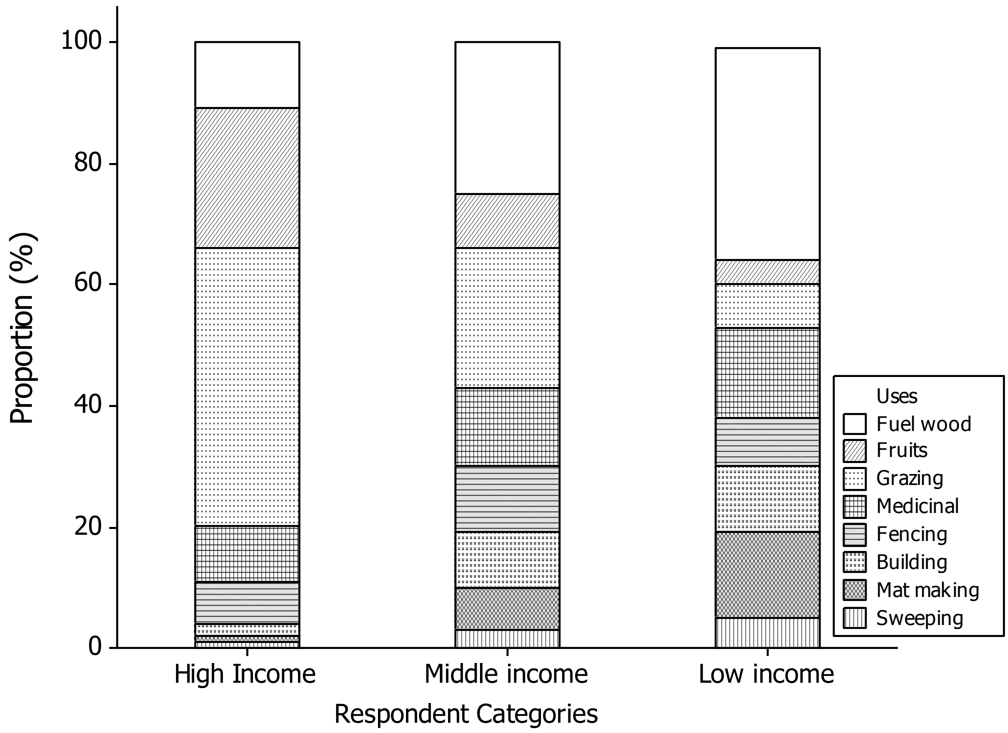


Figure 10: Use - Respondent category association within the riparian community of Chepkoilel River swamp

4.3 Macrophyte diversity and Abundance

4.3.1 Macrophyte Diversity

A total of 108 plant species belonging to 86 genera and 35 families were identified within the Chepkoilel river swamp during the study period (Table 5). Family Poaceae had the highest number of species (20), followed by Asteraceae and Cyperaceae which had a total of 15 and 13 species respectively. The genus *Cyperus* had the highest number of species (5) viz. *C. papyrus, C. rotundus, C. rigidofolius, C. nitidus, C. triandra, and C. laevigatus*.

Table 5: Species Distribution of species collected among plant Family in Chepkoilel river swamp

Family	No. of Species	Family	No. of Species	Family	No. of Species
Acanthaceae	3	Euphorbiaceae	1	Poaceae	20
Amaranthaceae	1	Fabaceae	4	Polygonaceae	4
Asclepiadaceae	1	Hydrocharitaceae	1	Potamogetonaceae	1
Asteraceae	15	Lamiaceae	6	Rosaceae	1
Basellaceae	1	Lentibulariaceae	1	Rubiaceae	1
Branchieriae	1	Linaceae	2	Solanaceae	2
Fabaceae	1	Malvaceae	3	Typhaceae	2
Chlorophyceae	1	Onagraceae	2	Umbeliferaceae	2
Commelinaceae	3	Orchidaceae	1	Verbenaceae	2
Convolvulaceae	1	Oxathdaceae	1	Vitaceae	1
Cucurbitaceae	2	Papilionaceae	5	Xanthornhoeceae	1
Cyperaceae	13	Phytollaceae	1		
Total	108				

Station 1 had the highest number of species followed by S6 while the lowest number of species was recorded at S3 (Table 6).

Table 6: Plant distribution per station

Stations	Number of Families	Number of Genera	Number of Species
S1	32	74	83
S2	14	21	31
S3	9	16	19
S4	26	38	49
S5	22	27	33
S6	29	71	76

The results for diversity and evenness indices are shown in Figure 10. Species diversity was highest at S1 (2.71 ± 0.15) and differed significantly ($p \leq 0.05$) from all the other stations except S6. Station 3 had the lowest diversity (1.32 ± 0.21) and it varied significantly $p \leq 0.05$ from all the other stations except S5. Species diversity in stations 2, 4 and 5 did not vary significantly $p \leq 0.05$. The evenness index was highest at S6 (0.39 ± 0.07). However, it did not vary significantly ($p>0.05$), from that in the other stations except S3 which had the lowest value of 0.18 ± 0.03.

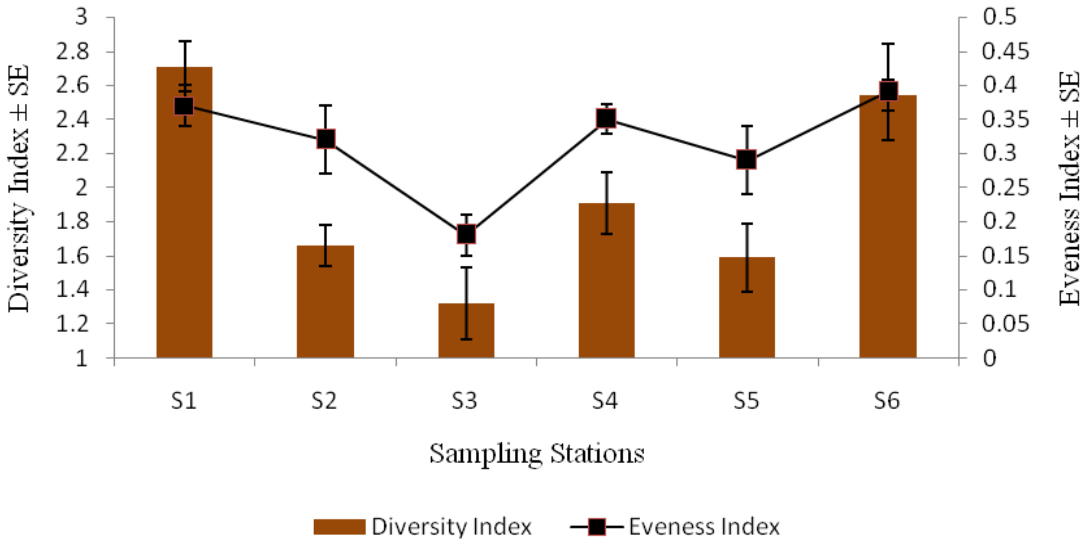


Figure 11: Shannon-Weiner diversity index and Evenness index per sampling station along Chepkoilel River swamp

For non-taxonomic groupings, the herbs had the highest number of species (41) followed by the grasses (20 species). Sedges and shrubs had 13 and 17 species respectively. The creepers, trees, climbers, and algae had 6, 5, 3 and 2 species respectively. In terms of relative abundance, the grasses accounted for 37.4% while algae and climbers accounted for 1.2% and 2.7% respectively (Figure 12).

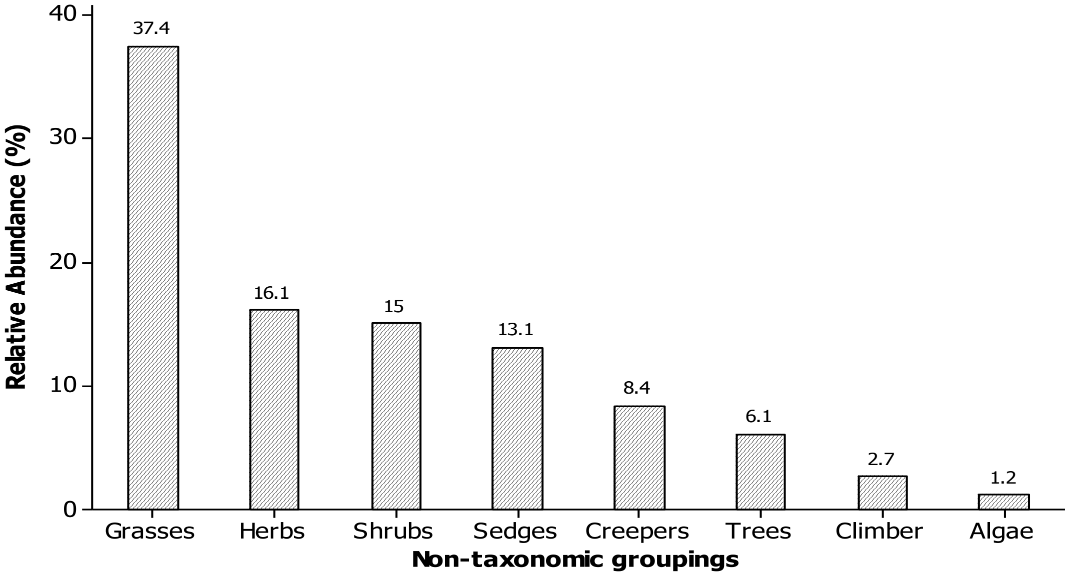


Figure 12: Relative abundance of macrophytes within Chepkoilel river swamp

Most macrophytes were terrestrial while the semi aquatic ones were the least in terms of number of species. Station 4 had the highest number of true aquatic plants (Figure 13) while S3 had the lowest. The number of terrestrial plants was highest in S1 and lowest in S3.

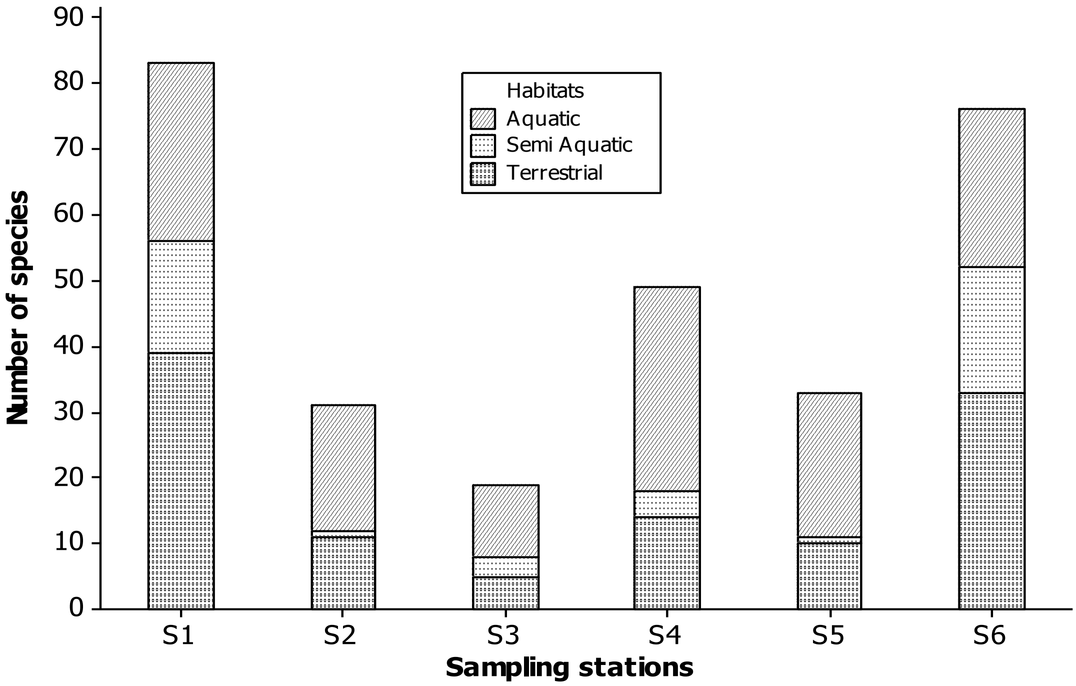


Figure 13: Composition of macrophytes per station in terms of their habitats along the swamp

Station S1 had the highest total number of macrophytes with relative abundance of 1,204 which accounted for 25.2% followed by station S6 which a total macrophyte abundance of 1,019 plants. Station 2 had the lowest total abundance among the stations sampled (483 plants). Stations S3 and S4 had total macrophyte abundances of 512 plants and 943 plants respectively.

There were significant differences in total abundance between the stations (F = 169.302, p = 0.000). Total plant abundance in S1 was significantly higher ($P \leq 0.05$) than in the other stations. Multiple comparisons of abundance between the various stations along the swamp (Table 6), station S1 varied significantly from all the stations at 95% confidence limits. Station 6 had the second highest total abundance but it did not differ significantly ($P \geq 0.05$) from that in S4. Station 2 had the lowest abundance which did not differ significantly from that in S3.

4.4 Water Quality

4.4.1 Physico-chemical parameters

Results on physico-chemical parameters are shown in Table 7 below. Temperature did not vary significantly between the stations along the swamp ($p > 0.05$) and it ranged between 18.7 ˚C and 22.7 ˚C with the highest value at S3 and the lowest at S5.

Dissolved oxygen and BOD differed significantly ($P \leq 0.05$) between the sampling stations. Station 1 had the highest DO concentration which was significantly different from that in the other stations except S6 (Table 7). The lowest DO concentration was recorded at station 3 and it did not differ significantly from that in station 5. BOD was highest at station 6 and lowest at S3. It was significantly different from that recorded in all the other stations. Conductivity and Total Suspended Solids were both highest at S3 and lowest at S1 and the variation was significant. The pH showed significant spatial variation along the swamp with lowest and highest values recorded in stations 3 and 6 respectively.

Table 6: Physic-chemical parameter values (Mean ± SEM) for each sampling station along Chepkoilel River swamp

	Sampling Stations along Chepkoilel river Swamp						Test Statistics	
Parameters	**S1**	**S2**	**S3**	**S4**	**S5**	**S6**	**P**	**F**
Temperature ^{0}C	21.06 ± 1.54^{b}	20.01 ± 1.62^{b}	22.7 ± 2.19^{b}	19.3 ± 1.49^{b}	18.7 ± 1.71^{a}	20.6 ± 1.66^{b}	0.081	14.37
DO (mg/l)	3.98 ± 0.31^{d}	1.67 ± 0.35^{b}	0.42 ± 0.18^{a}	2.87 ± 0.15^{c}	0.51 ± 0.21^{a}	3.85 ± 0.41^{d}	0.003	3.41
BOD (mg/l)	2.65 ± 0.58^{c}	1.19 ± 0.71^{b}	0.39 ± 0.09^{a}	2.77 ± 0.41^{c}	0.48 ± 0.19^{a}	3.26 ± 0.67^{d}	0.027	2.97
TSS (mg/l)	1.27 ± 0.06^{a}	1.93 ± 0.11^{b}	2.29 ± 0.17^{c}	1.97 ± 0.14^{b}	2.21 ± 0.09^{c}	1.44 ± 0.03^{a}	0.001	2.62
Conductivity (µS/cm)	123 ± 7.97^{a}	206 ± 9.43^{b}	279 ± 11.82^{c}	211 ± 8.91^{b}	255 ± 11.51^{bc}	126 ± 6.29^{a}	0.003	78.43
Ph	7.21 ± 0.67^{d}	6.2 ± 0.53^{b}	5.89 ± 0.83^{a}	6.88 ± 0.54^{c}	5.99 ± 0.72^{a}	7.32 ± 0.55^{d}	0.012	2.57

N/B Means with different superscripts across rows are significantly different at $p \leq 0.05$

4.4.2 Nutrients

One way Analysis of variance revealed significant differences between the sampling stations along the Chepkoilel River swamp during the study period in total phosphorus (F = 5.29, p = 0.001) and total nitrogen (F = 1.66, p= 0.007). Total phosphorus concentrations were highest at S3 (1.34 ± 0.23) and lowest at S1 (0.59 ± 0.09) whereas total nitrogen levels were highest at S5 (0.49 ± 0.11) and lowest at S1 (0.072 ± 0.009) (Figure 13).

Total phosphorus did not differ significantly between S1 and S6, Stations, S2, S4 and S5 also did not differ significantly from each with respect to Total Phosphorous and so did S3 and S5 did. Total nitrogen also exhibited an almost similar trend with stations S1 and S6; and S2 and S4 not showing any significant differences with respect to total Nitrogen. Stations S3 and S5 however showed significant differences in total nitrogen levels with all the other stations (Figure 14).

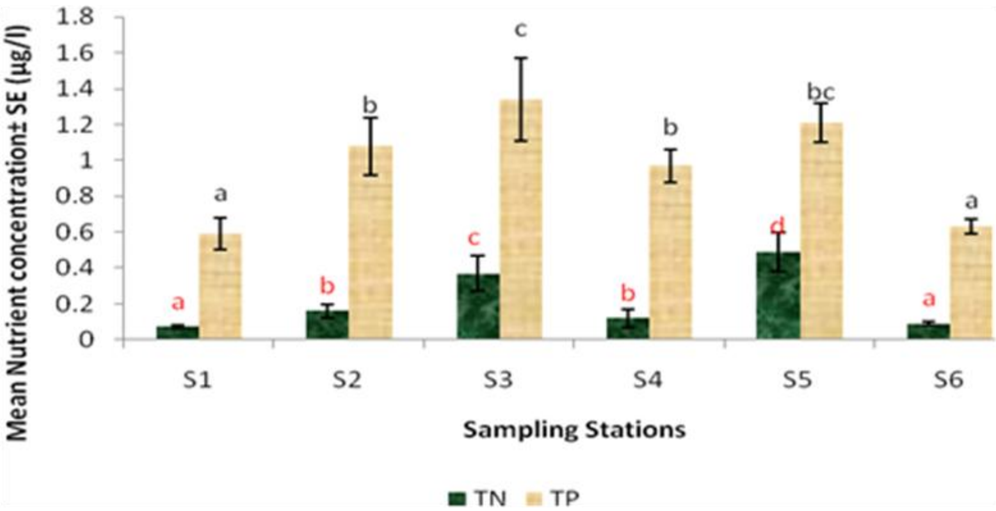


Figure 14: Nutrient (Total Nitrogen and Total Phosphorus) concentrations along Chepkoilel River swamp. Different letters show stations with significantly different means.

4.4.3 Correlation between macrophyte attributes and water quality parameters

The correlations between macrophyte attributes and water quality parameters are presented in Table 8 below. Total abundance, diversity index, eveness index, abundance of grasses, sedges, and vascular plants showed a strong and significant negative correlation with total suspended solids (TSS), conductivity, total nitrogen and total phosphorus. The same attributes showed a strong and significant positive correlation with dissolved oxygen (DO) and biochemical oxygen demand (BOD). Cover of plants with persistent litter on the other hand showed a strong and significant positive correlation with TSS and conductivity. Temperature however showed a unique characteristic. Its relationship with all the attributes was insignificant except with the eveness index with which it had a weak negative significant correlation at 95% confidence level.

Table 7: Correlation coefficients between macrophyte community attributes and water quality parameters within Chepkoilel River swamp

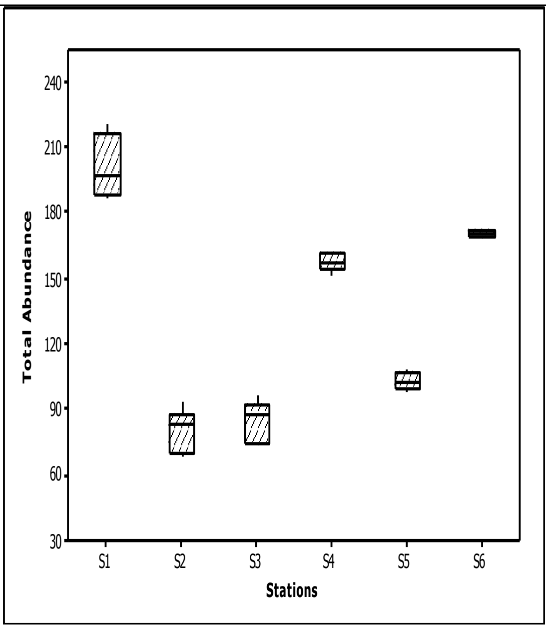
	Water Quality Parameters							
Macrophyte Attributes	Temperature	DO (mg/l)	BOD (mg/l)	TSS (mg/l)	Conductivity (µS/cm)	pH	TN (mg/l)	TP (mg/l)
Abundance	-0.031 (0.856)	0.855** (0.000)	0.823** (0.000)	-0.808** (0.000)	-0.782** (0.000)	0.882** (0.000)	-0.653** (0.000)	-0.845** (0.000)
Diversity	-0.016 (0.925)	0.921** (0.000)	0.842** (0.000)	-0.954** (0.000)	-0.947** (0.000)	0.923** (0.000)	-0.750** (0.000)	-0.946** (0.000)
Evenness	-0.392* (0.018)	0.709** (0.000)	0.704** (0.000)	-0.663** (0.000)	-0.713** (0.000)	0.702** 0.000	-0.622** (0.000)	-0.716** (0.000)
Grasses	0.020 (0.909)	0.914** (0.000)	0.853** (0.000)	-0.913** (0.000)	-0.891** (0.000)	0.932** (0.000)	-0.708** (0.000)	-0.933** (0.000)
Sedges	0.225 (0.188)	0.861** (0.000)	0.768** (0.000)	-0.911** (0.000)	-0.868** (0.000)	0.868** (0.000)	-0.687** (0.000)	-0.986** (0.000)
Plants with Persistent litter	-0.065 (0.707)	-0.284 (0.094)	-0.129 (0.454)	0.535** (0.001)	0.554** (0.000)	-0.247 (0.146)	0.205 (0.231)	0.448** (0.006)
Vascular plants	-0.038 (0.828)	0.882** (0.000)	0.837** (0.000)	-0.852** (0.000)	-0.830** (0.000)	0.902** (0.000)	-0.683** (0.000)	-0.883** (0.000)
Non-vascular plants	0.004 (0.984)	0.558** (0.000)	0.605** (0.000)	-0.415* (0.012)	-0.375* (0.024)	0.620** (0.000)	-0.373* (0.025)	-0.488** (0.003)

***** Shows significant correlation at 95% while ** shows significant correlation at both 99% and 95 %. (Values in brackets are p values)

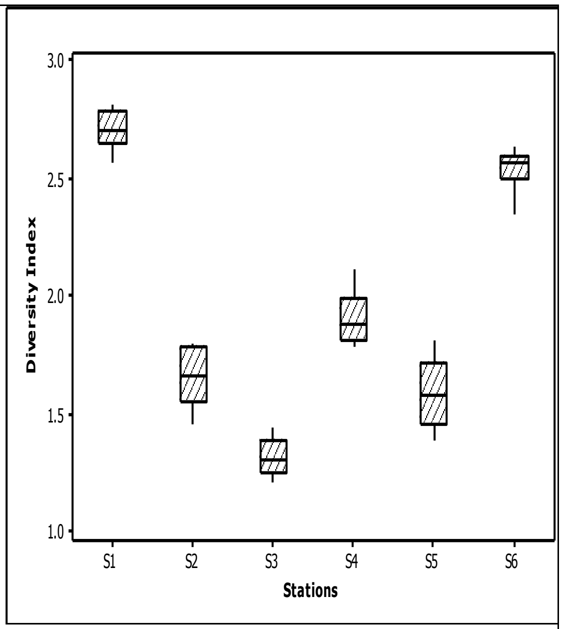
4.5: Macrophyte-Based Index of Biotic Integrity

4.5.1 Metric testing and selection

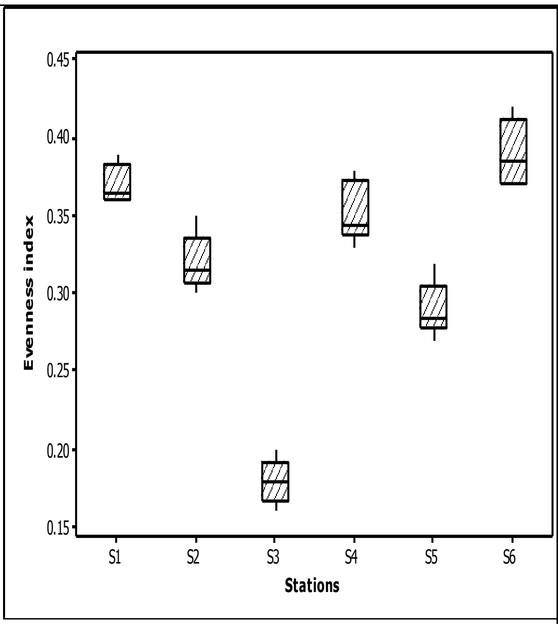
All the metrics were first examined for their relevance within the swamp. The Metric which measured presence or absence of bladderwort was found to be irrelevant since *Utricularia prehensilis* was the only species in this category observed in the wetland and this was in one station (S1) in very low abundance. It was therefore eliminated on the basis of irrelevance. The remaining ten metrics were tested using box and whisker plots for the purpose of developing an IBI. They were examined for intersection of whiskers among the station scores and those that did not have possibilities of common points qualified for use in this study. Out of the ten metrics tested, nine met the criteria (Figure 15) and were therefore used in the final IBI development. The ratio of tree to macrophyte abundance (Figure 15j) did not meet the criteria since the values did not vary between all the stations.



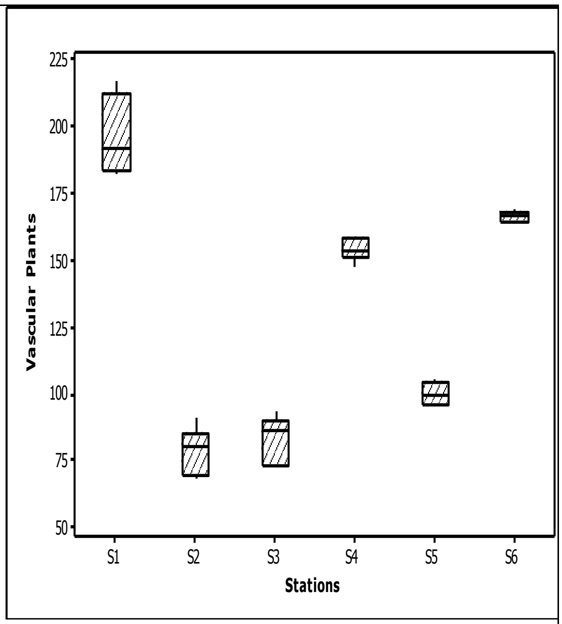
A



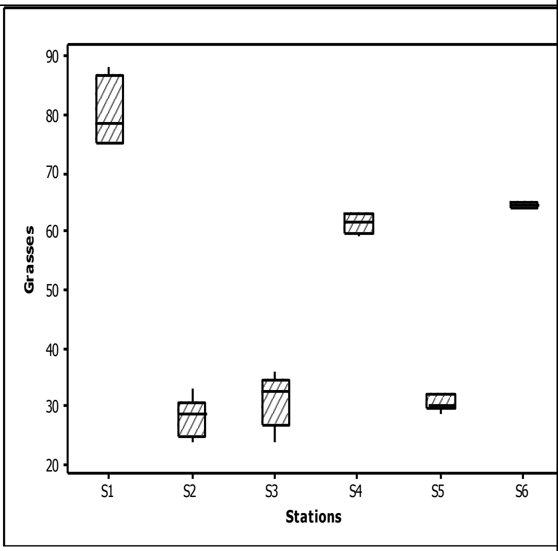
B


Evenness index
0.45
0.40
0.35
0.30
0.25
0.20
0.15
S1
S2
S3
S4
S5
S6
Stations

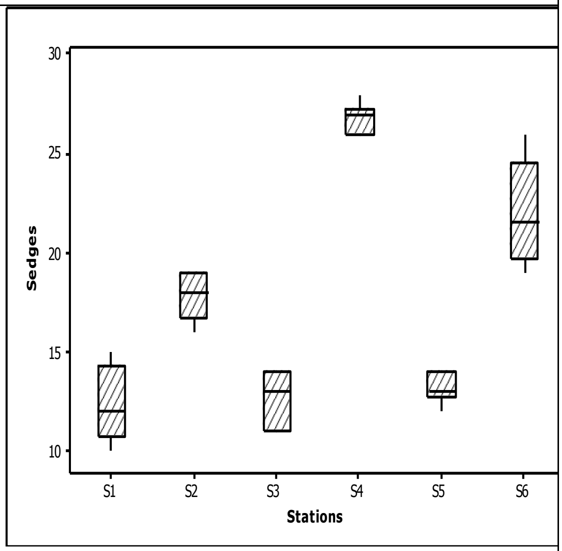
C


Vascular Plants
225
200
175
150
125
100
75
50
S1
S2
S3
S4
S5
S6
Stations

D


Grasses
90
80
70
60
50
40
30
20
S1
S2
S3
S4
S5
S6
Stations

E


Sedges
30
25
20
15
10
S1
S2
S3
S4
S5
S6
Stations

F

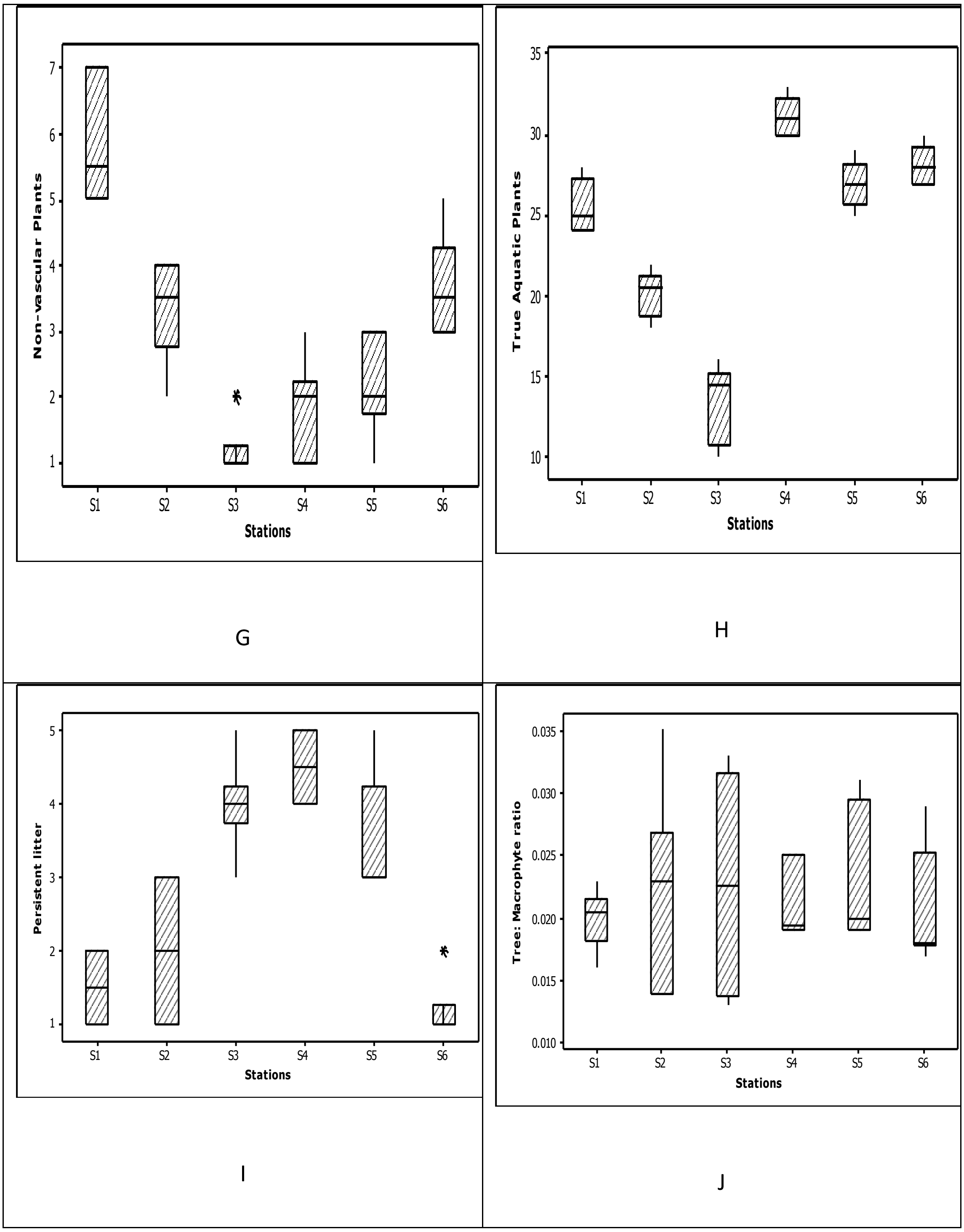


Figure 15: Box and Whisker plots shows intersection points of the six stations to be used to test the variability of metrics.

4.5.2 Reference and Metric Status

4.5.2.1. Reference Site Condition

Reference site is defined as a pristine or undisturbed site within an ecosystem. This kind of site is hardly available in the current time due to overexploitation of different habitats by human activities hence the study resorted to use minimally disturbed site as a reference site (Masese *et al*., 2009). For the purpose of this study, this site was selected after sampling and field observation in order to put into consideration field observations (*apriori)*. After sampling, a station was identified between S2 and S3. This station had low human interference and consequently minimal disturbance (Figure 16).

Figure 10: Showing minimal human disturbance at the reference station along Chepkoilel River swamp (Source: Author, 2015)

Macrophyte attribute values for the metrics that met the criteria of non-intersection of all or some 50% of the whiskers had the highest values at this station (Table 9) and were trisected for use in IBI development following the criteria used by Karr (1981).

Table 8: Values of Macrophyte attributes at the reference station

Metric	Reference Station
1. Total abundance	1, 551
2. Diversity Index	3.47
3. Evenness Index	0.498
4. Vascular plant genera	79
5.Grasses and grass like species	18
6. Sedges	11
7.Non-vascular plant abundance	20
8.True Aquatic plant species	41
9. Plants with Persistent litter	1

4.5.2.2 Metric Status

Metric 1 measured total abundance per station. Based on the reference site threshold, the abundance of 1551 plants was trisected to allow scoring. Any station with a total abundance above 1,158 individuals was given a score of 5; those with plants ranging between 771 and 1,158 plants got a score of 3. A score of 1 was then awarded for stations with abundance below 771.

Metric 2 measured the diversity index (H^I) while metric 3 measured evenness index. A healthy/undisturbed wetland normally has a diversity index of 3.5 while the reference site for this study had an index of 3.47. A score of 5 was thus given to the station with an index greater than 2.63, a score of 3 for stations with an index ranging between 1.75 and 2.63. A score of 1 was given to those stations with an index below 1.75. The maximum evenness index is 1 but the reference site within the swamp had an index of 0.498. An index above 0.375 got a score of 5 while those between 0.25 and 0.375 were given a score of 3. Any station with an index below 0.25 was given a score of 1.

Metric 4 was based on a principle that measures the richness of vascular plant genera; there were 86 genera for the sampled plants in the entire swamp of which 85 were vascular plants. The reference site had 79 genera of vascular plants and when trisected, areas with over 60 plant genera of vascular plants got a score of five; stations with genera between 40 and 60 got a score of 3 while those below 40 got a score of 1.

Metric 5 measured the diversity of grass and grass-like plants. There were 18 species of grass sampled in the reference station. The number of grass and grass-like plants for each station was calculated as a percentage of 18 and scoring done. Stations with 13 or more were given a score of 5 grasses, and stations with 9 to 13 were given score 3 and score 1 for those below 9.

Metric 6 measured the diversity of sedge species. There were 11 sedge species in the reference station and the number at each station was also calculated as a percentage of the reference station value and scoring done. Stations with more than 8 sedge species got a score of 5; those with between 5 and 8 sedge species got a score of 3 and for those with less than 5 sedge species got a score of 1.

Metric 7 measured the number of non-vascular plants (Mosses, liverwort, lichens and macroscopic algae). Only (two) algae species with an abundance of 20 were sampled at the reference station. The score of 5 was given for abundance greater than 15, 3 for an abundance ranging between 10 and 15 while the lowest score of 1 was given for abundance below 10.

Metric 8 measured the cover of the true aquatic plants species sampled in the wetland. From the non-taxonomic classification, 41 aquatic plant species were identified at the reference station. The scoring was calculated as follows. A score of 5 for a site with over 30 true aquatic plant species, 3 for stations with aquatic plants ranging between 20 and 30 whereas a score of 1 for those with less than 20 true aquatic plant species.

Metric 9 measured the cover of plants whose annual leaves and stems decompose very slowly after senescence (low abundance of these plants suggests rapid nutrients and mineral cycling and therefore a healthy wetland (papyrus and trees stems decompose slowly). In total, 5 tree species of this nature were identified in the wetland. However, the reference station only had 1 plant species from this category. A station with less than two of these species got a score of 5, those between 2 and 4 got a score 3 while stations with over four species of this kind got a score of 1.

4.5.3 Total IBI Scores

The general wetland health was evaluated by summing up the total scores for each sampling station and finding an average total score; which represented the overall index of biotic integrity for the wetland. From the summation results, station 1 had the highest score (37) whereas station 3 had the lowest total score of 11 (Table 10).

Table 9: Metric Scores per Station along Chepkoilel River swamp

	Sampling Stations						
Metrics	S1	S2	S3	S4	S5	S6	Maximum
Total abundance	5	1	1	3	1	3	5
Diversity Index	5	1	1	3	1	3	5
Evenness index	3	3	1	3	3	5	5
Number of Vascular plant genera	5	1	1	3	1	5	5
Grasses and grass-like plants	5	1	1	3	3	3	5
Sedges	3	3	3	5	3	5	5
Non vascular plants	5	3	1	1	1	3	5
True Aquatic plants	3	3	1	5	3	3	5
Plants with persistent litter	3	3	1	1	1	3	5
Total	37	19	11	27	17	33	45

4.5.4 Class Categories

After summing the IBI scores, the stations were then categorized into integrity classes as shown in Table 11 below. Station1 was categorized as excellent while S3 as very poor. In general the swamp belongs to the moderate integrity category with a score of 24 (53.3%) which was an average of the total scores from all the stations.

Table 10: Integrity Classes for the Stations sampled along Chepkoilel River swamp

Station	Percentage Score	Category/ Class
S1	82.2%	Excellent
S2	42.2%	Poor
S3	24.4%	Very Poor
S4	60.0%	Good
S5	37.8%	Poor
S6	73.3%	Good
Average	53.3%	Fair/Moderate Integrity

CHAPTER FIVE

DISCUSSION

5.1 Macrophyte Uses

Chepkoilel River swamp is of great socio-economic value to the community. Some of the activities that received high use preference like fuel wood collection, grazing and macrophyte harvesting have great effect on wetland structure and functioning.

5.1.1 Fuel wood Collection

Fuel wood collection in this swamp was high and this could be attributed to its availability and use as compared to other sources of energy among rural dwellers. Wood has multiple uses in the rural areas where the swamp is found and is not only easily available but also relatively cheap (Kalff, 2002). The high costs of alternative energy sources like gas and electricity, existing alternative uses of fuel wood for non-domestic purposes like fish smoking, ceramics, pottery, and preparation of "street food", and widespread cutting of small- and medium-sized branches from wild-growing trees were some of the reasons for fuel wood over-exploitation in the area. The commercial production of charcoal, which is in very high demand in urban areas, could also have led to over-exploitation of fuel wood. Apparently, over-exploitation of fuel wood has resulted in a reduction in the size of fuel wood harvested, and the use of less-preferred materials like twigs, cassava sticks, and tree stumps.

According to Orfanibis *et al.*, (2001), fuelwood provides the main energy source for both rural and urban households throughout the entire West African sub-region, with estimates of about 50% of total energy consumption. Fuelwood plays an important role in human activities like fish smoking and charcoal production by the wetland community. Wuver and Attuquayefio, (2006) in their study showed that the collection of fuelwood in the coastal wetlands of Ghana had assumed such alarming proportions that even certain tree species like *Milletia spp*. which was previously left intact because of its soil fertility rejuvenating qualities, was now being harvested for fuelwood.

5.1.2 Animal grazing

Studies have shown that livestock grazing has numerous effects not only on vegetation but also on other biodiversity like invertebrates and birds (Clary and Kinney, 2002; Matherson *et al.*, 2002). Some of the direct impacts of animal grazing include biomass removal, trampling, soil compaction, introductions and dispersal of seeds, altered micro-topography, and altered soil nutrient status.

Extensive grazing of riparian zones in Australia has resulted in loss of ecosystem function and major impacts on biodiversity (Geothal *et al.*, 2001). Surveys of riparian bird communities showed that, as grazing intensity increased, there was a shift in bird communities, such that small, insectivorous birds dependent on under-storey vegetation, fallen logs and leaf litter declined, while common, open country birds increased (Jansen and Robertson, 2001). The bird species which declined under higher intensity grazing include a number of species which are threatened or declining throughout the agricultural regions of the world.

Animal grazing affects plant diversity, abundance, richness, composition, and biomass which in return affect other dependent organisms. Grazing further alters nutrient levels which affects water quality of an ecosystem (Geothal *et al.*, 2001). All the above factors have had the potential of affecting the integrity, structure and function of Chepkoilel river swamp hence there is need to be checked or regulated.

5.1.3 Macrophyte harvesting

Apart from livestock grazing, almost all the other uses involved direct removal of plants from the swamp. Macrophyte removal has been shown to have ecological impacts on wetland structure and some of the direct impacts include reduced abundance of one species leading to dominance of the less attractive plants (Geothal *et al.*, 2001). Whichever way, the act leads to reduced plant cover which affects not only plant diversity but also the diversity of animals that depend on plants (Ervis and Wetzel, 2002). The high cost of living has forced the community to seek for alternative raw materials such as fencing materials to build shops and small kiosks hence creating a good market for macrophyte harvesters. The Chief of the area reported that there were laws governing the use and conservation of the swamp which prohibited individuals from farming near the swamp to prevent pollution. The laws however have never been implemented.

Plants play a key role in an ecosystem and any change in their diversity and abundance may cause far reaching implications and some ecological impacts of harvesting of macrophytes arrive from plant removal and habitat disturbance during the process and can lead to high turbidity due to suspended substances (Carpenter *et al.*, 1998) and removal of juvenile fishes from the breeding sites. Ervis and Wetzel, (2002) showed that most fishes breed under vegetation and invertebrates not only feed on microscopic plants but also live under the cover of plants. While investigating the impact of macrophyte harvesting on fish (Ervis and Wetzel, 2002) found that over 21,000 fishes were removed by harvesting macrophytes in a year and concluded that this activity has the potential of dislodging plant dwelling fish and invertebrates thus reducing faunal diversity within an ecosystem. The biodiversity within Chepkoilel river swamp reduced and could still reduce further since the macrophyte harvesting practice is on the increase.

5.2 Macrophyte Diversity and Abundance

Chepkoilel river swamp has a higher diversity compared to other swamps where the same study was conducted which is an indication of good health or availability of favorable conditions for plant growth and survival. In a study conducted on the 5 great lakes (Superior, Michigan, Huron and Ontario) in Canada to evaluate 127 wetlands, 94 macrophyte species were recorded (Croft and Chow-Fraser, 2007) while in another study in the South Brazil wetland; a total of 105 plant species were collected (Rolon *et al.,* 2008); in Loboi swamp (Rift Valley; Kenya) a total of 36 vascular plants in 13 families were recorded (Muasya *et al.,* 2004) while in Kingwal wetland (Achieng', 2011) 110 species, 83 genera and 39 families were recorded. From the above information it is evident that despite the current state of Chepkoilel river swamp being affected by human activities, there is still an indication of that there is good health and availability of favorable conditions for plant growth and survival compared to other swamps especially in the reference site.

In this study the low number of species recorded in S3 could be attributed to high human activity within the station. This station was located in an area where a bridge was recently repaired and was characterized by cattle grazing, crop cultivation and car washing.

Such activities not only reduce plant cover but also affect diversity negatively (Wilcox *et al.,* 2002, Croft and Chow-Fraser, 2007). Achieng' (2011) in his study within Kingwal wetland and Rolon *et al* (2008) found reduced diversity at disturbed sites and attributed it to activities such as animal grazing, brick making and macrophyte harvesting which concurs with the observations in this study.

5.3 Water quality

The differences in water quality between the different stations sampled in this study can largely be attributed to land-use practices. Dissolved Oxygen for instance was lowest in station 3 probably due to higher water temperature compared to the other stations sampled. The higher temperatures resulted from high human activities like car washing and use of inorganic fertilizers for crop cultivation and as a result high turbidity came with high human presence. High temperature reduces the solubility of oxygen while turbidity reduces light penetration thus low primary productivity which in turn affects the availability of DO (Kalff, 2002). High water temperature facilitates the release of ions, consequently leading to high conductivity (Bowman *et al*., 2006). During this study, it was recorded that TSS increased with an increase in temperature. A similar observation was made by Bailey *et al*., (1994).

Low pH values were recorded at Stations 3 and 5. This can be attributed to the higher temperatures at these stations due to reduced vegetation cover. High temperatures have been shown to increase evaporation thus inducing re-acidification of aquatic systems (Bowman *et al*., 2006) which in turn lowers the pH. The probable re-acidification due to temperature coupled with accidental oil spillage from the vehicles being washed at these stations of high human activity could be the likely cause of low pH.

Station 1 which had dense vegetation cover recorded the lowest temperature values. Vegetation cover limits direct solar radiation reaching the water thus contributing to minimal fluctuations of temperature. High solar radiation as a result of low macrophyte cover and little water volume can explain high water temperature in the areas experiencing high macrophyte harvesting and grazing as in S3 and S6 (Bowman *et al*., 2006).

The nutrient levels varied significantly among the stations. Station 3 recorded the highest concentrations of total phosphorus which could be due to the difference in the magnitude of animal grazing, and car washing since it was the only station where car washing took place. Total nitrogen was highest at station 5, an area experiencing high crop cultivation since most farmers used fertilizers rich in Nitrogen. It was therefore evident that animal grazing, crop cultivation and car washing have an effect on the concentration of nutrients. Robert and Rankin (1998) similarly obtained higher Nitrogen and Phosphorousn nutrient concentrations at a site that anthropogenic impact seemed to be more.

5.3.1 Correlation between water quality and macrophyte attributes

There was a significant negative correlation between total abundance, diversity index, eveness index, abundance of grasses, sedges, and vascular plants and total suspended solids (TSS), conductivity, total nitrogen and total phosphorus. The negative correlation between macrophyte abundance and nutrient values differed from the findings of D'Aiunto *et al*., (2006) who found a strong positive correlation between the two variables in their study. Results similar to those of D'Aiunto *et al*., (2006) were also reported by Havens *et al*., (1999) and McCormick *et al*., (2001). In all the three studies, the researchers concluded that nutrient loading increased macrophyte diversity and abundance. However the results of this study revealed a contrast to which could be attributed to high nutrient levels along this swamp as a result of animal grazing,car washing and run-off from farms. The low nutrient levels at high abundance stations could also be attributed to the nutrient absorption by the plants that leads to self cleansing.

Cover of plants with persistent litter on the other hand showed a strong and significant positive correlation with total suspended solids (TSS) and conductivity. Litter is part of the material that constitutes total suspended solids thus an increase in litter leads to an increase in TSS value. The results of this study confirms that introduction of litter in water bodies increases conductivity. A study by Masese *et al*., (2009) along Moiben River found high conductivities at disturbed sites and attributed it to presence of litter from the riparian zone and substatnces from agricultural farms.

5.4 Macrophyte-based Index of Biotic Integrity (IBI)

Biomonitoring is the use of biological indicators or organisms whose presence, absence or condition provides information about the quality of an aquatic ecosystem (Mason, 2002). The IBI in this study and its component metrics derived from macrophytes has a potential of informing the resource users and conservation agencies the status of Chepkoilel river swamp. It has shown some similarity with IBI scores that have successfully correlated with human activities like urbanization and agriculture (Carpenter *et al.,* 1998; Griffith *et al.,* 2005), and riparian destruction (Cragg, 1961; Griffith *et al.,* 2005).

This is an indication that the index could probably be a preliminary estimate of the current biotic integrity of all the stations and for the entire swamp.

Metric variability and response of metrics to impaired sites indicated that this IBI responded to the range of biological conditions found in the ecoregion. For example, according to the calculated IBI, station 3 realized 11 points while station 5 obtained 15 points out of the total 45 points. Station 1 had the highest score of 41 which placed it in the excellent integrity class a complete contrast to S3 which was categorized as being of under very poor integrity. The high human presence at stations 3 and 5 such as animal grazing and crop cultivation could have contributed to the low IBI scores.

For management, the metrics are indicative of a changing environment under the influence of intermediate levels of degradation. With increasing human population on the catchment area, the situation is likely to be exacerbated. Therefore, there is a potential need to halt the current trend and improve the habitat integrity at the stations. However, as observed by Omukoto (2007); and Orwa *et al.*, (2012) development of IBI for tropical ecosystems faces the setback of inadequate reference information from which to construct indices. The IBI developed from this study provides the first score results to use this approach in Chepkoilel river swamp.

CHAPTER SIX

CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusion

There were high human disturbances in the swamp with over twenty human activities touching on macrophytes alone. Extractive uses have negative impacts on flora diversity and the overall structure and functioning of the wetland. Most community members prefer to use the macrophytes from this swamp for animal grazing, fuelwood collection, medicinal purposes and papyrus harvesting.

The diversity, abundance and richness of macrophytes along Chepkoilel river swamp reduce with human disturbance. There were significant spatial differences in the distribution of non-taxonomic macrophyte groupings and composition along the swamp. Community attributes such as diversity, abundance and richness have significant relationships with water quality parameters such as nutrients (TN and TP) and physic-chemical parameters such as Dissolved oxygen and conductivity.

The macrophytes within Chepkoilel river swamp responded to human disturbance which made it possible to develop an index for monitoring changes in ecological integrity arising from human disturbance. The response of these macrophytes is thus sufficient to be used to monitor changes in the integrity of the wetland and should be used to monitor it regularly.

6.2 Recommendations

Based on the results and conclusions from this study, the following recommendations are made to help conserve and manage the swamp which is seemingly under immense pressure.

i. The community around Chepkoilel river swamp use its macrophytes in large quantities for various uses which have resulted in degradation of the swamp. Measures should be taken to educate the members of the riparian community to use the macrophytes carefully and avoid overexploitation of the swamp.
ii. Buffer zones should be provided at points of carwash so that the nutrient laden water is first pre-treated before it gets into the main channel.
iii. The developed IBI should be put into use to constantly monitor the swamp and if possible regulate the uses.
iv. Further studies should be done to incorporate all bioindicators for a single unitary index to help in the fine tuning the responses observed in this study hence constant monitoring of the integrity of the swamp.

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Appendix I: List of Macrophytes Identified

Acanthaceae	*Asytacia sp* (L.)	Terrestrial
	*Dyschoriste randicans (*Nees)	Terrestrial
	*Justicia anselliana (*Nees.*)* T. Anders.	Terrestrial
Asteraceae	*Conyza floribunda* H.B.K.	Semi Aquatic
	Conyza gouanii (L.) Willd.	Terrestrial
	Conyza sricta Willd.	Terrestrial
	Conyza suscaposa (L.)	Terrestrial
	Tridax procumbens (L.)	Terrestrial
	Ajuga remota (L.)	Terrestrial
	Bidens pilosa (L.)	Terrestrial
	Helichrysum forskahlii (J.F.Gmel)Hiliard & B.L.Burtt	Terrestrial
	Helichrysum newii Oliv & Hiern	Terrestrial
	Helichrysum schimperi Sch.Bip.	Terrestrial
	Vernonia lasiopus (O. Hoffm.)	Terrestrial
	Vernonia syringifolia (O.Hoffm)	Terrestrial
	Sonchus asper (L.)	Terrestrial
	Sphaeranthus suaveolens (Forssk) DC	Aquatic
Basellaceae	*Basella alba* (L.)	Aquatic
Branchieriae	*Poeceae abysinica* (A. Rich) Munro	Terrestrial
Cyperaceae	*Cyperus papyrus* (L.)	Aquatic
	Cyperus rotundus (L.)	Aquatic
	Cyperus rigidofolius (Steud)	Aquatic
	Cyperus nitidus Lam	Aquatic
	Cyperus triandra (L.)	Aquatic
	Cyperus laevigatus (L.)	Aquatic
	Eleocharis sp (L.)	Aquatic
	Fuirena stricta (Steud.)	Aquatic
	Fibristylis dichotoma (L.)	Aquatic
	Kyllinga bulosa (P.Beauv)	Aquatic
	Schoenoplectus corymbosus (Reichenbach)	Aquatic
Commelinaceae	*Commelina africana* (L.)	Terrestrial
	Commelina beghalensis (L.)	Terrestrial
	Floscopa glomerata (Schult& Schult.f)Hassk)	Aquatic
Caesalpiniaceae	*Chamaecrista mimosoides* (L.) Greene.	Terrestrial
Cucurbitaceae	*Zahneria scabra* (L.f) Sond.	Semi Aquatic
	Momordica faetida (Schumach.et Thonn.)	Semi Aquatic
Convolvulaceae	*Ipomoea tenuilostris* (Rendle) Verdc	Semi Aquatic
Chlorophyceae	*Spirogyra sp* (H.)	Aquatic

Appendix I continued

Euphorbiaceae	*Ricinus cummunis* (L.)	Terrestrial
Fabaceae	*Trifolium baccarinii* (Chiov)	Terrestrial
	Indigofera hombei (L)	Terrestrial
	Sena occidentalis (L)	Terrestrial
Hydrocharitaceae	*Elodea densa* (Planch) Casp	Aquatic
Lentibulariaceae	*Utriculria prehensilis* (E.Mey)	Aquatic
Lamiaceae	*Setureia sp* (L.)	Terrestrial
	Chlorophytum sp (L.)	Terrestrial
Lamiaceae	*Blephilia ciliate* (L.)	Semi Aquatic
	Genlosporum rotundifolium (Roxb)	Terrestrial
	Plectranthus edulis (L)	Semi Aquatic
	Pycnostachys deflexifolia (L)	Semi Aquatic
	Pycnostachys stulmanii (L)	Aquatic
	Ocimum bacilium (C)	Terrestrial
Malvaceae	*Sida cuneifolia (*Roxb)	Terrestrial
	*Sida ovata (*Forsk.*)*	Terrestrial
	Pavonia urens (Cav)	Terrestrial
	Aenanthe palustris (L)	Terrestrial
Orchidaceae	*Disa welwtschi* (L)	Terrestrial
Oxallidaceae	*Oxalis circicnata* (L.)	Terrestrial
Onagraceae	*Ludwigia leptocarpa* (Nutt) Hara	Terrestrial
	Cissus rotundifolia Forrsk) Valh	Terrestrial
Poaceae	*Cynodon dactillum* (L.)	Aquatic
	Eragrostis chalarothyrsus (C.E) Hubbard	Aquatic
	Panicum hymeniochilum (Nees)	Semi Aquatic
	Panicum poeoides (L.)	Aquatic
	Setaria annua (L.)	Terrestrial
	Digitaria scalarum (Schweinf.) Chiov	Terrestrial
	Echinochloa pyramidalis (Lam) Hitch & Chase.	Aquatic
	Echinochloa colona (L.)	Aquatic
	Eleusine indica (L.) Gaetn.	Aquatic
	Bramcheria sp (L.)	Aquatic
	Sporobolus spicatus (T.)	Semi Aquatic
	Themeda triandra (Forssk)	Semi Aquatic
	Cenchrus ciliaris (L.)	Semi Aquatic
	Pennisetum clandestinum (Chiov)	Semi Aquatic
	Tricum aestium (L.)	Terrestrial
	Branchiataria sp (L.)	Terrestrial
	Aristida adoensis (Hochst.Graminae)	Semi Aquatic

Appendix 1 Continued

	Setaria verticillata (L.)	Semi Aquatic
	Cenchrus sp (L.)	Semi Aquatic
	Heteropogon contortus (L.)	Semi Aquatic
Pappilionaceae	*Sesbania sesban* (L.) Merril.	Semi Aquatic
	Desmodium sp (L.)	Semi Aquatic
	Rhynchosia minima (L.)	Terrestrial
	Trifolium cryptopodium (A. Rich)	Semi Aquatic
	Lutonosis sp (A. Rich)	Semi Aquatic
Polygonaceae	*Polygonum amphibium* (Willd)	Aquatic
	Polygonum pulchrum (Willd)	Aquatic
	Polygonum salicifolia (L.)	Aquatic
	Polygonum strigosum (R.Br)	Aquatic
Phytolacaceae	*Phytolacca dodecandra* (L.)	Aquatic
Potamogetonaceae	*Potamogeton richardii (*A.Bennett)	Aquatic
Rubiaceae	*Galium scioanum* (Chiov.Plate)	Terrestrial
Rosaceae	*Rubus niveus* (Poir)	Terrestrial
Solanaceae	*Solanum incanum* (L.)	Terrestrial
	Solanum naucase (L.)	Terrestrial
Typhaceae	*Typha latifolia* (L)	Aquatic
	Typha domingensis (Pers.)	Aquatic
Umbiliferaceae	*Hydrocotyle monticol* (L)	Terrestrial
Verbanaceae	*Lantana trifolia* (L)	Terrestrial
	Verbena bonariensis Bitter.	Terrestrial
Vitaceae	*Amolelicussys abyssinica* (L)	Terrestrial
Xanthornhoeaceae	*Aloe vera* (A)	Terrestrial

Appendix II: Questionnaires

General Purpose Questionnaire

SECTION A – Personal Details

1. Respondent Category

High income [ ]

Middle income [ ]

Low Income [ ]

2. Gender of the respondent

Male [ ]

Female [ ]

3. Age of the respondent

Below 20 years [ ]

21– 30 years [ ]

31– 40 years [ ]

41 – 50 years [ ]

Above 50 years [ ]

4. What is the highest level of education attained?

Primary school [ ]

Secondary school [ ]

Certificate courses [ ]

Diploma courses [ ]

First degree [ ]

SECTION B- Macrophyte uses, frequency of use and use preference

5. What do you use macrophytes from this wetland for? List all the uses in the table below and frequency of Use. You may write below the table if space is not enough

Use(s)	Frequency

Key Informant Questionnaire

Section A – Personal Details

1. Informant Category

Herbalist	[1]
Macrophyte harvester	[2]
Food Kiosk Operators	[3]
Provincial administration	[4]

2. Gender of the respondent

Male	[1]
Female	[2]

3. Age of the respondent

Below 20 years	[1]
21– 30 years	[2]
31– 40 years	[3]
41 – 50 years	[4]
Above 50 years	[5]

Section B- Herbalists Only

Which macrophytes do you use from Marura swamp for medicinal purposes and which diseases/ailments to each plant treat? You may write below the table if space is not enough

Macrophyte (s)	Disease treated

Section C – Harvesters

1. Which macrophyte (s) do you harvest from Marura swamp

   …………………………………………………………………………………………

   …………………………………………………………………………………………

2. For what reason do you harvest the macrophytes from Marura swamp

   For domestic use e.g. fencing, sweeping etc. [1]

   Sell to others [2]

   Process into other products for sale e.g. mats [3]

3. If you sell the macrophytes or the products, which is your market

………………………………………………………………………………………

………………………………………………………………………………………

Section D – Food Kiosk Operators

1 What fuel or energy do you use while preparing your meals?

Gas	[1]
Kerosene	[2]
Fuelwood	[3]
Charcoal	[4]

2 Where do you get your fuelwood or charcoal from?

………………………………………………………………………………………

………………………………………………………………………………………

………………………………………………………………………………………

Section E – Provincial Administration

1 Are there any laws you know either communal or governmental that govern the use of Marura Swamp?

Yes	[1]
No	[2]

2 If yes which ones are they and how are they implemented?

………………………………………………………………………………………

………………………………………………………………………………………