# ASSESSMENT OF THE EFFECTS OF EFFLUENT FROM MUMIAS SUGAR FACTORY ON WATER QUALITY AND PHYTOPLANKTON SPECIES DIVERSITY OF RIVER NZOIA, KENYA

**BY**

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# DECLARATION

## Declaration by the Candidate

This thesis is my original work and has not been presented for any academic award or degree in any other university. No part of this work may be presented, reproduced or published without the prior written permission of the author and/or University of Eldoret.

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# DEDICATION

I dedicate this thesis to my wife Ruth Chebet, my children Anthony, Cynthia, Diana and; my mama, for being a source of encouragement. Moreover, to my late father, who always inspired me to have courage to imbibe into the springs of the foundations of knowledge.

## ABSTRACT

Phytoplankton species diversity were used to assess the effects of effluent from Mumias sugar factory on water quality in Kenya, on River Nzoia downstream of the discharge point in relation to variations in water quality in the course of $1^{st}$ December 2009 to $31^{st}$ March 2010. The study sort to deal with the gap, insignificant exploration of the linkage between physico-chemical water quality parameters and phytoplankton species diversity in the riverine environments of Lake Victoria basin and Kenya. A randomized experimental design and convenient upstream-downstream, independently repeated random sampling design was used to determine response of phytoplankton species diversity to spatial physico-chemical quality of the Nzoia River. EXCEL 2007 for windows 7 and STATISTICA program version 8 were used for univariate and multivariate statistical analysis considered ($\alpha < 0.05$), used. The physico-chemical loads were measured using modified standard methods while phytoplankton species diversity and composition at species and community level was determined using Sedgewick Rafter(S-R) counting chamber. The process ($S_2$) was significantly more polluting than milling ($S_1$) and boiler ($S_3$) while physico-chemical loads upstream ($S_4$) and downstream ($S_6$) sampling stations after treatment and before discharge were beyond the NEMA and WHO limits. Bacillariophyceae had the highest number of species (5). The bioindicators were *Microcystis spp*, *Melosira spp*, *Closterium spp*, *Gomphenema spp* and *Synedra spp* while the most pollution tolerant species were *Euglena spp*, *Navicula spp* and *Nitzchia spp*. *Melosira spp* (464) was the most abundant bioindicator species while *Microcystis spp* (14) were all found at $RN_3$. Shannon-Weiner diversity ($H_s$), Margalef's species richness (d) and Pielou species evenness (J) had similar spatial patterns except Simpson's Diversity Index (1–Ds). All the realized $H_s$ values were in the range of 0-1 while $J < 0.4$, being closer to zero (0) than one (1). A multiple criteria selected four (4) most significant principal components that yielded 99.90% of the total variance of the physico-chemical parameters corresponding to combined industrial (sugar) and domestic effluents; physico-chemical; soil leaching and agricultural run-off process variability. SMLR identified the contribution of each variable with a value of R 0.72, $R^2$ 0. 52(52%), $F_0$ 1.08 > $F\alpha$ 0.003 and $p>0.05$ that confirmed a linear relationship between the variation of water quality explained on the phytoplankton diversity. The 48% may be due to partially decayed organic matters from domestic and sugar effluents discharge, soil leaching or agricultural run-off process variability, an imbalance of free metal availability for phytoplankton due to physico-chemical process and sand harvesting. Findings from the study can enable achievement of a more elaborate biomonitoring programs on the quality of Nzoia River. Management decisions by Mumias Sugar Company, National Environment Management Authority (NEMA) and the government made from the enhanced biomonitoring programs.

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# ABBREVIATIONS

**APHA: A**merican **P**ublic **H**ealth **A**ssociation

**BIS: B**ureau of **I**ndian **S**tandards

**COGEN: C**o**G**eneration

**CPCB: C**entral **P**ollution **C**ontrol **B**oard

**DWAF: D**epartment of **W**ater **Affairs** and **F**orestry

**DV: D**ependent **V**ariable

**ELDOWAS: E**ldoret **W**ater and **S**anitization **C**ompany

**ECMC: E**nvironmental **C**ost **M**anagement **C**entre

**FMENV: F**ederal **M**inistry of **ENV**ironment

**GEF: G**lobal **E**nvironment **F**und

**GIS: G**eographic **I**nformation **S**ystems

**GPS: G**lobal **P**ositioning **S**ystem

**I.V: I**ndependent **V**ariable

**KeSReF: K**enya **S**ugar **R**esearch **F**oundation

**KSB: K**enya **S**ugar **B**oard

**KMFRI: K**enya **M**arine and **F**isheries **R**esearch **I**nstitute

**LBDA: L**ake **B**asin **D**evelopment **A**uthority

**LVBC: L**ake **V**ictoria **B**asin **C**ommission

**LWRRDC: L**and and **W**ater **R**esources **R**esearch and **D**evelopment **C**orporation

**MDSP: M**umias **D**istrict **S**trategic **P**lan of 2008 to 2012

**MVSP: M**ultivariate **S**tatistical **P**ackage

**NEMA: N**ational **E**nvironment **M**anagement **A**uthority

**NRBMI: N**zoia River Basin Management Initiative

**PCA: P**rincipal **C**omponent **A**nalysis

**PGH:** **P**ost **G**raduate **H**ealth

**RoSA:** **R**epublic **o**f **S**outh **A**frica

**SES:** **S**chool of **E**nvironmental **S**tudies

**SOP:** **S**tandard **O**perating **P**rocedures

**UNEP:** **U**nited **N**ations **E**nvironmental **P**rograms

**UNESCO:** **U**nited **N**ations **E**ducational **S**cientific and **C**hildren's **O**rganization

**USEPA:** **U**nited **S**tates **E**nvironmental **P**rotection **A**gency

**WAC:** **W**ashington **A**dministrative **C**ode

**WRC:** **W**ater **R**esearch **C**ommission

**WRMA:** **W**ater **R**esource **M**anagement **A**uthority

**WHO:** **W**orld **H**ealth **O**rganization

# OPERATIONAL DEFINITION OF TERMS

**Algae:** Algae are a group of unrelated simple organisms that contain chlorophyll living in aquatic habitats and in moist situations on land (Hine *et al*., 2004). The study used the term phytoplankton interchangeably with algae and in proportion to the physico-chemical values of sugar effluent discharges in the Nzoia River.

**Assessment:** Assessment is the value or amount at which something is calculated (Walker M.B P, 1990). The study's assessment implied a means of determining the Nzoia River's water quality monitoring activities as indicated in the appendix IV.

**Biota:** These are all the organisms living in a particular region, including plants, animals and microorganisms (Hine *et al*., 2004). The study area's biota was the phytoplankton.

**Bioindicators:** These are particular species or communities, which, by their presence, provide information on the surrounding physical and/or chemical environment at a particular site (Bellinger *et al*., 2010). In this study, some of the phytoplankton species were considered as bioindicators in relation to physico-chemical quality of the Nzoia River.

**Biological monitoring:** Wan (2010) explains biological monitoring as the specific application of biological response for the evaluation of environmental change for purpose of using this information in quality control program. The study's biological response entailed variability of the phytoplankton species diversity due to the Nzoia river physico-chemical quality arising from Mumias sugar effluents discharge.

**Diversity:** This is an Index of the number of species in a defined area, often represented mathematically (Walker M.B.P, 1990). The species and community composition, species indices, relative abundance, and spatial distribution defined the diversity of phytoplankton and hence the health status for Nzoia River.

**Effluent:** This is the liquid or gaseous waste from a chemical or other plant (Walker M.B.P, 1990). Effluent is an out-flowing of water from a natural body of water, or from a man-made structure like an industrial plant (USEPA, 2009). The study's effluents were the sugar liquid waste from Mumias sugar factory and its domestic liquid waste.

**Physico-chemical:** These were the attributes (pH, temperature, Chemical Oxygen Demand (COD), Biological Oxygen Demand ($BOD_5$), Dissolved Oxygen (DO), Electrical Conductivity (EC) and Total Dissolved Solids (TDS) for elements for data of water/effluent quality indicators in the study.

**Phytoplankton:** These are the photosynthetic members of the plankton floating in the waters of sea, river, ponds and lakes (Walker M.B.P, 1990). The study's phytoplankton were the plankton determined in the Nzoia River at the study area.

**Water Quality:** Edwards's aquifer glossary (2011) defines water quality as the chemical, physical, biological, radiological, and thermal condition of water. This was expressed as a comparison between the physico-chemical loads of Mumias factory effluents before and after discharge into the Nzoia River with those of NEMA and WHO.

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Finally, I would like to free every single one person and establishment mentioned in this thesis of any errors of omission and/or commission or any interpretational error(s). In view of these, the author remains exclusively liable.

# CHAPTER ONE
# INTRODUCTION

## 1.1: Background Information

Water as an elixir of life, is an essential natural resource that has declined at a faster rate causing a frightening global problem arising from industrial operations (Mahananda *et al*., 2010 and Mullai *et al*., 2013). Despite its importance, water is the most disappointingly managed resource in the world (Walakira, 2011). The state of management has arisen out of industrialization, urbanization and modern agriculture practices. These practices directly affect water resources quantitatively and qualitatively (Udoh, 2013).

Resultant global adverse effects on water resources threaten human and aquatic ecosystems' health (Meybeck *et al*., 1996a). This effectively reduces its availability while increasing competition for adequate water quality (Montse, 2009). Effluents discharged into receiving waters from the industrial operations and their cumulative adverse effects on the environment have received much interest due to rapid industrialization in contemporary society (Morrison *et al*., 2001). This vastly deteriorates water quality mainly due to unscientific waste disposal of domestic and industrial effluents that are far beyond most rivers' assimilative capacities (Agarrkar and Thombre, 2005 and; Baskaran and De Britto, 2010).

Therefore, indiscriminate discharge of industrial effluent from processing of sugar crops severely depletes water quality in rivers (Carminati, 2008 and Pratiksha *et al*., 2013). Monitoring physico-chemical characteristics of such a water body is vital for both short and long-term evaluation of its quality (Emeka, 2012). The purpose is to preserve the aquatic environment at a certain specified minimum quality.

The lack of knowledge of water resources modellers has resulted in minimal use of bioindicators (Yazdian *et al*., 2014). Furthermore, planning of water resources control and management is a challenge despite water quality assessment being an important tool for pollution control and assessment (Abdullahi *et al*., 2008). The importance is due to continuous monitoring of water quality to protect the river (Mullai *et al*., 2013). However, the effectiveness and dearth of water management measures, has led to water scarcity, its gradual drop and aggravated pollution.

Water pollution has intrinsic connection with anthropogenic activities with water being involved in the cycling of many of the nature's elements (Montse, 2009). These activities do occur in levels far beyond the environment's background levels, resulting in water pollution. Water, apart from being a vital requirement for biotic life and industrial processes, also works as a transport mechanism and a sink for domestic, agricultural and industrial waste causing pollution.

For developing countries like Kenya, surface water pollution is one of the most serious problems (Surindra *et al*., 2010). The sugar effluent discharges are a major component of this pollution and thus a pollution stress. The stress is due to sugar industries not being connected to the municipal sewerage network, dumping into rivers much of its effluents (Millette *et al*., 1991). Arising from this is the oxygen demand and nutrient loading of the water bodies that promote toxic phytoplankton blooms and leading to a destabilized aquatic ecosystem (Morrison *et al*., 2001). Phytoplanktons are one of the most rapid detectors of environmental changes owing to quick response to toxins and other chemicals (Suman *et al*., 2010). The presence or absence of phytoplankton species from the community indicates changes in physico-chemical attributes of an aquatic environment (Rissik, 2009).

Sensitive species are bound to act as bioindicators of water condition that are beyond the tolerance of many other biota used for monitoring (Nwankwo and Akinsoji, 1989 and Valdes-Weaver *et al*., 2006). The resultant pollution stress reduces the number of phytoplankton species but increases the number of individuals (Desai *et al*., 2008). The species diversity is severely affected due to a marked change in phytoplankton community that serves as natural oxygenator of rivers (Ishaq and Khan, 2013).

Phytoplankton remain important in ensuring ecological balance and the basis of the food chain in open water, as some species release toxic substances into the water harming human and other vertebrates (Kitner and Poulickova, 2003 and Rey *et al*., 2004). They are sensitive to ecological alterations, requiring monitoring particularly at species level in determining the health status of the receiving waters. Its water quality due to effluents discharge greatly affects rate of production of phytoplankton (Schindler, 1978 and; Dahl and Wilson, 2000).

Species diversity indices when correlated with physico-chemical water parameters provide one of the best ways to detect and evaluate the effects of pollution on aquatic communities (Margalef, 1958). Although their capacity as indicator species is commonly recognized, there are few studies from Turkey as is typical of the current Kenyan situation (Abuzer and Okan, 2006). Studies in Kenya have remained scarce on the effects of land use on the water quality of riverine environments within the Lake Victoria basin (Raburu and Okeyo, 2005). Species diversity remains one of the most widely adopted metrics for assessing patterns and processes of biodiversity.

The scarce studies imply little consideration being placed on phytoplankton species diversity as alternative to water quality assessment in Kenya. Despite a theoretical consensus on diversity metrics, standardized methods for their measurement are lacking, especially at the scales needed to monitor biodiversity for conservation and water quality management (Chiarucci *et al*., 2011). However, phytoplankton species diversity and physico-chemical water parameters remain an important criterion for evaluating the suitability of water for drinking and other purposes (Ishaq and Khan, 2013). Phytoplanktons are recurrently used as indicators in temperate systems, but not much is known about their application to impacted African tropical systems (Bellinger *et al*., 2006). This is notwithstanding the importance of the phytoplankton as bioindicators of water quality.

Bangladesh had few reports on the effects of industrial effluents on phytoplankton (Tahmida *et al*., 2009). The use of physico-chemical properties of water has been indicated to assess water quality in providing a good impression of the status, productivity and sustainability of such water body (Mustapha, 2008). A better impression would be to link physico-chemical water parameters to phytoplankton diversity. Similarly, there is need to have baseline data that will be of profound importance and necessary for a monitoring program on pollution studies to the aquatic biota of Kenyan inland waters (Onyari *et al*., 1981).

From the foregoing, there is need for more exploration of the linkage between physico-chemical water quality parameters and phytoplankton species diversity of the

Nzoia River. The specific issues the study focused upon and assessed were pH; Electrical Conductivity(EC); Total Dissolved Solids(TDS); temperature; Biological Oxygen Demand ($BOD_5$); Chemical Oxygen Demand(COD) and Dissolved Oxygen(DO) for the physico-chemical water parameters while the species and community composition, bioindicators, species indices and relative species abundance. This was basis for the study as phytoplankton are the most commonly used community characteristics in environmental monitoring due to ability to summarize the data generated in biotic studies (Glenn, 1990).

### 1.2: Statement of the Problem

There continues to be insignificant exploration of the linkage between physico-chemical water quality parameters and phytoplankton species diversity in the riverine environments of Lake Victoria basin and Kenya (LVBC, 2011). This has made phytoplanktons to persist in playing minimal integral role as pollution assessment tools and lack of incorporation into standardized monitoring methods (McCormick and Cairns, 1994; Amany and Mohamed, 2003 and; Chiarucci *et al*., 2011). Such disregard has made bioindicators of the aquatic ecosystems to be ignored, despite their importance as a pollution assessment tool (McCormick and Cairns, 1994; Amany and Mohamed, 2003 and; Chiarucci *et al*., 2011).

Focus has apparently been on Webuye paper mills and associated aquatic health effects (Simiyu *et al*., 2006). This presents challenges to the local communities, arising from pollution due to the effects of intensified agriculture and the changing livelihood systems in the sugarcane growing areas and the flood plain areas of Nzoia River at Budalangi (Onywere *et al*., 2007). The importance of pairing physico-chemical analyses with the bioindicators provides emphasis for need to link physico-chemical and biological monitoring, thus allowing a better assessment of the water pollution as in the present study (Cairns, 2002 and Dlamini, 2009).

To close this gap, one approach, which was the focus of this study, was to find a statistical relationship between phytoplankton species diversity, which can reflect the overall environmental condition of a river and the physico-chemical water quality in the study area. Finding this relationship will help in determining the quality of aquatic environments.

## 1.3: Research Overall and Specific Objectives

### 1.3.1: Overall Objective of Study

The overall objective of this study was to assess the effects of effluent from Mumias sugar factory on water quality and phytoplankton species diversity of Nzoia River.

### 1.3.2: Specific Objectives

The specific objectives of the study are:

1. To determine the physico-chemical water parameters loads of Mumias sugar factory effluents before and after treatment.
2. To assess the physico-chemical water quality parameters in the Nzoia River upstream and downstream Mumias factory effluent discharge points.
3. To determine phytoplankton species diversity in the Nzoia River upstream and downstream Mumias factory effluent discharge points.
4. To compare effects of the physico-chemical water quality parameters in the Nzoia River on the phytoplankton species diversity upstream and downstream Mumias sugar factory effluent discharge points.

### 1.4: Null Hypotheses

The null hypotheses are:

1. **$H_0$:** All the mean values of the physico-chemical parameter loads of Mumias factory effluents before and after treatment are equal.
2. **$H_0$:** All the mean values of the physico-chemical water quality in the Nzoia River due to Mumias factory effluents discharge are equal.
3. **$H_0$:** There are no effects of the physico-chemical water quality of the Nzoia River on the phytoplankton species diversity upstream and downstream of Mumias factory effluent discharge points.

### 1.5: Significance of the Study

The aggregated and fused data developed from the study would be simpler to understand and easy for interpretation of water quality (Bharti and Katyal, 2011). This study is fundamental, as previous research works on phytoplankton species diversity on the same sites are sparse with minimal data to make comparisons.

However the literature on the bioindicators and the criteria for understanding the quality of aquatic environment is rich, but there is a gap between these studies and those related to water resources planning and management (Yazdian *et al*; 2014). Thus, the findings from the current study will be important for any future use of phytoplankton species diversity as bioindicators and; biomonitoring programs on effluents generation and discharge into freshwater systems. The Mumias Sugar company management, National Environment Management Authority (NEMA) and the government can therefore achieve a more elaborate monitoring of the quality of

Nzoia River.

## 1.6: Justification of Study

Sugar industry is one of the largest globally (Khoram *et al*., 2013). The choice of Mumias sugar factory that contributes 60% of Kenya's annual cane crushing capacity was due to its production of high volume of sugar effluents discharged into the Nzoia River (KeSReF, 2009). The point source pollution, due to sugar milling activities generates effluents that results in the use Nzoia River without proper effluents treatment.

This poses challenges to the local communities, arising from pollution due to the effects of intensified agriculture and the changing livelihood systems in the Nzoia riverine basin. Management decisions made from the enhanced biomonitoring programs can address these challenges. This made the choice of sugar effluents appropriate for this study.

# CHAPTER TWO
# LITERATURE REVIEW

## 2.1: Aspects of River Health

The concept of river health has continued to draw different opinions. This is due to varied sectors having divergent opinions on what a healthy river is depending on the values held. Maheshwari *et al*., (2012) related a healthy river to **(i)** its visual appeal; **(ii)** sustaining ecological integrity; **(ii)** maintaining hydrologic balance; and **(iv)** river water fit for a purpose. Fellows *et al*., (2006) linked river health assessment to the measurement of the distribution and abundance of plant and animal species. This is despite the term ‘river health’ being inherently blurred as it encompasses the natural variations in form and function existing between all river systems (DWAF, 2010). This regrettably makes measurement of environmental conditions tricky, as what is healthy in terms of system dynamics in one system may not be true of another.

Rapport *et al*., (1998) and; Bunn and Davies (2000) reported that measurement of ecosystem health unendingly entails ecosystem organizations like biodiversity. Consequently, a healthy river may be described as undisturbed river similar to that found in pre-European times (DWAF, 2010). A healthy river is one that sustains its physical, chemical and biological structure and function, recovers after short-term natural disturbance, supports local biota, and maintains key processes, such as sediment transport, nutrient cycling, assimilation of waste products, and energy exchange (DWAF, 2010). Integration of physical, chemical and biological structure into physico-chemical and phytoplankton species diversity becomes therefore necessary to have a better perspective of river health. This makes the health of phytoplankton community an expression of quality of the water (Fella *et al*., 2013).

For the purpose of this study, river health referred to phytoplankton species diversity as a function of physico-chemical water quality parameters of the Nzoia River due to effluents discharged from Mumias sugar factory. Using this definition, a healthy river has the ability to support and maintain key ecological processes and a community of phytoplankton with a species composition, diversity and functional organization that is as similar as possible to that of an undisturbed ecosystem.

### 2.2: Ecological Indicators Used in Ecosystem Health

Roux (1999) defined ecosystem health as the capability of an ecosystem to support and maintain a balanced, integrated and adaptive community of organisms having a diversity of species, composition and functional organization comparable to that of the natural habitats of the region. Ecosystem health (ecosystem integrity) is the measure of the capacity of a system in its natural state to sustain and accomplish all its functions (Dlamini, 2009).

Ecosystem health is a growing concern in the riverine environments within the Lake Victoria basin. This is typified by the many studies done in the Nzoia river basin catchments indicating pollution levels. Results by Were (2000) pointed to the dynamic nature of the phytoplankton community within Kisumu Bay with the possibility of portraying the pollution status in the whole of Lake Victoria. These studies should aim at equating the aquatic ecosystem integrity status to the basic human needs as is the case with the Republic of South African' Water Act of 1998 (RoSA,1998 and Dlamini, 2009).

### 2.2.1: Physico-Chemical Water Quality Effects of Sugar Effluents on River Waters

The cumulative hazardous effects on the environment resulting from effluents discharged into receiving waters from industrial operations and agricultural production has continued to receive worldwide attention. The characterization of physico-chemical water parameters has equally been diverse with various studies combining both metallic and non-metallic elements with their compounds as physico-chemical. Meybeck *et al*., (1996a) characterized water bodies into hydrology, physico-chemistry and biology with a complete assessment of water quality based on appropriate monitoring of these components.

Tripathi *et al*., (2004) classified parameters as organic and inorganic; conservative (such as, chloride) and non-conservative (such as $BOD_5$); toxic and non-toxic; based on toxicity; radioactive and non-radioactive based on radioactivity; settleable, colloidal and dissolved based on the size of pollutants; absolute or effects imparting, such as, pH, hardness, alkalinity and pathogenic or non-pathogenic. Salequzzaman *et al*., (2008) characterized pH, Total Dissolved Solids (TDS), temperature and Electrical Conductivity (EC) as physical parameters while $BOD_5$, DO and COD as physico-chemical parameters. Analytical tests can classify pollution parameters as physical, chemical and biological (Tripathi *et al*., 2004).

Chapman (1996) restricts the ecological prevalence of species to specific physical and chemical water attributes requirements that vary throughout the species' life cycle. These parameters may also indicate some contamination from the landscape, and a variation in natural conditions of the area such as; geology, climate and morphology.

In South Africa, the water physico-chemistry parameters assessment used have been limited to DO, pH, EC and turbidity (Bongumusa, 2010).

Within most developing countries, point and nonpoint source pollution are major environmental problems affecting water quality (Jafari and Alavi, 2010). The point source pollution due to sugar milling activities causes numerous effects on the habitat quality of water systems (Carminati, 2008). The sugar effluent discharged result in organic pollution that depletes the oxygen content and increases the electrical conductivity of the water body. The effects compromises quality of the aquatic biota that is fundamental for the productivity, survival and support of aquatic organisms. This quality is an index of health and well-being of the ecosystem and has direct effects on human health (Udoh, 2013).

The sugar industry is at crossroads, facing difficult environmental challenges for the sugar mills, these being associated with liquid waste, gaseous emission and solid waste (Gunkel *et al*., 2006). This has prompted the need to have a critical evaluation of the effects of sugar industry on the rivers. Sugar processing requires hot water for a number of steps such as water for imbibitions, raw sugar, reheating and various washings (Kaur *et al*., 2010).

The effluents with high $BOD_5$ rapidly deplete available oxygen supply when discharged into water bodies endangering aquatic life. The high $BOD_5$ also creates septic conditions, generating foul-smelling hydrogen sulphide, which in turn can precipitate iron and any dissolved salts, turning the water black and highly toxic for aquatic life (Kaur *et al*., 2010). The various operations during processing sugar generate waste from washing, leakages, spillages, splashes and water used for cooling roller bearings and blow off (Goel, 2008 and Salequzzaman *et al*., 2008). The sugar factory waste generation processes realizes waste rich in sugar and oil from machines (Goel, 2008).

The main consideration in terms of sugar processing is the pollution arising from the discharge of mill wastewater into waterways. People living near the rivers are thus at greater risks as they use the water for domestic purposes besides the industrial activities. Relatively large volumes of water are abstracted from rivers for sugar processing and the wastewater discharged back into the river (Cheesman, 2005).

Unfortunately, there is no frequent and upto date monitoring and information providing facility on the quality of the industrial effluent discharged into the river and the quality of the water in the river for human use (Subin and Husna, 2013). Such information is important for the authorities to take proper action in preventing pollution of the environment for the good health of the population. Mumias sugar factory is a wet process industry consuming large quantity of water for manufacturing process (Mullai *et al*., 2013). This has established the estimated amounts of water to be at 7.5 $mlha^{-1}$ water for $100tha^{-1}$cane (Bakker, 1999). Sugar effluents, being a major pollutant in Kenya, would thus have its effects managed better as they alter the physico-chemical parameters, flora and fauna of receiving aquatic bodies (Onyari *et al*., 1981).

Since sugar, effluents are rich in organic matter, high in $BOD_5$ and COD and; they lead to lowered levels of DO in surface waters. Wastewater with varying levels of pollution load is generated at nearly all stages of sugar production. Wastewater generated from cane washing is muddy and has a high $BOD_5$. Water from cooling systems of barometric condensers is often contaminated with sugar and regarded as one of the major sources of potentially environmentally damaging waste in a cane mill (Cheesman, 2005 and Roy *et al*., 2007).

Relatively mild effluents from the mill house, containing oil, grease, and some sugar content, are generated from the lubricating and cooling systems, floor washings, the large quantity of water used for juice extraction, and some leakage and spill over (Cheesman, 2005 and Kaur *et al*., 2010). Washing the filter cloths used for sludge from the clarifier increases the suspended solids concentration and $BOD_5$ of the wastewater. Combined with floor washings, which also add washing chemicals and more sugar, the process house effluents are considered more contaminated than effluents from the mill house (Kaur *et al*., 2010).

For mills that have an attached distillery, the numerous distillation stages produce a highly contaminated effluent, with $BOD_5$ and COD concentrations of about 40,000–100,000 $mgl^{-1}$, called stillage. In general, sugar mill effluents contain acidic and alkaline compounds, a significant concentration of suspended solids and a high $BOD_5$, COD, and sugar concentration (Cheesman, 2005). However, Mumias sugar factory management has no mechanism for measuring quantity of industrial effluent leaving the factory through its treatment line (Akali *et al*., 2011).

Though the metering system for the respective water consumption points is absent, the Mumias sugar factory utilizes an average of 5,000$m^3$ and 4,000$m^3$ for domestic and other non-production purposes per day (ECMC, 2004). Similarly, the total treated effluent flowing back to the river is estimated at $0.0361 m^3 s^{-1}$ on daily basis (ECMC, 2004). The large volumes of water abstracted for processing likewise effects on habitat quality that leads to loss of valuable habitat for the biota (Carminati, 2008).

The Mumias sugar factory process house has the highest operations attributed to the highly significant effluent generation values due to washing leakages, spillages and blow off during various operations in the manufacturing process (Akali *et al*., 2011). Other key point sources of pollution include oil storage and diffuser tanks. These areas are characterized with high water spillage. Out of these operations, general characteristic of the sugar factory waste have been established as follows: pH, 4.6-7.1; COD, 600-4,380 ($mgl^{-1}$) and $BOD_5$, 300-2,000($mgl^{-1}$) (Goel, 2008). The rest of the factory effluent is channelled to process wastewater treatment plant having a set of six stabilization ponds located about 500 metres away from River Nzoia.

Similarly, Gunkel *et al*., (2006) identified pathways of contamination by evaluating cultivation and processing techniques of a bio-alcohol factory with annexed sugar cane cultivation. Millette *et al*., (1991) found sugar cane mill effluents to be concentrated typically containing biochemical oxygen demand ($BOD_5$) in the range of 15,000 $mgl^{-1}$. Gunkel *et al*., (2006) demonstrated that the effects of sewage discharge on the river's self-purification capacity indicated a severe change to the worse in the lower course. This was attributable to the use of stillage (wastewater from cane processing) for fertilization.

Results by Ale *et al*., (2009) from the Karnali distillery on effluent discharged measured effects on agricultural crops and environmental justice to the concerned people. The values for temperature, pH, DO, $BOD_5$ and COD for effluent stream were found to be different from those of dilute and the non-effluent stream water. Most were above the toxic level set by Nepal Bureau Standard. The distillery effluent was highly loaded with organic pollutants along with harmful heavy metals that showed significant effects on soil quality and the crop productivity that caused environmental injustice to the local people in terms of loss of crop productivity and environmental hazards.

Shivappa *et al*., (2007) determined physico-chemical profile of effluent from sugar mill in Bhadravathi Taluk, Karnataka. The averages and ranges for temperature, colour, turbidity, pH, electrical conductivity, $BOD_5$, COD, total dissolved solids, chloride, total alkalinity, total hardness, sulphate, phosphate, total acidity, calcium and magnesium were realized. A study of Ipojuca River in Pernambuco region of Brazil found that the dominant effects on water quality of the river were domestic sewage input in the upper catchment and sugar cane cultivation and processing in the lower catchment. Apart from desirability of clean rivers, this situation posed danger to health of aquatic life (Gunkel *et al*., 2006).

Meanwhile, monitoring of water quality in the Ba River during the sugarcane-crushing season in 1994 and 1995 showed very low DO at sites close to the discharge point of Rarawai mill (Fagan *et al*., 1995). Botelho *et al*., 2012 assessed the effects of sugar effluents in the Piracicamirim's creek water quality. Test organisms alongside physical and chemical analysis of water were used to perform toxicity tests on water samples upstream and downstream the industry. The physical and chemical parameters did not change during the sampling period. Though only dissolved oxygen changed during the sampling period, the water body was not affected.

Monitoring of water quality in Qawa River, Labasa during the 1995 sugar-crushing season showed water temperature and $BOD_5$ levels to be higher, and DO levels lower, than background levels near the FSC mill discharge outlet, which may be the cause of pollution reported in this river. The DO levels in the river downstream from the mill were also below that necessary to maintain healthy aquatic life (Tamata *et al*., 1996).

Olajumoke *et al*., (2010) investigated effects of brewery effluent on water quality in Majawe. The physico-chemical water parameters analysed were pH, temperature, alkalinity, electrical conductivity, total soluble solids(TSS),TDS, $BOD_5$,COD, DO and concentration of chloride, iron, magnesium, calcium, cadmium, lead, arsenic and mercury. The physico-chemical water parameters indicated that surface water and brewery effluent deviated from the WHO and FMENV standards. These findings showed contamination of surface water by brewery effluent.

Studies by Kumar *et al*., (2009) revealed severe pollution from sugar factories at Kashipur in River Kosi. The range of values for, DO, $BOD_5$ indicated mild organic pollution and COD industrial pollution. High EC indicated a larger quantity of dissolved mineral salts, making it sour and unsuitable for drinking.

According to Raburu and Okeyo (2005), rivers play a significant role at the receiving end of all the anthropogenic activities. A true reflection of these activities within the river catchment is provided through spatial studies of point or non-point sources of pollution. The character of streams and rivers from these studies should reflect an integration of physical and biological processes occurring in the catchment. Such studies on pollution of Nzoia River basin due to anthropogenic activities have been reported since the 1960s (Achoka, 1998; Mogere, 2000; GEF, 2004; and Aura *et al*., 2011).

The study by Achoka (1998) established tendency for oxygen concentrations to decrease downstream after wastewater from the Webuye Pulp and Paper Mills discharged into River Nzoia. Wastewater treatment systems in the catchment have been shown to be ineffective due to agricultural wastes discharged into the River Nzoia posing a substantial health hazard to aquatic life (Mogere, 2000). A similar study by Momanyi (2002) on selected water quality parameters and heavy metals in Nzoia River in relation to the Webuye Pulp and Paper Mills pointed out a significant increase in TDS downstream due to effluent discharge.

It is recommended that water quality monitoring with effluents of this nature be done using a combination of chemical analysis and biological indicators such as phytoplankton. This is to overcome the inadequacies such as time, cost and technical limitations of physico-chemical analysis (Makhlough, 2008). Their analysis as management regulatory factors in statutory bodies like NEMA of Kenya is made more difficult. This is due to rivers being very dynamic and potentially subject to great spatial heterogeneity (Makhlough, 2008). Meanwhile, integration with phytoplankton studies should provide continuous spatial information in surface waters without these limitations (Makhlough, 2008).

Studies on pollution levels in Kenya’s river systems have traditionally been through physical and chemical water analyses. Kenya’s National Environmental Management Authority (NEMA) discharge standards are based on physico-chemical water parameters to make management decisions on waters. Similarly, majority of Zimbabwe’s water quality studies have used physico-chemical parameters for water assessment (Mathuthu *et al*., 1997). Physico-chemical water parameters of water provide nutritional balance and ultimately govern the biotic relationships of organisms in an aquatic ecosystem; including ability to withstand pollution load. Chemical analyses can give very accurate measures of the amounts of individual substances in a river, but they only consider the water passing during collection time (Mackay and Masundire, 2002).

However, biological communities offer information on prolonged exposure of the stream to pollution and exhibit noticeably the effects of pollution on water quality (Mudyazhezha and Ngoshi, 2014). Likewise, biological communities reflect the overall ecological health, providing a holistic and an integrated measure of the river health as a whole (Chutter, 1998). This makes incorporation of the effects of chemical and physical disturbances on aquatic communities that occur over extended periods a viable option. Hence, there is need to increase use of biological conditions like phytoplankton species diversity in environmental monitoring. This enables a more precise characterization of the cumulative effects of anthropogenic activities on ecosystems (McCormick and Cairns, 1994).

According to Rey *et al*., (2004), it is reasonable to monitor aquatic biota because of the difficulty and cost of chemically analyzing every potential pollutant in a sample of water, and of interpreting results in terms of impact severity. Results from biological monitoring are cost effective, rapidly obtained and examination of organisms exposed to pollutants is continuous (Rott, 1991 and Rey *et al*., 2004).

**2.2.2: Phytoplankton Species Diversity as an Indicator of Ecosystem Health Due to Effluents Discharge**

Numerous studies together with the followings have revealed regularity between variations of bioindicators and oscillations in physico-chemical characteristics of water. The revelations occasioned the determination of phytoplankton species diversity resulting from effects of the physico-chemical water quality at effluent discharge points (Yazdian *et al.*, 2014).

There has been development of numerous biomonitoring methods of the ecosystem health for aquatic systems, most of them using attributes of whole assemblages of phytoplankton. Several researchers across the world have used biomonitoring as an important assessment tool (Lacdan *et al*., 2014). Biomonitoring has only as recently as 1996 become a standard tool in the management of South Africa's inland waters while Kenya is yet to encompass this as a standard biomonitoring tool in the management of inland waters (Hohls, 1996).

Dlammini (2009) has demonstrated the use of a functional approach to ecosystem health indicating species ecological functions and diversity. These confer information about the quality, amount of water accessible in the habitat and the ecosystem health as a whole (Day, 2000; Brainwood *et al*., 2004 and Samways *et al*., 2006). Equally, MacDonald and Niemi (2004) noted that resident species' diversity could also be used as indicators of change in ecosystem health as they can reveal an early decline of a habitat.

However, there has been a developing interest in Kenya to find out the linkage between biotic and physico-chemical water parameters to water quality. Aura *et al*., (2011) semi-quantitatively sampled benthic macro invertebrates from Rivers Kipkaren and Sosiani in the upper reaches of River Nzoia basin, Kenya. The results revealed a distinction between the benthic macro invertebrates from impacted, the less impacted sites, and the physico-chemical water parameters associated with this distinction.

Raburu *et al*., (2009) developed a macro invertebrate-based Index of Biotic Integrity (M-IBI) to monitor ecosystem health of the Nyando River and its tributaries. There were variations in ecosystem health among stations and this was reflected in community composition and structure of resident macro invertebrates. This is a deviation from the past, where the management of river systems had been based primarily on chemical water quality monitoring.

In Kenya, linking use of phytoplankton species diversity to physico-chemical water quality parameters has been marginal. This is despite the certainty that phytoplankton assemblage are useful bioindicators as mentioned by Reynolds (1984); Biggs, (1985); Dixit *et al*., (1992); McCormick and Cairns, (1994); Stolte *et al*., (1994); Richardson, (1997); Hötzel and Croome, (1999); Valdes-Weaver *et al*., (2006) Makhlough, (2008) and; Zakariya *et al*., (2013).

Dokulil, (2003) mentioned that Cohn in 1853 and 1870 first classified phytoplankton as indicators of water quality. The result was acknowledgement of phytoplankton as biomonitors and bioindicators. Since then, various systems deducing water quality from observations of indicators organisms have been developed, evolved, and diversified. Phytoplankton makes good indicators of the physico-chemical conditions prevailing in the aquatic environment since they receive most of their nutrition from dissolved chemicals in water (Kolayli and Sahin, 2009). This enables monitoring of eutrophication and pollution consequently assessing environmental health status of riverine ecosystem (Kolayli and Sahin, 2009; and Wan, 2010).

Ojunga *et al*., (2011) have been able to use phytoplankton and macroinvertebrate assemblages to assess the impact of a Kraft pulp and paper mill effluent in Kenya, on River Nzoia downstream of the discharge point in relation to changes in water quality. The findings indicated that taxon composition of phytoplankton and macroinvertebrates correlated with adverse environmental gradients resulting from the mill's effluent discharge. Overall, there was a shift in composition and abundance of both phytoplankton and macroinvertebrates, with the downstream site recording high numbers of tolerant taxa (i.e., *Microcystis* sp. and *Chironomus* sp.).

The presence of phytoplankton species in a given habitat will then indicate that one or more parameters are within tolerance limits of that species (Dokulil, 2003). This may relate to abundance and biomass due to environmental effects in general or specific stress symptoms. For example, a study by Makhlough (2008) recorded the presence of *Anabaena, Microcystis, Oscillatoria, Nostoc, Dinobryon, Chroococcus, Staurastrum paradoxum* and *Mallomonas* as indicators of toxic, unfavourable odours and pollution in aquatic ecosystems. The study showed the capacity of algological studies to provide early alert of water degradation and importance in water quality assessment. This is an indication that phytoplankton detect organic pollution because of their well-documented tolerance (Palmer, 1969). Moreover, phytoplankton species diversity indices are first order and measure stress in the environment, as a large number of species characterizes an unpolluted environment (Mason, 1996 and Wan, 2010). Phytoplankton species diversity and composition defines effects of various types of river degradation that was focus of current study (Patrick, 1973 and Wan, 2010).

## 2.3: Effects of the Physico-Chemical River Quality on the Phytoplankton Species Diversity in Surface Water

### 2.3.1: Effects of the Physico-Chemical Effluent Quality in Surface Water

According to Yazdian *et al*., 2014, there are two stages for linking a biotic index to a river water physico-chemical quality. The first stage is deciding an applicable index and attaining its feasible mathematical link with physico-chemical characteristics of river water. The parameters with significant effects on the water studied are characterised. Studying the mathematical relationship between variations of biotic indices with these physico-chemical characteristics is the second step. The proper biotic index that shows strongest statistical relation with the physicochemical parameters can be selected.

Pratiksha *et al*., (2013) undertook monitoring of water quality of Wardha River to assess its water pollution level. Higher values of several physico-chemical water parameters pointed to the pollution of riverine ecosystem in the study area. Domestic, municipal, and industrial effluents from paper and pulp industries as well as agricultural runoffs were directly or indirectly responsible for deterioration of water quality.

In a study conducted by Mullai *et al*., (2013) to find out the physico-chemical characteristics of Uppanar River in India, the various physico-chemical variables gave an outline of the river's water quality. This indicated a moderately polluted river likely to be due to the continuous discharge of municipal and industrial effluents.

A study by Monika (2013) concluded that water of Tapi River was moderately polluted due to discharges of industrial waste, domestic sewage and agricultural run-off in river water. The study had revealed temperature and electrical conductivity to be within the permissible limits of WHO and BIS while pH, DO, $BOD_5$ and COD, were observed beyond the permissible limits. A more efficient management to conserve this river was required.

Udoh (2013) investigated aspects of the physico-chemical water parameters of Eastern Obolo estuary, Nigeria whereby mean concentrations showed significant statistical differences for DO, temperature and EC. The variability of the concentrations of the parameters was lowest for DO and highest for hardness. The mean values of $BOD_5$ from all stations exceeded the recommended limits for aquatic life or potable water. Consequently, the estuary only assists in flushing out anthropogenic pollutants into the sea.

### 2.3.2: Effects of the Physico-Chemical Water Quality Parameters on the Phytoplankton Species Diversity Due to Effluent Discharge

Biological assessments can be used for identifying weaknesses in ecosystem environments caused by pollutants or degradation of habitats (Yazdian *et al*., 2014). They are also, in some cases, even more effective than physical and chemical measurement processes, because they are economical and need less time to be evaluated (Zakariya *et al*., 2013). Among the various components of the aquatic ecosystems, phytoplankton pave the way for one of the best and most efficient ways for biological assessments, for being useful bioindicators.

A number of workers have reported the use of phytoplankton species in assessing the degree of pollution or as indicators of water pollution and quality of different water bodies (Trivedy, 1986; Sudhaker *et al*., 1994; Dwivedi and Pandey, 2002; Naik *et al*., 2005 and; Nandan and Aher, 2005). Ho (1976) studied phytoplankton production in the disturbed Renggam Stream; Selangor carried one of the earliest phytoplankton studies conducted in relation to water pollution. Nather (1991) conducted studies on the pollution status of the Linggi River Basin, Seremban, and Negeri Sembilan using diatoms and reported a marked variation in species between the unpolluted and polluted stations. Anton (1981) recorded a decrease in phytoplankton species in the downstream stations due to heavy siltation in the Langat River, Selangor.

Sugar effluents discharged can produce changes in both aquatic fauna and flora besides endangering the human health for use of river water for domestic and agricultural purposes (Ayyasamy, 2008). In line with these changes, size, structure and biomass of phytoplankton population and production have been shown to be closely related to physico-chemical conditions of the water body (Mitchell-Innes *et al*., 1992). Mitchell-Innes *et al*., (1992) has further reported a general shift in the phytoplankton population size structure at about 15°C, high biomass with Bacillariophyceae-dominated populations being replaced by low biomass, flagellate-dominated populations. Suman *et al*., (2010) indicated changes in phytoplankton biomass and dominance by Bacillariophyceae followed by Dinophyceae and

Chlorophyceae in relation to DO, nutrients and turbidity. Species diversity in all the sampling stations indicated close correlation with ambient temperature. Bioindicator species like *Polykrikos schwartzil, Dinophysis norvegica* and *Prorocentrum concavum* pointed to moderately polluted water quality of the estuary.

Yeng (2006) reported that water pollution in the Ahning Reservoir, Kedah was associated with the appearance of certain species of phytoplankton, especially Dinoflagellates. Yap (1997) used the Shannon-Weiner diversity index and the saprobic index of phytoplankton for water quality assessment of a river ecosystem and concluded that ecological knowledge can be used in the management of a water body.

In Malaysia, the determination of trophic state has been conducted primarily by measurements of physico-chemical parameters, primary productivity and chlorophyll-a concentration. In a study conducted in the Muda and Pedu Reservoirs by Zulkifli (1980), the presence of *Anabaena, Microcystis, Oscillatoria, Nostoc, Dinobryon, Chroococcus*, *Staurastrum paradoxum* and *Mallomonas* was recorded, which are indicators of toxicity and pollution in aquatic ecosystems. This shows that algological studies are important for water quality assessment that can provide an early warning sign of water degradation.

A study by Dlamini (2009) showed the likely change of species diversity in the river due to complex interaction between physical, chemical and biological entities. Allan (2004) who supports this observation argues that the chemistry of the water flowing through a watershed is also directly affected by key elements determined by the terrestrial activities that contribute in shaping the distribution of aquatic species. The activities that release effluents have been associated with a number of impact types on water quality and aquatic biota in the receiving water bodies (Ojunga *et al*., 2011). This causes interaction by different environmental factors such as pH, light and temperature that have been noted to affect phytoplankton composition (Abuzer and Okan, 2006).

Lacdan *et al*., (2014), have illustrated the use of phytoplankton species diversity (Shannon-Wiener diversity index, and Palmer's pollution index) and the physico-chemical water parameters (temperature, pH, total dissolved solids, and dissolved oxygen) to assess the water quality. Identified phytoplankton from the study sites of Dao River were twenty-one genera belonging to six groups. Significant differences ($p<.05$) for physico-chemical water parameters were within the permissible limits for freshwaters. The recorded SW diversity index was highest in the midstream while lowest in the downstream of the river. Organic pollutants contaminate all the study sites as indicated by the Palmer's pollution index.

These interactions makes the phytoplankton useful as bioindicators of water quality, arising from their responses to changes in nutrients concentration, physical and chemical parameters (Rahmati *et al*., 2011). Therefore, changes in the phytoplankton community can reflect the occurrence of pollutants or other environmental stressors that causes dramatic increase of phytoplankton (Johnstone *et al*., 2008). This event can lead to low oxygen condition that affect other organisms in the aquatic food chain (Camargo and Alonso, 2006).

Investigations by Onyema *et al*., (2007) on the physico-chemical characteristics and phytoplankton showed notable variation. Salinity regime seemed a major determinant of variation in the composition and abundance of the phytoplankton encountered. Ekwu *et al*., (2006) showed that high levels of taxa richness and diversity of phytoplankton communities in the Cross River estuary provides a concise set of structural based criteria for assessment of the environmental status and productivity of an aquatic ecosystem. Furthermore, the high abundance of polluted water species of algae revealed indications of increasing environmental degradation.

A comparative study by Das (2010) on, "*Water Quality and Phytoplankton Population in Sewage Fed River of Mahanadi, Orissa, India",* revealed that, high temperature and better nutrient status harboured more phytoplankton. Higher phytoplankton populations corresponded to the fluctuation of prevailing conductivity, turbidity, dissolved oxygen, better organic load and chemical oxygen demand content of the said habitat. The composition of high concentration of Bacillariophyceae was of mainly *Navicula* and *Nitzchia* being indicative of polluted zone of the river. *Spirogyra ornata, Navicula cuspidate*, *Oscillatoria limnosa*, *Zygnema*, *Ulothrix, Nitzchia* and *Phormidium* were the most abundant species sequentially. *Oscillatoria* and *Nitzchia* species at sewage-affected sites can be used as an indicator of organic pollution in the river.

Ariyadej *et al*., (2004) studied the diversity of phytoplankton relationships to the physico-chemical environment (temperature, pH, dissolved oxygen, alkalinity, conductivity, water transparency, and nutrients). The study reported presence of one hundred and thirty five species in seven divisions of phytoplankton. The species variation in decreasing order due to the physico-chemical environment were

Chlorophyta (50%), Cyanophyta (21%), Bacillariophyta (13%), Pyrrophyta (6%), Cryptophyta (4%), Chrysophyta (3%) and Euglenophyta (3%). By applying a principal components analysis (PCA) using the multivariate statistical package (MVSP), it was found that *Cyclotella meneghiniana* and *Melosira varians* were the most abundant. The factors affecting the phytoplankton species by Canonical Correspondence Analysis Ordination (CCA-ORD program) were alkalinity, water temperature, water transparency, nutrients and conductivity.

Results by Zakariya *et al*., (2013) in the five sampling stations surveyed indicate that surface water temperature, Turbidity, Dissolved Oxygen and pH did not significantly vary between the stations ($p>.05$), whereas $BOD_5$ varied significantly between the stations ($p<.05$). Phytoplankton abundance showed the decreasing order of abundance: Bacillariophyta, Chlorophyta, Cyanophyta, Chrysophyta and Pyrrophyta. Based on SW diversity index, the water in the five stations surveyed is classified as moderately polluted. The results show that the multitude users of the lower Niger River have negatively affected its water quality.

Findings from Nzoia River basin on the effect of mill effluents on the aquatic environment show that the evaluation of biotic communities offers a comprehensive alternative to the use of physico-chemical water parameters (Etiégni *et al*., 2007; Merilainen and Oikari 2008). Previous studies have indicated a decrease in fish richness and deteriorated water quality downstream of the effluent discharge point (Balirwa and Bugenyi 1988; and Achoka 1998). However, because of their mobility, fish cannot give a comprehensive account on the effects of pulp mill effluent as opposed to the more sedentary phytoplankton.

Physical and chemical parameters remain inadequate to assess the damage of industrial wastewater in aquatic environments (Dalzell *et al*., 2001). It becomes essential to use phytoplankton as they interact with the pollutants giving an effective response on the water quality (Botelho *et al*., 2012). Chemical analyses when paired with the ecological indicators can show the state of the water over a long period (Dlamini, 2009).

Information on phytoplankton species diversity and abundance consequently carry great potential for inclusion in water physico-chemical assessments. The present study could provide insight into phytoplankton responses as indicators of water quality, assessing environmental health and changes in management practices respectively (Hötzel and Croome, 1999). This implies phytoplankton communities give more information on changes in water quality than mere nutrient concentrations (Medupin, 2011). Furthermore, phytoplankton species determined can reflect the present history of the water quality in the river, allowing detection of disturbances that might otherwise be missed (Eekhout *et al*., 1996). The data results could potentially add to an understanding of ecological health of the Nzoia River at the study site.

### 2.4: Approaches to Ecosystem Health Data Analysis

#### 2.4.1: Multivariate Analysis

According to Khattree and Naik (1999), the subject of multivariate analysis deals with the statistical analysis of data collected on more than one variable, while their statistical dependence is often taken into account during data analysis. Assessment of pollution effects in surface water as an aspect of river health requires univariate and multivariate statistical techniques for data analysis (Davies *et al*., 2001 and USDOE, 2012). The application of multivariate methods has increased tremendously in recent years for analyzing environmental data and drawing meaningful information (Alkarkhi *et al*., 2008). These techniques are useful for the evaluation and interpretation of convoluted water quality data sets (Sundaray, 2010).

The multivariate techniques have been widely used as unbiased methods in analysis of water quality data for drawing out meaningful conclusions, to characterize and evaluate water quality for analyzing spatio-temporal variations caused by natural and anthropogenic processes (Salim *et al*., 2014). The multivariate data analysis provides an overall evaluation of the significance of differences between groups, identify environmental parameters that are most strongly correlated with each other, assess similarities among phytoplankton communities and define environmental factors to phytoplankton species associations (Wold, 1991, Amany and Mohamed, 2003 and; Chia *et al*., 2011). The complex water quality data sets arise from the need to compare the distribution patterns of phytoplankton communities in the rivers with physico-chemical parameters. Researchers have found that multivariate statistical techniques allow for the analysis of the relationship between such biotic and abiotic variables (Wan, 2010).

Multivariate statistical techniques like principal component analysis(PCA) and Stepwise Multiple Linear Regression (SMLR) analysis have in recent years been used to assess the effects of anthropogenic activities and the interpretation of water quality parameters (Thareja *et al*., 2011 and Fataei *et al*., 2012). This illustrates the usefulness of multivariate statistical techniques in assessing overall water quality management.

According to Davies *et al*., (2001) multivariate and univariate statistical techniques can be used to calculate the statistical significance changes in the whole community and highlight the species or suite of species responsible for the changes in community composition. Multivariate techniques are relatively straightforward to interpret as they can present the extent of community change in a single visual graph. Univariate analyses should be used to assess the significance of any change in the abundance of an individual species, or any changes in the diversity of phytoplankton. A student's t-test can be used to assess the spatial change in abundance of individual species (Davies *et al*., 2001).

### 2.4.2: Biodiversity Indices

Biodiversity remains a significant attribute and core constituent of an ecosystem's health support system (Zhao *et al*., 2013). Indices are most often used to describe measures of community composition such as species abundance, diversity, evenness, richness, and dominance. Similarly, species occur in relatively low numbers in a community when an environment becomes stressed (Dlamini, 2009). Various biodiversity indices have been projected and manipulated by environmentalists in diverse countries. For instance species richness, evenness, species diversity and dominance indices (Yazdian *et al*., 2014).

According to Clarke and Warwick (1994) richness is the measure related to the total number of taxa, with a diverse sample having more species. It indicates the presence of various species and determined by the number of species in an area. An increasing number of taxons can be due to habitat diversity, suitability of water or its improved quality (Yazdian *et al*., 2014).

Evenness index demonstrate the distribution of the communities of species. The more even species distribution is, (i.e. the number of individual organisms or abundance of species are more similar), the higher stability is present which results in greater biodiversity (Yazdian *et al*., 2014). On the other hand, Shannon-Weiner's (SW) diversity index measures species numbers in a biological community. It is a fast and reliable tool to identify major changes in community structures (Pettersson, 1998).

Compared to species richness, SW diversity index provides extra information regarding community composition by taking the relative abundances of different species into account (Kumar and Sharma, 2014). Seasonal patterns of SW, species richness and evenness indices have indicated similarities to seasonal changes in species abundance and composition (Yazdian *et al*., 2014). Desai *et al*., (2008) likewise used SW's index and pollution level designed by Biligrami (1988) as shown in Table 2.1.

**Table 2. 1: Shannon-Weiner's Index Pollution**

| **Species Diversity** | **Pollution Level** |
|---|---|
| 3.0-4.5 | Slight |
| 2.0-3.0 | Light |
| 1.0-2.0 | Moderate |
| 0.0-1.0 | Heavy |

According to Clarke and Warwick (1994), SW diversity index indicates that uncertainty of occurrence increases as both the number of species increases with the resultant individuals being distributed more and more evenly among the species already present. It is further mentioned by Dlamini (2009) that community diversity value of Shannon-Weiner (H') ranges between zero (0) (low complexity) and four (4) (high complexity).

Elsewhere Nather (1991) states that Simpson's diversity index takes into account both richness and equitability and is the probability that a taxon is selected from different species. Higher diversity signifies greater value of the index. Diversity range for Simpson's index is between zero (0) and one (1) since it is about probability. The exponential form of the SW index and the inverse of the Simpson index have the property of equalling species richness when all species are equally abundant, and converging to unity as the set of species approaches maximum inequality (Chiarucci *et al*., 2011). Both the Simpson and SW indices are widely used, as they are composed of species richness and relative abundance within communities (Zhao *et al*., 2013). This has made them an integral part in determining the ecosystem's health (Jorgensen *et al*., 2005) and in water quality assessments (Kannel *et al*., 2007).

Studies on the structure and functioning of planktonic communities have provided opportunities to investigate patterns of responses to abiotic dynamics (Jafari and Alavi, 2010). This dynamism can be useful in evaluating the resilience of a river system. Linkage of the ensuing variability to the quality and quantity of resources like sugar effluents discharged into a river system is possible (Jafari and Alavi, 2010). Long-time variability in rivers using phytoplankton community are relatively easy to detect but protocols for surveys are yet to be developed (Dokulil, 2003). The interference of sugar effluents, through modifications in the physico-chemical conditions of the aquatic ecosystem can indirectly affect the structure of the phytoplankton community.

The approach based on the phytoplankton indices of community structure (diversity, evenness, richness, similarity), with the assumption that a pristine and healthy environment can be typified by a greater diversity of organisms than found in degraded environments, has been used for monitoring rivers. Although the study conducted at the Pinang River Basin demonstrated that the difference in species diversity could be related to changes in water quality, comparing diversity as a tool to discriminate water quality conditions was limited to sampling stations upstream from those near the estuary (Wan and Mansor, 2000). It is therefore imperative to have an accurate estimate of the water quality using species diversity by precisely defining the species that compose the community, as was the case with the present study (Wan, 2010).

Higher biodiversity indices indicate less stress in ecosystems, higher abundance and more even distribution of species in the ecosystem (Yazdian *et al.*, 2014). Pielou, (1975), Stirling, (2007) and Comín (2011) stated that diversity index increases by increased number of species or increasing the total number of organisms in populations; when the population of various species is distributed evenly, the diversity index increases as well.

Tolan *et al.*, 2011, acknowledges the historical derivation of indices, drawn from dynamic assessments of the planktonic assemblages. They summarize a series of diverse community measures into one or more quantitative variables. They also reveal much of the underlying information inherent in the vast amount of raw data a phytoplankton assessment generates.

Within the framework of data reduction, indices are much similar to the principal components and canonical correlations tests (Tolan *et al*., 2011). In order to find the relationship between biodiversity indices and the physico-chemical characteristics of river, Principal Component Analysis (PCA) and Stepwise Multiple Linear Regression (SMLR) was used in this study.

Principal components is parametric-based test used to reduce the quantity of variables down to a manageable subset that explains the greatest amount of total variation without discarding any useful information (Osborne *et al*., 1993). Principal component analysis (PCA) is a potent pattern recognition model that attempts to reveal the variance of a large data set of inter-correlated variables with a smaller set of independent variables (principal components) (Hopke, 1985, Simeonov *et al*., 2003 and; Mustapha and Abdu, 2012). This widely used, but poorly understood model and referred to as the black box, remains the mainstay of modern data analysis (Shlens, 2003).

In a complicated system, a large number of physico-chemical and biotic parameters are subject to constant changes. This makes it difficult to determine change characteristics of the waters in terms of a single factor. The PCA can therefore become an important tool that discovers linear combinations of the original variables that describe the significant variations in the data, and extract the latent factors, according to the Eigen value-more-than-one rule (Jun-De Dong *et al*., 2010).

Zhang *et al*., 2007, have indicated that the application of PCA aids in the interpretation of convoluted data matrices to better understand the water quality and ecological status of the studied systems. Simeonov *et al*., (2003) applied cluster analysis (CA), PCA and multiple regression analysis on principal components to interpret a large and complex data matrix obtained during a monitoring program of surface waters in Northern Greece. A multivariate receptor model was also applied for source apportionment estimating the contribution of identified sources to the concentration of the physico-chemical parameters. Missing data were completed by mean values of the neighbour data. Through the PCA, the sources of the pollutants were identified.

In recent years, many studies related with these methods have been carried out. For instance, Kayser and Tenke, (2003) determined how specific methodological choices affect "data-driven" simplifications of event-related potentials (ERPs) using principal components analysis (PCA). They concluded that unrestricted, unstandardized covariance-based PCA solutions have a better optimization of component identification and measurement when compared to correlation-based PCA solutions on the same data set. These studies indicate that these methods may be used to assess the relationships between variables and possible pattern in distribution of measured data.

## 2.5: Theoretical and Conceptual Framework

### 2. 5.1: Theoretical Framework

The study was based on the systems theory (Bertalanffy, 1968). The study of a system is transdisciplinary, consisting of abstract organization of phenomena, independent of their substance, type, or spatial or temporal scale of existence (Heylighen *et al*., 1992). The complex relationships among organizational and environmental variables that optimize decisions in a system can be understood by use of quantitative methods (Katz and Kahn, 1966). The modern scientific theory of analyzing complex phenomena into elementary particles and processes articulated by systems theory has arisen from emphasis on synthesis (Chen and Stroup, 1993).

The holistic approach of the systems theory explains a phenomenon whereby the whole cannot be more than the sum of its components without interaction of the parts (Tzagkarakis, 2009). The interaction results in a total input and total output without worrying which part of the input goes to which subsystem. The subsystems therefore require common language(s) for system integration. This connects systems theory and information theory. Furthermore, river basins, considered as an open system by Raj and Azeez (2012), are limited, as future status cannot be calculated with perfect accuracy, since all necessary information is unknown (Bertalanffy, 1968).

The system theory has been successful in helping understand processes that can readily be decomposed into simple causal chains. It has a unifying theoretical framework for science with the following five major strengths: the multidisciplinary nature, the ability to engage complexity, the capacity to describe system dynamics and change, the ability to represent the relationship between the micro-level and macro-level of analysis, and the ability to bring together the natural and human worlds (Chen and Stroup, 1993). Systems theory in the current study was meant to link the complex interactions between the physico-chemical water parameters of Mumias sugar effluents and those of the Nzoia River to the phytoplankton species diversity of the

Nzoia River.

The description of the dynamics and changes in the Nzoia River as a system, representation of the relationships between the components (micro-level) and the whole (macro-level) of Nzoia River for analysis, and the ability to bring together the natural and anthropogenic activities was found suitable to use the systems theory. Anthropogenic activities such as the sugar processing provide a system that interacts with the hydrological system like River Nzoia through water abstraction and sugar effluent discharges into the river water system.

Both the River Nzoia waters and water quality-assessment program were considered systems with River Nzoia waters being part of a bigger system, the hydrological cycle. The effluent discharged into the River Nzoia waters also serves as an input from the sugar effluent system into the River Nzoia water system whereby the mixing/interaction that resulted in the variation of its physico-chemical water parameters that affect the phytoplankton diversity. The National Water Quality Monitoring Council (NWQMC) (2004) does have a system for water quality monitoring. Similarly, South Africa has a drinking water quality system framework. All these are systems that interact with other systems outside themselves. Patten (1984) on “System theory formulation of site-specific water quality standards and protocols” has formulated a system theory approach to review current procedures of USEPA site-specific methodology, and a set of recommendations proposed for their improvement using the system theory formulation to guide further developments. The Dempster–Shafer theory also called theory of evidence was used as a potential methodology for interpreting water quality data (Sadiq, *et al*., 2004).

### 2.5.2: Conceptual Framework

Support for the study's theoretical significance stemmed from the system theory framework that resulted in modelling of conceptual framework indicated in figure 2.1.

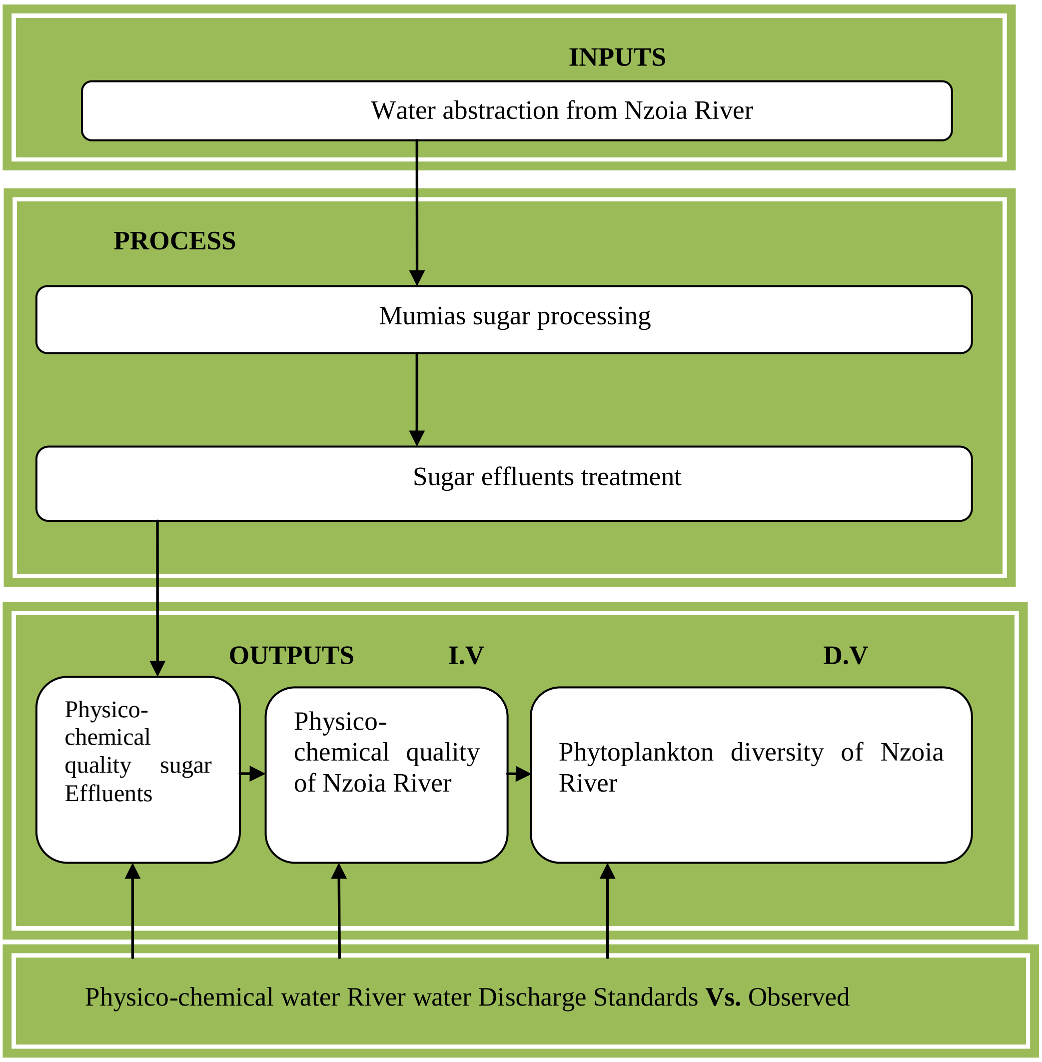


**Figure 2. 1: Conceptual Framework of the Study**

It is also similar to that of Thomann and Mueller's (1987) model, which observes water quality management problems, can be controlled by means of assignment of allowable discharges to a water body. The conceptual framework of the study is a modified model of the system theory derived from Van Belle (1995) indicated in figure 2.2.

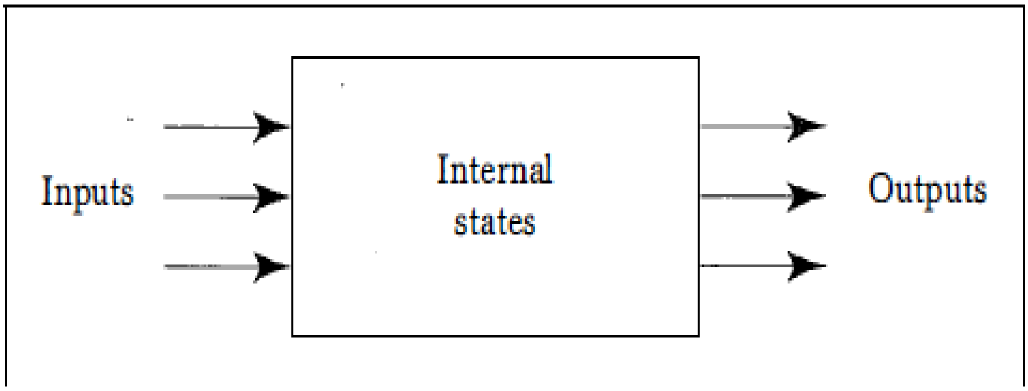


**Figure 2. 2: The Conceptual Framework of the System Theory (Source: Van Belle, 1995)**

The study's conceptual framework provides a way of looking at and describing reality of the interactions in the Nzoia River system due to sugar effluents discharged from Mumias sugar factory. This view offers a suitable springboard for constructing a scientific worldview of the study based on theoretical underpinnings (Van Belle, 1995). The framework proposes that phytoplankton species diversity of Nzoia River may vary due to the effects of physico-chemical loads of sugar effluent discharges on the physico-chemical quality of the Nzoia River (**I**ndependent **V**ariable-**I**.**V**). The quality may in turn have the potential s on the phytoplankton species diversity (**D**ependent **V**ariable-**D**.**V**) of the Nzoia River as seen in figure 2.1. The measured outputs involved determination of the physico-chemical parameter loads of sugar effluents before and after treatment, the s of the physico-chemical loads of sugar effluents discharge on the physico-chemical quality in the Nzoia River that affects the phytoplankton species diversity in the Nzoia River. These were basis for the assessment of the effects of effluent from Mumias sugar factory on water quality and phytoplankton species diversity of Nzoia River at the study locality.

### 2.5.3: Summary of Literature Review

The review indicates that literature on the bioindicators and the criteria for understanding the quality of aquatic environment is rich, but there is a gap between these studies and those related to water resources planning and management. There is still minimal use of phytoplankton due to lack of knowledge of water resources modellers. Planning of water resources control and management remains a challenge despite water quality assessment being an important tool for pollution control and assessment. This has made phytoplankton to persist in playing minimal integral role as pollution assessment tools and lack of incorporation into standardized monitoring methods.

However, literature has shown that phytoplankton species diversity and composition can define effects of various types of river degradation. Their sensitivity to ecological alterations makes them suitable in water quality monitoring programs in determining the health status of the receiving waters. Phytoplankton species diversity and physico-chemical water parameters remain an important criterion for evaluating the suitability of water for drinking and other purposes. Correlating species diversity indices with physico-chemical water parameters provides one of the best ways to detect and evaluate the effects of pollution on aquatic communities.

Standardized methods for measuring on diversity metrics are lacking, especially at the scales needed to monitor biodiversity for conservation and water quality management. Multivariate data analysis provides meaningful information on assessment of pollution effects in surface water. These techniques are useful for the evaluation and interpretation of convoluted water quality data sets.

To close this gap, one approach, which was the focus of this study, was to find a mathematical relationship between phytoplankton species diversity, which can reflect the overall environmental condition of a river and the physico-chemical water quality in the study area. Finding this relationship will help in determining the quality of aquatic environments. Since there are widespread databases about physico-chemical characteristics of water bodies in many basins around the world, achieving this relationship can facilitate in determining the quality of aquatic environments wherever no documentation on the quantity or diversity of species is available.

# CHAPTER THREE
# MATERIALS AND METHODS

## 3.1: Description of Study Area

### 3.1.1: Study Location

The study location is part of Mumias district that was curved out of the larger Butere/Mumias district in January 2007, covering 586.2 $Km^2$ (DDOM, 2008). The sampling stations mapped lie approximately within $0^o$25'N, $34^o$25E, $0^o$19'N and $34^o$ 30'E of the study location as shown in figure 3.1.

Global Positioning System (GPS), of type Garmin etrex vista HCx 16D3672296 series, was used to map sampling stations while Arc GIS program version 9.3 generated the map for accurate direction of the location and the sampling stations. The sugar factory is along Mumias-Bungoma road approximately two kilometres from Mumias town. The factory occupies 4,295 hectares of land (ECMC, 2004). Started in 1973, it is currently the largest sugar miller in Kenya, with several activities including production of sugar cane, sugar milling, electrical power generation, alcohol and marketing of sugar (KSB, 2009).

### 3.1.2: Geology and Hydrology

The DDOM, (2008) report states that Mumias district has a varying topography with a few hills and valleys dissected by a number of small streams. The geological formation of the study location is mainly of post-Kavirondian granites; pre-Cambrian volcanic and sedimentary rocks making for a good source of sand.

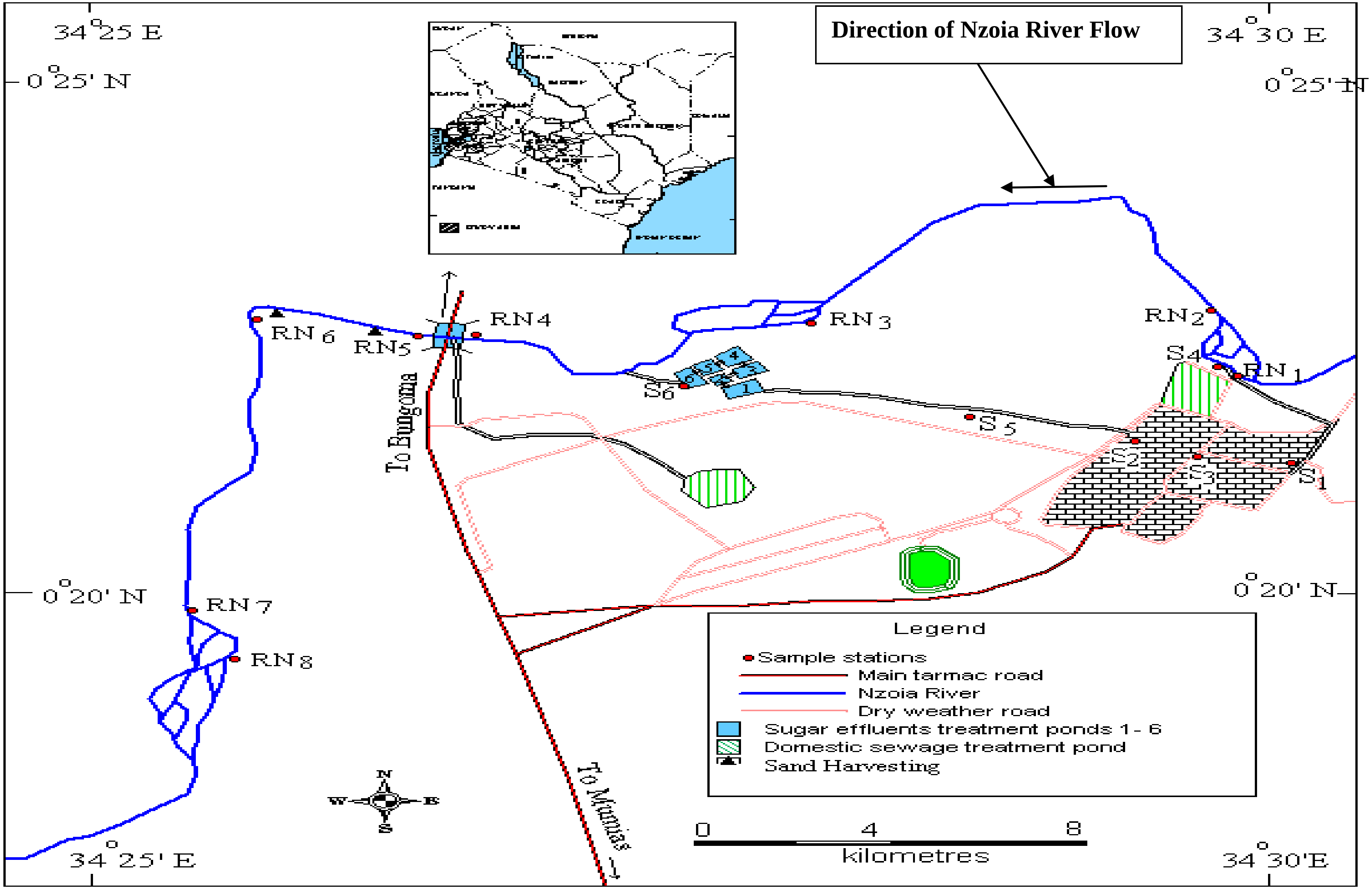


**Figure 3.1: Geographic Location of the Nzoia River and position of sampling stations. (Source: Geography Dept-Moi University)**

The soils in the sugar belt are red to dark brown ferrasols, nirosols and acrisols that are generally deep and well drained internally (KSB, 2009). A number of streams and rivers, including the Nzoia River, dissect the district resulting in ample surface water resources.

The Nzoia River system covers a catchment area of approximately 12,900 $km^2$, arising from Mount Elgon and draining into Lake Victoria, a distance of about 334 km (NRBMI, 2006; Simiyu *et al*., 2009). It is the largest among the rivers that drain from the Kenyan side of the lake (LBDA, 1987). The annual rainfall ranges between 900 and 2200 mm (Simiyu *et al*., 2009). The study location, through which the Nzoia River passes, has high temperatures all the year round with mean maximum being about 30˚C with a range of $24^0$C-$30^0$C.

There is high rainfall almost all the year round ranging between 1597–2873 $mmyear^{-1}$ becoming less intense between December and February (DDOM, 2008). Precipitation over a 10-year period (1993-2002) averaged 1900 mm in Mumias zone (KSB, 2009). The average district population density is over 593 persons $Km^2$ with the most populous areas in Mumias Municipality having 22% of the population. The high population is attributed to presence of Mumias Sugar Company.

### 3.1.3: Socio-Economic Activities

The Nzoia River is important to Western Kenya. Nationally, the river system supports major activities that include industrial and agricultural development, domestic water supplies and livestock watering. The good soils, high rainfall and the many river streams in the study location have led to farmers devoting almost 68 % of their arable land on cane production (DDOM, 2008).Similarly sand harvesting takes place within the study location between $RN_5$ and $RN_6$ while fishing and farming is along the riverbanks.

Mumias factory uses the Nzoia river waters for domestic purposes, sugar milling process and effluent discharge with likelihood of polluting and influencing its health status (Akali *et al*., 2011). Akali *et al*., 2011 attributes both point and non-point sources of water pollution the Nzoia River experiences to these major development activities in the catchment basin and the study location.

### 3.2: Selection Criteria of Water Quality Assessment Parameters

The study being an impact type formed the basis of selection criteria for the physico-chemical water parameters (Lehr *et al*., 2005 b). The physico-chemical water parameters in current study can be used to make inferences on the effects of other water attributes like alkalinity; salinity; dissolved organics; essential elements on water and phytoplankton diversity.

The use of four diversity indices achieved a more comprehensive description of the phytoplankton biodiversity (Zhao *et al*., 2013). The use of indices was based on the premise that when phytoplankton are stressed, the number of taxa and the evenness with which individuals are distributed among taxa are both reduced resulting in a lower index values (Pontasch *et al*., 1989). Data combination on abundance within phytoplankton species into diversity indices enabled easier determination of Nzoia river health status.

### 3.3: Research Design

A preliminary industrial survey involved data collection in the factory effluent generation points. A randomized experimental design was used to quantify the response of phytoplankton species diversity as a function of the physico-chemical quality of the Nzoia River due to Mumias sugar effluents (Festing, 2002). This was based on the three basic principles of experimental design that are replication, randomisation and local control (Jayaraman, 1999, Kothari, 2009).

The design enabled confirmation of cause-effect relationships, parameterization of relationships in the regression model, provision of quantitative, numeric description of data and to precisely characterize conditions (Kathuri, 1993 and; Jan, 2014). The purpose of the research was to assess the effects of effluent from Mumias sugar factory on water quality and phytoplankton species diversity of Nzoia River.

### 3.4: Sampling Design and Techniques

#### 3.4.1: Sampling Design

The study adopted a bi-weekly sampling schedule from 1$^{st}$ December 2009 to 31$^{st}$ March 2010. This was similar to Reeve *et al*., (1994), Chapman, (1996), Momanyi, (2002) and Smucker *et al*., (2014). A convenient upstream-downstream, independently repeated random sampling design was adjusted as taken up from Patton, (1990) APHA, (1999), Lehr *et al*., (2005) b, Goel, (2008) Igbinosa and Okoh (2009); and Ojunga *et al*., (2011). This overcame the problem of large number of variables that limits the sampling design while optimally satisfying the assumptions of many multivariate parametric procedures (Johnson and Wichern, 1992). The Table 3.1 indicates a profile of study sampling stations.

**Table 3.1: A Profile of Study Sampling Stations**

| Location | Sampling stations | Coordinates | Elevation (m) Asl | Remarks |
|---|---|---|---|---|
| Upstream | $S_1$ | 00.36376° N and 034.50626° E | 1266 | Milling house |
| Process | $S_2$ | 00.36478°N and 034.50299°E | 1298 | Process house |
| Boiler | $S_3$ | 00.36415°N and 034.50415°E | 1301 | Boiler house |
| Upstream | $S_4$ | 00.36729°N and 034.50478°E | 1284 | Before discharge |
| Before pond1 | $S_5$ | 00.36703° N and 034.49495° E | 1287 | Sugar effluents inlet to pond 1 of sugar effluents treatment |
| Downstream | $S_6$ | 00.36681° N and 034.49224° E | 1283 | Sugar effluents treatment outlet from pond 6 before discharge |
| Upstream | $RN_1$ | 00.36740° N and 034.50492° E | 1284 | Factory's water abstraction point & control sampling station |
| Upstream | $RN_2$ | 00.36980° N and 034.50433° E | 1283 | Impact station: Upstream Effluents discharge for $S_4$ |
| Mid-way | $RN_3$ | 00.36950° N and 034.49518° E | 1281 | Impact station: Mid-way between $RN_2$ and downstream discharge point |
| Downstream | $RN_4$ | 00.36922° N and 034.48750° E | 1277 | Impact station: Downstream sugar effluents discharge point for $S_6$ |
| Downstream | $RN_5$ | 00.36888° N and 034.48582° E | 1274 | Impact station: Downstream domestic effluents discharge point |
| Downstream | $RN_6$ | 00.36904° N and 034.48286° E | 1274 | Impact station |
| Downstream | $RN_7$ | 00.36846° N and 034.48117° E | 1270 | Impact station |
| Downstream | $RN_8$ | 00.36627° N and 034.48192° E | 1266 | Impact station |
| Change in Elevation from $RN_1$ **to** $RN_8$ (1284-1266) | | | 18 | A drop in altitude |

All the sampling stations on land were designated as S while those in the Nzoia River as RN with the subscript number differentiating the various sampling stations as indicated in Table 3.1. Sampling stations selected were from upstream, before effluent discharge at the Mumias factory water abstraction point ($RN_1$), to downstream along the Nzoia River upto $RN_8$ and on land for $S_{1-6}$. This was to determine spatial trends (Chapman, 1996; Gertraud *et al*., 1999; Lehr *et al*., 2005 b and Goel, 2008).

### 3.4.2: Sampling Techniques

A multistage sampling technique involved samples collection, 50 m after discharge points. The effluent discharges will have completely mixed with the river waters due to the high turbulence (Reeve *et al*., 1994, Jayaraman, 1999 and Ojunga *et al*., 2011).

The bottles were pre-cleaned by washing with non-ionic detergents, rinsed in tap water, 1:1 hydrochloric acid and finally with de-ionized water and labelled to indicate the date and sampling station. Each bottle was well rinsed with the respective sample before filling to ensure complete inertness.

The collecting 500 ml sample bottle and 20-µm silk plankton net were spread while standing at a safe edge on the riverbank. Oblique brisk drags were made against the river flow direction and a sample of 100 ml poured consecutively into each 500 ml polythene bottle, for the fourteen (14) sampling stations. This made the net not to touch the river bottom, resulting in subsurface sampling and vertical mixing of sample water from the river into the sample bottle (APHA, 1999, Igbinosa and Okoh, 2009 and; Ojunga *et al*., 2011). The span of the dragging rope was 2.5 m, permitting in effect a distance of about 2.0 m, thereby offering an efficiency of about 80%.

From each sampling station, a 500 ml duplicate of eight (8) independently repeated samples were collected, to measure physico-chemical water parameter loads and phytoplankton species diversity respectively. This was for maximum correlation of results and; adherence to the four fundamental principles of statistical design (APHA, 1999 and Vaux *et al*., 2012). These are: **(1)** $n > 1$ for reproducibility of knowledge; **(2)** Consideration of plausible alternative interpretations of an observed result; **(3)**

Application and generalization of conclusions to the population from the multiple measures and **(4)** The provision of important quality controls on the conduct of experiments and monitoring performance.

Phytoplankton samples, using 20-µm silk plankton net of 50cm diameter were collected. Each sample was collected in 500 ml bottle by multiple stages of sub-sampling (i.e., onsite → sample aliquot → microscopic field) which Britton *et al*., (1987), Round, (1993), Kelly *et al*., (1995) and APHA, (1999) have explained. Several sub-samples were collected from slightly different spots at sampling stations $RN_{1-8}$ and $S_6$. This was to achieve better phytoplankton representativeness and statistical results (Vollenweider, 1969 and Ojunga *et al*., 2011). Lugol's solution (0.15 ml) was added into the 500 ml bottles containing phytoplankton. The purpose was for fixing, preservation and staining (Britton *et al*., 1987). Homogenized filled up sample bottles resulted in desired water attributes to be measured and stored in cold dark environment to prevent reduction of counts as stated by Suman *et al*., (2010).

The samples were transported to the COGEN laboratory at the Mumias sugar factory immediately and analysis of $BOD_5$, COD and phytoplankton species diversity. This was 2 to 4 hours after collection and stored in refrigeration at $4^oC$ before transportation to ensure the species to be analyzed remain unchanged (Igbinosa *et al*., 2009). The ELDOWAS laboratories were used for determination of COD (Closed reflux Dichromate method) within 82 hours and $BOD_5$ after 5 days of incubation at $20\pm1^o$ C while University of Eldoret fisheries laboratories were used for phytoplankton diversity.

## 3. 5: Laboratory Procedures

For all analyses, calibration involved regular measurement of independent standards to confirm instrument accuracy (USDOE, 2012). Similarly, the Sedgewick-Rafter (SR) counting chamber was calibrated before use according to Gertraud *et al*., (1999), Sakset and Chankaew, (2013) and Biological Surveys, (2014).

### 3.5.1: Determination of pH, EC, TDS, DO and temperature

Hach portable meters from COGEN laboratory at the Mumias sugar factory were used to measure *in situ* liquid samples temperature, pH, electrical conductivity (EC) and Total dissolved solids (TDS) (Suman *et al*., 2010). A glass mercury-filled thermometer of -10 to +110 $^{0}$C in 1$^{0}$C divisions, 30 cm long and diameter of 7 mm determined the temperature. A calibrated pH meter (HACH-Sension 4) using buffers of pH 4.0, 7.0, and 10.0 was used to make the readings. The TDS/EC meter probe (HACH Sension7) key was changed to TDS to record readings after those for EC. An electrode method using Dissolved Oxygen (DO) meter (HACH-multi-HQ4od) determined the DO.

### 3.5.2: Determination of Biochemical Oxygen Demand ($BOD_5$)

Dilution method was adapted and an automated incubator (HACH $BOD_5$TrakTM) used according to Bartram *et al*., (1996), Hach *et al*., (1997), APHA (1999) and Lehr *et al*., (2005a). Each effluent samples were well homogenized and pipetted into twelve separate standardized dark bottles of 300 ml after 5 days under a set of standard environmental conditions (Lehr *et al*., 2005a). The $BOD_5$ read-out from the incubator as stated by Hach *et al*., 1997, was mathematically based the following equations:

mg/l BOD = (slope x 300) – Y intercept = undiluted sample DO…………**Equation 1**

mg/l BOD = (a x b) – c = d…………………………………………….. **Equation 2**

**a.** The slope of the line is equal to the mg/l DO consumed per ml of sample taken. At any point on the line, the mg/l DO remaining there is subtracted from the mg/l DO where the line crosses the DO scale. Dividing the difference by the ml of sample at the point chosen provides the slope.

**b.** The value 300 is the volume of the BOD bottle.

**c.** The "Y intercept" is the DO value where the line crosses the DO scale.

**d.** The sample DO is the DO of the undiluted sample.

The $BOD_5$Trak apparatus measured the drop in pressure and displayed the results directly as mg/l BOD.

### 3.5.3: Determination of Chemical Oxygen Demand (COD)

The closed reflux dichromate method (APHA, AWWA and WEF, 1998) was adopted, modified and used spectrophotometer (Hach DR, 2010 and, Lawson-Wood and Robertson). Samples of 100ml and 2,00ml deionized water were homogenized, preheated to 150$^o$C, 0.2ml pipetted into vials, digested for two(2) hours in vials using a dichromate reactor(s/n0106000087680 under closed reflux conditions and placed in racks to cool(LaPara et al.,2000). The Hach's COD vial kit provided colour change between orange to green based on the amount of oxidation to the COD concetration on the spectrophotometer. The Hydrogen Pthalate(KHP) was the reference standard. The spectrophotometer generated a standard calibration curve by measuring concetration of dichromate and their absorbance for COD determination in mgl$^{-1}$. The decrease in dichromate ion was measured spectrophotometrically at 445nm and was directly proportional to the mass of oxygen consumed per litre of solution as mentioned by Lawson-Wood and Robertson,(2016). The mg/LCOD is therefore equivalent to mg/l $O_2$.

### 3.5.4: Phytoplankton Species Diversity Determination

Determination of the phytoplankton composition at species and community level involved microscopic enumeration and identification of the lowest taxonomic unit using dichotomous identification keys and illustrations (Palmer 1959; Needham *et al*., 1962; Palmer 1969; APHA, 1998, Sanet *et al*., 2006, Hauer *et al*., 2007 and; Bellinger *et al*., 2010). A Sedgewick Rafter (S-R) counting chamber of a 50 mm by 20 mm by 1mm with a grid floor/graticule (1,000 small squares) and a raised-rim chamber holding 1 ml as seen in figure 3.2 was used (Greenberg *et al*., 1987, Gertraud *et al*., 1999, Suman *et al*., 2010, Rajagopal, 2010, Hossain *et al*., 2013, Sakset and Chankaew, 2013 and; Biological Surveys, 2014).

Each of the 1,000 grid floor/graticule fields thus contains 1µml of the 1ml (50 mm x 20 mm x 1mm). Each of the preserved 500ml sample bottles were gently shaken and inverted thoroughly for at least 30 seconds to mix well to avoid sub-sampling errors before sub-sampling.

A pipette was used to measure 1.0 ml homogenised sample and fill the chamber with the cover slip gently nudged to cover the chamber completely as shown in figure 3.2. It was then placed directly under an upright X100 monocular microscope Swift© model M3200 with a calibrated eyepiece.

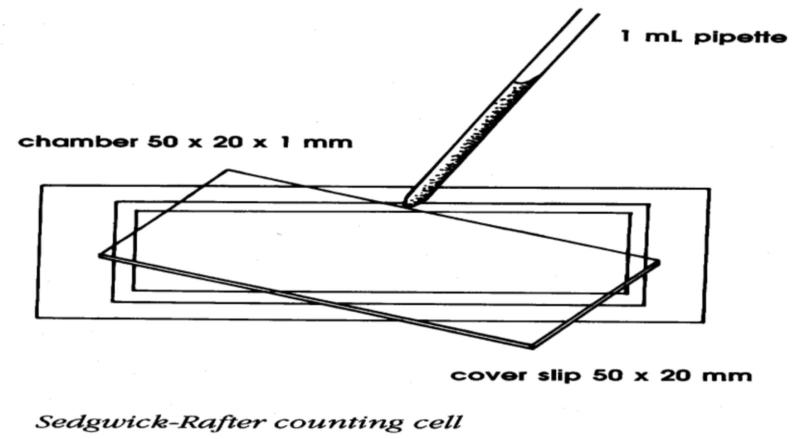


**Figure 3. 2: Sedgewick-Rafter Counting Chamber**
(**Source: Biological Surveys, 2014)**

Phytoplankton species mean countml$^{-1}$ by subsequent sub-sampling of the grid floor/graticule fields involved standard procedures (Gertraud *et al*., 1999 and Suman *et al.*, 2010). The size of S-R was calibrated then used to modify and derive equation1 for phytoplankton cell counts ml$^{-1}$ such that:

From the 1.0 ml homogenised sample, phytoplankton counts was made in upto thirty (30) Number of Fields (NF) constituting,

TC as Total Counts and DF the Dilution Factor.

While,

One field= (1 x TCNF$^{-1}$) cell counts of 17 small squares at

x100 magnification= (1 x TCNF$^{-1}$) cell counts.

Thus: 1,000 small squares in a 50mm x 20mm x 1mm (ml$^{-1}$) S-R counting chamber constituted:

1,000 x 1 x TC x 1 x DF (NF x17)$^{-1}$ countsml$^{-1}$…............................**Equation 3**

(APHA, 1998 and Ishaq and Khan, 2013).

The phytoplankton cell countsml$^{-1}$ determined phytoplankton species diversity

(Britton *et al*., 1987). The diversity of phytoplankton due to responses of species to pollution levels (Dlamini, 2009) was determined by use of equations 4 to 7 of the following four indices:

Shannon-Weiner (SW) species diversity ($H_s$) index:

$$H_s = - \sum_{i=1}^{S} (Pi)\ (\ln Pi)$$ ..........**Equation 4**,

Where:

$H_s$ is SW index in a sample of S species;

S the number of species in the sample;

$Pi = niN^{-1}$, the relative abundance of ith species;

N is the total individual number of all species in the community;

ni is the individual number of the ith species and

ln = the natural log (Shannon *et al*., 1949, Adelasu *et al*., 2008; Zhao *et al*., 2013 and; Kumar and Sharma, 2014).

Margalef's species richness (d): d=S-l/InN..........**Equation 5**,

Where:

d is Margalef's diversity index;

S is Total number of species and

N is number of individuals (Margalef, 1958).

Pielou species evenness (J): $J= H_S /\ln S$..........**Equation 6**,

Where:

$H_S$ is Shannon-Wiener index;

J is Evenness and

S is total number of species (Pielou, 1977).

Simpson's Diversity Index= (1–Ds)............................................................**Equation7,**

was derived from Simpson's index (Ds): Ds=Σn (n-1)/N (N-1),

Where:

Ds = Simpson diversity dominance after introducing bias correction,

N = the total number of organisms of all species and

n = the total number of organisms of a particular species.

(Magurran, 2004 and Zhao *et al*., 2013).

### 3.6: Statistical Data Analysis

The obtained data were subjected to univariate and multivariate statistical analysis considered significant at $\alpha < 0.05$ (Wold, 1991; Geladi and Dåbakk, 1995; Mugenda and Mugenda, 1999; Amany and Mohamed, 2003; Tavernini *et al*., 2009; Chia *et al*., 2011 and USDOE, 2012). All the means, standard errors, phytoplankton composition at species and community level; diversity indices were determined by univariate analysis on EXCEL 2007 for windows 7 while principal component analysis (PCA) and stepwise multiple linear regression (SMLR) used multivariate statistical data analysis on STATISTICA program version 8.0 (StatSoft, 2007 and Sabah, 2013).

One-way analysis of variance (ANOVA) and Duncan multiple range test (DMRT) compared the same physico-chemical parameters amongst different sampling stations in $S_{1-3}$ and $RN_{1-8}$ respectively (StatSoft, 2007; Igbinosa *et al*., 2009 and Miltonprabu and Sumedha, 2013). Two-way dependent t-test described significant spatial physico-chemical parameter trends between $S_1$ and $S_4$; $S_2$ and $S_5$ and; $S_5$and $S_6$ (Sabah, 2013; Farahnaky *et al*., 2014 and; Salim *et al*., 2014). One-way 2-tailed independent t-test compared Mumias sugar effluents physico-chemical quality before ($S_4$ and $S_6$) and at discharge ($RN_2$ and $RN_4$) upstream and downstream respectively with the hypothetical mean values of NEMA and WHO (NEMA, 2003; WHO, 2006; Polonsky and Waller, 2009 and; Fella *et al*., 2013).

Statistical Package for the Social Sciences Software (SPSS) version 11.5 for Windows was used to generate Kaiser-Meyer-Olkin (KMO) and Bartlett's test of sphericity values that determine data suitability for a good PCA (SPSS, 2007, Jagadeesan *et al*., 2011 and; Wuttichaikitcharoen and Babel, 2014). The PCA, based on multiple criteria, extracted possible factors of variability influencing physico-chemical quality between sampling stations $RN_{1-8}$ (Braak ter, 1986; Manly, 1986; Haan, 2002; Landau and Everitt, 2003; Pallant, 2005; Shrestha and Kazama, 2007; Bongumusa, 2010; Williams *et al*., 2010; Jagadeesan *et al*., 2011; Ogbuagu *et al*., 2012 and; Wuttichaikitcharoen and Babel, 2014). The most significant principal components (PCs) were selected to evaluate their contribution to the variation in phytoplankton species diversity at sampling stations $RN_{1-8}$ by SMLR for interpretation (Suman *et al*., 2010, Mendes 2011 and; Mustapha and Abdu, 2012).

**3.7: Study Assumptions**

The analytes of interest were assumed to be uniformly distributed at each sampling station during the study period. This is because analytes like dissolved oxygen, temperature and pH can fluctuate over a diurnal stage (Stumm and Morgan, 1996). Similarly, phytoplankton species diversity was assumed to be affected by the Nzoia river quality due to Mumias sugar factory effluents discharge. This is because absence of a species and phytoplankton species diversity is dependent on other factors like the effects of predation, competition or geographic barriers besides the pollution status of a water body (Ramakrishnan, 2003 and Baldwin *et al*., 2005).

**3.8: Ethical Issues**

The approval of the proposal by the SES Academic Board enabled granting of consent by the Mumias sugar company for an industrial attachment as seen in appendices II, III and IV.

# CHAPTER FOUR
# RESEARCH RESULTS

## 4.1: Introduction

The assessment of the effects of effluent from Mumias sugar factory on water quality and phytoplankton species diversity of River Nzoia was from 1st December 2009 to 31st March 2010. Data on the physico-chemical water parameters loads of Mumias sugar factory effluents was established and its quality before discharge assessed. Thereafter river Nzoia water quality was assessed, phytoplankton species diversity established and; a statistical relationship established between phytoplankton species diversity and the physico-chemical river Nzoia water quality. The spatial trends of the physico-chemical water parameter loads from $RN_{1-8}$ and effect on the phytoplankton species diversity is inclusive of the results presented.

## 4.2: Determination of the Physico-Chemical Water Parameters Loads of Mumias Sugar Factory Effluents Before and After Treatment

The results presented in Table 4.1 indicate physico-chemical loads for the effluent generation sample stations values for $S_1$(milling), $S_2$(process) and $S_3$(boiler), variability of the physico-chemical water parameter loads from $S_2$–$S_5$(before pond1) to treatment ponds 1-6 and through meanders from $S_1$-$S_4$ before upstream discharge point. The effluent quality before discharge at $S_4$ and $S_6$ (after pond 6) by comparison with NEMA and WHO values are presented. The determinations of the physico-chemical water parameters loads of Mumias sugar factory effluents before and after treatment are presented in Table 4.1 and described in sections 4.2.1 to 4.2.7.

### 4.2.1: The pH

The mean±SE pH values for $S_4$ (7.92±1.17) upstream before discharge and $S_6$ (7.52±0.09) downstream before discharge were both within both discharge limits. The process ($S_2$) had the highest mean±SE pH value of 9.48±1.91 while boiler ($S_3$) had the lowest at 6.40±0.4100.

The values for $S_1$, $S_2$ and $S_3$ were significantly different (one-way ANOVA, $p<.05$). The pH values between $S_2$ and; $S_1$ were significantly different (DMRT, $p<.05$) and $S_3$ (DMRT, $p<.05$) respectively. The pH decreased significantly from $S_2$ (9.48±1.91) to $S_5$ (8.52±1.89) (t= 14.57753; $p<.05$) while the increase from $S_1$ (6.89 ±1.18) to $S_4$ (7.92±1.17) and decrease from $S_5$ (8.52±1.89) to $S_6$ (7.52±0.09) was not significant.

### 4.2.2: Total Dissolved Solids (TDS)

The mean±SE TDS ($mgl^{-1}$) values for $S_2$ (1466.51±257.51) was the highest. The boiler ($S_3$) had the lowest value of 101.59±34.26. Highly significant values less than both discharge limits were at $S_4$ (164.49±25.27) (WHO: t=-205.417, $p<.05$ and NEMA: t= -115.887, $p<.05$) and $S_6$ (928.80±22.80) (WHO: t= -46.9394, $p<.05$and NEMA: t=-11.8838, $p<.05$).

The differences of TDS between $S_1$, $S_2$ and $S_3$ was highly significant (one-way ANOVA, $p<.05$). The differences between $S_2$ and; $S_1$ was highly significant (DMRT, $p<.05$) and $S_3$ (DMRT, $p<.05$) respectively. The TDS decrease from $S_2$ (1466.51±257.51) to $S_5$ (1191.20±128.81) (t= 6.050313; $p<.05$) and; $S_5$ (1191.20±128.81) to $S_6$ (928.80±22.80) (t=6.953993; $p<.05$) was a highly significant while an increase from $S_1$ (130.28±34.02) through the meanders to $S_4$ (164.49±25.27) (t=-5.28555; p<.05) was highly significant.

### 4.2.3: Biological Oxygen Demand ($BOD_5$)

The highest mean±SE $BOD_5$ ($mgl^{-1}$) values were at $S_2$ (7109.63±1460.52) and lowest at $S_6$ (172.38±14.46). Highly significant values more than both discharge limits were at $S_4$ (827.75±132.59) (WHO: t=16.590, p<.05 and NEMA: t=17.017, p<.05) and $S_6$ (172.38±14.46) (WHO: t=8.4635, p<.05and NEMA: t=9.8467, p<.05).

There was statistically highly significant differences between $S_1$, $S_2$ and $S_3$ (ANOVA: p=0.000000). There was highly significant differences of $BOD_5$ between $S_2$ and., $S_1$ (DMRT: p<.05) and $S_3$ (DMRT: p<.05) respectively. The $BOD_5$ indicated a highly significant increase from $S_1$ (646.88±137.16) to $S_4$ (827.75±132.59) (t=-8.35635; p<.05) but a highly significant decrease from $S_5$ (6600.00±1207.08) to $S_6$ (172.38±14.46) (t=15.12605; p<.05) and $S_2$ (7109.63±1460.52) to $S_5$ (6600.00±1207.08) (t=4.589135; p<.05).

### 4.2.4: Chemical Oxygen Demand (COD)

The mean±SE COD ($mgl^{-1}$) values were highest at $S_2$ (40762.13±25654.08) and lowest at $S_6$ (443.50±18.03). The sampling stations $S_4$ (1957.13±681.56) (WHO: t= 3.972, p<.05 and NEMA: t=7.914, p<.05) and $S_6$ (443.50±18.03) (WHO:t=-30.8657, p<.05 and NEMA: t=21.8251, p<.05) had highly significant values beyond both discharge limits. There was statistically significant differences betweenS$_1$, $S_2$ and $S_3$ (one-way ANOVA: p<.05). There was highly significant differences of COD between $S_2$ and; $S_1$ (DMRT: p<.05) and $S_3$ (DMRT: p<.05) respectively. The COD significantly decreased from $S_2$ (40762.13±25654.08) to $S_5$ (33669.25 8155.42) ($t=2.105720$; $p<.05$) while having a highly significant decrease from $S_5$ (33669.25±23067.02) to $S_6$ (443.50±18.03) ($t=4.068856$; $p<.05$). There was no significant difference of COD mean± SE values between $S_1$ and $S_4$.

### 4.2.5: Dissolved Oxygen (DO)

The mean±SE DO ($mgl^{-1}$) values were highest at $S_6$ (4.04±0.57) and the lowest at $S_2$ (1.65±1.86). There was significant DMRT difference between $S_2$ and $S_3$ DMRT: ($p<.05$). This was despite the difference between $S_1$, $S_2$ and $S_3$ (one-way ANOVA: $p>0.05$) not being statistically significant. There was significant DO increase from $S_1$ (2.99±0.61) to $S_4$ (4.50±1.25) ($t=-3.18253$; $p<.05$) and $S_5$ (2.88±0.61) to $S_6$ (4.04±0.57) ($t=-3.22661$; $p<.05$) whileS$_2$ (1.65±1.86) to $S_5$ (2.88±1.71) ($t=-4.48396$; $p<.05$) had a highly significant increase.

### 4.2.6: Electrical Conductivity (EC)

The mean±SE EC ($\mu scm^{-1}$) values were highest atS$_6$ (1466.85±197.12). The lowest were at $S_1$ (240.64±57.37). The mean±SE values at $S_4$ (306.83±175.19) were less than WHO and highly significant ($t=-11.191$, $p<.05$) while at $S_6$ (1466.85±197.12) ($t=2.3684$, $p<.05$) were significantly more than WHO discharge limits. There was statistically highly significant EC differences between $S_1$, $S_2$ and $S_3$ (one-way ANOVA: $p<.05$) and; between $S_2$ and; $S_1$ (DMRT: $p<.05$) and $S_3$ (DMRT: $p<.05$) respectively. There was no significant change from $S_1$ to $S_4$; $S_5$ to $S_6$ and; $S_2$ to $S_5$.

**Table 4. 1: The Physico-Chemical Parameter Loads for $S_{1-6}$ of Mumias Sugar Factory Effluents Before and After Treatment**

| Mean±Standard Error (M±SE) | | | | | | | ANOVA | Discharge Standards | |
|---|---|---|---|---|---|---|---|---|---|
| **Variables** | **$S_1$** | **$S_2$** | **$S_3$** | **$S_4$** | **$S_5$** | **$S_6$** | **$S_{1-3}$** | **NEMA** | **WHO** |
| pH | 6.89 ±1.18 | 9.48±1.91*** | 6.40±0.4100 | 7.92±1.17 | 8.52±1.89 | 7.52±0.09 | 0.000244*** | 6.5-8.5 | 6-9 |
| TDS $mgl^{-1}$ | 130.28±34.02 | 1466.51±257.51*** | 101.59±34.26 | 164.49±25.27 | 1191.20±128.81 | 928.80±22.80 | 0.000000*** | 1,200 max | 2,000 max |
| $BOD_5$ $mgl^{-1}$ | 646.88±137.16 | 7109.63±1460.52 | 488.63±205.77 | 827.75±132.59 | 6600.00±1207.08 | 172.38±14.46 | 0.000000*** | 30 max | 50 max |
| COD $mgl^{-1}$ | 1775.13±840.05 | 40762.13±25654.08 | 1809.50±854.40 | 1957.13±681.56 | 33669.25±23067.02 | 443.50±18.03 | 0.000024*** | 50 max | 1,000 max |
| DO $mgl^{-1}$ | 2.99±0.61 | 1.65±1.86** | 3.52±1.93 | 4.50±1.25 | 2.88±1.71 | 4.04±0.57 | **NS** | - | 5.00 max |
| EC $\mu scm^{-1}$ | 240.64±57.37 | 1204.97±160.37*** | 190.49±63.92 | 306.83±175.19 | 948.71±444.83 | 1466.85±197.12 | 0.000000*** | - | 1,000 max |
| Temp $^0C$ | 36.88±7.75 | 52.00±11.26*** | 46.13±11.03 | 37.88±2.03 | 36.13±1.81 | 24.88±0.44 | 0.023263** | - | 40 |

**Note that the following denotes: ***Highly Sig. DMRT and ANOVA values of p<.005; **Sig.DMRT and ANOVA values of p<.05 and NS: Not Sig.**

### 4.2.7: Temperature

The mean±SE values at $S_4$ (37.88±2.03) ($t=-2.959$, $p<.05$) were significant and less while at $S_6$ (24.88±0.44) ($t=-34.3222$, $p<.05$) were highly less significant than WHO discharge limits. The highest temperature mean value was at $S_2$ (52.00±11.26). The differences between $S_1$, $S_2$ and $S_3$ were statistically significant (one-way ANOVA: $p<.05$). Subsequent DMRT showed highly significant differences between $S_2$ and; $S_1$ ($p<.05$). Highly significant temperature decrease were from $S_2$ (52.00±11.26) to $S_5$ (36.13±1.81) ($t=-3.590257$; $p<.05$) and; $S_5$ (36.13±1.81) to $S_6$ ( 24.88±0.44) ($t=-24.82548$; $p<.05$).

These results indicate variations for the mean values of the physico-chemical parameter loads of Mumias sugar factory effluents before and after treatment.

### 4.3: Assessment of the Physico-Chemical Water Quality Parameters in the Nzoia River Upstream and Downstream Mumias Factory Effluent Discharge Points

The Table 4.2 shows the Mean±SE, range, one-way ANOVA and DMRT of physico-chemical water quality parameters for $RN_{1-8}$ of the Nzoia River upstream and downstream of effluent discharge points. Upon effluents discharge, the Nzoia River physico-chemical water quality at $RN_2$ (upstream) and $RN_4$ (downstream) discharge sample stations was determined.

### 4.3.1: The pH

Figure 4.1 and Table 4.2 indicates that all the pH mean±SE values for $RN_{1-8}$ were within the stated NEMA (6.5-8.5) and WHO (6-9) discharge limits.

The mean±SE values of pH at $RN_2$ (7.67±0.18) and $RN_4$ (7.25±0.08) were not significantly different from WHO discharge standards while domestic discharge point ($RN_5$) (7.28±0.06) was significantly lower than NEMA values (t=-3.816) (p<.05). There was an observed increase in pH mean±SE values from the reference sampling station $RN_1$ (7.59±0.19) to the upstream sugar milling effluents discharge point $RN_2$ (7.67±0.18) thereafter decreasing to the lowest level at $RN_8$ (7.11±0.21). The pH mean±SE values had statistically significant differences between $RN_{1-8}$ (ANOVA: p<.05**).

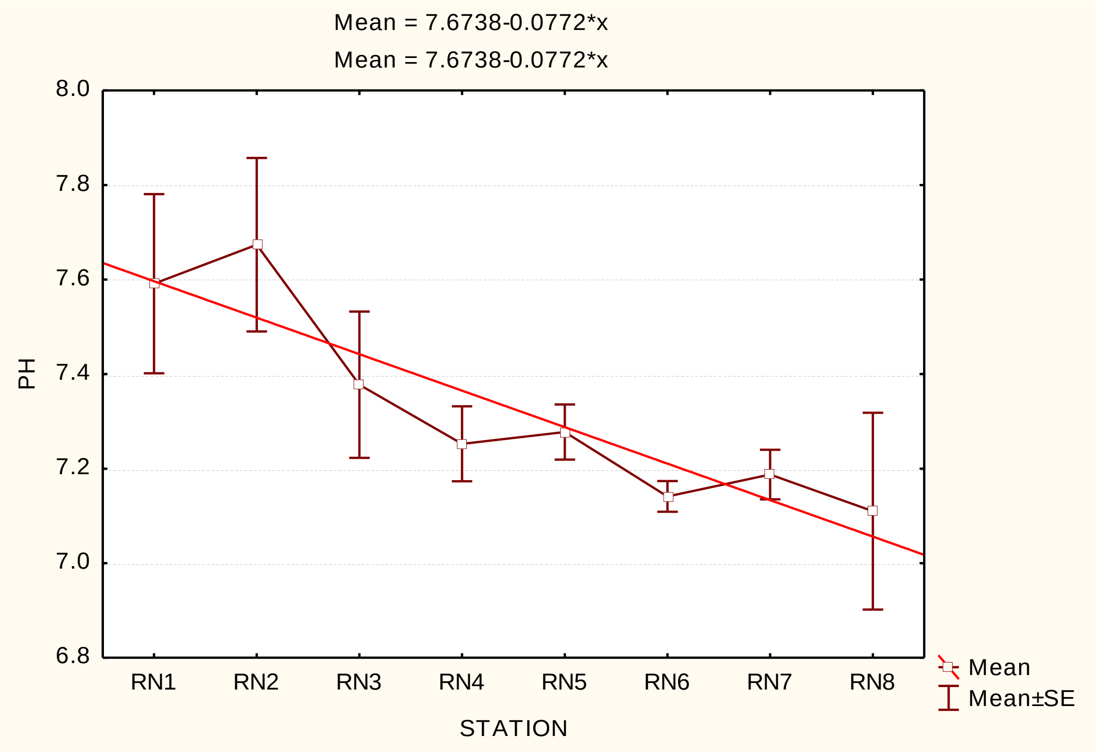


**Figure 4. 1: Mean±SE pH quality variability in the Nzoia River**

There was significant differences between $RN_6$ and; $RN_1$ (DMRT: p<.05**) and $RN_2$ (DMRT: p<.05**) respectively while $RN_7$ showed with $RN_2$ (DMRT: p<.05**).

**Table 4. 2: The Physico-Chemical Water Quality In The Nzoia River Upstream And Downstream Of Mumias Sugar Factory Effluent Discharge Points**

| Parameters | Mean±Standard Error (M±SE) and Range | | | | | | | | Discharge Stds | | ANOVA |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | $RN_1$ | $RN_2$ | $RN_3$ | $RN_4$ | $RN_5$ | $RN_6$ | $RN_7$ | $RN_8$ | NEMA | WHO | $RN_{1-8}$ |
| pH | 7.59±0.19 | 7.67±0.18 | 7.38±0.16 | 7.25±0.08 | 7.28±0.06 | 7.14±0.03 | 7.19±0.05 | 7.11±0.21 | 6.5-8.5 | 6-9 | 0.039733** |
| TDS $mgl^{-1}$ | 52.45±2.22 | 53.65±2.19 | 52.83±2.15 | 53.05±2.59 | 56.08± 3.03 | 53.95± 2.60 | 53.99±2.59 | 53.59±2.50 | 1,200 max | 2,000 max | **NS** |
| $BOD_5$ $mgl^{-1}$ | 16.75± 0.53 | 18.00± 0.54 | 17.00± 0.57 | 25.63± 4.08 | 31.63± 4.99 | 18.88± 1.49 | 19.50± 1.38 | 17.50±1.306 | 30 max | 50 max | 0.000388*** |
| COD $mgl^{-1}$ | 24.38± 1.48 | 28.63± 1.00 | 24.75± 0.45 | 51.00± 9.97 | 53.88± 9.96 | 45.88± 7.48 | 44.63± 9.86 | 39.25±7.24 | 50 max | 1,000 max | 0.016885** |
| DO $mgl^{-1}$ | 6.14± 0.31 | 5.55± 0.32 | 5.76± 0.34 | 5.85± 0.34 | 5.62±0.32 | 5.86± 0.30 | 5.60±0.32 | 5.34± 0.36 | | 5.00 max | **NS** |
| EC $\mu scm^{-1}$ | 99.97± 4.19 | 102.50± 5.36 | 100.95± 5.09 | 100.40± 4.92 | 115.89± 12.47 | 104.20± 6.91 | 107.03±6.86 | 106.35± 6.67 | | 1,000 max | **NS** |
| Temp $^0C$ | 24.38± 0.32 | 24.50± 0.33 | 24.50± 0.33 | 24.50± 0.33 | 24.50± 0.93 | 24.50± 0.33 | 24.50± 0.33 | 24.50± 0.33 | | 40 | **NS** |

**Note that the following denotes**: ***: Highly significant values of p<0.005., **: Significant values of p<.05 and NS: Not Significant

### 4.3.2: The Total Dissolved Solids (TDS)

The TDS ($mgl^{-1}$) $RN_{1-8}$ mean±SE values were less than both NEMA (1,200) and WHO (2,000) limits, having minimal changes as indicated in Table 4.4 and figure 4.2. The low mean±SE values of TDS when compared to both NEMA and WHO limits at $RN_2$ (53.65±2.19) for NEMA (t=-522.497) ($p<.05$) while (t=-887.130) ($p<.05$) for WHO; $RN_4$ (53.05±2.59) for NEMA (t=-443.389) ($p<.05$) while (t=-752.654) ($p<.05$) for WHO and $RN_5$ (53.05±2.59) for NEMA (t=-378.017) ($p<.05$) while (t=-642.382) ($p<.05$) for WHO were highly significant.

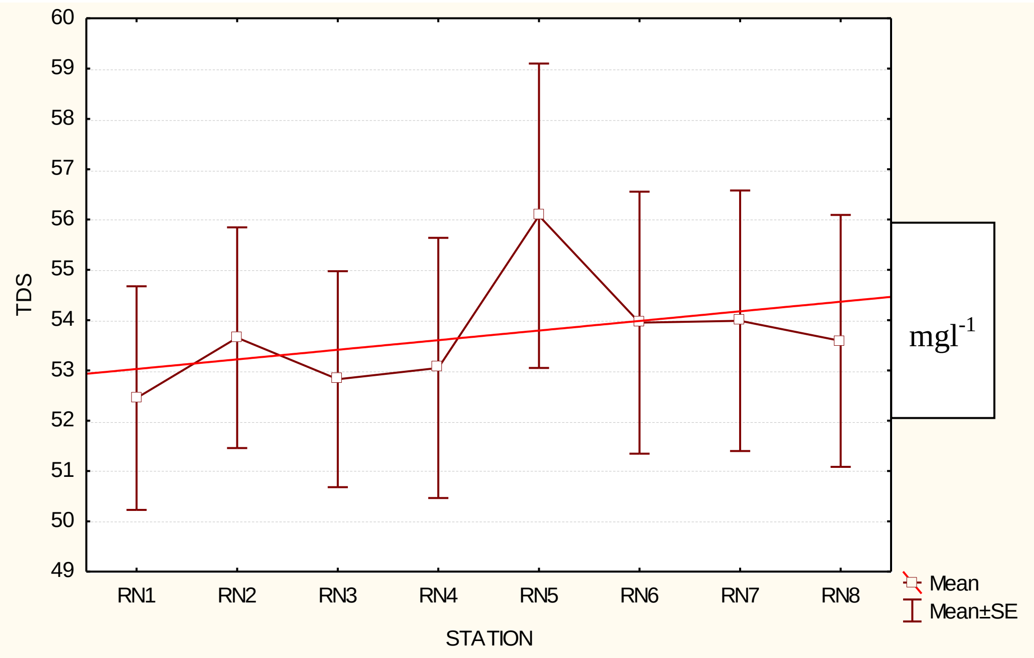


**Figure 4. 2: Mean±SE TDS quality variability in the Nzoia River**

The low mean±SE values of TDS when compared to both NEMA and WHO limits at $RN_2$ (53.65±2.19) for NEMA (t=-522.497) ($p<.05$) while (t=-887.130) ($p<.05$) for WHO; $RN_4$ (53.05±2.59) for NEMA (t=-443.389) ($p<.05$) while (t=-752.654) ($p<.05$) for WHO and $RN_5$ (53.05±2.59) for NEMA (t=-378.017) ($p<.05$) while (t=-642.382)

(p<.05) for WHO were highly significant. There was an observed increase of TDS mean±SE values from downstream sugar ($RN_4$:53.05±2.59) to domestic ($RN_5$:56.08±3.03) effluents discharge points. There was no statistically significant differences for TDS between $RN_{1-8}$ (ANOVA: p>0.5).

### 4.3.3: The $BOD_5$ and COD

From Table 4.2 and figure 4.3, the $BOD_5$ ($mgl^{-1}$) $RN_{1-8}$ mean±SE values were all below NEMA (30) and WHO (50) discharge standards.

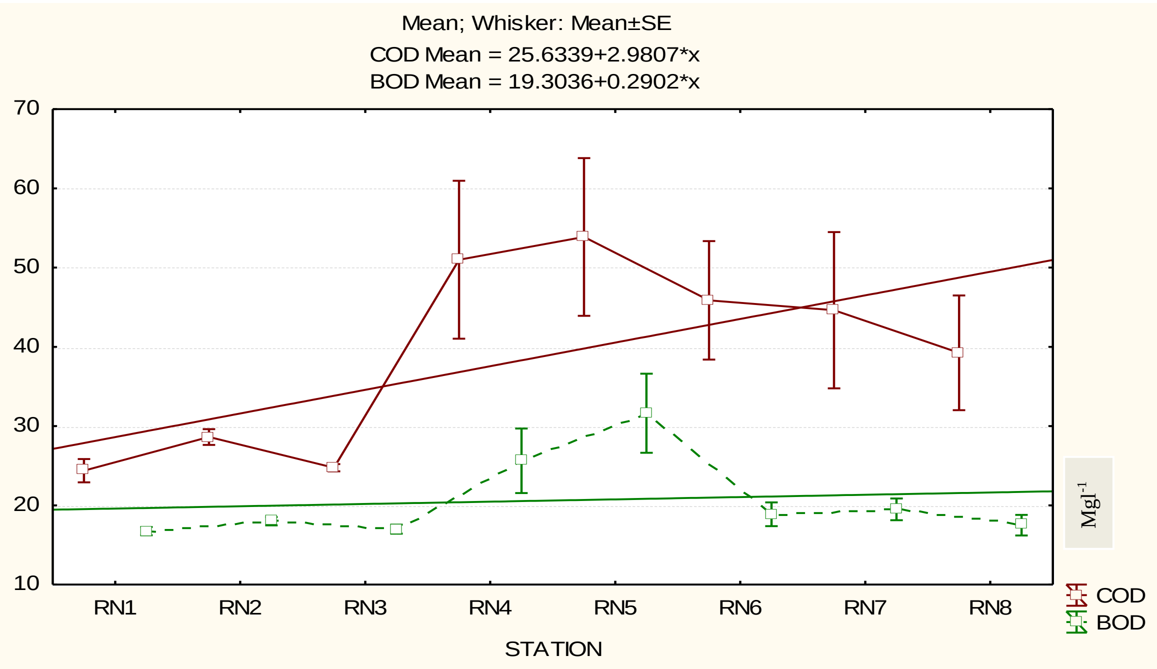


**Figure 4. 3: Mean±SE $BOD_5$ and COD quality variability in the Nzoia River**

The mean±SE values at $RN_2$ (18.00±0.54) (NEMA: t=-22.450: p<.05and WHO: t=-59.867; p<.05), $RN_4$ (25.63±4.08) (WHO: t=-5.975; p<.05) and $RN_5$ (31.63±4.99) (WHO: t=-3.683; p<.05) were highly significant and less than the indicated discharge limits.

The COD ($mgl^{-1}$) $RN_{1-8}$ mean±SE values were all below NEMA (50) and WHO

(1,000) discharge standards except at $RN_4$ (51.00±9.97) and $RN_5$ (53.88±9.96) that were more than NEMA Discharge limits. The COD mean±SE values at $RN_2$ (28.63±1.00) (NEMA: t=-21.399: $p<.05$and WHO: t=-972.461; $p<.05$); $RN_4$ (51.00±9.97) (WHO: t=-95.207; $p<.05$) and $RN_5$ (53.88±9.96) (WHO: t=-95.207; $p<.05$) were highly significant and less than WHO.

The $BOD_5$ and COD mean±SE values at $RN_4$ and $RN_5$ respectively were not significantly different from the NEMA discharge limits. Figure 4.3 further indicates an increase of both $BOD_5$ and COD from $RN_1$ to $RN_2$ while decreasing at $RN_3$. Thereafter an increase occurs from $RN_3$ to $RN_5$ with a gradual decrease to RN8. The $BOD_5$ showed statistically significant differences between $RN_{1-8}$ (ANOVA: $p<.05$***). There was statistically significant differences between $RN_1$ with $RN_4$ (DMRT: $p<.05$**) and $RN_5$ (DMRT: $p<.05$***); $RN_2$ with $RN_4$ (DMRT: p=0.047909**) and $RN_5$ (DMRT: $p<.05$***) and $RN_3$ with $RN_4$ (DMRT: $p<.05$**) and $RN_5$ (DMRT: $p<.05$***); RN4 with $RN_8$ (DMRT: $p<.05$**); $RN_5$ with $RN_6$ (DMRT: $p<.05$***), RN7 (DMRT: $p<.05$***) and $RN_8$ (DMRT: $p<.05$***). One-way analysis of variance (ANOVA) of COD showed statistically significant differences between $RN_{1-8}$ (ANOVA: $p<.05$**). There was statistically significant differences of COD mean±SE values between $RN_1$ with $RN_4$ (DMRT: $p<.05$**) and $RN_5$ (DMRT: $p<.05$**); $RN_2$ with $RN_5$ (DMRT: $p<.05$**) and; $RN_3$ with $RN_4$ (DMRT: $p<.05$**) and $RN_5$ (DMRT: $p<.05$**).

**4.3.4: The Dissolved Oxygen (DO)**

The entire DO ($mgl^{-1}$) $RN_{1-8}$ mean±SEvalues seen in Table 4.2 and figure 4.4 are greater than WHO (5.00) discharge standards with the highest (6.14±0.31) being at the reference sampling station ($RN_1$). These values decreased at $RN_2$ (5.55±0.32), increasing upto $RN_4$ (5.85±0.34) while decreasing at $RN_5$ (5.62±0.32).

The mean±SE values of DO at $RN_2$ (5.55±0.32) and RN5 (5.62±0.32) did not have statistically significant difference with WHO ($p>0.05$) while those at $RN_4$ (5.85±0.34) ($t=2.465$, $p<.05$) were significantly more than WHO discharge standards. There was subsequent increase at $RN_6$ (5.86±0.30) and thereafter an observed continued decrease upto $RN_8$. There were no statistically significant DO differences between $RN_{1-8}$.

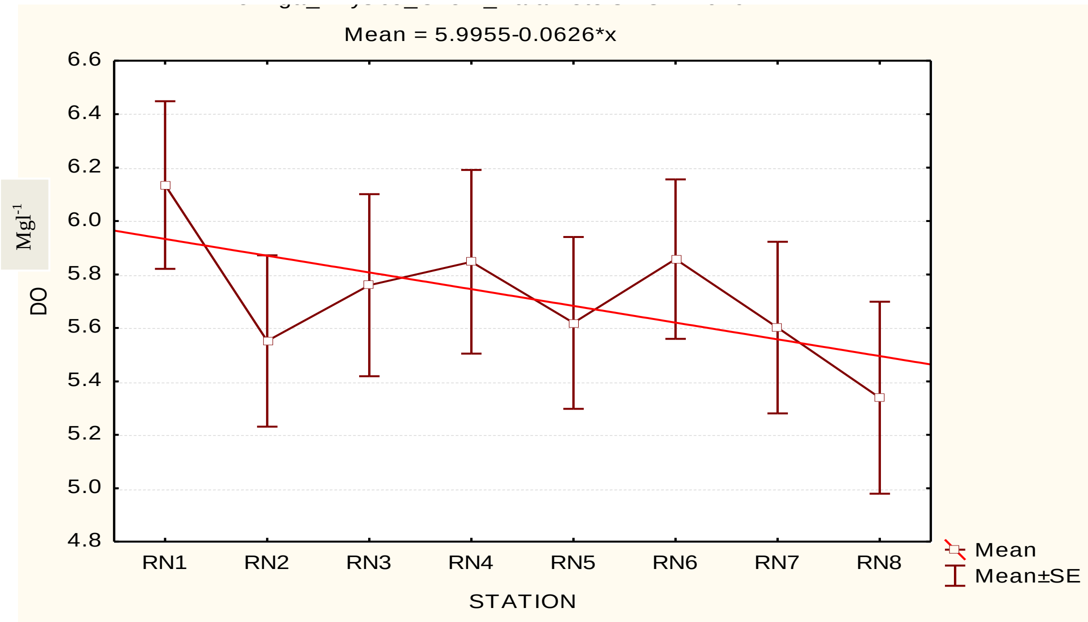


**Figure 4. 4: Mean±SE DO quality variability in the Nzoia River**

### 4.3.5: The Electrical Conductivity (EC)

The EC (µscm$^{-1}$) $RN_{1-8}$ mean±SE values in Table 4.2 and figure 4.5 were all less than WHO (1,000) discharge limits. The differences with WHO values at $RN_2$

(102.50±5.36) (t=-167.548) (p<.05), $RN_4$ (100.40±4.92) (t=-182.898) (p<.05) and $RN_5$ (115.89±12.47) (t=-70.907) (p<.05) were statistically highly significant.

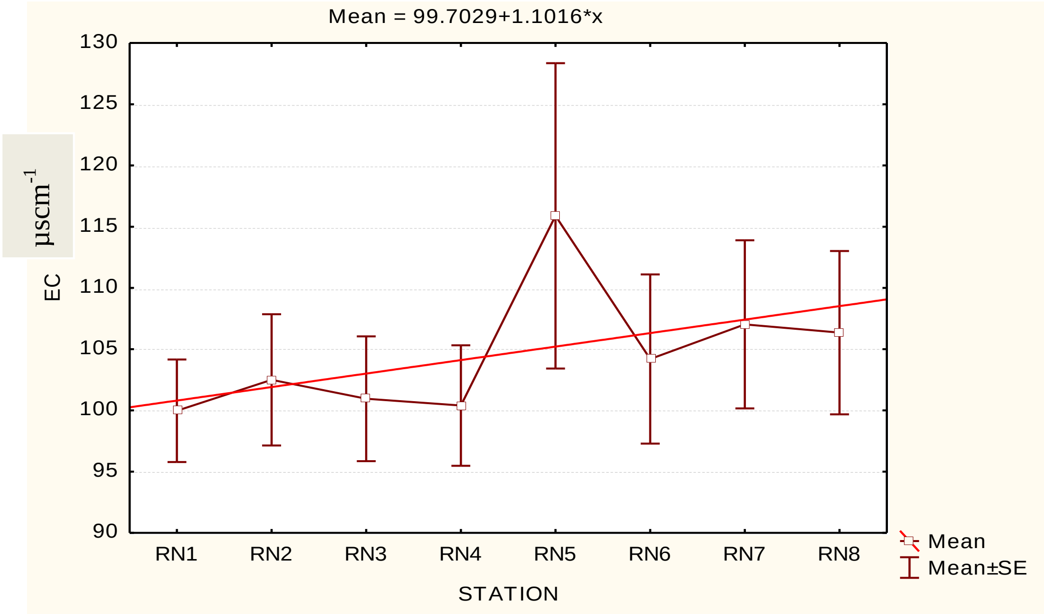


**Figure 4. 5: Mean±SE EC quality variability in the Nzoia River**

There was a subtle decrease of EC mean±SE values from $RN_2$ (102.50±5.36) to downstream sugar effluent discharge point $RN_4$ (100.40±4.92) while increasing to the domestic effluent discharge $RN_5$ (115.89±12.47). Similarly, an increase was observed at $RN_5$ having the largest difference between the maximum and minimum EC values thereafter decreasing towards $RN_6$. However, the changes in the mean±SE values after $RN_6$ and for $R_{1-8}$ were statistically not significant (NS) as indicated in Table 4.2.

### 4.3.6: The Temperature

The temperature ($^0$C) $RN_{1-8}$ mean±SE values were all were below WHO (40) discharge standards as seen in Table 4.2.The mean±SE values at $RN_2$ (upstream discharge point) (24.50± 0.33) (t=-47.353) (p<.05), $RN_4$ (downstream discharge point) and $RN_5$ (24.50±0.33) (t=-47.353) (p<.05) respectively were highly significant in comparison to WHO. The lowest mean value was at $RN_1$ (24.38±0.32) while the rest of the Nzoia River had 24.50± 0.33. The changes in the temperature mean±SE values for $RN_{1-8}$ were therefore not significant (NS) (Table 4.2). The upstream ($RN_2$) and downstream ($RN_4$) physico-chemical quality due to Mumias sugar factory effluents discharge were still beyond the NEMA and WHO hypothetical values (discharge standards) respectively. This is despite the sugar effluents undergoing treatment process before discharge into the Nzoia River.

## 4.4: Comparison of the Effects of the Physico-Chemical Water Quality Parameters in the Nzoia River on the Phytoplankton Species Diversity Upstream and Downstream Mumias Sugar Factory Effluent Discharge Points

### 4.4.1: Principal Component Analysis

The Table 4.3 and figure 4.6 presents principal component analysis (PCA) indicating extracted possible factors of variability influencing physico-chemical quality between sampling stations $RN_{1-8}$.

Each significant loading (factor) of more than 0.60 likely represents the type of pollution at the respective principal component. The largest positive/negative loading implies the meaning of the dimensions; with contribution of the variables increased with the increasing loading in dimension for positive loading; and the converse for negative loading. The factor loadings >0.75, 0.75-0.50, and 0.50-0.0 were explained as strong, moderate, and weak loading respectively and only strong factors were considered for analysis.

**Table 4.3: Loading of seven (7) physico-chemical water quality parameters on principal component analysis, e, f & v**

| Parameters | PC 1 | PC 2 | PC 3 | PC 4 | PC 5 | PC 6 | PC 7 | Summary Statistics | |
|---|---|---|---|---|---|---|---|---|---|
| COD | **-0.797**$^{e}$ | -0.553$^{e}$ | -0.216$^{e}$ | -0.106$^{e}$ | -0.003$^{e}$ | 0.014$^{e}$ | 0.011$^{e}$ | 37.34 | 20.76 |
| $BOD_5$ | -0.178$^{e}$ | -0.127$^{e}$ | **0.975**$^{e}$ | 0.018$^{e}$ | 0.008$^{e}$ | -0.028$^{e}$ | -0.010$^{e}$ | 19.90 | 7.51 |
| DO | 0.025$^{e}$ | -0.006$^{e}$ | 0.036$^{e}$ | -0.080$^{e}$ | -0.549$^{e}$ | **0.787**$^{e}$ | 0.269$^{e}$ | 5.70 | 0.90 |
| TDS | -0.216$^{e}$ | 0.143$^{e}$ | -0.037$^{e}$ | **0.956**$^{e}$ | -0.130$^{e}$ | 0.022$^{e}$ | -0.017$^{e}$ | 53.79 | 6.73 |
| pH | -0.002$^{e}$ | 0.005$^{e}$ | 0.001$^{e}$ | 0.037$^{e}$ | 0.113$^{e}$ | -0.247$^{e}$ | **0.962**$^{e}$ | 7.34 | 0.44 |
| EC | -0.534$^{e}$ | **0.811**$^{e}$ | 0.013$^{e}$ | -0.241$^{e}$ | 0.002$^{e}$ | -0.001$^{e}$ | 0.004$^{e}$ | 105.27 | 19.97 |
| Temp | -0.017$^{e}$ | 0.015$^{e}$ | 0.007$^{e}$ | 0.094$^{e}$ | **0.818**$^{e}$ | 0.565$^{e}$ | 0.045$^{e}$ | 24.48 | 0.87 |
| **Eigen values** | 484.165 | 395.656 | 36.349 | 15.798 | 0.421 | 0.342 | 0.140 | **Mean** | **Std. Dev. (SD)** |
| **Cumulative Eigen value** | 484.165 | 879.821 | 916.170 | 931.969 | 932.390 | 932.732 | 932.872 | | |
| **%Total variance** | 51.90 | 42.41 | 3.90 | 1.69 | 0.05 | 0.04 | 0.02 | | |
| **% Cumulative variance** | 51.90 | 94.31 | 98.21 | 99.90 | 99.95 | 99.99 | 100.00 | | |
| COD | **-0.845**$^{f}$ | -0.530$^{f}$ | -0.063$^{f}$ | -0.020$^{f}$ | -0.000$^{f}$ | 0.000$^{f}$ | 0.000$^{f}$ | | |
| $BOD_5$ | -0.523$^{f}$ | -0.337$^{f}$ | **0.783**$^{f}$ | 0.009$^{f}$ | 0.000$^{f}$ | -0.002$^{f}$ | -0.001$^{f}$ | | |
| DO | 0.610$^{f}$ | -0.138$^{f}$ | 0.237$^{f}$ | -0.352$^{f}$ | -0.395$^{f}$ | 0.511$^{f}$ | 0.112$^{f}$ | | |
| TDS | -0.707$^{f}$ | 0.424$^{f}$ | -0.033$^{f}$ | **0.565**$^{f}$ | -0.013$^{f}$ | 0.002$^{f}$ | -0.001$^{f}$ | | |
| pH | -0.024$^{f}$ | 0.247$^{f}$ | 0.019$^{f}$ | 0.340$^{f}$ | 0.168$^{f}$ | -0.331$^{f}$ | 0.828$^{f}$ | | |
| EC | -0.588$^{f}$ | **0.807**$^{f}$ | 0.004$^{f}$ | -0.048$^{f}$ | 0.000$^{f}$ | -0.000$^{f}$ | 0.000$^{f}$ | | |
| TEMP | -0.436$^{f}$ | 0.337$^{f}$ | 0.047$^{f}$ | 0.426$^{f}$ | 0.608$^{f}$ | 0.378$^{f}$ | 0.020$^{f}$ | | |
| COD | **0.636**$^{v}$ | 0.306$^{v}$ | 0.046$^{v}$ | 0.011$^{v}$ | 0.000$^{v}$ | 0.000$^{v}$ | 0.000$^{v}$ | | |
| $BOD_5$ | 0.032$^{v}$ | 0.016$^{v}$ | **0.950**$^{v}$ | 0.000$^{v}$ | 0.000$^{v}$ | 0.001$^{v}$ | 0.000$^{v}$ | | |
| DO | 0.000$^{v}$ | 0.000$^{v}$ | 0.001$^{v}$ | 0.006$^{v}$ | 0.301$^{v}$ | **0.619**$^{v}$ | 0.072$^{v}$ | | |
| TDS | 0.047$^{v}$ | 0.021$^{v}$ | 0.001$^{v}$ | **0.914**$^{v}$ | 0.0170$^{v}$ | 0.001$^{v}$ | 0.000$^{v}$ | | |
| pH | 0.000$^{v}$ | 0.000$^{v}$ | 0.000$^{v}$ | 0.001$^{v}$ | 0.013$^{v}$ | 0.061$^{v}$ | **0.925**$^{v}$ | | |
| EC | 0.285$^{v}$ | **0.657**$^{v}$ | 0.000$^{v}$ | 0.058$^{v}$ | 0.000$^{v}$ | 0.000$^{v}$ | 0.000$^{v}$ | | |
| TEMP | 0.000$^{v}$ | 0.000$^{v}$ | 0.000$^{v}$ | 0.009$^{v}$ | **0.670**$^{v}$ | 0.319$^{v}$ | 0.002$^{v}$ | | |

**Note:** e, f **and v** respectively denote Eigen vectors of covariance, factor loadings and Variable Factor (VF) contributions of the variables. The bold values denote significant loading Eigen vectors (>0.60) (Stevens, 1996), factor loadings and VF contributions (Shrestha and Kazama 2007) included in the PCs.

The 0.69 Kaiser-Meyer-Olkin (KMO) value was more than 0.50 while Bartlett's test of sphericity of 192.276 showed significance at 95% ($p< 0.05$) which indicated variables as uncorrelated, making the data suitable for a good PCA/FA. A multiple criteria, that used eigenvalue>1, Eigen vectors, factor loadings, variable factor contributions and scree plot, selected four (4) most significant principal components into decreasing order of PC1, PC2, PC3 and PC4.

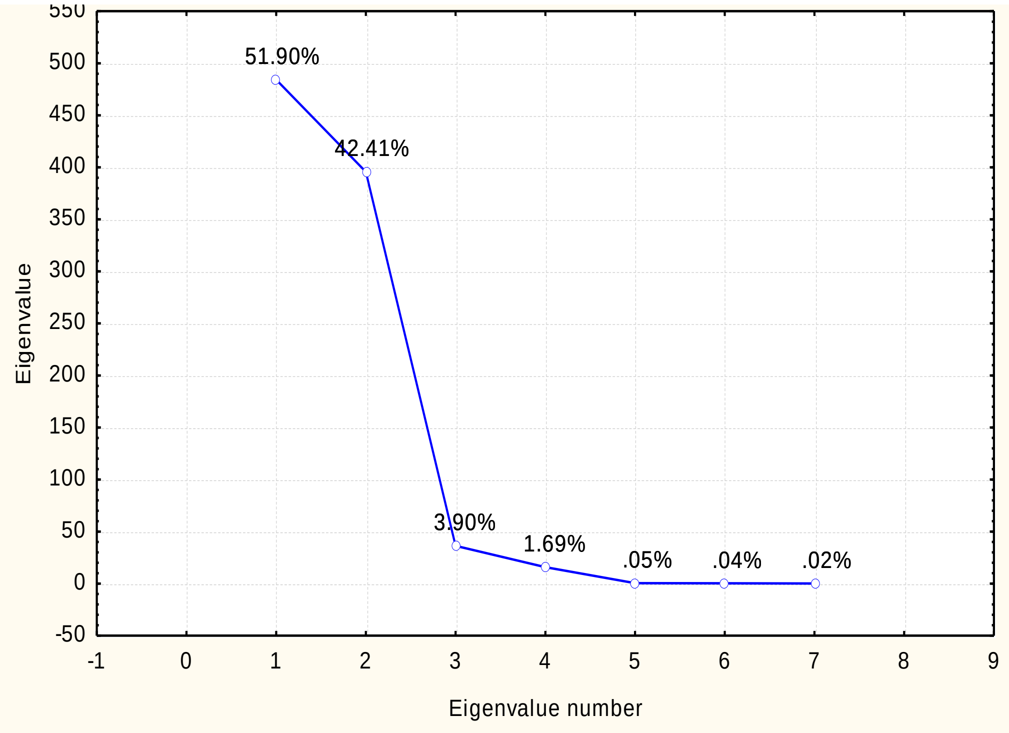


**Figure 4. 6: Scree plot for eigenvalues against the component number**

They all yielded upto 99.90% total variance of the physico-chemical parameters (Table 4.3 and figure 4.6). This explained data set of physico-chemical water quality parameters for $RN_{1-8}$.

The PC1 accounted for the highest total variance and Eigen value of 51.90% and 484.165 respectively. These correlated with COD that had the highest, Eigen vector (-0.797), variable factor (VF) contributions (0.636) and strongest negative factor loading (-0.845) within PC1. The PC2 accounted for the second highest total variance and Eigen value of 42.41% and 395.656 respectively. They correlated with EC that had the highest Eigen vector (0.811), VF contributions (0.657) and strongest positive factor loading (0.807) within PC2.

The PC3 had the third highest total variance and Eigen value of 3.90% and 36.349 respectively. They correlated with $BOD_5$ that had the highest Eigen vector (0.975), VF contributions (0.950) and strongest positive factor loading (0.783) within PC3. The PC4 accounted for the fourth highest total variance and Eigen value of 1.69% and 15.798 respectively. They correlated with TDS that had the highest Eigen vector (0.956), VF contributions (0.914) and strongest positive factor loading (0.565) within PC4.

The rest of the physico-chemical parameters within PC1, PC2, PC3 and PC4 had insignificantly less than 0.05 VF contributions respectively. The sampling station $RN_5$ had the largest SD(20.76) and a standard error (SE) of 9.96 for COD in PC1 followed by SD(19.97) and SE of 12.47 for EC in PC2 (Tables 4.2 and 4.3; and figures 4.3, 4.5 and 4.6).

All the significant positive and negative correlations were greater than 0.3. The COD had a stronger significant positive correlation with $BOD_5$ than TDS and a significant negative correlation with DO. The DO had the strongest negative correlations with TDS followed by temperature, electrical conductivity (EC) and pH. The TDS had the strongest correlations EC followed by temperature and lastly pH. The EC was only correlated with temperature.

**Table 4. 4: Physico-chemical parameter correlations arising from PCA results between sampling stations $RN_{1-8}$**

| Parameters | COD | $BOD_5$ | DO | TDS | pH | EC | TEMP |
|---|---|---|---|---|---|---|---|
| **COD** | 1.00 | **0.57**[a] | **-0.45**[a] | **0.36**[a] | -0.12 | 0.07 | 0.18 |
| **$BOD_5$** | | 1.00 | -0.09 | 0.21 | -0.05 | 0.04 | 0.16 |
| **DO** | | | 1.00 | **-0.69**[a] | **-0.31**[a] | **-0.45**[a] | **-0.50**[a] |
| **TDS** | | | | 1.00 | **0.31**[a] | **0.73**[a] | **0.68**[a] |
| **pH** | | | | | 1.00 | 0.20 | 0.23 |
| **EC** | | | | | | 1.00 | **0.51**[a] |
| **TEMP** | | | | | | | 1.00 |

[a]Significant correlations above 0.30 are bold-faced.

The most significant principal components (PCs) were selected to evaluate their contribution to the variation in phytoplankton species diversity by multiple stepwise regression. The variations of the Nzoia River water quality indicated by the PCA findings could have been caused by changes in the quality of any of the inputs to the water body in the present study.

### 4.4.2: Determination of Phytoplankton Species Diversity in the Nzoia River Upstream and Downstream Mumias Factory Effluent Discharge Points

The overall phytoplankton composition, distribution and abundance at the study stations indicated in Table 4.5 and figure 4.1 respectively.

The Table 4.5 indicates that twelve (12) species comprising of 39,590 individuals were collected from the sampled stations $RN_{1-8}$ and $S_6$. From the entire sampled stations, the five major orders and number of their respective species recorded in decreasing order were Bacillariophyceae (*Synedra*, *Nitzchia*, *Melosira*, *Navicula*, *Asterionella* and *Tabellaria*) (5), Euglenophyceae (*Euglena*, *Phacus* and *Gomphenema*) (3) while Cyanophyceae (*Microcystis*), Chlorophyceae (*Botryococcus*) and Chrysophyceae (*Closterium*) had 1 species each.

The pollution indicating phylankton (the bioindicators) were *Microcystis spp, Melosira spp, Closterium spp, Gomphenema spp* and *Synedra spp* while the most pollution tolerant species were *Euglena spp, Navicula spp* and *Nitzchia spp*. The number of species in each of the sampled stations in decreasing order were 6 ($RN_7$); 5 ($RN_2$); 4($RN_1$, $RN_3$, $RN_5$, $RN_6$ and $RN_8$); and 3($RN_4$). The overall abundance of bioindicators of the sampled stations in decreasing order was *Melosira spp* (464), *Synedra spp* (286); *Closterium spp* (73), *Gomphenema spp* (15) and *Microcystis spp* (14). All the *Microcystis spp* was found at $RN_3$. The *Melosira spp* was completely absent at $RN_4$ and $RN_5$ and; highest at $RN_7$ (295).

From Table 4.5 and figure 4.7, the control sampling station $RN_1$ had the highest numerical abundance of 8,786 individuals at 22.18% with $RN_3$ having the lowest of 80 individuals at 0.21% while $RN_7$ had the highest number of species (6) at 50% of the number of species collected. Only the *Closterium spp* was found at $S_6$ (73). The *Gomphenema spp* were found at $RN_3$ (6) and $RN_4$ (9) while *Synedra spp* were found at $RN_2$ (66), $RN_6$ (7), $RN_7$ (206) and $RN_8$ (7). From $S_6$ and $RN_{1\text{-}8}$, the relative numbers of the pollution tolerant species in decreasing order were *Navicula spp* (199); *Euglena spp* (110) and *Nitzchia spp* (103). Sampling station $RN_4$ had the lowest number of species (3) which is about 25 % of the total number of species.

**Table 4.5: Phytoplankton Community (Order) and Species Composition, Distribution and Abundance at the Indicated Sampling Stations**

| Phytoplankton Order | Species Name | Spatial Mean±SE values (counts ml$^{-1}$) | | | | | | | | | Grand Population Total and % |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | $RN_1$ | $RN_2$ | $RN_3$ | $RN_4$ | $RN_5$ | $RN_6$ | $RN_7$ | $RN_8$ | $S_6$ | |
| Cyanophyceae **Total of the Order** | *Microcystis* | 0 | 0 | 14±0.28 | 0 | 0 | 0 | 0 | 0 | 0 | **14** |
| **% of Total at Each Sampling Station** | | 0 | 0 | 17.97 | 0 | 0 | 0 | 0 | 0 | 0 | **0.04** |
| Bacillariophyceae | *Synedra* | 0 | 66±0.21 | 0 | 0 | 0 | 7±0.20 | 206±0.23 | 7±0.68 | 0 | 286 |
| | *Nitzchia* | 0 | 0 | 0 | 0 | 0 | 0 | 88±0.24 | 15±0.71 | 0 | 103 |
| | *Melosira* | 29±0.43 | 110±0.17 | 22±0.24 | 0 | 0 | 7± 0.26 | 295±1.89 | 0 | 37±0.86 | 464 |
| | *Navicula* | 7±0.12 | 88±4.38 | 37±0.36 | 0 | 29±0.20 | 8±0.36 | 29±0.37 | 0 | 36±1.18 | 199 |
| | *Asterionella* | 0 | 0 | 0 | 0 | 29±0.24 | 0 | 0 | 0 | 0 | 29 |
| | *Tabellaria* | 0 | 66±0.31 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 66 |
| **Total of the Order** | | 36 | 331 | 59 | 0 | 59 | 22 | 618 | 22 | 73 | **1220** |
| **% of Total at Each Sampling Station** | | 0.41 | 21.73 | 74.03 | 0 | 0.72 | 1.12 | 7.17 | 0.79 | 3.36 | **3.08** |
| Euglenophyceae | *Euglena* | 14.67±0.13 | 0 | 0 | 0 | 88.29±0.25 | 0 | 0 | 7.43±0.17 | 0 | 110 |
| | *Phacus* | 0 | 0 | 0 | 9.08± 0.42 | 0 | 0 | 58.9± 0.26 | 0 | 36.91± 0.514760 | 68 |
| | *Gomphenema* | 0 | 0 | 6.41± 1.05 | 8.55± 1.42 | 0 | 0 | 0 | 0 | 0 | 15 |
| **Total of the Order** | | 15 | 0 | 6 | 18 | 88 | 0 | 59 | 7 | 37 | **230** |
| **% of Total at Each Sampling Station** | | 0.17 | 0 | 7.99 | 0.32 | 1.09 | 0 | 0.68 | 0.26 | 1.7 | **0.58** |
| Chlorophyceae **Total of the Order** | *Botryococcus* | 8735± 0.62 | 1191± 0.87 | 0 | 5494± 2.09 | 7941 ± 1.60 | 1986±1.82 | 7939±2.40 | 92±2.32 | 1984± 0.55 | **38051** |

| | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| **% of Total at Each Sampling Station** | | 99 | 78 | 0 | 99.68 | 98 | 99 | 92 | 99 | 91.55 | **96.11** |
| **Chrysophyceae Total of the Order** | *Closterium* | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 73± 0.98 | **73** |
| **% of Total at Each Sampling Station** | | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 3.39 | **0.19** |
| **Number of species at Each Sampling Station** | | 4 | 5 | 4 | 3 | 4 | 4 | 6 | 4 | 5 | **12** |
| **Grand Total of Individuals** | | **8786** | **1522** | **80** | **5512** | **8088** | **2009** | **8616** | **2810** | **2167** | **39,590** |
| **% of Grand Total of Individuals** | | 22.18 | 3.84 | 0.21 | 13.92 | 20.43 | 5.07 | 21.76 | 7.10 | 5.47 | **100** |

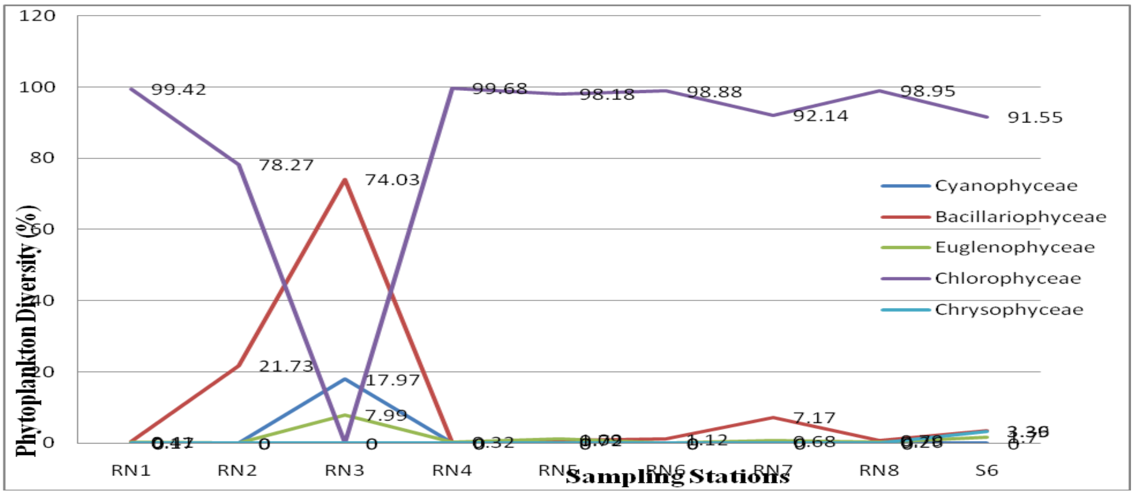


**Figure 4. 7: Spatial Phytoplankton Abundance at the Sampling Stations**

The upstream discharge point $RN_2$ had the highest number (4) of species accounting for 33% of the total number of species. In the entire sampling stations $RN_{1-8}$ and $S_6$, the relative number and percentage of individuals in decreasing order was Chlorophyceae (38,051)96.11%; Bacillariophyceae (1,220)3.08%; Euglenophyceae (230)0.58%; Chrysophyceae (73)0.19%; and Cyanophyceae (14)0.04%. The percentage of individuals for Chlorophyceae was more than 90% in each of the sampled stations with a maximum of 99.68% at $RN_4$ except at $RN_3$ that had 0%.

The Table 4.6 and figure 4.8 indicate that Shannon-Weiner diversity ($H_s$), Margalef's species richness (d) and Pielou species evenness (J) have similar spatial patterns except Simpson's Diversity Index (1–Ds). All the realized Shannon-Weiner diversity ($H_s$) values in Table 4.6 were in the range of 0-1 while those for evenness were less than 0.4, being closer to zero(0) than one(1).

**Table 4.6: Phytoplankton Indices at the Sampling Stations**

| Parameters | Sampling stations | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | **$RN_1$** | **$RN_2$** | **$RN_3$** | **$RN_4$** | **$RN_5$** | **$RN_6$** | **$RN_7$** | **$RN_8$** | **$S_6$** |
| ($H_s$) | 0.04 | 0.68 | 0.90 | 0.02 | 0.11 | 0.05 | 0.22 | 0.03 | 0.40 |
| (d) | 1.21 | 1.50 | 2.51 | 1.28 | 1.22 | 1.45 | 1.21 | 1.39 | 1.43 |
| (J) | 0.02 | 0.28 | 0.36 | 0.01 | 0.04 | 0.02 | 0.09 | 0.01 | 0.16 |
| (1-Ds) | 0.99 | 0.63 | 0.32 | 0.99 | 0.96 | 0.98 | 0.85 | 0.98 | 0.84 |

**Note:** ($H_s$) =Shannon-Weiner diversity; (d) =Margalef's species richness; (J) =Pielou species evenness and (1-Ds) =Simpson's Diversity Index

The Shannon-Weiner diversity was in the range of 0-1 and species evenness followed the same spatial trend at most sampling stations as indicated in Table 4.6 and figure 4.8. High Shannon-Weiner diversity was associated with high species evenness and richness. Simpson's Diversity Index showed a different trend, decreasing with increasing Shannon-Weiner diversity, species evenness and richness.

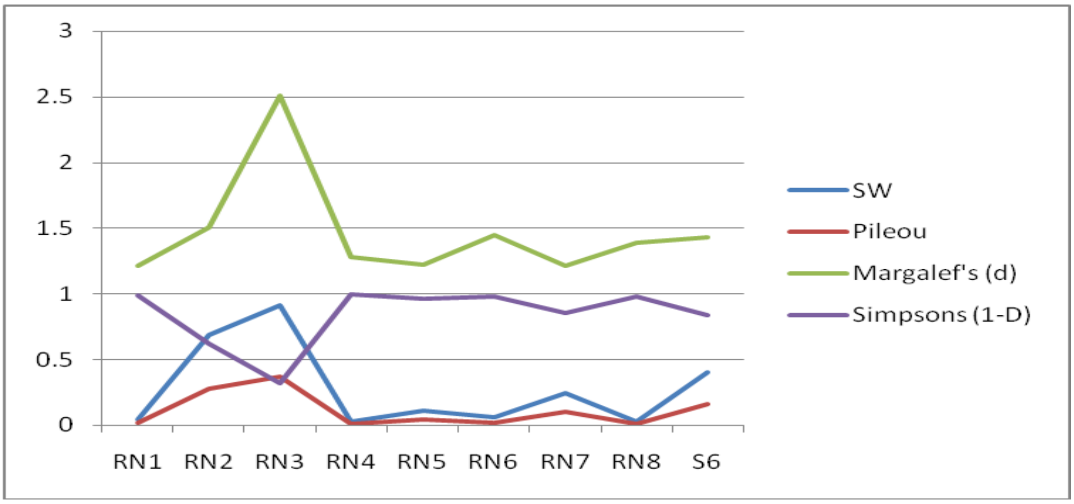


**Figure 4. 8: Spatial variation of Diversity Indices at the Sampling Stations**

The lowest number of species at $RN_4$ resulted in the lowest values of Shannon-Weiner diversity (0.02), species evenness (0.01), species richness of 1.28 despite not being the lowest as expected; and Simpson's diversity index of 0.99 being among the highest as indicated in Table 4.6. The spatial variation of diversity indices at $RN_2$; $RN_3$; $RN_5$ and $RN_7$ were in the decreasing order of species richness; Shannon-Weiner diversity; species evenness and Simpson's Diversity. The mean values for Shannon-Weiner diversity; species richness and evenness increased from $RN_1$ upto $RN_3$ that had the highest values of 0.90, 2.51 and 0.36 respectively while Simpson's Diversity of 0.32 was lowest compared to the other sampling stations. The highest total species (6) was at $RN_7$ while Simpson's Diversity Index of 0.99 was highest at $RN_1$and $RN_4$.

### 4.4.3: Mathematical Relationship between Phytoplankton Species Diversity and The Physico-Chemical Water Quality Upstream and Downstream Mumias Sugar Factory Effluent Discharge Points

The Stepwise Multiple Linear Regression (SMLR) function between the phytoplankton species diversity and the physico-chemical parameters as the observed variable and predictor variables respectively are expressed as:

$Y= \beta_0+\beta_1X_1+\beta_2X_2+\beta_3X_3+\beta_4X_4+\beta_5X_5+\beta_6X_6+\beta_7X_7+ \varepsilon i$……………………**Equation 8**

(Mustapha and Abdu, 2012).

Where: Y represented the dependent variable (phytoplankton species diversity using the Shannon Weiner (SW) diversity- $H_s$) that is being predicted or explained (Kumar and Sharma, 2014);

$\beta_0$ (Beta) as the constant or intercept;

$\beta_1$, $\beta_2$, $\beta_3$, $\beta_4$, $\beta_5$, $\beta_6$ and $\beta_7$ as the slopes (a set of Beta coefficients) for $X_1$ (pH), $X_2$ (TDS), $X_3$ ($BOD_5$), $X_4$ (COD), $X_5$ (DO), $X_6$ (EC) and $X_7$ (Temperature) respectively;

$X_1$, $X_2$, $X_3$, $X_4$, $X_5$, $X_6$ and $X_7$ as the first to the seventh independent variables explaining the variance in Y respectively and; $\varepsilon$i the standard error.

Equation 8 becomes the theoretical model and used to derive the estimated model for the study.

**$H_0$:** $\mu_1 = \mu_2 = \mu_3 = \mu_4 = \mu_5 = \mu_6$ (All of the means are equal) (Bless and Kathuri, 1993; Meir and Zünd, 2000).......................................................................**Equation 9**.

The realized the null hypothesis for $RN_{1-8}$ that states:

**$H_0$**: There are no effects of the physico-chemical quality of the Nzoia River on the phytoplankton species diversity upstream and downstream of Mumias sugar factory effluent discharge points.

Then: **$H_0$** $= \beta_0 = 0$ (constant or intercept is equal to zero)..........................**Equation 10**.

**$H_0$** $= \beta_1 = \beta_2 = \beta_3 = \beta_4 = \beta_5 = \beta_6 = \beta_7 = 0$

(All the regression partial slopes are equal to zero)..................................**Equation 11**.

Equation 10 was used to predict the phytoplankton species diversity (SW) due to the physico-chemical parameters and to generate hypotheses about the causes of variation in SW (equations 10 and 11).

Before interpreting the result, classical assumptions of linear regressions were checked using scatter plot (figure 4.9) and normal p-p plot (figure 4.10).

The figure 4.9 for standardized predicted values against observed values shows that, the relationship between the phytoplankton species diversity ($H_s$) and the predictors ($\beta_0$, COD, TDS, EC, and $BOD_5$) is linear and the residuals variances are equal or constant on both sides of line of the best fit. The increase of predictors results in increase of the phytoplankton species diversity that is linear and distinct.

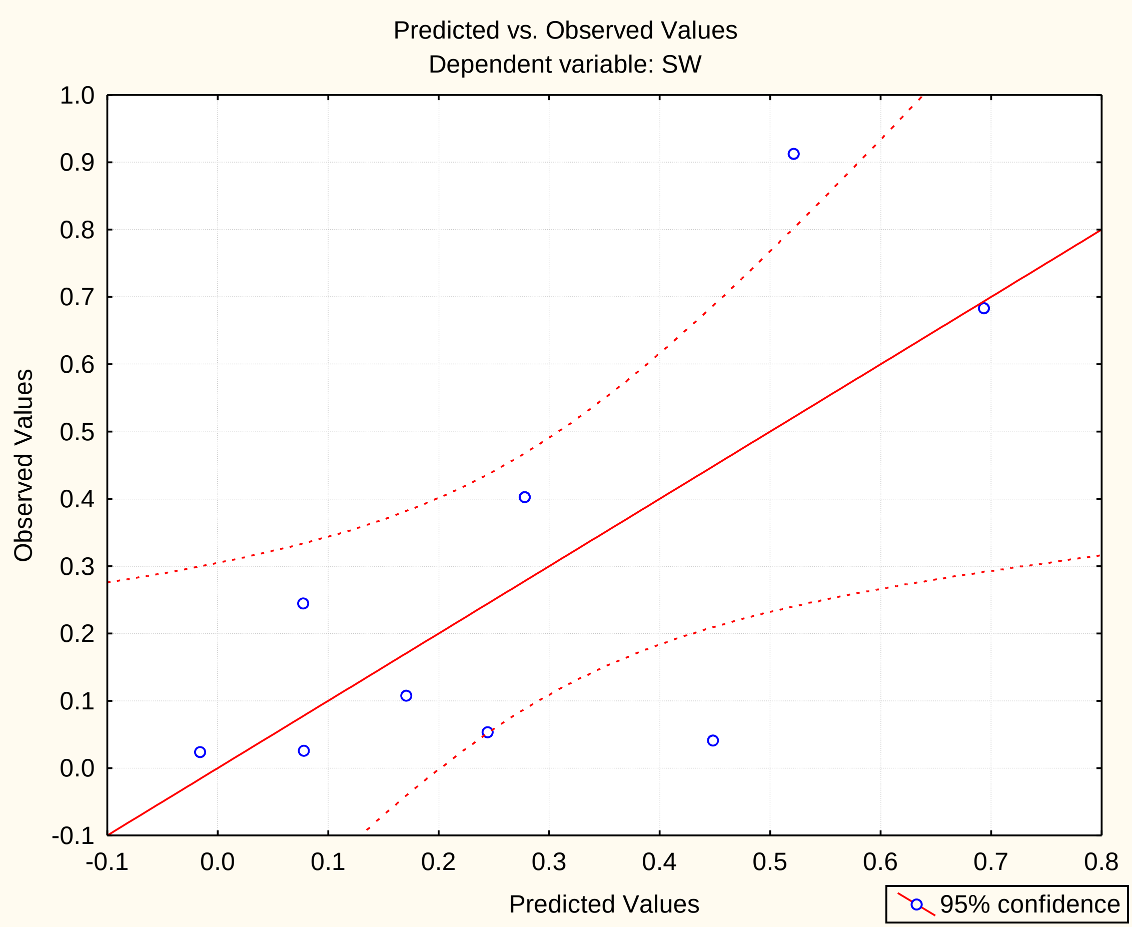


**Figure 4.9: Observed values (circles) and predicted values (connected by line) for the stepwise multiple regression for the relationship between the phytoplankton species diversity (Hs) and the predictors. Dashed lines are 95% confidence intervals for the diversity**

The figure 4.10 reveals that there is direct relationship between the predictors and the phytoplankton species diversity ($H_s$), whereby as the predictors increase, phytoplankton species diversity ($H_s$) increases. All the observed values fall roughly along the straight line indicating that the residuals are from normally distributed population.

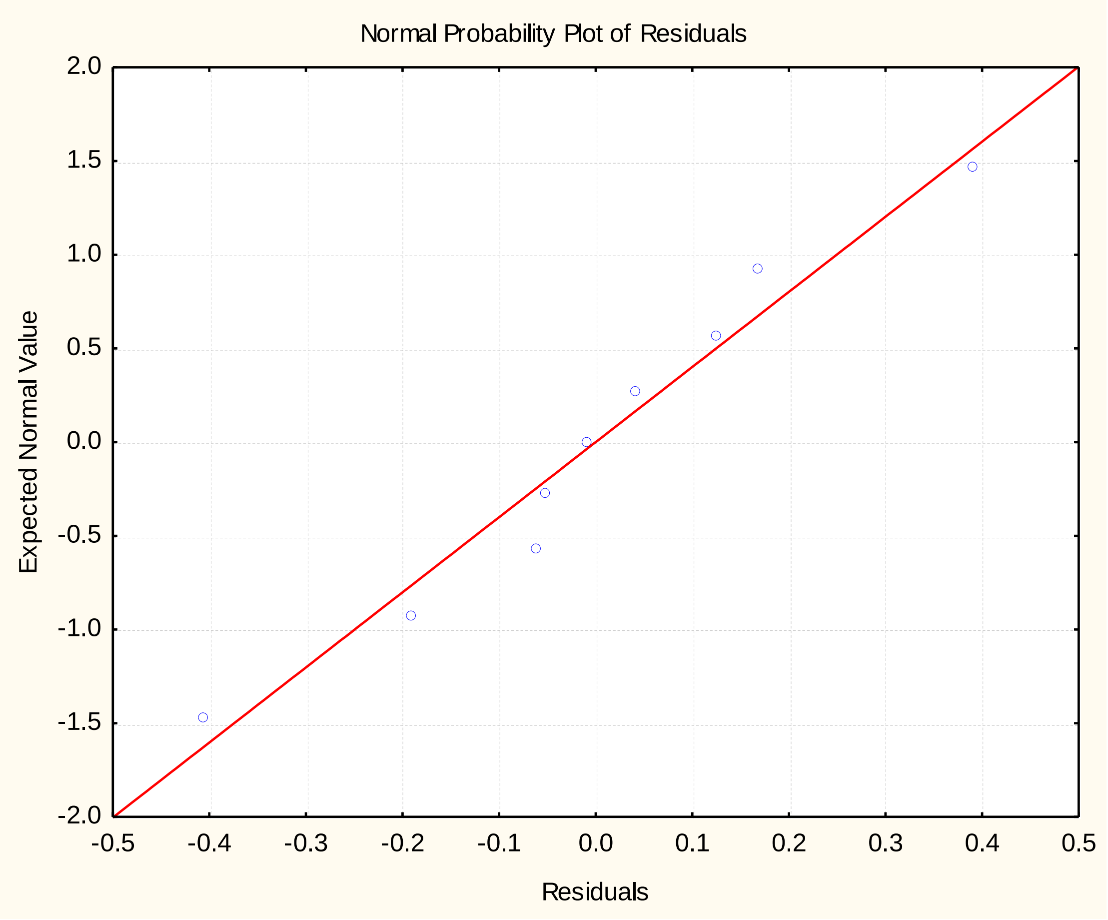


**Figure 4.10: Normal p-p plot of stepwise multiple regression for standardized residuals**

The Table 4.7 indicates results for estimated regression models for the study using stepwise multiple regression analysis derived from Equation 12 whose predictor variables were derived from most significant principal components (PCs). The Table 4.8 shows summary of the stepwise multiple regression analysis.

**Table 4.7: The Stepwise Multiple Regression Analysis**

| Equation No. | **Estimated Regression Models** | $R^2$ | Std error |
|---|---|---|---|
| 12 | Y= 0.94-0.58*COD+ 0.36 | 0.34 | 0.36 |
| 13 | Y=-4.95-0.83*COD + 0.37*TDS+ 6.93 | 0.41 | 6.93 |
| 14 | Y=-16.72-0.90*COD+1.56*TDS-I.2*EC+13.07 | 0.52 | 13.07 |
| 15 | Y=-16.43-0.92*COD+1.53*TDS-1.2*EC+ 0.03*$BOD_5$ +15.67 | 0.52 | 15.67 |

[a]**Predictors:** $\beta_0$ (Constant/Intercept), COD, TDS, EC, and $BOD_5$.

**Table 4. 8: Summary of the Stepwise Multiple Regression Analysis**

| | **SE of β** | **$\beta_0$** | **$\varepsilon_i$** | **t(4)** | **p-level** | |
|---|---|---|---|---|---|---|
| | | -16.43 | 15.67 | -1.05 | 0.35 | |
| **COD** | 0.57 | 0.48 | 0.44 | 1.08 | 0.34 | |
| **TDS** | 1.41 | 0.002 | 0.04 | 0.05 | 0.96 | |
| **EC** | 1.27 | -0.03 | 0.02 | -1.61 | 0.18 | |
| **$BOD_5$** | 0.63 | -0.08 | 0.08 | -0.93 | 0.41 | |
| | **R** | **$R^2$** | **$R^2$ – change** | **F-to-entr/rem** | **p-level** | **Equation No.** |
| **COD** | 0.58 | 0.34 | 0.34 | 3.62 | 0.1 | 15 |
| **TDS** | 0.64 | 0.41 | 0.07 | 0.72 | 0.43 | 16 |
| **EC** | 0.72 | 0.52 | 0.11 | 1.12 | 0.34 | 17 |
| **$BOD_5$** | 0.72 | 0.52 | 0.0003 | 0.003 | 0.96 | 18 |

Fitting the stepwise multiple regression analysis from Table 4.7 into theoretical model (Equation 8) realized the following estimated model:

Y= -16.43 -0.92*COD+1.53*TDS -1.2*EC + 0.03*$BOD_5$ +15.67…...**Equation 15**.

The Nzoia River quality variation at the study site represented by Shannon Weiner (SW) diversity-$H_s$ was explained by the four most significant principal component (PC) predictor variables namely: COD, TDS, EC and $BOD_5$.

The table 4.7 and the estimated model (Equation 15) revealed that: among the parameters calibrated by stepwise regressions analysis, the Beta coefficient for TDS (1.53) provided the strongest positive unique contribution to the phytoplankton diversity. The second highest negative contribution was EC (-1.2), followed by COD (-0.92) while $BOD_5$ (0.03) was the least positive contributor to the phytoplankton diversity. A positive or negative contribution either respectively increased or decreased the phytoplankton species diversity upstream and downstream of Mumias sugar factory effluent discharge points as indicated in equation 15.

The $\beta_0$ (-16.43) and Beta coefficients for β4 (COD), $\beta_2$ (TDS), $\beta_6$ (EC) and $\beta_3$ ($BOD_5$) in the model estimate (Equation 15) had a corresponding t-value of -1.05 at p>0.05(Tables 4.7 and 4.8). This meant that for the estimate model:

$\mathbf{H_0} = \beta_0 \neq 0$ (constant or intercept is not equal to zero) while,

$\mathbf{H_0} = \beta_4 \neq \beta_2 \neq \beta_6 \neq \beta_3 \neq 0$ (All the regression partial slopes are not equal to zero).

The $R^2$ values increased from 0.34 in equation 12 to 0.52 in equation 15 during building of model estimate (Tables 4.5 and 4.6).

The $R^2$ value in the ANOVA model fit revealed that that estimated model accounted for 52% of the variation of water quality explained by COD, TDS, EC and $BOD_5$(Equation 15) on the phytoplankton species diversity of the Nzoia River . The 48% may be due to error, some other factors identified in the principle component analysis alongside other factors at the study site.

The $F_0$ 1.08 > $F_\alpha$ 0.003 and p>0.05 indicates that, the slope of the estimated linear regression model is not equal to zero, confirming that, there is linear relationship between the variation of water quality explained on the phytoplankton species diversity in the estimated model as seen in figures 4.9 and 4.10.

Since $\mathbf{H_0} = \beta_4 \neq \beta_2 \neq \beta_6 \neq \beta_3 \neq 0$ while $\mathbf{H_0} = \beta_0 \neq 0$, at least one physico-chemical parameters is linearly related to SW.

The $R^2$ in of equation 15 are comparable to the variable factor (VF) contributions of the variables realized in PCA in Table 4.3. Both show similar variable contribution of more than 60% to the model estimate ($R^2$) and the respective four (4) principal components. The values for both COD-$R^2$ (0.63): VFs (0.636) and TDS-$R^2$ (0.94): VFs (0.914) were similar while EC-$R^2$ (0.93) > VFs (0.657) and $BOD_5$-VFs (0.950) > $R^2$ (0.70) were greater than VFs and $R^2$ respectively.

The $R^2$ of 52%, comparison of $R^2$ with VFs, $\varepsilon i$ of 15.67 and; the linearity of relationship in the estimated model as seen in figures 4.10 and 4.11 provided a measure of the model's "goodness of-fit".

# CHAPTER FIVE
# DISCUSSIONS AND LIMITATIONS

## 5.1: Discussions

In the present study, there was effort to assess of the effects of effluent from Mumias sugar factory on water quality and phytoplankton species diversity of river Nzoia. Randomized experimental design quantified the response of phytoplankton species diversity as a function of Mumias sugar mill effluents discharges influencing the physico-chemical quality of the Nzoia River. This entailed convenient upstream-downstream independently repeated sampling design involving experimental field and laboratory analytical tasks. The study reveals the extent of the linkage between physico-chemical quality of Nzoia River and phytoplankton species diversity due to effects of Mumias sugar factory effluents discharge.

### 5.1.1: Determination of the Physico-Chemical Water Parameters Loads of Mumias Sugar Factory Effluents Before and After Treatment

Determination of the physico-chemical water parameters loads of Mumias sugar factory effluents before treatment involved $S_1$, $S_2$, $S_3$ and $S_5$ sampling stations. $S_4$ and $S_6$ were sampling stations after treatment.

The statistical design was such that the null hypotheses for $S_{1\text{-}6}$ states that:

**$H_0$:** All the mean values of the physico-chemical parameter loads of Mumias factory effluents before and after treatment are equal.

Then:

It follows that; Mumias Sugar effluents discharge does not affect physico-chemical quality of water of Nzoia River

Hence for: One-way Analysis of Variance (ANOVA) for $S_2$, $S_1$ and $S_3$,

**$H_0$:** $\mu_1 = \mu_2 = \mu_3$..........................................................................................**Equation 16**;

In the present study, results indicated significantly more elevated levels of water physico-chemical parameter loads at $S_2$ (the process house) than $S_1$ (the milling house) and $S_3$ (boiler). This may have been due to combined floor washings from added washing chemicals and more sugar (Kaur *et al*., 2010).

Two-way dependent t-test of $S_2$ and $S_5$; $S_1$ and $S_4$ and; $S_5$ and $S_6$,

**$H_0$:** $\mu_2 = \mu_5$; $\mu_1 = \mu_4$ and $\mu_5 = \mu_6$…………………………………..………**Equation 17**

and;

One-way 2-tailed independent test for $S_4$ and $S_6$,

**$H_0$:** $\mu_4 = \mu_N$; $\mu_4 = \mu_W$; $\mu_6 = \mu_N$; $\mu_6 = \mu_W$.......................................................**Equation 18**,

Where,

$\mu_N$ and $\mu_W$ are NEMA and WHO hypothetical value (discharge standards) respectively.

As was the case a study by Akali *et al*., (2011), effluents from milling ($S_1$) flowed through meanders for purpose of attaining minimum discharge standards before ending up into the Nzoia River. The $BOD_5$, COD and DO values at upstream ($S_4$) and downstream ($S_6$) before discharge points were not ideal for supporting aquatic life, full contact recreation and within recommended levels for drinking waters (Boyd and Lichtkoppler, 1979; DWAF, 1996; WHO, 2006 and NEMA, 2013). This is consistent to observations mentioned by Goel (2008) and Salequzzaman *et al*., (2008). The spatial trends documented by Doke *et al*., (2011) for physico-chemical characteristics of sugar effluents are similar to those in Tables 4.1., 4.4 and figures 4.1 to 4.7.

The Chemical oxygen demand (COD) is the amount of oxygen required for the oxidation of inorganic matter using a strong chemical oxidant (Subin and Husna, 2013). The concentration of COD in surface water range from $20mgl^{-1}$ in unpolluted water to $200mgl^{-1}$ in waters receiving effluents (Chapman, 1996).The sampling stations $S_{1-6}$ exhibited COD mean±SE greater than WHO and NEMA. This study finding indicates high oxygen demanding materials in sugar effluents produced at $S_4$ and $S_6$ that can deplete oxygen in the Nzoia River (Subin and Husna, 2013).

Low value of DO and high COD is associated with high organic matter content and sewage disposal in rivers (Rai, 1974 and; Mishra and Ram, 2007). The determined physico-chemical values at $S_6$ are in line with the observed trends of inadequately treated industrial effluents in the Nzoia River (Achoka, 1998).

The pH is an important parameter that affects aquatic life of any water body and used in evaluating the acid-base balance of water (Rai, 2011 and Mullai *et al*., 2013). The pH may indirectly have effects on human health by causing variations of other water quality parameters such as solubility of metals and survival of pathogens (Zabed *et al*., 2014). The sugar effluent pH for $S_2$ indicates a high inorganic and organic solutes present in the sugar effluents (Rai, 2011 and Doke, *et al*., 2011). Studies by Akali *et al*., (2011) indicated that pH values are raised through lime stabilization to untreated wastewater from process towards pond 1. This may have attributed the rise in pH values from $S_2$ to $S_5$.

Total Dissolved Solids (TDS) is a convenient measure of the total ionic concentration in water (Udoh *et al*., 2013). There was significant increase of TDS from $S_1$ to $S_4$ and decrease from $S_2$ to $S_5$ probably due to surface runoffs from the surrounding and prevention of entry from runoffs by concrete channels respectively (Akali *et al*., 2011). The decrease of TDS in the sugar effluent treatment ponds from $S_5$ to $S_6$ could be attributable to the dissolved substances precipitating on merging with the factory effluent (Akali *et al*., 2011). The significantly lower TDS mean±SE values than both NEMA and WHO limits may check growth and lead to the death of many aquatic life forms (Subin and Husna, 2013).

The highest $BOD_5$ loads at $S_2$ (Table 4.1) were due to organic waste discharges from the sugar process house (Rai *et al*., 2011). All the mean±SE values for $S_{1-6}$ were more than WHO and NEMA discharge limits may be due to floor washing, wastewater and condensate water, leakage in valves and glands of the pipeline, added sugarcane juice, syrup and molasses in the sugar effluent (CPCB, 2000). The observed significant increase in $BOD_5$ from $S_1$ to $S_4$ was may be due to severe pollution from the spray ponds and the domestic treatment pond adjacent to $S_4$ (Kumar *et al*., 2009).

Dissolved oxygen in water is an important factor determining the occurrence and abundance of aquatic organisms, as it has the ability to support aquatic life (Conte and Cubbage, 2001 and; Subin and Husna, 2013). The amount of DO in water depends on the source, temperature, chemical and biological process taking place in a water body (WHO, 2006). Studies by Pratiksha *et al*., (2013) attributed low DO values to high levels of sugars in the process water capable of excessive oxygen demand loading. This may have been the case low values at $S_2$. The increase of DO from $S_1$ to $S_4$, $S_2$ to $S_5$ and; $S_5$ to $S_6$ may have been due to abundant oxygen contributed by oxygenation of the water column from turbulent flow patterns in the channels (Kegley and Andrew, 1998; and Mullai *et al*., 2013).

The electrical conductivity represents the total concentration of soluble salts and a measure of water capability to transmit electric current in water bodies (Trivedy and Goyal, 1986). In the current study, all the mean±SE values for $S_{1-6}$ were less than WHO except at $S_2$ and the highest at $S_6$. This may result in sour water unsuitable for drinking upon discharge (Trivedy and Goyal, 1986).

The temperature decrease from $S_2$ through $S_5$; to $S_6$ may have been due to sugar effluents movement from process towards the effluents treatment ponds and movement through the six ponds during treatment respectively.

Investigations by Akali *et al*., (2011) similarly indicate wide variations of physico-chemical water quality parameters from Mumias sugar factory's lagoons at pond 6 of the sampling station $S_6$. This is due to stagnation in technology for effective adoption of modern Environmental Management System (EMS) in waste management at Mumias sugar factory (Omuterema, 2005).

### 5.1.2: Assessment of The Physico-Chemical Water Quality Parameters In the Nzoia River Upstream and Downstream Mumias Factory Effluent Discharge Points

Since the null hypothesis for $RN_{1-8}$ states that:

**$H_0$:** All the mean values of the physico-chemical water quality in the Nzoia River due to Mumias factory effluents discharge are equal,

Then:

**$H_0$:** $\mu_1=\mu_2=\mu_3=\mu_4=\mu_5=\mu_6=\mu_7=\mu_8=\mu_N= \mu_W$ (All of the means are equal)...**Equation 19**.

For the one-way 2-tailed independent test of $RN_2$, $RN_4$ and $RN_5$,

**$H_0$:** $\mu_2 = \mu_N$; $\mu_2 = \mu_W$;............................................................................ **Equation 20**,

**$H_0$:** $\mu_4 = \mu_N$; $\mu_4 = \mu_W$ and; …....................................................................... **Equation 21**,

**$H_0$:** $\mu_5 = \mu_N$; $\mu_5 = \mu_W$............................................................................ **Equation 22**,

Where $\mu_{1\text{-}8}$ represents the mean values of the physico-chemical parameter loads for $RN_{1\text{-}8}$ while $\mu_N$ and $\mu_W$ were NEMA and WHO hypothetical values (discharge standards) respectively.

Three discharge effluent points on the Nzoia River were characterized as upstream ($RN_2$) and downstream ($RN_4$ and $RN_5$) at the study area. Establishment of a statistical relationship compared the physico-chemical water quality on the determined phytoplankton species diversity.

The obtained physico-chemical values for $RN_{1\text{-}8}$ were within or below both discharge limits of NEMA and WHO. The exception was DO values that were greater than WHO (5.00). Such values convert complex organic substances to simple dissolved inorganic salts may be utilized by the phytoplankton. This concurs with observations made by Howayda *et al*., (2012).

Prakash *et al*., (2007) attributed high pH to increased photosynthesis of the phytoplankton blooms resulting into the precipitation of carbonates of calcium and magnesium from bicarbonates. This is similar findings in the current study at upstream discharge point ($RN_2$) (Table 4.2 and figure 4.1) that had the highest mean±SE of pH. The near neutral to alkaline pH values obtained in most sampling stations along the Nzoia River is an indication to the rapidly deterioting state of river in the present study (Adesalu *et al*., 2008). The pH values outside 6.5 to 8.5 ranges may strongly suggest pollution (Lagler, 1956). The pH values in the present study are similar to those reported by Igbinosa and Okoh (2009).

While treated domestic and sugar effluents discharge in the present study may have caused minimal variations in TDS mean±SE along $RN_{1-8}$, findings by Subin and Husna (2013) showed wide spatial variations along the river waters. This was due to dissolution of higher total ionic concentrations of organic and other inorganic particles arising from sewage, industrial and solid waste discharge (Subin and Husna, 2013). The $BOD_5$ ($mgml^{-1}$) is the amount of oxygen required by the microorganisms during their growth in wastewater and has a value of 5 $mg^{-l}$ or less in unpolluted, natural waters (Ayoola and Ajani, 2009 and; Rai *et al*., 2011). It is an important water quality variable included in water quality studies despite not being identified in the causative factor matrix (Deas *et al*., 1999).

The $RN_{1-8}$ mean±SE values were all below NEMA and WHO discharge standards except at $RN_5$.The $BOD_5$ statistical significant differences between $RN_{1-8}$ may be due to the observed anthropogenic activities such as washing, dumping of refuse and sewage into the Nzoia River. This is similar to findings by Zakariya *et al*., (2013). From figure 4.3, increase in $BOD_5$ mean±SE values from $RN_3$ to $RN_5$ attests to the effects of the sugar effluent discharges that can severely depress DO (Deas *et al*., 1999).

Thereafter a gradual decrease to $RN_8$ was observed suggesting a significant relationship between $BOD_5$ loading and distance downstream after the effluent discharge point (Akali *et al*., 2011). The present study had $BOD_5$ values were above 1mgl$^{-1}$ and more than 10.0 mgl$^{-1}$ respectively, making Nzoia River to be considered wastewater contaminated and heavily polluted (UNESCO/WHO/UNEP, 1996, Maria, 1983 and Adakole, 2002). Akali *et al*., (2011) also observed this as an indicator that effluent from the factory had significant effects on river's $BOD_5$. Similar organic waste discharges from various sources have been observed on the Ganga river waters (Bhargava, 1982).

The COD mean±SE values at $RN_4$ and $RN_5$ were more than NEMA discharge limits but below the average of 60.30 mgl$^{-1}$ reported for Nzoia River (Davies, 1996). The higher mean±SE levels of COD than NEMA discharge limits were likely to point to highly polluted waters at $RN_4$ and $RN_5$. Subin and Husna, (2013) attributed this to highly polluted effluent discharge point as in the present study. Similarly, the high demand on dissolved oxygen by the wastes discharged, may water render unfit for drinking, irrigation and decreased recreation. This view is similar to those of Tyagi and Mehra, (1990) and., Reddy and Baghel, (2010).

Despite the DO mean±SE values for $RN_{1-8}$ seen in Table 4.4 and figure 4.4 being greater than WHO (5.00) discharge standards, they were within the 5 mg$^{-l}$ to 9 mg$^{-l}$ limits for drinking water (Mimoza, 2007). Both the recommended standards for drinking purpose of 6 mg$^{-l}$ and; sustaining fish and aquatic life of 4-5 mg$^{-l}$ suggested by Rao (2005) are similarly within the limits for drinking water. However, the reference sampling station ($RN_1$) DO value of 6.14±0.31 mg$^{-l}$ was less than the normal range of 8-10 mg$^{-l}$ for unpolluted water (Pearce *et al*., 1999, Rao, 2005). The use of water quality criteria for dissolved oxygen by WHO becomes a challenge as it is meant to ensure the maintenance of biological function of a river system (Igbinosa and Okoh, 2009).

The narrow increase of EC mean±SE values from downstream sugar effluent discharge point $RN_4$ to the domestic effluent discharge $RN_5$ may result in the corresponding higher levels of anions and cations in the water (Subin and Husna, 2013). This possibly implies the domestic effluents are likely to alter the chelating properties of the Nzoia River and create an imbalance of free metal availability for flora and fauna (Akan *et al*., 2008). The narrow variation of EC in the River Ganga reported by Rai *et al*., (2011) is in tandem with the present study. Results by Igbinosa and Okoh (2009) indicate higher conductivities upstream and downstream of the discharged points suggesting for other point sources pollution entering into the receiving water body. This is similar to the findings in the current study whereby upstream ($RN_2$) and downstream ($RN_4$) sugar effluent and; domestic ($RN_5$) discharge points are comparatively higher than the rest along the Nzoia River study area.

Temperature is an important factor for determining the rate of photosynthesis affecting the chemical and biological reactions in aquatic ecosystems (Monika, 2013 and Mullai *et al*., 2013). The temperature variation may affect the physico-chemical characteristics, distribution and abundance of phytoplankton (Manikannan *et al*., 2011; Soundarapandian *et al*., 2009).

The temperature ($^0$C) $RN_{1-8}$ mean±SE values were all were below WHO discharge standards as seen in Table 4.4 and figure 4.6. The changes in the temperature mean±SE values for $R_{1-8}$ are therefore not significant (NS) (Table 4.2). The lack of statistically significant variation in water temperature between sampling stations on the Nzoia River may be due to the sun around the tropics during the sampling duration of 1st December 2009 to 31st March 2010 (Zakariya *et al*., 2013). Chia *et al*., 2011 and Tanimu *et al*., 2011 have reported similar results.

Therefore:

**$H_0$:** $\mu_1 \neq \mu_2 \neq \mu_3 \neq \mu_4 \neq \mu_5 \neq \mu_6 \neq \mu_7 \neq \mu_8 \neq \mu_N \neq \mu_W$ (All of the means are not equal)……………………………………………………………**Equation 23**.

For the one-way 2-tailed independent test of $RN_2$, $RN_4$ and $RN_5$,

**$H_0$:** $\mu_2 \neq \mu_N$; $\mu_2 \neq \mu_W$;........................................................................ **Equation 24**,

**$H_0$:** $\mu_4 \neq = \mu_N$; $\mu_4 \neq \mu_W$ and; …................................................................ **Equation 25**,

**$H_0$:** $\mu_5 \neq \mu_N$; $\mu_5 \neq \mu_W$......................................................................... **Equation 26**,

### 5.1.3: Comparison of the Effects of the Physico-Chemical Water Quality Parameters in the Nzoia River on the Phytoplankton Species Diversity Upstream and Downstream Mumias Sugar Factory Effluent Discharge Points

Principal component analysis (PCA) is a potent pattern recognition model that attempts to reveal the variance of a large data set of inter-correlated variables with a smaller set of independent variables (principal components-PC) (Hopke, 1985, Simeonov *et al*., 2003 and; Mustapha and Abdu, 2012). It is a pure mathematical technique without any assumption whose results demonstrate reliable information with respect to reality in fields of scientific research (Mazlum *et al*., 1999).

The PCA postulates an objective way of attaining indices of this type so that the variation in the data can be accounted for as succinctly as possible (Lei Lei, 2013). PCA provides information on the most meaningful parameters that describe the majority of the data set, affording data reduction with minimum loss of original information (Helena *et al*., 2000).

The PC can be articulated mathematically as indicated in the equation 23:

zij = ai1x1j + ai2x2j + ai3x3j + … + aimxmj........................................... **Equation 27,**

Where z is the component score, a is the component loading, x the measured value of variable, i is the component number, j the sample number and m the total number of variables (Lei Lei, 2013). The variable factor (VF) contributions constructed by PCA include unobservable, hypothetical, latent variables while PC is a linear combination of observable water quality variables (Shrestha and Kazama, 2007).

Findings from Simeonov *et al*., (2003) and Gwayali *et al*., (2012) indicate that COD and $BOD_5$ are likely drivers of variability of the combined industrial (sugar) and domestic effluents discharge point sources. The correlation of PC1 with the organic factor COD and PC3 with another organic factor $BOD_5$ from Table 4.3 and figure 4.6 may have been due to similar drivers of variability at $RN_4$ and $RN_5$ respectively. The strong negative loading PC1 has with COD indicates the loading of partially decayed organic matters from domestic and sugar effluents discharge (Gyawali *et al*., 2012). The correlations between PC2 and EC and; PC4 and TDS are likely to be due to physico-chemical and; soil leaching or agricultural run-off process variability respectively. This agrees with findings by Simeonov *et al*., (2003) and Srivastava *et al*., (2012). This concurs with Yazdian *et al*., (2014) who asserts that TDS and EC correlate highly in the environment. The results by Salim *et al*., 2014 from PCA also suggested that the natural soluble salts, nonpoint source nutrients, and anthropogenic organic pollutants explain most of the variations in water quality.

The stronger significant positive correlation between COD and $BOD_5$ (Table 4.4) is a measure of oxygen demand by both biodegradable and non-biodegradable pollutants (Mustapha and Abdu, 2012). This is confirmed by the significant negative correlation between COD and DO. The inverse relationship between DO and temperature is a natural process since warmer water becomes saturated more easily with oxygen (Gwayali *et al*., 2012). All the significant positive and negative correlations were greater than 0.3, making the water quality data collected reliable and valid (Gwayali *et al*., 2012).

The largest standard deviation for COD in PC1 followed by EC in PC2 at $RN_5$ corresponds to the largest principal component vector displacement and thus the most polluted sampling station (Ngodhe *et al*., 2014). Documentation of PCA patterns similar to the current study concur to those by Simeonov *et al*., 2003; Plata-Díaz and Pimienta, 2011; Gwayali *et al*., 2012; Ngodhe *et al*., 2014 and; Wuttichaikitcharoen and Babel, 2014.

The variations of the Nzoia River water quality indicated by the PCA findings due to $BOD_5$ and COD could have been caused by changes in the quality of any of the inputs to the water body (WHO/UNEP, 1996). Increased distance from the polluting sources, results in longitudinal mixing smoothing out irregularities with fewer sample parameters needed to meet given confidence limits (Lehr *et al*., 2005 b). However, as the distance between the source of variability and sampling point increases, there was also dilution with some variables being reduced by self-purification, deposition and adsorption (Lehr *et al*., 2005 b).

Phytoplanktons are useful for control of the physico-chemical and biological conditions of a water system and important biological indicator of the water quality (Ariyadej *et al*., 2004). The environmental physico-chemical variations may affect particular species and induce the growth and abundance of other species (Thamizh and Sivakumar, 2011). This resulted in the total number of species listed in the present study to vary noticeably.

The presence of *Euglena spp*, *Navicula spp*, *Nitzchia spp*, toxic *Microcystis spp* and *Closterium spp* as the most pollution tolerant species in the present study may imply that the Nzoia River are organically polluted, likely to be indicative of pre-high nutrient (eutrophic) status and are typical of phosphate enriched waters (Palmer, 1969; Gunale and Balakrwashnan, 1981; APHA, 1999; Bellinger *et al*., 2006, Desei *et al*., 2008; Suman *et al*., 2012 and; Sakset and Chankaew, 2013). This can serve as an early-warning signal that reflects the ‘health’ status of the Nzoia River. Patrick (1965) concluded that *Euglena spp* was highly pollution tolerant genus and, therefore, reliable bioindicator of eutrophication in the present study.

The comparatively less polluted upstream control sampling station ($RN_1$) was characterized by the highest species abundance of *Botryococcus spp.* This coincides with findings by Ogbuagu and Adelapo, (2012), probably being due to less sand mining activities than downstream sugar effluent discharge sites. The most pollution tolerant species of *Navicula spp* was found at all the sampling stations except at $RN_4$, a probable indication of heavy polluted sites having highest degree of organic pollution. This situation is comparable to outcomes by (Jafari and Gunale, (2006) and; Thi, (2006). This has been qualified in other countries for saprophilous or tolerant phytoplankton (Thi, 2006). This concurs with Mbao *et al*., 2013 who observed that *Navicula spp* had no significant correlation with nutrient concentration.

In this study *Gomphonema spp* was found only at $RN_3$ and $RN_4$, sites that likely receive high agricultural wastes (Sabater *et al*., 1988). The *Nitzchia spp* and *Gomphonema spp* have been found to be resilient to organic and heavy metal pollution and typically recorded in nutrient rich and poorly oxygenated waters (Bere and Tundisi, 2011). Extreme pollution resistant *Gomphonema spp* and *Navicula spp* have similarly been found to be dominant and diverse in distribution in Nyangores tributary of the Mara River, Kenya due to the intensive farming activities (Mbao *et al*., 2013).

The presence of *Phacus spp*, which is likely to tolerate organically polluted waters, may be a strong indication of the high pollution status at the study area (Adesalu *et al*., 2008). The *Nitzchia spp* displayed no particular site preference, being found downstream sampling stations $RN_7$ and $RN_8$ due being cosmopolitan and insensitive to environmental change (Lung'ayiah, 2002).

The abundance of *Microcystis spp* only at $RN_3$ may be due to tolerance to high level of nutrients and usually being found in high phosphate waters (Sakset and Chankaew, 2013). This may likely be due to intensive application of N: P fertilizers (Osano *et al*., 2003 and Raburu *et al*., 2009). Makhlough (2008) recorded the presence of *Microcystis*, as indicator of toxic, unfavourable, odours and pollution in aquatic ecosystems. In the present study, it appears that taxon (species level) presence/abundance can be a valid and useful tool for inferring local water quality conditions (Bellinger *et al*., 2006).

The results indicate that Shannon-Weiner diversity indices of less than1.0 at $S_6$, $RN_2$ and $RN_4$ for sugar effluents and $RN_5$ domestic effluents discharge points respectively could be a pointer to effluents coming from the sugar mills significantly inhibiting the phytoplankton growth (Khwaja, 2001). Such low values may account for high pollution levels of the effluents due to species occurring in relatively low numbers in an environment that was stressed (Desei *et al*., 2008 and Dlamini, 2009). This may generally be interpreted as characteristic of polluted conditions over time, where a few tolerant genera dominate the community. Ngodhe *et al*., (2014), have also highlighted such a possibility. Likewise, the current study findings concur with those of Yazdian *et al*., (2014). They showed that species diversity and distribution is clearly related to water quality and the more contaminated water is, the less the diversity index will be.

The highest mean values for Shannon-Weiner diversity; species richness and evenness at $RN_3$ while having lowest mean values for Simpson's Diversity is a phenomenon known in all types of natural aquatic conditions (Amany and Mohamed, 2003). However, the Shannon-Weiner diversity values in the present study were in the range of 0-1, making the Nzoia River at the study site to qualify as heavily polluted with low phytoplankton community complexity (Biligrami, 1988; Desai *et al*., 2008; Dlamini 2009 and; Kumar *et al*., 2012).

The lowest number of species at $RN_4$ resulted in the lowest values of Shannon-Weiner diversity and species evenness, a likely pointer to a degraded ecosystem (Suman *et* *al*., 2010). However, species richness indicated no clear relation with the diversity index. This is comparable to what Reed (1978) found, that diversity indices were closely related to evenness, whereas species numbers (richness) were unimportant in determining species diversity for phytoplankton.

Species diversity indices when correlated with physical and chemical parameters provide one of the best ways to detect and evaluate the effects of pollution on aquatic communities (Margalef, 1968). The presence of phytoplankton in polluted and unpolluted waters at the study site is a useful determinant of water quality (Jafari and Gunale, 2006). The water quality variation represented by Shannon Weiner (SW) diversity-$H_s$ was chosen as it is widely used while composed of species richness and relative abundance within communities (Zhao *et al*., 2013). The SW was explained by the four most significant principal component (PC) predictor variables using stepwise multiple linear regression (SMLR). To employ SMLR for statistical modelling, metric nature and a prior decision on selection of dependent and remaining independent variables were considered using PCA (Hair *et al*., 2010).

The generated estimated model (Equation 18) was used to predict the variability of phytoplankton species diversity due to the predictor variables. The estimated model (Equation 18) selected was based on the highest $R^2$ and magnitude of the F-ratio (Chenini and Khemiri, 2009). The standardized predicted value against observed values in figure 4.9 shows a linear relationship whereby an increase of predictors results in increase of the phytoplankton species diversity (figure 4.10), accounting for evaluation of normality assumption (Manoj, 2014).

Despite the non-significant SMLR results of p=0.47, logical spatial patterns within $RN_{1-8}$ sampling stations were still recognized from correlations between species diversity indices with physico-chemical parameters (Margalef, 1968). The use of $R^2$ value of 52% as a model goodness-of-fit criteria implied the major effects on the phytoplankton species diversity was due to physico-chemical loads (Mendes 2011).

The $R^2$ value in Table 4.9 reveals that estimated model may have accounted for 52% of the variation of water quality explained by COD, TDS, EC and $BOD_5$(Equation 18) on the phytoplankton species diversity of the Nzoia River . The 48% may be due to partially decayed organic matters from domestic and sugar effluents discharge (PC1 and PC3), soil leaching or agricultural run-off process variability (PC4), an imbalance of free metal availability for phytoplankton due to physico-chemical process (PC2) and sand harvesting (Simeonov *et al*., 2003, Gwayali *et al*., 2012, Srivastava *et al*., 2012 and; Subin and Husna, 2013).

The Table 4.7 and the estimated model (Equation 18) reveal that: TDS provided the strongest positive unique contribution to the phytoplankton species diversity likely to be due to soil leaching or agricultural run-off process variability (Simeonov *et al*., 2003 and Srivastava *et al*., 2012). The second highest negative contribution for EC may create an imbalance of free metal availability for phytoplankton due to physico-chemical process (Simeonov *et al*., 2003, Srivastava *et al*., 2012 and; Subin and Husna, 2013). The COD loading may be due to partially decayed organic matters from domestic and sugar effluents discharge (Gwayali *et al*., 2012).

Observations elsewhere indicate that effluent discharges from sugar industries are a major component of water pollution, contributing to oxygen demand and nutrient loading of the water bodies and promote toxic algal blooms and leading to a destabilized aquatic ecosystem (Morrison *et al*., 2001). This is similar to the global trend of using biological criteria in environmental assessment and pollution monitoring (Wu *et al*., 2005). Furthermore, phytoplankton species diversity and spatial distribution are an expression of the environmental health or biological integrity of a particular water body (Ekwu *et al*., 2006).

The present study has shown the linkage between physico-chemical water quality and phytoplankton species diversity using mathematical expressions. This was based on the system theory that has been successful in helping understand processes that can be readily decomposed into simple causal chains (Chen and Stroup, 1993). The PCA examined variable importance in a decreasing order and create variables of importance.

The regression model was grounded in the system theory. This made the regression equations answer three types of theoretical research questions: **(a)** can specific combinations of independent variables predict or explain variance in the dependent variable?; **(b)** is a specific variable in a set of independent variables necessary to predict or explain variance in the dependent variable; and **(c)** can specific combinations of independent variables predict or explain variance in the dependent variable, given a strong theoretical rationale for including control variables as predictors? (Thayer, 2002). Thus, phytoplankton species diversity (SW) through regression was linked to the physico-chemical water quality in the Nzoia River aquatic system due to Mumias factory effluents discharge.

The inclusion of the systems theory in the current study enabled linkage between the complex interactions of all the physico-chemical water parameters and the phytoplankton species diversity of the Nzoia River. The interactions realized from the data collected made it possible to add effects of domestic effluents to the study design. The description of the dynamics and spatial variations in the Nzoia River as a system, representation of the relationships from the data analysis, and the ability to bring together the interactions and linkages was suitable through use of the systems theory.

Anthropogenic activities such as the sugar processing provided a system that interacted with the hydrological system the Nzoia River through water abstraction and sugar effluent discharges into the river water system. The limitations of river basins as open systems (Raj and Azeez, 2012), was overcome through linkages made between physico-chemical quality of Nzoia River and phytoplankton species diversity due to effects of Mumias sugar factory effluents discharge in the present study.

The various data analysis tools used in the current study were aimed at synthesising complex data collected (Chen and Stroup, 1993). These quantitative scientific methods are used in understanding complex relationships among organizational and environmental variables to optimize decisions (Katz and Kahn, 1966). The conceptual framework of the current study incorporated the input-process-output model that is well grounded in the systems theory.

In summary, the methods used in the present study can offer an effective solution to water quality management for the cases involving complexity in quality data. These statistical tools provide more objective interpretation of surface water quality variables. They disclose most important parameters responsible for variation in the dataset and identify origin of pollutants and all possible water pollution sources. Therefore, there is need to properly manage effluents discharge in the Mumias sugar factory and monitor anthropogenic activities, to ensure minimal negative effects on the Nzoia River.

The findings may be included into the estimation of criteria of the discharge standards during standard-setting process of the Nzoia River water system. These findings will enhance the linkage between physico-chemical water parameters and phytoplankton species diversity in the riverine environments of Lake Victoria basin for effective river water quality management.

## 5.2 Limitations

The interpretation of the identified principle components (PCs) in the current study was limited due to lack of adequate information on their activities. This is similar to findings made by Mazlum *et al*., 1999 who found that some activities might have lasted for short periods and not for the entire duration of the observation period.

Uncertainties arose in explaining the physical nature of significant variables in some components (e.g. $BOD_5$, DO, TDS and EC in the PC1 in Table 4.3). This perspective may imply that the principal components (PCs) were not obtained well enough for interpretation of the components with respect to variables. Insufficient data and errors in analyzing the quality variables in the laboratory may have contributed to this failure (Mazlum *et al*., 1999). Since this was a data-driven study, the duration may have limited the availability of data, restraining its applicability. The proposed SWR model (Equation 14) was developed under natural conditions, without a water regulating structure on the Nzoia River; hence, the use of model in other basins with infrastructure may not be defensible (Wuttichaikitcharoen and Babel, 2014).

Furthermore, the procedures of SWR assume a linear relationship between the variables or their functions. This is rather rare for environmental data that is normally non-linear while methods are based on linear principles (Abdallaoui and Badaoui, 2014). The nonlinear responses were resolved by use of sufficiently high sample size of a multistage sampling, independently repeated random sampling design and use of unbiased multivariate statistical techniques (Reeve *et al*., 1994, Jayaraman, 1999, Jan, 2014 and; Salim *et al*., 2014). This provided a representative and consistent analysis of the water quality in the present study (Salim *et al*., 2014).

# CHAPTER SIX
# CONCLUSION AND RECOMMENDATIONS

## 6.1: Conclusion

The study findings indicate that Mumias sugar effluent had effects on the phytoplankton diversity. It can be concluded that:

**(1)** The process ($S_2$) was significantly more polluting than milling ($S_1$) and boiler ($S_3$);

**(2)** The sugar effluent quality upstream ($S_4$) and downstream ($S_6$) sampling stations after treatment and before discharge were above the NEMA and WHO limits;

**(3)** The physico-chemical water quality parameters in the Nzoia River upstream and downstream Mumias factory effluent discharge were not within NEMA and WHO limits.

**(4)** The COD, $BOD_5$, EC and TDS may have accounted for 52% in the variation of water quality, corresponding to the following likely pollution sources: (a) The combined industrial (sugar) and domestic effluents discharge point sources at $RN_4$ and $RN_5$ respectively and; (b) The physico-chemical and; soil leaching or agricultural run-off process variability respectively. Both **(4)** (a) and (b) may have accounted for 48% in the variation of water quality.

**(5)** The Nzoia river waters were likely to be organically polluted at the study site.

**(6)** The domestic effluents discharge sampling station ($RN_5$) was the principal sampling station having most effects on quality of water, than other non-principal sampling stations.

**(7)** The COD, EC and $BOD_5$ were the most important physico-chemical water quality parameters that had an effect on phytoplankton species diversity.

The statistical relationship between phytoplankton species diversity and the physico-chemical water quality reflected the overall environmental condition in the study area.

## 6.2: Recommendations

### 6.2.1: Determination of the Physico-Chemical Water Parameters Loads of Mumias Sugar Factory Effluents Before and After Treatment

The company should consider a closed circle process in order to have all liquid wastes subjected to integrated waste management principles. This can reduce environmental impacts. Monitoring of effluent generation points can enhance the decision making processes on waste management.

### 6.2.2: Assessment of the Physico-Chemical Water Quality Parameters in the Nzoia River Upstream and Downstream Mumias Factory Effluent Discharge Points

The company should consider improving the current technology for sugar and domestic effluent treatment. The constructed wetlands maybe of an immediate advantage for secondary treatment in re-use of the resultant liquid.

### 6.2.3: Comparison of the Effects of the Physico-Chemical Water Quality Parameters in the Nzoia River on the Phytoplankton Species Diversity Upstream and Downstream Mumias Sugar Factory Effluent Discharge Points

The company can enhance the biomonitoring programs on effluents generation and discharge into Nzoia River by applying the mathematical linkage between phytoplankton species diversity and the physico-chemical water quality. This can reflect the overall environmental health of the river. Finding this relationship will help in determining the quality of aquatic environments.

The company can to undertake monitoring of the soils which likely impact on the Nzoia River from the run-offs.

The government through NEMA can review the current use of physico-chemical water quality to determine discharge standards and incorporate phytoplanktons as bioindicators

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### 6.2.4: Further Research

Further study can be dedicated to finding similar SWR model within the riverine environments of Kenya and Lake Victoria basin. This may assess to compare conclusion on the choice of phytoplankton species diversity and physico-chemical parameters. The SWR model in the present study may be verified within a broader geographical area and a broader range of variables identified in the four (4) most significant PCs besides the physico-chemical parameters. This will provide an adequate long period of data and more stations with similar hydrological and geo-morphological conditions, and re-generating a new set of regression model.

Findings similar to the current study may be incorporated as a sub-index of the water quality index for the monitoring program. This will entrench the theoretical consensus on measurements of diversity metrics about the quality, sampling design and analytical protocols in the riverine environments within the Lake Victoria basin and Kenya.

It would be important to conduct a study that examines more closely, whether or not anthropogenic activities across the Nzoia river basin have similar or different effects on the phytoplankton species diversity, as was the case in the present study.

Finally, the Mumias Sugar company management, National Environment

Management Authority (NEMA) and the government can therefore incorporate phytoplankton species diversity into indices based on multimeritcs. This may achieve a more elaborate water quality-monitoring program within the Nzoia river water basin and other similar freshwater bodies.

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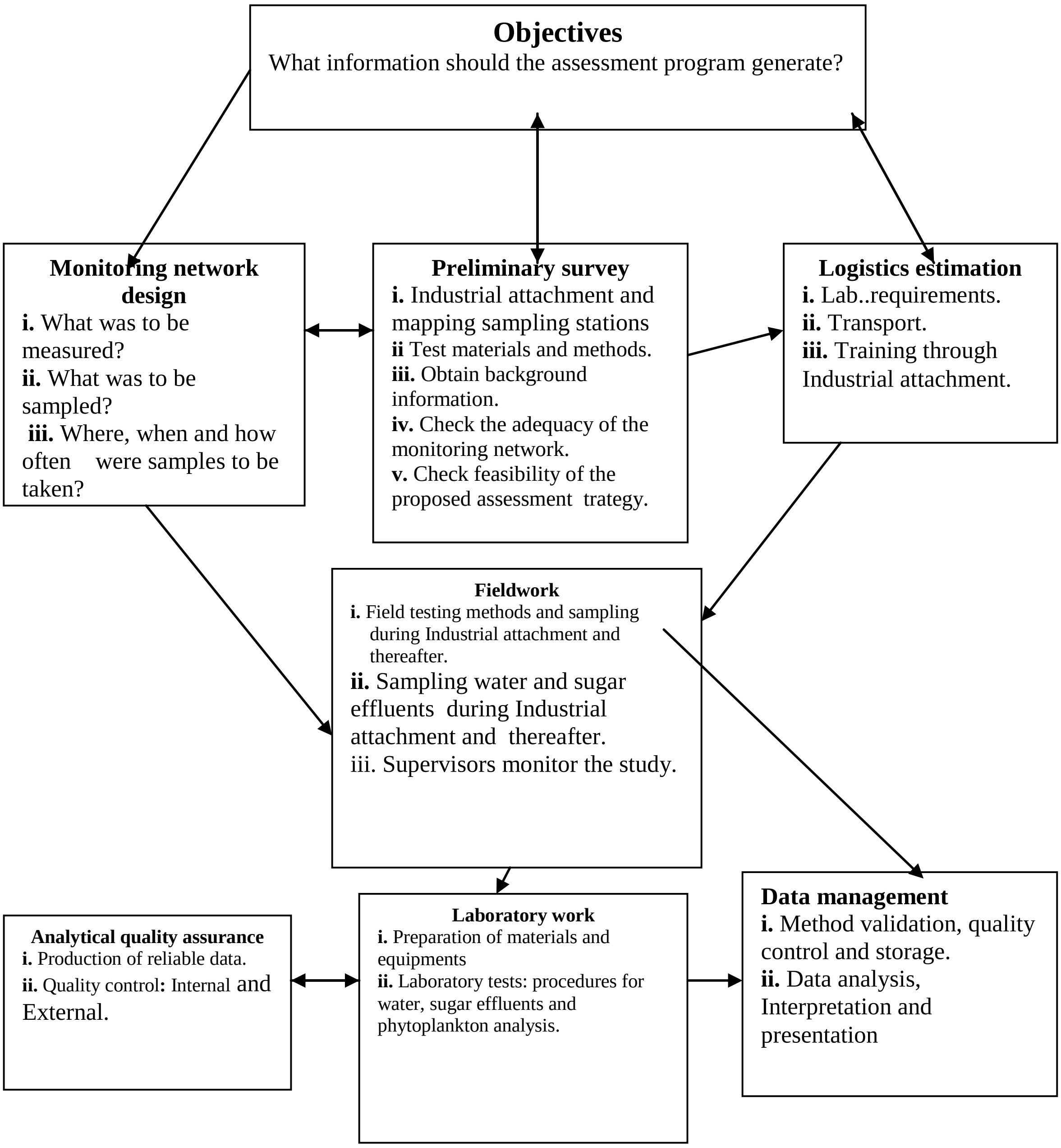
# APPENDICES

## Appendix I: Elements of the Study's Design for the Nzoia River



## Appendix II: Consent Letter for Permission to Carry out Research

**MOI UNIVERSITY**

**SCHOOL OF ENVIRONMENTAL STUDIES**

Tel: (053) 43013
Fax No: (053) 43292/43149
Telex No. 35047 MOIVARSITY

P. O. Box 3900
Eldoret
KENYA

**REF: SES/PGH/11/08** **2$^{ND}$ DECEMBER, 2009**

The Office of the President
Department of Research
P.O BOX 30510
**NAIROBI**

Dear Sir,

**SUBJECT: PERMISSION TO CARRY OUT RESEARCH.**

This is to introduce to you Mr Welinga Alwang'a Martin who is a bonafide student registered for Master of Philosophy in Environmental Studies ( Environmental Health) in the Department of Environmental Biology and Health, School of Environmental Studies , Moi University.

Mr Alwang'a is carrying out a research on:

**"Assessment of Physico-Chemical Quality Parameters of Mumias Sugar Factory Effluents and Potential Impacts on River Nzoia Waters."**

This letter is to request you to kindly allow him to carry out the research .

Your assistance will be highly appreciated.

Yours faithfully,

DEAN
SCHOOL OF ENVIRONMENTAL STUDIES
MOI UNIVERSITY

**S.N. MBUGUA**
**FOR: DEAN, SCHOOL OF ENVIRONMENTAL STUDIES**
/hn

## Appendix III: Acknowledgement Letter from Mumias Sugar Factory for Industrial Attachment during Time of Data Collection

MUMIAS SUGAR
COMPANY LIMITED

MUMIAS SUGAR COMPANY LIMITED

Head Office
P.O: Private Bag Mumias, Kenya
Tel: +254 56 641620, 641621
Cell: +254 722 203891-5, 0734 600334/5
Fax: +254 56 641234
email: msc@mumias-sugar.com

Nairobi Office
P.O. Box: 57092 City Square 00200 Nairobi, Kenya
Tel: +254 20 271 2317-8
Cell: +254 720 140080
Fax: +254 20 271 2316
email: msc@mumias-sugar.com

Our Ref : PEA/E/128

Date : 20 November 2009

WELINGA MARTIN
Moi University

Dear Sir/Madam,

**RE: INDUSTRIAL TRAINING ATTACHMENT ACKNOWLEDGEMENT**

This is to acknowledge receipt of your enquiry dated 20-NOV-09 in connection with the above subject.

We would like to inform you that we can only consider trainees for practical training attachment after the following conditions have been fulfilled:-

1. First of all a preliminary application must be channeled well enough in advance through the Principal of your institution, enclosing a detailed proposal outlining your training needs and indicating the period of attachment. Practical training attachment is, however, restricted to a maximum period of 12 weeks.

2. Trainees must be self supporting i.e they must provide their own accommodation, meals and all other expenses. The Company will not be in a position to provide financial assistance in any way.

3. Trainees must provide for their own stationery, tools and protective wear i.e overalls or dust coats, gloves, safety belts and head gear.

4. The Institution concerned must accurately complete and sign the enclosed indemnity forms in triplicate, the original which must be signed over Two Kenya Shilling Revenue stamp. In certain special cases the parent or guardian of the trainee can sign the indemnity form in which case an advocate of the High Court of Kenya witnesses the indemnity. This is to indemnify the Company against injury to the trainee and also against damage to our equipment and is valid for a period of 12 months.

Yours faithfully,
for: MUMIAS SUGAR COMPANY LIMITED

LYDIAH MARIA [illegible]
HEAD OF HUMAN RESOURCES

## Appendix IV: Industrial Training Attachment Indemnity Form

**MUMIAS SUGAR**

Natural Kenyan Sweetness

**TRAINING CENTRE** **REF: PER/E/12B**

**INDUSTRIAL TRAINING ATTACHMENT INDEMNITY FORM**

**THIS AGREEMENT was made on the** 30th Nov **day of the year** 2009
**BETWEEN Mumias Sugar Company Limited as the training party and** Mr.WELINGA A. MARTIN
**As the trainee party.**

**WHEREBY it is AGREED as follows:-**

1. **Mumias Sugar Company Ltd. shall at its absolute discretion take and accept as trainees such number of persons as may in writing be agreed up**
2. **on from time to time between the Mumias Sugar Company Ltd. and** Mr.WELINGA A. MARTIN

2. **the Training of such persons shall be for such period as may from time to time be agreed upon between the Mumias Sugar Company Ltd. and** Mr. WELINGA A. MARTIN **and under such Conditions, rules, regulations, orders by laws or the like as the Mumias Sugar Company Ltd. shall from time to time impose.**

3. **without prejudice to Clause 2 above** Mr. WELINGA A. MARTIN **shall at all times save and keep the Mumias Sugar Company Ltd., its agents and or servants harmless and indemnified in respect of all costs, claims, damages, expenses and liabilities of whatever nature and kind arising out of or in connection with or in respect of any form or programmes of training within a period of 12 months of any person agreed upon by** Mr. WELINGA A. MARTIN **and the Mumias Sugar Company Ltd. to be trained by the Mumias Sugar Company Ltd. on behalf of** MOI UNIVERSITY & ELDORET POLYTECHNIC.

4. **This agreement shall be valid for a period of 12 months from** 1st December 2009
**IN WITNESS whereof the parties to these presents have hereunto set their hands the day and year first above written.**

**SIGNED FOR AND ON BEHALF OF**
**MUMIAS SUGAR COMPANY LIMITED**

**Date:**

**WITNESS**

**SIGNED FOR AND ON BEHALF OF**

**Date:**

Mr. WELINGA A. MARTIN

Alubala A. Andambi
Advocate and Commissioner for Oaths
P. O. Box ... Eldoret

**WITNESS**

# Appendix V: Monitoring for Discharge of Treated Effluent into the Environment

(r. 14)

SIXTH SCHEDULE

**MONITORING FOR DISCHARGE OF TREATED EFFLUENT INTO THE ENVIRONMENT**

Lead Agency: ..........................................................................................................

Name of organization ..........................................................................................

Nature of work .....................................................................................................

Sample No ............................................................................................................

Description of sample ..........................................................................................

Date and time sample received in lab ..................................................................

Date and time sample was examined ...................................................................

**Average* Flow Rate ($m^3$/day)**

| **Parameter** | **RESULTS** | | | | |
|---|---|---|---|---|---|
| | Sample upstream | Sample at discharge point | Sample downstream | Guide value | Remark |
| pH | | | | 6.5-8.5 | |
| Biological Oxygen Demand (5 days at 20 °C) | | | | 30 (mg/L) max | |
| Chemical Oxygen Demand | | | | 50 (mg/L) max | |
| Suspended solids | | | | 30 (mg/L) max | |
| Ammonia –$NH_4$ + Nitrate-$N0_3$ + Nitrite –$N0_2$ | | | | 100 (mg/L) max | |
| Total Dissolved Solids | | | | 1200 (mg/L) max | |
| E.Coli | | | | Nil/100 ml | |
| Total coliform | | | | 1000/100 ml | |

***Based on sampling analysis monitoring frequency. (daily/weekly/monthly/quarterly)**

Others

1. ..........................................................................................................
2. ..........................................................................................................
3. ..........................................................................................................
4. ..........................................................................................................

As guided by the Fourth Schedule or as may be directed by the Authority

## Appendix VI: WHO Discharge Standards

| | WHO Discharge Standards | |
|---|---|---|
| **S/N** | **Parameters** | **Standards** |
| 1 | pH | 6-9 |
| 2 | TDS($mgl^{-1}$) | 2,000 |
| 3 | COD($mgl^{-1}$) | 1,000 |
| 4 | $BOD_5$($mgl^{-1}$) | 50 |
| 5 | DO($mgl^{-1}$) | 5 |
| 6 | EC($\mu scm^{-1}$) | 1,000 |
| 7 | Temp($^oC$) | 40 |

**Source:** WRMA (2010), courtesy of Nyamori Rose, head of water quality and pollution control (WRMA, Nairobi)