



**UNIVERSITY OF NAIROBI**

**ASSESSMENT OF POLYCHLORINATED BIPHENYLS AND  
ORGANOCHLORINE PESTICIDES RESIDUE LEVELS IN FISH FROM  
NANDI-LOWER NYANDO RIVER SUB-CATCHMENT, KENYA**

**BY**

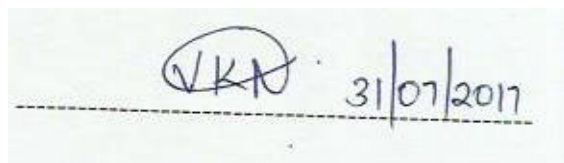
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THESIS SUBMITTED IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE  
AWARD OF DEGREE OF MASTER OF SCIENCE IN ENVIRONMENTAL CHEMISTRY  
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## DECLARATION

This thesis is the original work of the author except where due references are made. It has not been submitted partially or wholly for the award of degree to this or any other institution of learning.

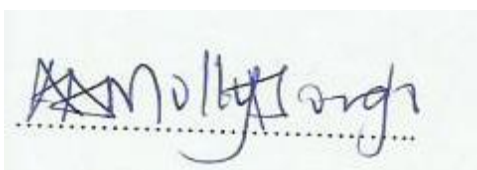


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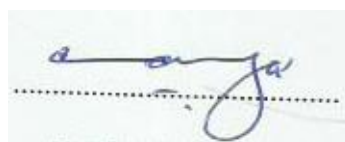
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## **DEDICATION**

Dedicated to my creator, the most Gracious. A special feeling of sincere gratitude goes to my loving parents David and Ruth Nthusi, my brothers: Nathan and Gift Nthusi and my friends for their words of encouragement throughout.

I dedicate this work and give special thanks to my wife Jane Wanjiru and my wonderful son Elvis Mutua for being there for me throughout the masters program. Both have been my greatest cheerleaders.

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## ABSTRACT

On the Kenyan side of Lake Victoria lies its largest extension, Winam Gulf, averaging 10 meters in depth and covering area of 1350 km<sup>2</sup>. It drains to the open waters of the lake through Rusinga channel. The long rains season in the gulf is generally from March to May with November being the short rainy season, followed by the dry season. Nyando drainage basin has been divided into five main sub-catchment areas namely Nyando-Nandi, Nyando-Kericho, Awach-Kano and Nyando-Kano. It has been noted to be the most polluted drainage basin owing to the numerous farming activities and a wide range of pesticide application in farms along its banks.

This study aimed at determining the fish species of lower Nyando, correlating their occurrence to water quality parameters and quantifying the chemical pollution in the basin. In this regard, baseline data that can enable development of a monitoring plan for lower Nyando was generated. A total of 23 fish species from 5 orders and 7 families were collected from the thirteen sampling sites in the Nandi-Lower Nyando River sub-catchment. The family cyprinidae had the majority of the fish species (48%). Six major species were collected in the study sites and these included *Barbus altianalis*, *Barbus nyanzae*, *Clarias gariepinus*, *Clarias leocephalus*, *Labeo victorianus* and *Oreochromis niloticus*. Two indices were used in this study to give emphasis to the different aspects of diversity; the Shannon index that stresses the richness component and the Simpson index that lays greater emphasis on the evenness component. Sites are at the lower reaches of the river; Nyando at Dykes, Awach Kano and Ahero Irrigation Channel (Sites 17, 18 and 33), had the highest richness of species compared to sites in the middle reaches of the river; Ainopngetuny, Ainopisiwa and Kapngorium (Sites 22, 23 and 26). In general, the high diversity and richness sites are served with urban discharge and water from channels in the irrigated rice growing areas in Ahero and Awach Kano whereas the low diversity and richness sites are characterised by large scale coffee and sugarcane farming.

There was significant variation ( $P < 0.05$ ) in most physico-chemical parameters between sampling sites in the four sampling periods as determined by one-way ANOVA. Correlation redundancy analysis (RDA) biplot showed a good relationship between fish species distribution and environmental variables. The most influential and explanatory environmental variables were altitude, temperature and dissolved oxygen concentration (DO). pH, TDS, turbidity, COD, BOD, temperature and phosphorus concentration positively influenced fish diversity.

Altitude and dissolve oxygen negatively influenced fish diversity but were associated with the presence of *Barbus neumayeri* and *Clarias liocephalus*.

Organochlorine pesticides investigated were  $\gamma$ -HCH,  $\delta$ -HCH,  $\alpha$ -HCH,  $\beta$ -HCH, aldrin, dieldrin, p,p'-DDT, p,p'-DDE, p,p'-DDD,  $\alpha$ -Endosulfan,  $\beta$ -Endosulfan, endosulfan sulphate, heptachlor, heptachlor epoxide, endrin, endrin aldehyde and methoxychlor with hexachlorohexanes as the most predominant pesticides. Total OCPs detected were highest in September at site 18 name (109.53 ng/g ww) and lowest in December at site 27 name (7.49 ng/g ww). Residue levels of aldrin, endrin and heptachlor were higher than their metabolites dieldrin, endrin aldehyde and heptachlor epoxide, indicating recent exposure of the fish species. Six indicator non-dioxin-like polychlorinated biphenyls (Congener numbers: 28, 52, 101, 138, 153, and 180) were analysed. Total PCBs detected were highest in September at site 17 (6.13 ng/g ww) and lowest in July at site 21 (0.34 ng/g ww). The results obtained showed higher concentrations of  $\sum$ PCBs in carnivorous fish species than in bottom feeders, indicating bioaccumulation in the food web of the river basin. Complimentary studies, incorporating biomagnification and bioaccumulation investigations, should be conducted to determine point and non-point sources of POPs and their fate in the aquatic environment especially around Ahero Irrigation Scheme.

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## LIST OF ACRONYMS

AMAP	Arctic Monitoring and Assessment Programme
ANCAP	African Network for Chemical Analysis of Pesticides
ANOVA	Analysis of Variance
BDL	Below Detection Limit
BOD	Biological Oxygen Demand
CIFA	Committee of Inland Fisheries in Africa
COD	Chemical Oxygen Demand
COP	Conference of the Parties
DDD	Dichlorodiphenyltrichloroethane
DDE	Dichlorodiphenyldichloroethylene
DDT	Dichlorodiphenyltrichloroethane
DO	Dissolved oxygen
EU	European Union
GC-ECD	Gas Chromatography Electron Capture Detector
GC-MS	Gas Chromatography–Mass Spectrometry
GEF	Global Environment Facility
GMP	Global Monitoring Plan
HCH	Hexachlorocyclohexane
ICH	International Conference on Harmonisation
IFS	International Foundation of Science
IPEP	International POPs Elimination Project
IUCN	International Union for Conservation of Nature
KMFRI	Kenya Marines and Fisheries Research Institute
LOD	Limit of Detection
LOQ	Limit of Quantification
LVEMP	Lake Victoria Environmental Management Project
LVFO	Lake Victoria Fisheries Organization
LVFRP	Lake Victoria Fisheries Research Project
LVWR	Lake Victoria Water Resources
NDL	Not Detectable
OCPs	Organochlorine Pesticides
PCBs	Polychlorinated Biphenyls
PCDDs	Polychlorinated Dibenzodioxins
PCDFs	Polychlorinated Dibenzofurans
PCPB	Pest Control Product Board
POPs	Persistent Organic Pollutants
RDA	Redundancy Analysis
TDS	Total Dissolved Oxygen
TSS	Total Suspended Solids
UNEP	United Nations Environment Programme
USEPA	United States Environmental Protection Agency
WHO	World Health Organisation

## UNITS OF MEASUREMENT

g	Gram
Kg	Kilogram
L	Litre
ml	Millilitre
ng	Nanogram
NTU	Nephelometric Turbidity Units
ppb	Parts per billion
ppm	Parts per milion
ppt	Parts per trillion
$\mu\text{g}$	Microgram
$\mu\text{L}$	Microlitre
$\mu\text{S}$	Micro Siemens

# CHAPTER ONE

## INTRODUCTION

### 1.1 Lake Victoria Basin

Kenya's is a diverse country as is seen through her mountains, flat lands, landscapes and coastlines. From the snow-capped mountains in the eastern and western highlands; Mount Kenya (5,200 metres), Mount Elgon (about 4,300 metres), Aberdares Range (about 4,000 metres) [Mathu and Davies, 1996], to the the Indian Ocean. The topography gradually shifts from low-lying coastal plains to highlands and into the Great Rift Valley. The Nyando, Lolgorien and Nandi Escarpments separate the western highlands from the Lake Victoria Basin [Mathu and Davies, 1996].

With a basin area of 184,000 km<sup>2</sup> and a total area of 68,800 Km<sup>2</sup> [Swallow *et al.*, 2009], Lake Victoria is the second largest fresh water lake after Lake Superior. The Lake's waters span three East African countries as follows: Tanzania (51%), Uganda (43%) and Kenya (6%) [Nyeko-Ogiramoi *et al.*, 2013]. The Lake Victoria basin is comprised of 11 river basins [Swallow *et al.*, 2009]. The major rivers draining into the Lake Victoria basin, based on geopolitical sources, are Mara, Kagera, Grumeti, Issanga, Mirongo, Mbalageti and Simiyu from Tanzania and Uganda [Nyeko-Ogiramoi *et al.*, 2013]. On the Kenyan side, six major rivers drain into the Lake Victoria, including the Kuja-Migori (with a total area of 6600 km<sup>2</sup>), Nyando (3,450 km<sup>2</sup>), Nzoia (12,842 km<sup>2</sup>), Sio (1,437 km<sup>2</sup>), Sondu-Miriu (3,508 km<sup>2</sup>) and Yala (3,357 km<sup>2</sup>) [Raburu and Masese, 2012], contributing over 37% of its surface water inflows [Nyeko-Ogiramoi *et al.*, 2013].

The largest river basin is the Kagera, contributing about 33% inflows into the Lake, and drains parts of Rwanda, Burundi, Tanzania and Uganda west of Lake Victoria [Swallow *et al.*, 2009; Nyeko-Ogiramoi *et al.*, 2013]. Draining Lake Victoria is the River Nile; with an average outflow of about 23.4 km<sup>3</sup> year<sup>-1</sup>, therefore the Lake basin contributes the largest volume to the upper River Nile [Nyeko-Ogiramoi *et al.*, 2013]. Lake Victoria has high ecological value and offers ecosystem services in the form of hydroelectric power and water used in the tourism and fisheries sectors for transport, domestic, agricultural and industrial use [Swallow *et al.*, 2009].

Various studies carried out in the basin have found that the high pollutant load to the basin from the use and discharge of pesticides into the lake may cause severe degradation of the environmental quality [Werimo *et al.*, 2009]. Most of the agricultural transformations are taking place in Lake Victoria basin mainly because of its fertile soils and a favorable climate [Wasswa, 2004]. The lake sustains important fisheries and is relied upon by local communities for their livelihood and as one of the major sources of income. A variety of fish is widely distributed throughout the lake with most species found at a depth of 30 metres [Wasswa, 2004].

## **1.2 The Winam Gulf**

On the Kenyan side of Lake Victoria lies its largest extension, Winam Gulf, averaging 10 meters in depth [Romero *et al.*, 2005] and covering area of 1,350 km<sup>2</sup>. It drains to the open waters of the lake through Rusinga channel. The long rains season in the gulf is generally from March to May with November being the short rainy season, followed by the dry season [Fusilli *et al.*, 2013]. These favorable climatic conditions have led to rapid urban growth and consequently environmental problems.

The Committee for Inland Fisheries of Africa (CIFA) noted the pollution problems in Kenya's side of Lake Victoria since the catchment basin is large with six rivers draining toxic chemicals into the lake [Abongo *et al.*, 2014]. Winam Gulf was noted to be the most polluted catchment area [Calamari *et al.*, 1995] within the lake basin. Its catchment has an area of 11,994 km<sup>2</sup> and is constituted by the Nyando and Sondu-Miriu River basins alongside the North and Southern Lakeshores [Abongo *et al.*, 2014]. The Northern and Southern lakeshores comprise of seasonal rivers. Because of the CIFA concerns, it would be crucial to conduct a study focusing on the chemical pollutants entering the Winam Gulf through its tributaries upstream [Abongo *et al.*, 2014].

The large river basins, Sondu-Miriu and Nyando, carry nutrient and sediment load from agricultural activities upstream such as tea [Abongo *et al.*, 2014] and sugar cane farming [Romero *et al.*, 2005] into Winam Gulf. This excessive organic material promotes growth of unwanted plant life such as weeds and results in eutrophication of the lake [Fusilli *et al.*, 2013]. Domestic waste effluent discharged into the Gulf from surrounding counties (Kisumu, Homa Bay) further increase the nutrient load and consequently the biological oxygen demand of the waters [Scheren *et al.*, 2000].

### **1.3 The Lake Shore Basin**

The lakeshore basin is a dry area with small seasonal rivers draining from it [Calamari *et al.*, 1995]. The Northern Lakeshore consists of Awach- Seme, Kibos, Nyamasaria with Kibos, Asembo-Kisumu and Kadimu-Uyoma as sub-basins. Awach-Kibuon, Awach Tende and Olambwe are seasonal rivers draining the southern sub-basin [Calamari *et al.*, 1995].

#### **1.3.1 The Sondu-Miriu Basin**

Sondu-Miriu basin covers an area of 3,489 km<sup>2</sup>. Tributaries flowing into the basin originate from Kericho and the eastern side of Kisii highlands with Kipsonoi River draining most parts and extending to the southwestern parts of the escarpment. From parts of the main escarpment River Itare joins Kipsonoi to form Sondu-Miriu River. Tributaries flowing into Itare include Chemosit River. Before entering Lake Victoria, Sondu-Miriu River drains the Nyabondo and Nyakach areas.

The river has an annual and monthly average run-off of about 42.2 m<sup>3</sup>/s and 43.0m<sup>3</sup>/s respectively. It drains the high potential agricultural areas of Kericho and Kisii highlands with livestock farming, tea and coffee as the major activities. The Sondu-Miriu basin is divided into sub-basins namely; upper Itare, lower Itare, Kitoi, Kabianga, Sisei, Kapsonoi and Miriu [Calamari *et al.*, 1995].

#### **1.3.2 Nyando River Basin**

Kenya is known to be the main polluter of the lake with highest number of water systems draining into Lake Victoria compared to Uganda and Tanzania. Among the rivers draining into the lake from Kenya are Nzoia, Sio, Sondu-Miriu, Yala, Nyando and Guch-Migori [Abong'o *et al.*, 2015a].

Lower Nyando River sub-catchment originates from Nandi County and is of particular concern since this is a major agricultural and industrial area. With a catchment that spans 3,450 km<sup>2</sup> and a length of 170 km, Nyando river basin has been divided into five sub-catchment areas namely Nandi-Nyando, Awach-Kano, Nyando-Kano and Kericho-Nyando [Abong'o, 2009]. More than 50% of the total water discharge of the Nyando comes from the Nyando-Nandi sub-catchment. The Nyando basin traverses Nandi Hills and Londiani, Kipkelion and Sigowet areas of Kericho County as well as Koru, Muhoroni, Chemelil, Lower Nyakach and Kano plains of Kisumu county.

Industrial and agricultural activities involving application of fertilizers and pesticides in farms in these areas influence the environment. Consequent effluent discharge from coffee, tea and sugar factories within the Nyando catchment area is a major source of pollution load to the Kenya side of Lake Victoria [Calamari *et al.*, 1995]

The Nyando River has been noted to be the most polluted drainage basin owing to the numerous farming activities and a wide range of pesticide application in farms along its banks and sedimentation [Abong'o *et al.*, 2014]. It also has the following characteristics that qualify its choice; average sediment transfer index of 0.30 and average % slope of 5.0 [Odada *et al.*, 2004] thus making its impact on Lake Victoria very high.

#### **1.4 Lake Victoria Water Quality and Pollution Sources**

Pollution impact by municipal and industrial discharge and rain run-off from agricultural fields are evident in rivers flowing in to Lake Victoria and extends to large inlets such as Winam Gulf. Urbanisation and population growth of Kisumu town, in its 100 years of existence celebrated in 2002, has led to routine discharge of raw, untreated sanitation and industrial effluent directly into Winam Gulf.

Various studies [IUCN, 1992; Cru Ruud, 1995; Henry *et al.*, 2000; Tole *et al.*, 2000] have reported increased pollution from pesticide use in the Tanzania and Kenyan sides of the lake due to their use to boost agricultural productivity. Tea, coffee, rice, sugar cane and horticultural products are farmed in large-scale estates spread within the vast catchment. Surface run-off from these farms contains pesticides and fertilizers that reach rivers draining in to the basin. Other reported uses of pesticides in the basin include killing bird pests [Aryamanya-Mugisha, 1993] and for fishing [Orgaram, 1992].

Following press reports in May 1999 alleging fish harvesting in Lake Victoria by use of endosulfan insecticide, the European Union banned all fish import from the East African region. Commercial fishing activities around the three East African countries were greatly affected resulting in an estimated loss of income of more than 300 million dollars. This greatly affected economies in the region. Use of dichlorodiphenyltrichloroethane (DDT) for vector control and other organochlorine pesticides in the East Africa region has continued despite international efforts through the Stockholm convention to prohibit their production and use. Trace amounts of DDT residues have been reported in sampled lake waters [Calamari *et al.*, 1995].



#### **1.4.1 Programmes that serve Lake Victoria**

The three countries sharing Lake Victoria waters have plans to use it for different purposes. In order to properly manage this resource, understanding the water regime in the lake basin is important. This has resulted in joint concerted efforts to preserve the lake's environment and fisheries. The spread of invasive species *Eichhornia crassipes* (water hyacinth) further attracted the attention of the international community and resulted in inception of programmes aimed at understanding the lake's ecosystem such as: the Lake Victoria Water Resources (LVWR) project, Lake Victoria Environmental Management project (LVEMP) and the Lake Victoria Fisheries Organization (LVFO).

Lake Victoria Water Resources (LVWR) project helped establish a network of rain gauges for monitoring hydro meteorological parameters in the lake. The Lake Victoria Fisheries Organization (LVFO) helped create a common resource management approach amongst the three East African countries to ensure sustainability in lake management while the Lake Victoria Fisheries Research Project (LVFRP) focused on developing a coordinated action plan for the fisheries sector in riparian governments. The lake is solely drained by the Nile River from the Uganda side. Managing the lake for the benefit of communities downstream remains a cause for tension.

Lake Victoria Environmental Management project (LVEMP) aimed to improve resource efficiency while conserving the lake's biodiversity and genetic resources in order to reverse environmental degradation. Phase I of the project began in 1997 and was completed in 2004. Phase II of the project began in 2009 and is scheduled to end in 2017. The focus in the current phase is to improve environmental management of the lake amongst the five East Africa community partner states (Kenya, Tanzania, Uganda, Burundi and Rwanda) through institutional capacity building targeting pollution hotspots in sub-catchments.

Despite strong commitment to maintain the ecological integrity of the lake from the countries sharing its waters, most of these programmes focus on aspects of fisheries. There has been limited research done on chemical pollutants, most of which are pesticides.

This study therefore aims to quantifying polychlorinated biphenyls and organochlorine pesticide residue levels in fish from Nandi-Lower Nyando River sub-catchment and make recommendations for sustainable ways to manage pollution in order to conserve safe water and ecology in Kenya's Lake Victoria.

## 1.5 Hypothesis

Two hypotheses were tested in this study;

- i) There is no variation of fish species in the pristine Nandi-Lower Nyando River sub-catchment.
- ii) Detection of the banned and restricted organochlorine and PCB pesticide residue levels in fish along Nandi-Lower Nyando is due to bioaccumulation and biomagnification.

## 1.6 Problem Statement

In general, studies on residue levels and effects of the various agrochemicals on Kenya's environment are very limited [Werimo *et al.*, 2009], particularly organochlorine pesticides. This category of pesticides has been widely regulated in most countries because of their high persistence and their propensity for bioaccumulation [ANCAP, 2004]. In spite of this rather limited information and research on pesticides, importation of pesticides in Kenya is largely uncontrolled. The increased pesticide use within the country has continuously resulted into misuse mainly due to lack of information in the population about pesticide handling. The situation has been aggravated by violation of pesticide use regulations by unscrupulous people as previously seen in 1999 when the European Union banned fish import from Lake Victoria following allegations of use of endosulfan to capture fish in the lake. Local and international markets experienced reduced sales as several cases of endosulfan use in fishing were reported that year [Werimo *et al.*, 2009]. As lower Nyando traverses industrial towns, PCBs are released to the aquatic environment from improper disposal of waste equipment such as transformers [Kenya NIP, 2014] and can bioaccumulate up the food chain.

Because of the European Union fish import ban and Central Institute of Freshwater Aquaculture (CIFA) concerns it would be crucial to conduct a study focusing on the chemical pollutants entering the Winam Gulf and in particular from Nyando River drainage basin - the major Kenyan river source of sedimentation and pollution into the lake. Previous studies in the lake basin have shown a heavy bias towards fisheries with limited research done on chemical pollution from pesticides and industrial effluent.

There is need therefore, to establish baseline data in pesticide residue levels in fish species within the Nyando basin in a bid to develop a policy on safe use of pesticides in the region.

## **1.7 Objectives of the Study**

### **1.7.1 *General Objectives***

The previous studies on water, sediments, soil and aquatic weeds showed that organochlorine pesticides are detected in the Nyando River drainage basin [Abong'o, 2009] and could have negatively impacted the ecosystem health of the basin. The current study aimed at generating baseline data on polychlorinated biphenyls and organochlorine pesticide residue levels in fish in the Nandi-Lower Nyando River sub-catchment in order to improve knowledge for action by the Kenya Government to institute remedial measures such as stringent control over the application of persistent and banned pesticides and to encourage agricultural practices which integrate pest management.

### **1.7.2 *Specific Objectives***

- 1) Determine the types, nature, occurrence, abundance and distribution of fish species found along the Nandi-Lower Nyando River sub-catchment.
- 2) Link the occurrence, abundance and distribution of fish species to effects of some measured physico-chemical parameters along the Nandi-Lower Nyando River.
- 3) Quantify the levels of polychlorinated biphenyls and organochlorine pesticide residue levels in the fish species along the sub-catchment.

## **1.8 Justification**

The three East African countries that share Lake Victoria have, in recent times and with support from the international community, promoted joint efforts to manage and preserve the lake's fisheries and water resources. Analysis of pesticide use, distribution and fate in fish in the major tributaries of Lake Victoria has not been a priority in these efforts. A study carried out in one drainage system, for instance, could provide baseline data and sound methodology to catalyse similar studies in other waterways and the lake.

Products from the region have suffered reduced consumer confidence in international markets due to the detection of high pesticide residue levels. Results of the proposed project will improve the quality of products of agricultural and fishing activities that are a major source of income within the lake basin region and therefore fetch higher prices in world markets.

The Lower Nyando River sub-catchment originates from Nandi County and is of particular concern since this is a major agricultural and industrial area. Agricultural activities and effluent from tea, coffee and sugar factories are a major source of pollution to the river.

The rate of sedimentation due to River Nyando's slope is high compared with all the rivers draining into Lake Victoria [LVEMP, 2003]. This coupled with the fact that analysis of 48 soil samples from six farms growing vegetables, maize, sugar cane, tea and rice in Nyando River drainage basin revealed presence of all the targeted pesticides [Abong'o *et al.*, 2015a]. The mean concentrations recorded decreased in the order methoxychlor ( $138.97 \pm 1.517 \mu\text{g/kg}$ ),  $\Sigma$ endosulfan ( $30.267 \pm 2.098 \mu\text{g/kg}$ ),  $\Sigma$ DDT ( $17.513 \pm 1.689 \mu\text{g/kg}$ ), dieldrin ( $14.073 \pm 0.440 \mu\text{g/kg}$ ), endrin ( $10.155 \pm 0.860 \mu\text{g/kg}$ ), lindane ( $8.985 \pm 1.318 \mu\text{g/kg}$ ) and  $\Sigma$ Heptachlor ( $0.681 \pm 0.021 \mu\text{g/kg}$ ), respectively [Abong'o *et al.*, 2015a]. The distribution showed that dieldrin was in use in vegetable farms in Kedowa area, tea farms in Nandi District and in Ahero rice paddies; while  $\beta$ -endosulfan was commonly used on tea farms in Nandi.

Water analysis from the 26 sampling sites showed the highest mean concentrations detected for methoxychlor ( $8.817 \pm 0.020 \mu\text{g/L}$ ),  $\Sigma$ endosulfan ( $1.648 \pm 0.04 \mu\text{g/L}$ ), dieldrin ( $1.1561 \pm 0.042 \mu\text{g/L}$ ), endrin ( $0.281 \pm 0.003 \mu\text{g/L}$ ),  $\Sigma$ DDT ( $0.242 \pm 0.009 \mu\text{g/L}$ ),  $\Sigma$ heptachlor ( $0.148 \pm 0.011 \mu\text{g/L}$ ) and lindane ( $0.144 \pm 0.006 \mu\text{g/L}$ ) respectively [Abong'o, 2009]. Sediment residue levels were higher than those found in water in the order, methoxychlor ( $92.893 \pm 3.039 \mu\text{g/kg}$ ), lindane ( $33.917 \pm 2.360 \mu\text{g/kg}$ ), aldrin ( $26.676 \pm 0.981 \mu\text{g/kg}$ ), dieldrin ( $23.62 \pm 4.810 \mu\text{g/kg}$ ) and  $\beta$ -endosulfan, ( $10.502 \pm 0.800 \mu\text{g/kg}$ ), respectively [Abong'o, 2009]. The analysis of aquatic weeds recorded methoxychlor ( $39.641 \pm 3.045 \mu\text{g/kg}$ ) as the highest residue concentrations, followed by aldrin ( $15.519 \pm 3.756 \mu\text{g/kg}$ ). These higher levels may be as a result of by accumulation of the pesticide in the drainage basin. The levels of pesticides were higher in sediment, weeds and soil than in water [Abong'o, 2009].

It has been argued that poor land-use management practices, use of agrochemicals and high nutrients load negatively impact River Nyando and Lake Victoria ecosystems [Abong'o *et al.*, 2014, Peters and Meyback, 2000]. It was therefore important to monitor organochlorine pesticides residue levels and PCBs pollution problems in fish in Nyando River basin.

## CHAPTER TWO

### LITERATURE REVIEW

#### 2.1 Persistent Organic Pollutants and the environment

The Stockholm Convention restricts the production, use, release and unsafe disposal of Persistent Organic Pollutants (POPs) in order to protect against the harmful impacts of exposure on human health and on the environment. POPs are organic chemical compounds that resist degradation in the environment, accumulate in fatty tissue where they bioaccumulate through the food chain and are transboundary being distributed geographically through moving air masses and water currents [UNEP-AMAP, 2011]. Due to the transboundary nature of POPs releases to the environment, significant concentrations are found in the aquatic environment from a variety of sources such as surface runoff from farms and point sources such as sewage treatment plant effluent [UNEP, 2013]. The Convention also promotes the use of alternatives in vector and pest control.

A lot of scientific evidence has revealed that POPs are among the most hazardous pollutants released into the environment due to their toxic, lipophilic and persistent nature [Burreau *et al.*, 2004]. Persistent chemicals bioaccumulate and biomagnify through the food web, resulting in increased concentration levels at higher trophic levels in predator species [Ondarza *et al.*, 2012]. Their high affinity to tissue with high lipid content, such as is the case with some fish species in the aquatic ecosystem, enables these toxic contaminants to concentrate upwards in various trophic levels of the food chain; from zooplankton to fish and eventually to humans. Fish tissue monitoring therefore serves as an indicator of aquatic ecosystem quality.

Persistent organic pollutants (POPs) are synthesised organic compounds which include organochlorine pesticides (OCPs), polychlorinated biphenyls (PCBs), and other by-product substances originating from human activities (dioxins and furans) [Deribe *et al.*, 2011]. Past and current use of produced POPs leads to primary release into the environment. That is, direct dispersion into soils and air (pesticides), volatilization into air from initial application (semi-volatile chemicals like PCBs) and leaching into water from initial application [UNEP-AMAP, 2011].

With increase in population and growing demand for food, production of pesticides and fertilizers has been on the rise and has coincided with the development of sensitive analytical equipment (GC-ECD, GC-MS) for trace analysis. With this new technology constantly increasing sensitivity to lower limits of detection (ppt – parts per trillion) trace levels of chemicals can be detected in the environment giving much more information on their toxicity, pathways and environmental fate [UNEP, 2013]. Due to their persistent nature and lipophilic properties, the potential risks of human exposure to POPs are a growing concern. To address this issue, monitoring programs have been established globally and regionally to investigate health risks associated with exposure and determine prevailing levels of POPs in different environmental compartments.

One such initiative is the UNEP/GEF funded Global Monitoring Plan (GMP) that focuses on monitoring POPs in core media (soil, sediment, water, fish, mother's milk and ambient air). The 2012 Kenya national report found presence of POPs in fish and human milk. Environmental samples revealed high levels of Aldrin and dieldrin in fish. Relatively low levels of POPs were found in ambient air and were attributed to transboundary transport rather than point source origin. The Stockholm Convention initially covered 12 chemicals and focused on restriction of use of POPs.

Under the convention, compounds that are prevalent in the environment are listed under Annex A for elimination. They include hexachlorobenzenes, aldrin, dieldrin, endrin, chlordane, mirex, toxaphene, heptachlor and PCBs. Annex B for restriction has DDT only. Annex C for Unintentional Production includes hexachlorobenzenes, polychlorinated dibenzo-p-dioxins (PCDDs), polychlorinated dibenzofurans (PCDFs) and polychlorinated biphenyls (PCBs) [UNEP-POPs Convention Text, 2010].

Pesticides have adverse health impacts on non-target organisms [Gil *et al.*, 2012] despite being developed to kill rodents, insects and weeds. Dietary intake of food items and prey is the main exposure route for humans and wildlife to POPs in the environment. They readily accumulate in human fat tissue after ingestion of contaminated foodstuffs due to their high affinity for lipids. Toxicity and mode of action has been associated with targeting the endocrine system in humans and wildlife [Qing Li *et al.*, 2006] where they can have carcinogenic effects.

Transboundary movement by wind and water currents has led to geographic distribution of persistent organic chemicals as far as the Arctic and Antarctic [Scheringer, 2009]. Over 30% of the world's supply of surface fresh water can be found in the three largest African lakes: Victoria, Tanganyika and Malawi [Hecky, 2003]. This presents a paradox of wealth in a region where getting water can be a daily struggle for millions of households and where devastating droughts can affect national economies. The same three lakes are thought to hold about 10% of all known species of freshwater fishes and nearly all the species in each lake are endemic to that lake [Hecky, 2003]. Lake Victoria, for instance, provides inexpensive animal protein to its population.

## **2.2 Pesticides**

Synthetic pesticides were first used in 1940. It was considered desirable for pesticides to be persistent - so that a single application would last long - and toxic for killing target organisms. However, these qualities also cause harm to non-target organisms since breakdown products (metabolites) are also present in the environment. When introduced in various environmental compartments - such as soils, sediments, and water bodies - these pesticides bioaccumulate [Qing Li *et al.*, 2006] upwards in the food chain.

### **2.2.1 Organochlorine Pesticides (OCPs)**

Organochlorine hydrocarbons are organic compounds containing at least one covalently bonded chlorine atom. They are very stable compounds [Afful *et al.*, 2010] with divergent chemical properties and consequently a broad range of applications. Degradation of dichlorodiphenyltrichloroethane (DDT), for instance, takes 4 to 30 years in the temperate climates [Afful *et al.*, 2010]. Since they persist in the environment, their occurrence at various trophic levels in the food chain has been reported since the 1960s [Frederick, 1991].

Organochlorine pesticides enter the environment through direct application on land for agricultural and disease vector control. Examples include: hexachlorobenzene - widely used as a fungicide; DDT - mostly used as an insecticide; chlordane - used as a soil insecticide; lindane - used as an insecticide; endrin - used as a rodenticide; and heptachlor - used to kill termites.

### **2.2.2 Polychlorinated Biphenyls (PCBs)**

PCBs (Polychlorinated Biphenyls) are commercially produced organic compounds [WHO, 2003] with the chemical formula  $C_{12}H_{10-x}Cl_x$ . A PCB molecule is composed of two benzene rings (biphenyl) around which 2 to 10 chlorine atoms can attach in many different ways to result in a total of 209 different molecules known as congeners.

PCBs are hydrophobic but are soluble in oils and fats making aquatic mammals at high risk of bioaccumulation. They are all man-made compounds that can be transported over long distances [Li *et al.*, 2007] and enter the environment mainly through waterways [Fitzpatrick, 2006] where they bioaccumulate in the fatty tissue of living organisms [Bordajandi *et al.*, 2008]. PCBs have long been used in electrical equipment such as transformers due to their ability to maintain thermal stability.

When burnt at high temperatures they can generate dioxins which are highly toxic. PCB congeners with at most one chlorine atom in the ortho-position exhibit dioxin-like toxicity whereas those with chlorine atoms at two or more ortho-positions express non-dioxin-like toxicity.

### **2.3 Studies on pesticide use in Kenya**

Substantial data on pesticide residue levels in fish has so far been gathered in Kenya. For instance, a study conducted by Madadi (2005) in River Sio showed dieldrin (236.37  $\mu\text{g}/\text{kg}$ ), p, p'-DDD (205.06  $\mu\text{g}/\text{kg}$ ),  $\alpha$ -endosulfan (11.94  $\mu\text{g}/\text{kg}$ ), p, p'-DDT (40.39  $\mu\text{g}/\text{kg}$ ) as the highest residue levels detected during the rainy season. Wandiga *et al.*, (2002) found concentrations of 1011 ng/g and 418 ng/g of p, p'-DDT and p, p'-DDD respectively in sampled fish tissue from the Kenya coastal region.

A study conducted in Athi River by Mugachia *et al.*, (1992) found organochlorine pesticides residues in six species of fish. The concentrations decreased from p, p'-DDE, p, p'-DDT, p, p'-DDD,  $\beta$ -HCH,  $\alpha$ -HCH, heptachlor to o, p'-DDD. In a similar study carried out in the Hola irrigation scheme by Munga (1985) DDT and endosulfan tissue residues showed strong correlation with fat content of sampled fish species. High concentrations were found in the liver and egg of sampled fish than in juvenile fillets.



Lincer *et al.*, (1981) found a bottom feeding fish, *Labeo Cylindricus*, in Lake Baringo with DDE levels of  $7.4 \times 10^{-2}$  mg/kg while Koeman *et al.*, (1972) found total DDT residue levels in fish of  $1.0-7.0 \times 10^{-3}$  mg/kg wet mass, all of which are much lower than concentrations found in marine fish species [Everaats *et al.*, 1996; Barasa, 1998]. Agricultural activities in rivers upstream of the lake contribute to this observed difference.

Ssebugere *et al.* (2014) reported moderate concentrations of PCBs (ranging from 41 to 670 pg g<sup>-1</sup> lipid weight) in two fish species, *Lates niloticus* and *Oreochromis niloticus*, from Napoleon Gulf with higher concentrations found in the predatory species. Omwoma *et al.* (2015) found higher concentrations of non-dioxin-like PCBs ranging from 421 to 958 pg g<sup>-1</sup> dry weight in surface sediment at Kisumu site of Winam Gulf. Oluoch-Otiego *et al.* (2016) analysed seven indicator PCBs in sediment, fish and parasite samples in Winam Gulf. Concentrations ranged from 2.2 to 96.4 µg/kg dry weight in sediment and 300 to 3,000 µg/kg lipid weight in fish. Fish species higher up in the trophic level (carnivorous species) had higher concentrations of PCBs compared to omnivorous species hence suggesting biomagnification.

#### **2.4 Preliminary Studies on agrochemicals used in the Nyando Basin**

In Nandi-Lower Nyando River sub-catchment, Methoxychlor ( $1.088 \pm 0.086$  µg/L) was detected at highest concentration at Chemwanabei [Abong'o, 2009] in water at concentrations lower than WHO drinking water guideline values [WHO, 2003]. Concentrations of heptachlor and lindane were below the detection limits in all the samples analysed. Dieldrin and endosulfan sulfate were detected at all sampling sites except Nyando at Dykes and Chebirirkut at Tinderet Dam. The highest dieldrin concentration was detected at Chemwanabei ( $0.078 \pm 0.006$  µg/L) and endosulfan sulfate at Kapngorium ( $1.558 \pm 0.166$  µg/L). However these concentrations are all above the WHO and Australian guidelines of daily intake for drinking water [WHO, 2003]. For sediment samples, aldrin was detected at Ahero Irrigation Channel ( $16.048 \pm 1.484$  µg/kg) and methoxychlor ( $73.702 \pm 7.893$  µg/kg) at Nyando at Ogilo and lindane ( $10.530 \pm 1.871$  µg/kg) at Chebirirkut, were the highest frequencies of detection in sediment samples. In weed samples the highest concentration was obtained for methoxychlor ( $77.279 \pm 8.661$  µg/kg) and p,p'-DDT ( $38.025 \pm 1.959$  µg/kg) at Kapngorium. These sites are further defined in the next section and form the basis for this study, which seeks to find what the case for fish in Nandi-Lower River.

## CHAPTER THREE

### METHODOLOGY

#### 3.1 Study Area

Nyando River is divided into two sub-catchments; Kericho-upper Nyando and Nandi-lower Nyando) [Abong'o, 2009]. Lower Nyando River sub-catchment originates from Nandi County. With a catchment that spans 3450 km<sup>2</sup>, a length of 170 km [Abong'o *et al.*, 2015a] and an average slope (5%) [LVEMP, 2003], the Nyando basin drains is of particular concern since it traverses major agricultural and industrial areas of Rift Valley highlands of Kenya (Figure 1).

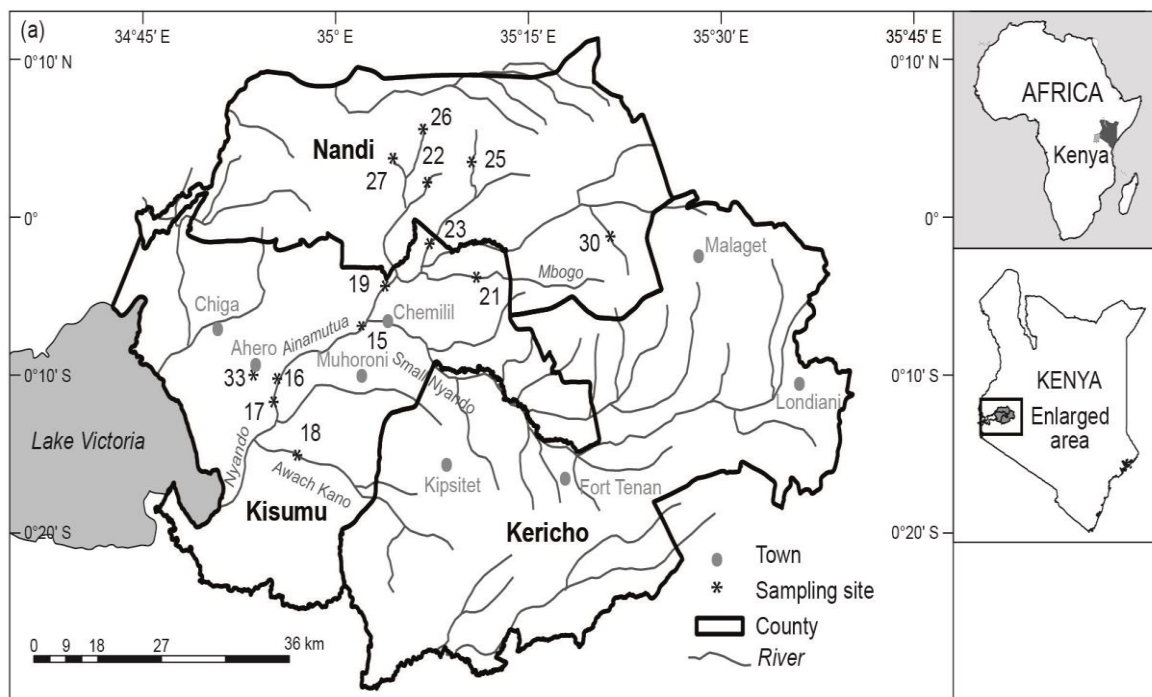
The Ainamotua tributary comprises of streams rising from the Nandi Hills and Tindiret areas in Nandi County and the small Nyando River (Kericho-upper Nyando) itself comprise of streams rising from the Londiani and Mau Forest in Kericho County.

The Nyando River basin traverses Nandi Hills of Nandi County, Londiani, Kipkelion and Sigowet areas of Kericho County and Koru, Muhoroni, Chemelil, Lower Nyakach and Kano plains of Kisumu County. Agricultural activities involving the use of fertilizers and pesticides and industrial activities where effluents from tea and sugar factories are directly emitted into the river are the main source of pollution in these areas [Abongo *et al.*, 2014].

#### 3.2 Environmental conditions in the sampling area

This study is part of the research works on the Nyando River catchment area with a special forecast on lower Nyando sub-catchment area (Figure 1). The Nandi-Lower Nyando River comprises, Sites 15 to 33 (Table 1). Predominant activities in the river basin include small-scale maize, sorghum, tea, coffee and rice farming [Abong'o *et al.*, 2015b]. Land degradation in the Nyando River basin is estimated to affect 1,444 – 1,932 Km<sup>2</sup> of the catchment area [Odada *et al.*, 2009].

Sampling was conducted four times (July, September, December 2011 and March 2012) in order to investigate the seasonal effects of anthropogenic activities on pesticide residue levels in fish. The coordinates, altitude and human activities around the sampling sites were recorded (Table 1).



**Figure 1: Map of Sampling Sites in Lower Nyando**

These sampling sites had been marked out by Lake Victoria Environmental Management Project [LVEMP, 2003]. Site 30 was the reference point designed to provide baseline data on natural water quality. Site 20 is the confluence of rivers from sampling sites 21, 22 and 23 and was not sampled. Sites 24, 28 and 29 (Figure 1) are seasonal streams and therefore were not sampled.

### 3.3 Materials and chemicals

Analytical grade chemicals were purchased from Fisher Scientific (USA), Aldrich Chemical Company and BDH (United Kingdom). HPLC grade solvents (99% purity) that were bought include isooctane and diethyl ether. Acetone and hexane used for sample extraction were triple distilled before use.

Sodium chloride and activated charcoal were baked at 120 °C in a Mermert oven for two hours and cooled in desiccators before use whereas anhydrous sodium sulphate was activated at 200 °C before use in the cleanup process [UNESCO, 1993].

Reference standards for polychlorinated biphenyls (using PCB 103 as the internal standard) and organochlorine pesticides (using Isodrin as the internal standard) obtained from Dr. Ehrenstorfer GmbH Company (Germany) were used for instrument calibration by injecting (1  $\mu$ L) of the calibration standard mixtures into the Agilent gas chromatography (Model 6890N equipped with a detector).

### **3.4 Fish sampling and identification**

Fish were caught from each of the sampling sites with the use of a 400 V (10 A) electro-fisher operated with a Honda GX 240 8 HP generator and identified at the site to species level. During the electro fishing activity a pulsed mode of discharge of electro-fisher was adopted for electrocution lasting 10 minutes at each attempt. The samples were taken from a cross section of the river (50 meters downstream and upstream at each sampling site) at different depths. The samples were identified, counted, the weight and length measured for each fish species at each site. Each sample species were wrapped in sterilized aluminum foil, fastened by masking tape, labeled and placed in self-sealing black polythene bags to avoid contamination. The samples were immediately stored in a refrigerator at -4 °C in the field vehicle and transported to the Kenya Marines and Fisheries Research Institute (KMFRI) laboratory in Kisumu.

Samples from the field were temporarily stored at -19 °C in deep freezers at Kenya Marines and Fisheries Research Institute (KMFRI) during the sampling period awaiting transportation to University of Nairobi laboratory. At the University of Nairobi's pesticide analytical research laboratory, all samples awaiting analysis were stored at -20 °C. Samples stored under refrigeration were brought to room temperature before pesticide extraction and analysis was done.

**Table 1: Description of sampling sites in the Nandi-Lower Nyando River sub-catchment area**

Site Number	Site Name	ALTITUDE	GIS (Position)		Human Activities around sites
			N(+)/S(-)	E	
15	Nyando at Ogilo	1204	-0.166°	35.162°	Human settlement, subsistence agriculture, recreation
16	Nyando at Ahero Bridge	1163	-0.172°	34.921°	Human settlement, subsistence agriculture, sugarcane farming
17	Nyando at Dykes	1160	-0.201°	34.929°	Human settlement, recreation
18	Awach Kano	1162	-0.234°	34.957°	Human settlement, subsistence agriculture
19	Ainamutua-Kibigori	1194	-0.076°	35.056°	Subsistence agriculture, livestock rearing, sugarcane farming
21	Mbogo	1266	-0.061°	35.148°	Large scale coffee and sugarcane farming
22	Ainopngetuny	1324	+0.030°	35.118°	Large scale coffee and sugarcane farming
23	Ainopisiwa	1323	-0.028°	35.118°	Large scale sugarcane farming, river sand harvesting and recreation
25	Chemwanabei	1819	+0.065°	35.188°	Human settlement, subsistence farming
26	Kapngorium at Bridge	1847	+0.054°	35.100°	Human settlement, large scale tea farming, subsistence agriculture, raw effluent discharge from tea factory
27	Kundos at bridge	1844	+0.051°	35.062°	Subsistence agriculture, tea farming, livestock rearing
30	Chebirirkut at Tinderet Dam	2190	-0.037°	35.348°	Tea farming
33	Ahero Irrigation Channel	1161	-0.172°	34.908°	Irrigated rice farming, cattle watering and recreation

### **3.5 Measurement of physico-chemical parameters**

Six parameters namely; temperature, dissolved oxygen (DO), conductivity, turbidity, pH and total dissolved solids (TDS) were measured on site at depths of about 5 – 10 cm below the water surface using a precalibrated Hydrolab YSI 610 instrument.

For the remaining 5 parameters; total suspended solids (TSS), nitrogen, phosphorous, biological oxygen demand (BOD) and chemical oxygen demand (COD), river water was sampled in triplicates (three 1-litre plastic containers). The containers were thoroughly rinsed with nitric acid followed by repeated washing with de-ionised water and thrice rinsed with sample water before collection. Samples were then placed in cooler boxes and taken to KEMFRI Kisumu laboratory where they were stored in freezers at -18 °C awaiting analysis. The method of Mackereth *et al.* (1989) was used for the determination of TSS, nitrogen, phosphorous, BOD and COD.

### **3.6 Pesticide extraction, cleanup and analysis**

The samples were removed from the deep freezer; left to thaw and the lengths and weights of the individual fish was taken.

#### **3.5.1 Extraction procedure**

Na<sub>2</sub>SO<sub>4</sub> was prepared by baking out for 16 hrs at 250 °C to remove contaminants. 20g of the fish samples were weighed in a pre-cleaned 50ml beaker which had been rinsed with acetone.

For each gram of wet sample 3g of baked-out Na<sub>2</sub>SO<sub>4</sub> was added and the sample homogenized using pestle and mortar, covered with aluminum foil and left overnight to dry. The dried samples were then transferred to soxhlet extraction thimbles and 100µl of 1 ppm of isodrin added as the internal standard. Each sample was soxhlet extracted (5 cycles per hour) in 175 ml of triple distilled Hexane: Acetone (3:1, v/v) in a 500 ml round bottomed flask. Glass pearls were added to allow smooth boiling and the extraction was done for at least 16 hours (overnight).

After extraction the apparatus were allowed to cool then rinsed three times with 1 ml of extraction solvent and the rinse added to the sample extract. 2 ml of isooctane was added as a keeper and extract evaporated with a LABCONCO rotary evaporator to 3 ml.

The keeper has a higher boiling point than the solvents (Hexane: Acetone) and prevents the extracts from volatilizing. The extracts were then transferred to labeled graduated tubes and flasks rinsed 3 times with 1 ml Hexane: Acetone. The samples were reduced to 1 ml under a gentle stream of white-spot Nitrogen.

### ***3.5.2 Clean up and fractionation of the samples***

Sample clean-up was done using alumina chromatographic column 25 cm x 1.5 cm diameter packed with 1g of activated anhydrous sodium sulphate (drying agent), followed by 15 g of deactivated alumina ( $\text{Al}_2\text{O}_3$ ) and finally another 1g layer of activated anhydrous sodium sulphate. The sample extract was then eluted through the column and the elute collected in a 250 ml round bottom flask after pre-conditioning with 15 ml HPLC grade Hexane. The soxhlet extractor was rinsed with 1 ml HPLC grade Hexane and the rinse transferred to the column as soon as the sample extract had eluted. The rinse was repeated 4 times with 1 ml HPLC grade Hexane. The column was not allowed to dry completely. A last elution was done with 165 ml of HPLC grade Hexane.

The extract was transferred into a clean pre-weighed vial and the flasks rinsed 3 times with 1 ml HPLC grade Hexane. The samples were then evaporated to 0.5 ml under a gentle stream of white spot Nitrogen. The extracts were ready for fractionation. An appropriate column was taken for  $\text{SiO}_2$  and plugged with silanised glass wool in the fume hood. 1.8g of 1.5%  $\text{SiO}_2$  was added and 1g of baked  $\text{Na}_2\text{SO}_4$  added on top. The sides of the column were ticked to allow the  $\text{SiO}_2$  to settle in the column. The column was conditioned with 4 ml of Isooctane. After the Isooctane had eluted the sample extract was added and the eluate collected in a 50 ml graduated measuring cylinder. The sample extract vial was rinsed with 1 ml Isooctane and the rinse transferred to the column after the elution of sample extract. The rinse was repeated 2 times after which elution was done with 11 ml Isooctane and all the eluate was collected in the same sample vial.

When all the Isooctane had eluted 10 ml of Isooctane: Diethylether (85:15) was added and the eluate collected in a separate sample vials. Both extracts were evaporated to 1 ml under a gentle stream of Nitrogen. The extracts were then analysed using a gas chromatograph equipped with a micro-electron capture detector (micro-ECD).

### 3.5.3 GC-ECD Analysis

Fish tissue pesticide residue analysis was done using a gas chromatograph (Agilent 6890N) equipped with an auto sampler (Agilent 7683 Series injector) and a micro-electron capture detector (Agilent  $\mu$ ECD). Detector and injector temperatures were maintained at 300 °C and 250 °C respectively. Nitrogen was used as make-up gas with a constant flow rate of 2 ml/min and Helium was used as the carrier gas. The injection volume was 1  $\mu$ l with a pulsed splitless injection mode. The following injection temperature program was applied: 90 °C (3 min), 90 °C to 200 °C (at 30 °C/ min and hold time of 15 min), 200 °C to 275 °C (at 30 °C/min and hold time of 5 min).

A high performance fused silica capillary column (dimensions; length of 30 m, internal diameter of 0.25 mm and film thickness of 0.25  $\mu$ m and phase composition; 5% Phenyl 95% Methyl polysiloxane) was used. Data processing was done using Chemstation software. The calibration curves were prepared from stock solutions containing the PCB and organochlorine pesticide standards. The concentrations were calculated by exactly weighing all stock solution additions and Isooctane. Internal standards calibration using authentic reference standards was used to normalize peak areas of the analytes in the sample extracts.

Recovery studies were carried out by spiking some samples with pesticide standards and extracting as described above. Test injections were carried out and concentration or dilution factors determined. The chosen syringe standard was then added to the sample extract, treated with sulphuric acid and top layer quantitatively transferred into sample vials. Isooctane was injected once into the GC with the normal temperature programme for organochlorine pesticide analysis.

### 3.7 Quality Assurance and Quality Control

A blank was run every fifteen samples for quality control and the sensitivity and linearity of the calibration curves was monitored daily. The sample analyses and recoveries resulted from triplicate determinations (N = 3) for each. In order to validate the analytical methodology, pesticide recovery experiments were done using a spiked representative blank and for sample extracts in accordance with Miensah *et al.*, 2015.



Limit of detection (LOD) was calculated as three times the signal-to-noise ratio. Log transformations were done during statistical analysis in order to perform parametric tests (2-way ANOVA) for the non-parametric (not normally distributed) data collected. Non-detectable (BDL) data was assigned a value of zero.

### **3.8 Statistical Analysis**

Results were considered significant at a 5% level ( $p \leq 0.05$ ). The data on fish species collected were analyzed using CANOCO for Windows Version 5, a program for multivariate statistical analysis [Ter Braak and Šmilauer, 2012]. Using Redundancy Analysis (RDA) option, sampling date and species data were added in order to show how fish species composition was affected by environmental parameters.

The mean values for the measured PCB and organochlorine pesticide residue levels were calculated using Microsoft Excel. Further statistical analysis of variance (ANOVA) was done using Statistical Package for the Social Sciences (SPSS 22) for windows.

## CHAPTER FOUR

### RESULTS

#### 4.0 The types, nature, occurrence, abundance and distribution of fish species along Nandi-Lower Nyando River sub-catchment

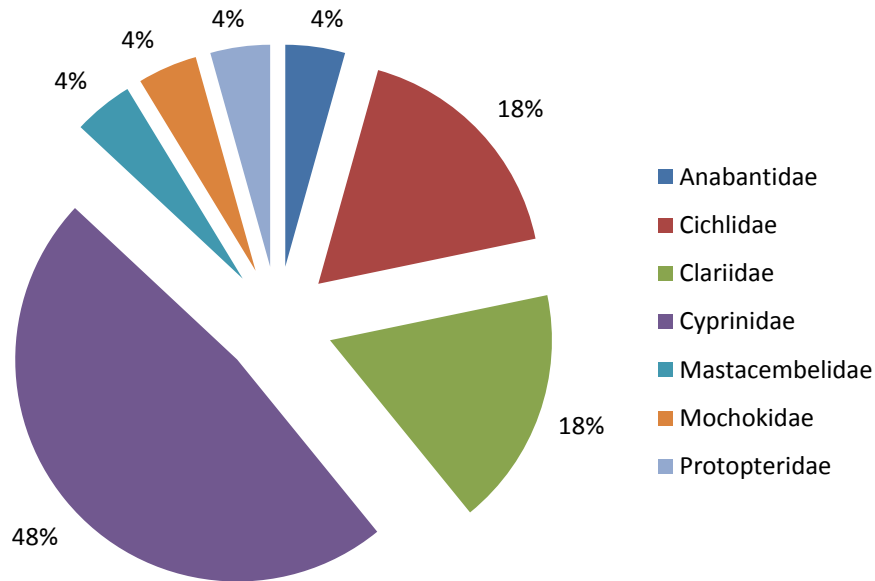
#### 4.1 Fish assemblage characteristics

##### 4.1.1 *The families of fish*

Rivers flowing in and consequent differences in pollutant load occasioned by seasonal variation (high in the rainy season) influence the water quality in Lake Victoria. These changes are normally as a result of anthropogenic forces and alter the ecological integrity of the lake [Katunzi *et al.*, 2010]. At least 300 species formed the original fish community of the lake of which a significant proportion were haplochromine cichlids with at least 14 cyprinids [Ogutu-Ohwayo, 1990].

Fish are aquatic vertebrates that vary greatly in size, length and are the main aquatic organisms of direct value to man. Rivers and streams supply lakes with water for various uses. Nyando River, for instance, has important fisheries such as *Labeo victorinus* and *Barbus sp.* catfishes [Raburu, 2003] which specifically migrate up the river to breed and return to Lake Victoria to feed. Hence the Nyando River is important in biodiversity conservation since some of the species that have been depleted in Lake Victoria still survive in it [Balirwa, 1998]. 23 fish species from 5 orders and 7 families were recorded in the Nandi-Lower Nyando River sub-catchment (Tables 4). The fish collected were dominated by family Cyprinidae which were mainly species of *Barbus altianalis* that showed the highest occurrence, followed by *Labeo victorinus* and *Barbus nyanzae* respectively.

The family Cyprinidae had the majority of the fish species (48%) and was followed by the other six families; clariidae (18%), cichlidae (18%), mochokidae (4%), protopteridae (4%), mastacembelidae (4%) and anabantidae (4%) (Figure 2).



**Figure 2: Fish Families of Lower Nyando**

#### **4.1.2 The orders of fish**

Nandi-Lower Nyando River sub-catchment has 5 orders and 23 species of fish namely Perciformes, Siluriformes, Cypriniformes, Lipidosirenformes and Synbranchiformes

##### **4.1.2.1 Perciformes**

Perciformes, which means perch-like, are considered a dominant order of vertebrates, with over 10,000 species, most of which are marine fishes and some that inhabit freshwater rivers and lakes. They are classified as ray-finned fish and are characterised by diversity in habitat, feeding and breeding behaviour.

The habitats found across the perch-like fish distribution range are brackish waters, lakes, estuaries, rivers, streams, shallow seas and the open ocean.

In Nandi-Lower Nyando River sub-catchment, Perciformes belonged to the families Anabantidae (1 species; *Ctenopoma muriei*) and Cichlidae (4 species; *Oreochromis leucostictus*, *Oreochromis niloticus*, *Haplochromis Sp.* and *Pseudocrenilabrus multicolor*) (Table 2).

#### **4.1.2.2 *Siluriformes***

Siluriformes are a rich order of fish, with over 2,800 species commonly known as catfish for their prominent barbels that resemble a cat's whiskers. They are widely distributed geographically, on every continent except Antarctica, and elicit special interest in biogeography studies. Mostly found in fresh waters, they are nocturnal and are adapted to live in deep waters.

Catfish are of economic value as many of the larger species, reported to grow upto 300 kg in weight, are farmed or fished for human consumption. Their habitat ranges from lakes and ponds to wetlands and rivers.

In Nandi-Lower Nyando River sub-catchment, Siluriformes found belonged to the families Clariidae (4 species; *Clarias alluaudi*, *Clarias gariepinus*, *Clarias liocephalus* and *Xenoclarias eupogon*) and Mochokidae (1 species; *Synodontis victoriae*) (Table 2).

#### **4.1.2.3 *Cypriniformes***

The order Cypriniformes contains 6 families and more than 3,250 species including the carps, minnows, loaches and relatives. Two-thirds of the order's diversity is constituted by the family *Cyprinidae* (carps and minnows). Few species in this order are found in marine waters with most species inhabiting fresh waters.

In Nandi-Lower Nyando River sub-catchment, Cypriniformes found belonged to the family Cyprinidae and constituted 11 species; *Barbus altianalis*, *Barbus apleurogramma*, *Barbus carstino*, *Barbus cercops*, *Barbus jacksoni*, *Barbus kerstenii*, *Barbus neumayeri*, *Barbus nyanzae*, *Barbus paludinosus*, *Barbus Sp* and *Labeo victorianus* (Table 2).

#### **4.1.2.4 *Lepidosireniformes***

Lepidosireniformes are freshwater fish commonly known as lungfish due to their ability to breathe air. This enables adult species to survive the dry season by aestivating and burrowing into mud as is the case for African and South American lungfish.

In Nandi-Lower Nyando River sub-catchment, Lepidosireniformes found belonged to the family Protopteridae and constituted 1 species; *Protopterus aethiopicus* (Table 2).

#### **4.1.2.5 *Synbranchiformes***

Synbranchiformes are an order of eel-like fish that predominantly inhabit fresh waters with only three species found in marine waters. They are widely distributed geographically with as many as three families: Synbranchidae – found mostly in Central and South America, Mastacembelidae – found mostly in Asia and Chaudhuriidae - found mostly in India. Their diet includes benthic invertebrates and fishes.

In Nandi-Lower Nyando River sub-catchment, Synbranchiformes found belonged to the family Mastacembelidae and constituted 1 species; *Aethiomastacembelus frenatus* (Table 2).

#### **4.1.3 *Fish species of Lower Nyando***

Nandi-Lower Nyando River sub-catchment has 23 species (Table 2). A number of rare occurrences of some fish were observed comprising 11 species; *Aethiomastersembelus fretanus*, *Barbus apleurogramma*, *Barbus carstino*, *Barbus paludinosus*, *Barbus Sp.*, *Ctenopoma murei*, *Haps Sp.*, *Protopterus aethiopicus*, *Pseudocrenilabrus multicolour*, *Synodontis victoriae* and *Xenoclarias eupogon* all recorded in small numbers therefore in this section only 12 abundant species have been discussed.

**Table 2: Average weights, lengths and habitats of fish species**

Species Name	Common Name	Habitat	Status	Average fresh body weight $\pm$ SD (g)	Average fresh body length $\pm$ SD (cm)
<b>Perciformes</b>					
<b>Anabantidae</b>					
<i>Ctenopoma murei</i>	Sia	Benthopelagic	Native	2.2 $\pm$ 0.40	5.3 $\pm$ 0.83
<b>cichlidae</b>					
<i>Oreochromis leucostictus</i>	Blue spotted tilapia	Benthopelagic	Introduced	12.1 $\pm$ 1.66	7.5 $\pm$ 0.65
<i>Oreochromis niloticus</i>	Ngege	Benthopelagic	Introduced	61.9 $\pm$ 3.65	11.9 $\pm$ 1.24
<i>Haplochromis Sp.</i>		Benthopelagic	Endemic	1.2 $\pm$ 0.07	4.1 $\pm$ 0.16
<i>Pseudocrenilabrus multicolour</i>		Demersal	Native	2.7 $\pm$ 0.32	5.0 $\pm$ 0.35
<b>Siluriformes</b>					
<b>clariidae</b>					
<i>Clarius alluaudi</i>	Oludhe	Demersal	Native	17.1 $\pm$ 2.29	11.9 $\pm$ 2.59
<i>Clarias gariepinus</i>	Mumi	Benthopelagic	Native	293.0 $\pm$ 6.36	27.9 $\pm$ 1.27
<i>Clarias liocephalus</i>	Nduri	Demersal	Native	21.5 $\pm$ 2.88	14.7 $\pm$ 1.03
<i>Xenoclarias eupogon</i>	Lake Victoria deepwater catfish	Demersal	Native	1.5 $\pm$ 0.13	5.7 $\pm$ 0.98
<b>mochokidae</b>					
<i>Synodontis victoriae</i>	Okoko	Benthopelagic	Native	22.4 $\pm$ 1.44	13.2 $\pm$ 1.55
<b>Cypriniformes</b>					
<b>cyprinidae</b>					
<i>Barbus altianalis</i>	Adel	Benthopelagic	Native	29.3 $\pm$ 2.41	11.0 $\pm$ 1.46
<i>Barbus apleurogramma</i>	Adel	Benthopelagic	Native	1.2 $\pm$ 0.23	4.4 $\pm$ 1.15
<i>Barbus carstino</i>		Benthopelagic	Native	0.4 $\pm$ 0.09	3.6 $\pm$ 1.17
<i>Barbus cercops</i>	Luambwa barb	Benthopelagic	Native	0.4 $\pm$ 0.08	3.6 $\pm$ 1.17
<i>Barbus jacksoni</i>	Jackson's barb	Benthopelagic	Native	7.4 $\pm$ 1.46	8.9 $\pm$ 1.38
<i>Barbus kerstenii</i>	Kersten's barb	Benthopelagic	Native	1.6 $\pm$ 0.56	5.4 $\pm$ 0.58
<i>Barbus neumayeri</i>	Neumayer's barb	Benthopelagic	Native	11.7 $\pm$ 1.06	9.6 $\pm$ 1.17

Species Name	Common Name	Habitat	Status	Average fresh body weight $\pm$ SD (g)	Average fresh body length $\pm$ SD (cm)
<i>Barbus nyanzae</i>	Adel	Benthopelagic	Native	3.9 $\pm$ 1.06	7.3 $\pm$ 1.28
<i>Barbus paludinosus</i>	Straightfin barb	Benthopelagic	Native	4.3 $\pm$ 0.95	7.6 $\pm$ 0.76
<i>Barbus Sp.</i>		Benthopelagic	Native	8.4 $\pm$ 2.02	8.4 $\pm$ 1.79
<i>Labeo victorianus</i>	Ningu	Benthopelagic	Native	54.3 $\pm$ 2.02	15.6 $\pm$ 1.24
<b>Lepidosireniformes</b>					
<b>protopteridae</b>					
<i>Protopterus aethiopicus</i>	Kamongo	Demersal	Native	329.8 $\pm$ 4.61	43.3 $\pm$ 2.81
<b>Synbranchiformes</b>					
<b>mastacembelidae</b>					
<i>Aethiomastersebelus fretanus</i>	Okunga	Demersal	Native	15.1 $\pm$ 2.70	18.6 $\pm$ 5.11

#### 4.1.3.1 *Barbus altianalis*

*Barbus altianalis*, of the family Cyprinidae, is a ray-finned fish species mostly inhabiting fresh waters such as those of Lake Victoria [De Vos and Thys, 1990b]. This species is omnivorous with juveniles mostly feeding on plant material and adults feeding on other fishes. It is a large fish species that can grow upto 90 cm in length. This makes it particularly important for breeding in ponds for human consumption and recreation [Robins *et al.*, 1991]. It has wide geographic distribution due to its ecological tolerance but takes long to mature to its full size [Eccles, 1992] when grown in fisheries for food. Despite its high tolerance to the changing environmental conditions, it is known to be temporarily affected by increasing water turbidity.

In Nandi-Lower Nyando River sub-catchment, the largest *Barbus altianalis* was found at site 15 (Nyando at Ogilo) in July. It weighed 253.0 g with a length of 21.9 cm (Table 2a, Appendix II) while the temperature at site 15 in July was  $25.45 \pm 0.07$  °C and the pH was  $7.90 \pm 0.04$  (Table 3).

#### 4.1.3.2 *Barbus cercorps*

*Barbus cercorps* is a ray-finned freshwater fish mostly found in rivers of Nyanza Province in Kenya [De Vos and Thys, 1990b] and has a maximum recorded length of 7.0 cm [De Vos and Thys, 1990b].

In Nandi-Lower Nyando River sub-catchment, the largest *Barbus cercorps* was found at site 22 (Ainopngetuny) in September. It weighed 5.2 g with a length of 4.5 cm (Table 2b, Appendix II) while the temperature at site 22 in September was  $18.65 \pm 0.07$  °C and the pH was  $7.95 \pm 0.02$  (Table 4).

#### 4.1.3.3 *Barbus jacksoni*

*Barbus jacksoni*, of the family cyprinidae, is a ray-finned fish species that can grow up to 11.6 cm in length. It survives best at pH range of 6.8 - 7.8 and temperatures of 22 °C – 28 °C [Baensch and Riehl, 1995].

In Nandi-Lower Nyando River sub-catchment, the largest *Barbus jacksoni* was found at site 17 (Nyando at Dykes) in September. It weighed 14.6 g with a length of 11.1 cm (Table 2b, Appendix II) while the temperature at site 17 in September was  $21.60 \pm 0.01$  °C and the pH was  $7.62 \pm 0.04$  (Table 4).



#### **4.1.3.4 *Barbus kerstenii***

*Barbus kerstenii* is a ray-finned fish found in mountain streams and along vegetated fringes of large rivers [Skelton, 1993]. Presence of a brilliant orange spot on the operculum is characteristic for this species. It has a maximum length of 9.0 cm, survives at pH range of 6.5 - 7.5 and temperatures of 23 °C – 26 °C [Baensch and Riehl, 1991].

In Nandi-Lower Nyando River sub-catchment, the largest *Barbus kerstenii* was found at site 21 (Mbogo) in December. It weighed 2.9 g with a length of 6.6 cm (Table 2c, Appendix II) while the temperature at site 21 in December was 21.10±0.01 °C and the pH was 7.47±0.02 (Table 5).

#### **4.1.3.5 *Barbus neumayeri***

*Barbus neumayeri*, of the family cyprinidae, is a ray-finned freshwater fish that can grow up to 11.8 cm in length and survives at temperatures of 23 °C – 27 °C [Baensch and Riehl, 1991].

In Nandi-Lower Nyando River sub-catchment, the largest *Barbus neumayeri* was found at site 18 (Awach Kano) in September. It weighed 197.4 g with a length of 24.4 cm (Table 2b, Appendix II) while the temperature at site 18 in September was 25.40±0.01 °C and the pH was 7.72±0.01 (Table 4).

#### **4.1.3.6 *Barbus nyanzae***

*Barbus nyanzae*, of the family Cyprinidae, is a ray-finned fish species that can grow upto 7.0 cm in length, survives best at pH range of 6.5 - 7.5 and temperatures of between 18 °C – 28 °C [Baensch and Riehl, 1997].

In Nandi-Lower Nyando River sub-catchment, the largest *Barbus nyanzae* was found at site 16 (Nyando at Ahero Bridge) in July. It weighed 60.7 g with a length of 4.9 cm (Table 2a, Appendix II) while the temperature at site 16 in July was 25.30±0.42 °C and the pH was 7.72±0.18 (Table 3).

#### **4.1.3.7 *Clarias alluaudi***

*Clarias alluaudi* is a ray-finned fish in the family clariidae (air breathing catfishes) found mainly in marginal water-lily and papyrus swamps. It has a maximum length of 35.0 cm [Eccles, 1992] and normally feeds on insects [Teugels, 1986].

In Nandi-Lower Nyando River sub-catchment, the largest *Clarias alluaudi* was found at site 17 (Nyando at Dykes) in September. It weighed 23.0 g with a length of 14.7 cm (Table 2b, Appendix II) while the temperature at site 17 in September was  $21.60 \pm 0.01$  °C and the pH was  $7.62 \pm 0.04$  (Table 4).

#### **4.1.3.8 *Clarias gariiepinus***

*Clarias gariiepinus*, of the family clariidae, is a ray-finned fish species that can grow up to 170 cm in length [IGFA, 2001], 60.0 kg in weight [Robins *et al.*, 1991] and 15 years in age [Weyl and Booth, 2008]. This species survives at pH range of 6.5 - 8.0 [Riede, 2004], depth range 0 - 80 m [Witte and de Winter, 1995] and temperatures of 8 °C – 35 °C [De Moor and Bruton, 1988]. It is omnivorous, mostly feeding on prey such as insects [De Moor and Bruton, 1988] and also migrates to spawn in streams [Witte and de Winter, 1995]. This species is able to survive harsh environmental conditions due to its ability to breathe under such conditions through an accessory breathing organ.

In Nandi-Lower Nyando River sub-catchment, the largest *Clarias gariiepinus* was found at site 21 (Mbogo) in July. It weighed 2.5 kg with a length of 73.2 cm (Table 2a, Appendix II) while the temperature at site 21 in July was  $20.45 \pm 0.07$  °C and the pH was  $7.89 \pm 0.01$  (Table 3).

#### **4.1.3.9 *Clarias liocephalus***

*Clarias liocephalus*, of the family clariidae, is a ray-finned fish species that can grow up to 32.0 cm in length [Teugels, 1986] and survives at temperatures of 22 °C – 28 °C [Baensch and Riehl, 1995]. It is an omnivorous species found mostly in swamps.

In Nandi-Lower Nyando River sub-catchment, the largest *Clarias liocephalus* was found at site 22 (Ainopngetuny) in July. It weighed 59.7 g with a length of 20.0 cm (Table 2a, Appendix II) while the temperature at site 22 in July was  $20.95 \pm 0.07$  °C and the pH was  $7.76 \pm 0.02$  (Table 3).

#### **4.1.3.10 *Oreochromis niloticus***

*Oreochromis niloticus*, of the family cichlidae, is a ray-finned fish species that can grow up to 60.0 cm in length [Eccles, 1992], 4.3 kg in weight [IGFA, 2001] and survives at temperatures of 14 °C – 33 °C [Philippart and Ruwet, 1982]. It is mostly found in freshwater habitats like rivers, lakes and irrigation channels [Bailey, 1994].

In Nandi-Lower Nyando River sub-catchment, the largest *Oreochromis niloticus* was found at site 16 (Nyando at Ahero Bridge) in December. It weighed 128.6 g with a length of 18.5 cm (Table 2c, Appendix II) while the temperature at site 16 in December was  $22.15 \pm 0.07$  °C and the pH was  $7.29 \pm 0.02$  (Table 5).

#### **4.1.3.11 *Oreochromis leocostictus***

*Oreochromis leocostictus*, of the family cichlidae, is a ray-finned freshwater fish that occupies inshore zones and is common in lagoons [Lowe-McConnell, 1982], can grow up to 36.3 cm in length [Welcomme, 1967] and survives at temperatures of 26 °C – 28 °C [Baensch and Riehl, 1985]. It can tolerate considerable deoxygenation and warm temperatures [Baensch and Riehl, 1985] and is known to occur at 38.0 °C [Trewavas, 1983].

In Nandi-Lower Nyando River sub-catchment, the largest *Oreochromis leocostictus* was found at site 33 (Ahero Irrigation Channel) in September. It weighed 12.7 g with a length of 7.3 cm (Table 2b, Appendix II) while the temperature at site 33 in September was  $32.00 \pm 0.01$  °C and the pH was  $6.88 \pm 0.10$  (Table 4).

#### **4.1.3.12 *Labeo victorianus***

*Labeo victorianus* is a ray-finned freshwater fish that is endemic to the Lake Victoria drainage [Seegers *et al.*, 2003]. It has a maximum length of 41.0 cm [van Oijen, 1995]. It is known to ascend both large rivers and streams during floods and spawn in floodwater pools or inundated grasses at margins of rivers [Fryer and Whitehead, 1959].

In Nandi-Lower Nyando River sub-catchment, the largest *Labeo victorianus* was found at site 18 (Awach Kano) in September. It weighed 118.3 g with a length of 17.0 cm (Table 2b, Appendix II) while the temperature at site 18 in September was  $25.40 \pm 0.01$  °C and the pH was  $7.72 \pm 0.01$  (Table 4).

**Table 3: Physico-chemical parameters in Lower Nyando in July 2011**

PARAMETER / SITE	TEMP (°c)	CONDUCTIVIT Y (µS/cm)	DO (mg/L)	TURBIDITY (NTU)	pH	TDS (mg/L)	TSS (mg/L)	NITRATES (µgN/L)	PHOSPHATES (µgP/L)	BOD (mg/L )	COD (mg/L )
15	25.45±0.07	353.0±2.83	4.63±0.72	232.60±2.12	7.90±0.04	260.5±0.70	135.0±0.01	75.69±32.05	125.00±47.29	17.0	56.0
16	25.30±0.42	360.5±36.06	7.45±0.33	244.05±21.28	7.72±0.18	245.0±0.01	145.0±0.01	38.60±5.60	91.25±.043	16.0	48.0
17	26.40±0.14	327.0±12.73	6.72±0.75	630.65±35.74	7.99±0.23	272.5±3.53	263.0±9.89	34.06±1.79	95.00±0.43	15.0	48.0
18	27.00±0.01	110.0±4.24	7.13±0.54	136.25±0.49	8.00±0.07	140.0±0.01	58.0±4.24	15.46±2.48	11.25±1.32	6.0	20.0
19	20.80±0.01	241.0±0.01	7.69±0.01	198.15±0.49	7.85±0.07	160.0±0.01	90.0±0.01	27.31±9.39	30.63±0.44	4.0	15.0
21	20.45±0.07	275.5±0.70	7.86±0.07	118.45±0.07	7.89±0.01	164.5±0.70	47.5±3.53	14.87±2.31	43.75±0.43	1.0	8.0
22	20.95±0.07	212.0±0.01	6.92±0.35	217.60±7.49	7.76±0.02	155.0±0.01	115.0±0.01	76.27±13.83	28.75±3.97	1.0	8.0
23	23.60±0.14	321.5±0.70	7.95±0.04	135.25±3.60	8.24±0.03	214.5±0.70	100.0±0.01	25.34±8.23	30.00±2.20	7.0	8.0
25	18.60±0.14	156.5±10.60	6.51±0.35	285.25±42.35	7.34±0.01	150.0±0.01	125.0±0.01	33.83±1.16	11.88±0.44	6.0	16.0
26	19.00±0.01	83.0±0.01	7.94±0.01	180.30±7.77	7.44±0.26	110.5±0.70	75.0±0.01	54.18±9.22	4.06±0.01	7.0	16.0
27	19.75±0.35	62.5±0.70	7.02±0.01	116.00±0.01	7.33±0.14	116.5±2.12	45.0±0.01	32.55±1.98	2.19±0.88	5.0	16.0
30	17.60±0.01	64.5±0.70	10.90±0.31	15.05±0.07	6.81±0.28	70.50±0.70	10.5±0.70	257.32±9.83	1.88±0.44	8.0	32.0
33	23.45±0.07	439.5±7.77	5.90±0.29	351.65±26.23	7.25±0.07	81.50±2.12	140.5±0.70	21.62±2.65	4.07±0.88	9.0	32.0

**Table 4: Physico-chemical parameters in Lower Nyando in September 2011**

PARAMETER / SITE	TEMP (°c)	CONDUCTIVITY (µS/cm)	DO (mg/L)	TURBIDITY (NTU)	pH	TDS (mg/L)	TSS (mg/L)	NITRATES (µgN/L)	PHOSPHATES (µgP/L)	BOD (mg/L)	COD (mg/L)
15	21.50±0.01	155.5±0.70	7.76±0.07	721.35±1.34	7.82±0.01	366.0±1.414	425.0±7.07	507.52±0.21	124.14±9.09	25.5±0.70	80.5 ±0.71
16	21.65±0.07	188.5±0.70	8.09±0.55	999.99±0.01	7.90±0.12	310.5±0.707	2287.0±66.46	220.55±0.64	99.15 ±4.03	28.0±1.41	81.0 ±1.41
17	21.60±0.01	164.0±2.82	7.60±0.16	999.99±0.01	7.62±0.04	330.0±7.071	755.0±7.07	690.25±1.93	148.58±5.25	22.5±3.53	81.5 ±2.12
18	25.40±0.01	89.0±0.01	7.55±0.01	447.00±0.01	7.72±0.01	410.5±20.506	115.5±0.70	338.73±0.64	89.86±7.07	8.0±1.41	33.5 ±2.12
19	20.80±0.01	241.0±0.01	7.69±0.01	198.15±0.49	7.85±0.07	245.5±0.707	365.0±28.28	462.06 ±9.92	146.29±16.15	1.5±0.70	9.0 ±1.41
21	19.10±0.01	158.0±0.01	8.10±0.02	277.50±4.80	7.71±0.07	232.5±3.536	20.0±0.01	142.67±1.50	81.29±1.01	4.0±1.41	7.5 ±0.71
22	18.65±0.07	244.5±0.70	8.40±0.01	155.65±0.91	7.95±0.02	254.5±0.707	41.5±2.12	267.52±2.78	99.86±9.09	2.0±1.41	7.0 ±1.41
23	19.21±0.14	175.0±4.24	7.90±0.07	261.75±3.18	7.77±0.01	242.0±2.828	30.0±0.01	194.03±1.28	74.86±6.06	8.0±1.41	15.0 ±1.41
25	19.60±0.01	143.5±0.70	7.73±0.14	163.10±0.99	7.65±0.01	162.5±3.536	305.0±7.07	196.76±5.57	37.00±3.02	4.5±0.70	14.0±2.82
26	20.35±0.07	62.0±0.01	7.82±0.07	165.10±11.73	7.91±0.07	308.5±4.950	60.0±0.01	467.82±6.64	22.00±10.19	8.0±1.41	19.5±2.12
27	21.15±0.07	430.0±28.28	7.12±0.14	169.65±0.35	7.31±0.14	86.0 ±1.414	15.5±0.70	154.79±4.07	25.58±1.09	6.0±1.41	16.5±0.70
30	15.55±0.07	55.0±0.01	8.27±0.04	32.25±0.35	7.80±0.01	134.5±0.707	169.5±7.77	27.36±3.00	15.57±7.07	12.0±1.41	48.5±0.70
33	32.00±0.01	216.5±3.53	5.89±0.02	386.30±15.13	6.88±0.10	315.0±0.001	77.5±3.53	182.06±0.21	209.86±41.41	13.0±1.41	49.0±1.41

**Table 5: Physico-chemical parameters in Lower Nyando in December 2011**

PARAMETER / SITE	TEMP (°c)	CONDUCTIVITY (µS/cm)	DO (mg/L)	TURBIDITY (NTU)	pH	TDS (mg/L)	TSS (mg/L)	NITRATES (µgN/L)	PHOSPHATES (µgP/L)	BOD (mg/L)	COD (mg/L)
15	20.35±0.07	152.5±0.70	7.84±0.14	950.75±2.33	7.75±0.00	347.5±3.53	382.5±3.53	98.43±1.21	148.21±6.97	16.0±1.41	47.0±1.41
16	22.15±0.07	156.0±1.41	8.00±0.13	999.99±0.01	7.29±0.02	257.0±9.89	1209.5±106.77	120.86±1.41	166.00±0.01	12.5±0.70	31.5±0.70
17	21.47±0.09	162.0±0.01	7.81±0.14	999.99±0.01	7.37±0.03	347.0±9.89	750.5±21.92	64.57±59.19	151.71±4.04	12.0±1.41	31.5±0.70
18	22.50±0.01	90.5±0.70	7.70±0.09	571.75±7.42	7.57±0.18	280.0±7.07	159.5±7.77	113.14±0.20	73.86±5.05	6.5±0.70	24.5±0.70
19	19.70±0.01	167.5±10.60	7.85±0.10	984.25±12.65	7.66±0.02	285.0±0.01	440.0±0.01	82.86±2.22	113.86±1.01	2.5±0.70	11.5±0.70
21	21.10±0.01	150.0±1.41	8.17±0.14	471.95±50.70	7.47±0.02	300.0±0.01	93.5±4.95	53.29±0.80	78.86±0.01	2.0±1.41	8.5±0.70
22	19.60±0.01	240.5±0.70	8.03±0.17	176.00±0.99	7.53±0.11	135.5±0.70	60.0±0.01	39.71±0.20	58.86±2.02	1.0±0.01	7.5±0.70
23	19.90±0.01	218.0±0.01	8.93±0.07	237.75±37.83	7.68±0.01	100.0±0.01	19.5±0.70	66.71±0.40	49.57±3.03	1.5±0.70	8.5±0.70
25	18.65±0.07	138.5±6.36	7.92±0.09	219.85±17.74	7.16±0.01	185.5±0.70	55.0±0.01	213.14±3.88	103.14±6.06	1.5±0.70	7.0±1.41
26	18.60±0.01	64.5±0.70	7.78±0.13	258.55±14.07	7.25±0.07	160.5±6.36	54.5±0.70	119.57±0.01	18.86±4.04	1.5±0.70	8.5±0.70
27	20.95±0.07	41.0±8.48	7.73±0.21	230.15±9.68	7.19±0.26	110.5±0.70	5.0±0.01	70.57±0.20	17.43±0.01	4.5±0.70	11.5±0.70
30	16.05±0.91	40.0±9.89	7.09±0.46	42.55±11.38	6.86±0.02	35.0±0.01	5.0±0.01	196.00±2.62	128.86±0.01	3.5±0.70	12.5±0.70
33	21.40±0.01	218.0±8.49	5.04±0.07	999.99±0.01	7.48±0.38	105.0±7.07	382.5±3.53	63.86±4.44	101.71±14.14	8.5±0.70	25.0±1.41

**Table 6: Physico-chemical parameters in Lower Nyando in March 2012**

PARAMETER/ SITE	TEMP (°c)	CONDUCTIVITY ( $\mu$ S/cm)	DO (mg/L)	TURBIDITY (NTU)	pH	TDS (mg/L)	TSS (mg/L)	NITRATES ( $\mu$ gN/L)	PHOSPHATES ( $\mu$ gP/L)	BOD (mg/L)	COD (mg/L)
15	26.20 $\pm$ 0.14	456.5 $\pm$ 4.95	2.50 $\pm$ 0.35	192.65 $\pm$ 3.60	8.16 $\pm$ 0.09	295.0 $\pm$ 7.07	157.5 $\pm$ 3.53	13.12 $\pm$ 3.85	480.57 $\pm$ 2.02	13.0 $\pm$ 1.41	32.5 $\pm$ 0.70
16	29.40 $\pm$ 0.01	749.0 $\pm$ 0.01	6.12 $\pm$ 0.01	144.50 $\pm$ 0.01	7.49 $\pm$ 0.01	252.5 $\pm$ 3.53	210.0 $\pm$ 7.07	48.58 $\pm$ 17.14	432.00 $\pm$ 0.01	11.0 $\pm$ 1.41	33.0 $\pm$ 1.41
17	23.65 $\pm$ 0.07	373.5 $\pm$ 0.70	7.76 $\pm$ 0.04	228.00 $\pm$ 4.38	7.76 $\pm$ 0.07	255.0 $\pm$ 7.07	142.5 $\pm$ 3.53	50.85 $\pm$ 7.50	454.14 $\pm$ 3.03	10.5 $\pm$ 0.70	31.5 $\pm$ 0.70
18	28.00 $\pm$ 0.14	135.5 $\pm$ 0.70	6.43 $\pm$ 0.09	531.25 $\pm$ 19.72	8.39 $\pm$ 0.17	215.0 $\pm$ 7.07	307.5 $\pm$ 3.53	40.09 $\pm$ 1.71	48.43 $\pm$ 1.01	3.5 $\pm$ 0.70	7.0 $\pm$ 1.41
19	23.10 $\pm$ 0.01	247.0 $\pm$ 1.41	7.92 $\pm$ 0.07	249.75 $\pm$ 69.36	7.15 $\pm$ 0.04	190.0 $\pm$ 7.07	182.5 $\pm$ 3.53	70.85 $\pm$ 32.35	94.14 $\pm$ 7.07	1.5 $\pm$ 0.70	8.5 $\pm$ 0.70
21	24.05 $\pm$ 0.21	336.0 $\pm$ 7.07	7.39 $\pm$ 0.11	16.75 $\pm$ 1.48	7.86 $\pm$ 0.07	212.5 $\pm$ 3.53	30.0 $\pm$ 7.07	11.45 $\pm$ 1.07	97.00 $\pm$ 13.13	1.5 $\pm$ 0.70	7.5 $\pm$ 0.70
22	19.75 $\pm$ 0.07	270.5 $\pm$ 3.53	8.93 $\pm$ 0.02	17.70 $\pm$ 1.41	7.66 $\pm$ 0.01	192.5 $\pm$ 3.53	20.0 $\pm$ 7.07	54.18 $\pm$ 21.64	84.86 $\pm$ 8.08	1.0 $\pm$ 0.01	7.0 $\pm$ 1.41
23	21.30 $\pm$ 0.14	352.5 $\pm$ 7.77	8.77 $\pm$ 0.07	22.25 $\pm$ 0.07	7.67 $\pm$ 0.01	255.0 $\pm$ 7.07	17.5 $\pm$ 3.53	25.85 $\pm$ 2.57	87.71 $\pm$ 2.02	2.5 $\pm$ 0.70	7.5 $\pm$ 0.70
25	17.65 $\pm$ 0.35	143.0 $\pm$ 4.24	8.14 $\pm$ 0.50	94.20 $\pm$ 0.14	8.31 $\pm$ 0.07	215.0 $\pm$ 7.07	52.5 $\pm$ 3.53	132.21 $\pm$ 33.85	37.71 $\pm$ 6.06	3.5 $\pm$ 0.70	8.5 $\pm$ 0.70
26	18.60 $\pm$ 0.14	70.5 $\pm$ 4.95	8.55 $\pm$ 0.07	83.00 $\pm$ 1.69	8.07 $\pm$ 0.08	140.0 $\pm$ 7.07	42.5 $\pm$ 3.53	149.94 $\pm$ 19.92	25.57 $\pm$ 5.05	2.0 $\pm$ 1.41	7.5 $\pm$ 0.70
27	21.15 $\pm$ 0.21	56.5 $\pm$ 4.95	7.86 $\pm$ 0.07	59.75 $\pm$ 3.60	7.28 $\pm$ 0.02	105.0 $\pm$ 7.07	37.5 $\pm$ 3.53	73.58 $\pm$ 0.21	25.57 $\pm$ 9.09	2.5 $\pm$ 0.70	7.0 $\pm$ 1.41
30	16.10 $\pm$ 0.01	42.5 $\pm$ 0.70	7.57 $\pm$ 0.02	7.20 $\pm$ 0.70	7.84 $\pm$ 0.07	95.0 $\pm$ 7.07	17.5 $\pm$ 3.53	104.48 $\pm$ 13.49	12.00 $\pm$ 4.04	4.5 $\pm$ 0.70	15.5 $\pm$ 0.70
33	27.70 $\pm$ 0.14	440.5 $\pm$ 4.95	3.40 $\pm$ 0.43	27.30 $\pm$ 2.97	8.23 $\pm$ 0.14	302.5 $\pm$ 3.53	20.0 $\pm$ 7.07	8.27 $\pm$ 3.85	46.29 $\pm$ 12.12	7.5 $\pm$ 0.70	17.0 $\pm$ 1.41

#### **4.1.4 Average weights, lengths and habitats of fish species**

The 7 families in Nandi-Lower Nyando River sub-catchment belong to the 5 orders viz perciformes (2 families), siluriformes (2 families), cypriniformes (1 family), synbranchiformes (1 family) and lepidosireniformes (1 family). *Protopterus aethiopicus* species under the order Lepidosireniformes was the longest with a mean length of  $43.3 \pm 8.83$  cm, followed by *Clarias gariepinus* species under the order Siluriformes with a mean length of  $27.9 \pm 14.27$  cm (Table 2).

The two fish species have mean weights of  $329.8 \pm 4.61$ g and  $293.0 \pm 6.36$  g and reside in demersal and benthopelagic habitants respectively and are natives in this sub-catchment (Table 4). *Barbus cercops* species under the order Cypriniformes had the smallest mean weight and length of  $0.4 \pm 0.08$ g and  $3.6 \pm 1.17$ cm respectively among the fish species collected (Table 2).

#### **4.2 Occurrence, abundance and distribution of fish species**

Occurrence and distribution of fish families in the Nandi-lower Nyando were recorded. Detailed data are presented in Tables 7-10. Sites 22, 18 and 33 had the most number of the fish species respectively. These sites have large scale sugarcane (Site 22) and irrigated rice (Site 18 and 33) farms on which pesticides are applied regularly [Abong'o *et al.*, 2014].

*Barbus altianalis* was the most dominant species, contributing to 83% and 78% of total fish catch at Site 22 in July and September 2011, respectively (Tables 7 and 8, respectively). This was followed by *Barbus cercops* (68%) and *Haplochromis Sp* (40%) at Site 18 and 33 in July respectively. *Protopterus aethiopicus* species of the family Protopteridae was only found at Site 33 contributing 1% and 4% of total fish catch in July and March respectively.

December sampling period (Table 9) had the lowest contribution of the total fish numbers collected, followed by September and July while March (Table 10) had the highest number during the sampling periods, respectively.



**Table 7: Occurrence, abundance and distribution of fish species in Nandi-Lower Nyando in July 2011**

<b>TAXON/SITE</b>	<b>15</b>	<b>16</b>	<b>17</b>	<b>18</b>	<b>19</b>	<b>21</b>	<b>22</b>	<b>23</b>	<b>25</b>	<b>26</b>	<b>27</b>	<b>30</b>	<b>33</b>
<b>Perciformes</b>													
<b>anabantidae</b>													
Ctenopoma muriei													1
<b>cichlidae</b>													
Oreochromis leucostictus	2												
Oreochromis niloticus		1	4	1									1
Haplochromis Sp.													38
Pseudocrenilabrus multicolor													24
<b>Siluriformes</b>													
<b>clariidae</b>													
Clarias alluaudi				6		1							1
Clarias gariepinus			2	3		2							
Clarias liocephalus							5	6		2			
Xenoclaris eupogon													
<b>mochokidae</b>													
Synodontis victoriae													
<b>Cypriniformes</b>													
<b>cyprinidae</b>													
Barbus altianalis	3	7	1	10		41	192	97					
Barbus apleurogramma													
Barbus carstino						2							
Barbus cercops			4	122		11							25
Barbus jacksoni				10		2							
Barbus kerstenii													4
Barbus neumayeri						2		2		13			
Barbus nyanzae	24	30	7	9									
Barbus paludinosus													
Barbus Sp.							36	37		3			
Labeo victorianus	7	17	31	25									
<b>Synbranchiformes</b>													
<b>mastacembelidae</b>													
Aethiomastacembelus frenatus		1		3									
<b>Lepidosireniformes</b>													
<b>protopteridae</b>													
Protopterus aethiopicus													1
<b>TOTAL</b>	<b>36</b>	<b>56</b>	<b>49</b>	<b>189</b>	<b>0</b>	<b>61</b>	<b>233</b>	<b>142</b>	<b>0</b>	<b>18</b>	<b>0</b>	<b>0</b>	<b>95</b>

**Table 8: Occurrence, abundance and distribution of fish species in Nandi-Lower Nyando in September 2011**

<b>TAXON/SITE</b>	<b>15</b>	<b>16</b>	<b>17</b>	<b>18</b>	<b>19</b>	<b>21</b>	<b>22</b>	<b>23</b>	<b>25</b>	<b>26</b>	<b>27</b>	<b>30</b>	<b>33</b>
<b>Perciformes</b>													
<b>anabantidae</b>													
Ctenopoma muriei													2
<b>cichlidae</b>													
Oreochromis leucostictus	1												10
Oreochromis niloticus		12	18	2									1
Haplochromis Sp.													86
Pseudocrenilabrus multicolor													19
<b>Siluriformes</b>													
<b>clariidae</b>													
Clarias alluaudi			7										1
Clarias gariepinus				11									2
Clarias liocephalus						1	1	3		2			
Xenoclaris eupogon													2
<b>mochokidae</b>													
Synodontis victoriae				7									
<b>Cypriniformes</b>													
<b>cyprinidae</b>													
Barbus altianalis	49	4	26	16		15	136	57					
Barbus apleurogramma													
Barbus carstino		4											
Barbus cercops	13		7	11		13	23						
Barbus jacksoni			2	13									
Barbus kerstenii				1									
Barbus neumayeri						1	13						
Barbus nyanzae	2		6	13									3
Barbus paludinosus													
Barbus Sp.						2		11		78	13		81
Labeo victorianus	1		2	14									
<b>Synbranchiformes</b>													
<b>mastacembelidae</b>													
Aethiomastacembelus frenatus			1	9									
<b>Lepidosireniformes</b>													
<b>protopteridae</b>													
Protopterus aethiopicus													
<b>TOTAL</b>	<b>66</b>	<b>20</b>	<b>69</b>	<b>97</b>	<b>0</b>	<b>32</b>	<b>173</b>	<b>71</b>	<b>0</b>	<b>80</b>	<b>13</b>	<b>0</b>	<b>207</b>

**Table 9: Occurrence, abundance and distribution of fish species in Nandi-Lower Nyando in December 2011**

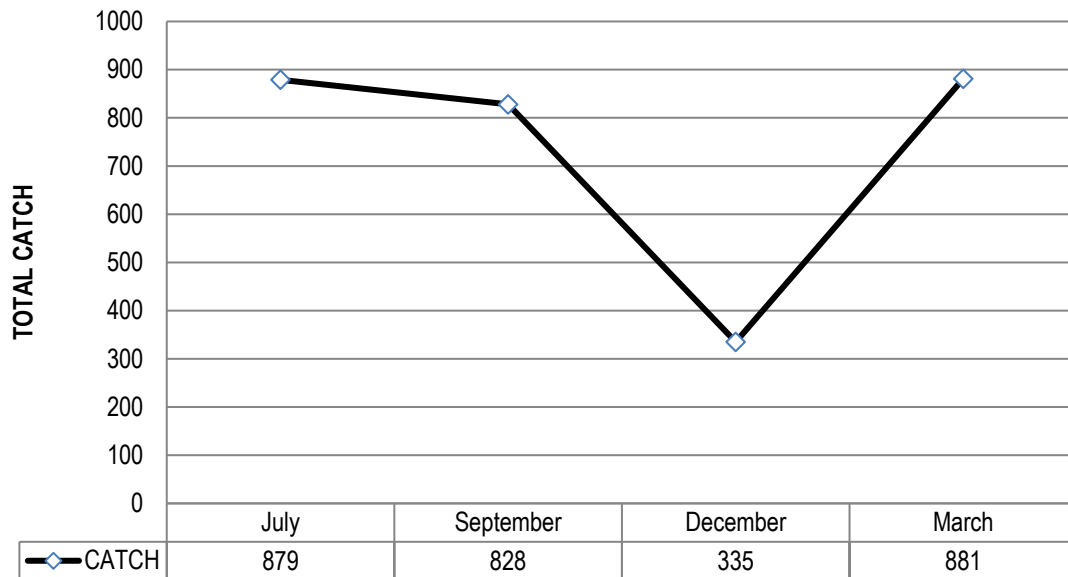
<b>TAXON/SITE</b>	<b>15</b>	<b>16</b>	<b>17</b>	<b>18</b>	<b>19</b>	<b>21</b>	<b>22</b>	<b>23</b>	<b>25</b>	<b>26</b>	<b>27</b>	<b>30</b>	<b>33</b>
<b>Perciformes</b>													
<b>anabantidae</b>													
Ctenopoma muriei													2
<b>cichlidae</b>													
Oreochromis leucostictus													8
Oreochromis niloticus	1	1											8
Haplochromis Sp.													8
Pseudocrenilabrus multicolor													12
<b>Siluriformes</b>													
<b>clariidae</b>													
Clarias alluaudi													3
Clarias gariepinus													4
Clarias liocephalus		1	3			1				1			
Xenoclarias eupogon													
<b>mochokidae</b>													
Synodontis victoriae													
<b>Cypriniformes</b>													
<b>cyprinidae</b>													
Barbus altianalis	22	7	4	11		6	11	34					
Barbus apleurogramma													8
Barbus carstino													
Barbus cercops	9		2	15									
Barbus jacksoni													
Barbus kerstenii						2							2
Barbus neumayeri				1			4	2		31	30		
Barbus nyanzae	3	4	2	11									
Barbus paludinosus													
Barbus Sp.			1	11									37
Labeo victorianus	1		5	5									
<b>Synbranchiformes</b>													
<b>mastacembelidae</b>													
Aethiomastacembelus frenatus				1									
<b>Lepidosireniformes</b>													
<b>protopteridae</b>													
Protopterus aethiopicus													
<b>TOTAL</b>	<b>36</b>	<b>13</b>	<b>17</b>	<b>55</b>	<b>0</b>	<b>9</b>	<b>15</b>	<b>36</b>	<b>0</b>	<b>32</b>	<b>30</b>	<b>0</b>	<b>92</b>

**Table 10: Occurrence, abundance and distribution of fish species in Nandi-Lower Nyando in March 2012**

<b>TAXON/SITE</b>	<b>15</b>	<b>16</b>	<b>17</b>	<b>18</b>	<b>19</b>	<b>21</b>	<b>22</b>	<b>23</b>	<b>25</b>	<b>26</b>	<b>27</b>	<b>30</b>	<b>33</b>
<b>Perciformes</b>													
<b>anabantidae</b>													
Ctenopoma muriei													2
<b>cichlidae</b>													
Oreochromis leucostictus													1
Oreochromis niloticus	7	4	3	0									
Haplochromis Sp.				4									
Pseudocrenilabrus multicolor													4
<b>Siluriformes</b>													
<b>clariidae</b>													
Clarias alluaudi													1
Clarias gariepinus	2	8	6	20				1					6
Clarias liocephalus						3	2	12		2	9		
Xenoclaris eupogon													
<b>mochokidae</b>													
Synodontis victoriae			2										
<b>Cypriniformes</b>													
<b>cyprinidae</b>													
Barbus altianalis		4	8	18			183	48					
Barbus apleurogramma			12										8
Barbus carstino													
Barbus cercops			3	8		6							
Barbus jacksoni													
Barbus kerstenii													
Barbus neumayeri							26	8		41	196		
Barbus nyanzae		3		75									
Barbus paludinosus			1	24									2
Barbus Sp.	3						1						
Labeo victorianus	6	12	34	43									
<b>Synbranchiformes</b>													
<b>mastacembelidae</b>													
Aethiomastacembelus frenatus		1		5									
<b>Lepidosireniformes</b>													
<b>protopteridae</b>													
Protopterus aethiopicus													1
<b>TOTAL</b>	<b>18</b>	<b>32</b>	<b>69</b>	<b>197</b>	<b>0</b>	<b>9</b>	<b>212</b>	<b>69</b>	<b>0</b>	<b>43</b>	<b>205</b>	<b>0</b>	<b>25</b>

#### 4.2.1 Variation of fish population during the sampling periods

A significant seasonal variation in fish assemblages was observed in July, September, December and March in Nandi-Lower Nyando catchment (Figure 3). The total population was highest in the the dry season (March) and least in the short rains season (December). July and September had relatively high fish population as was the case in March (Figure 3).



**Figure 3: Variation of fish population in July, September, December and March in Nandi-Lower Nyando sub-catchment**

#### 4.2.2 Richness of fish species

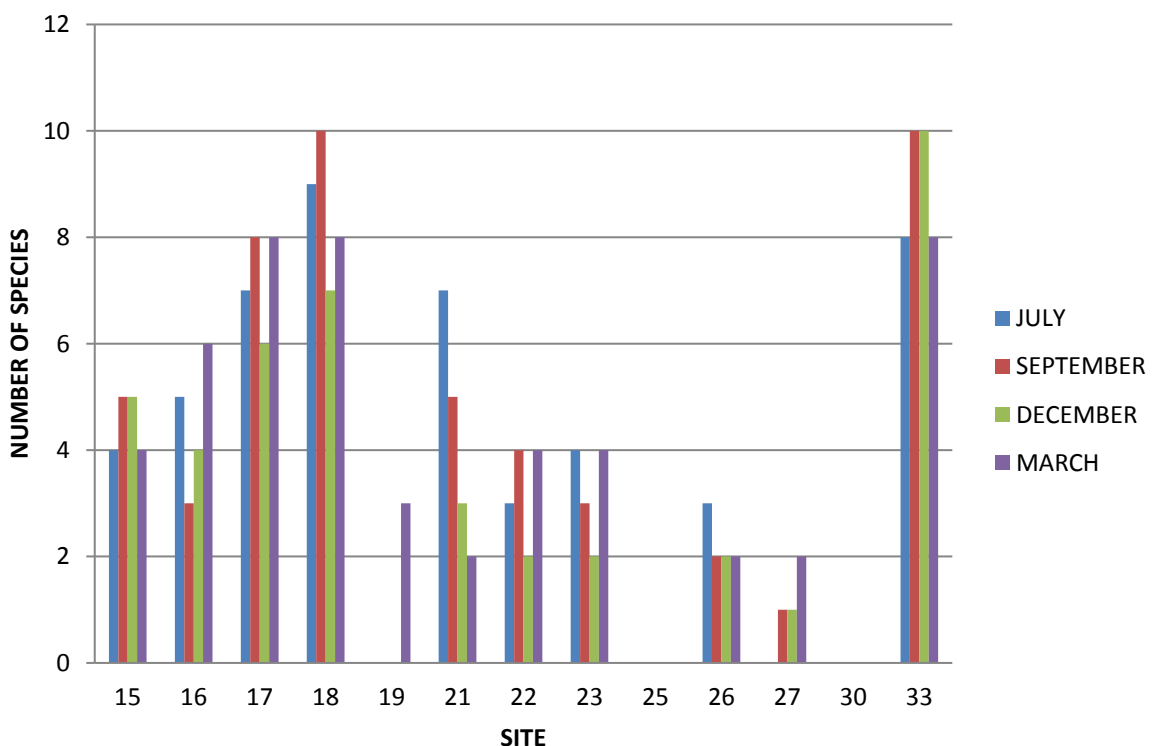
The sampling sites for this study had been marked out by the Lake Victoria Environmental Management Project (LVEMP). At sites 25 and 30 only physico-chemical parameters were measured. No fish samples were collected since there were no fish species in the two sites. Site 30 was the reference point designed to provide baseline data on natural water quality and had low water temperature in the range of  $15.5 \pm 0.071$  °C –  $17.60 \pm 0.001$  °C. Sites 25 also had low water temperatures in the range  $17.65 \pm 0.354$  °C –  $19.60 \pm 0.001$  °C (Tables 3-6).

In July 2011, site 18 (Awach Kano at Bridge) showed the highest richness of species (9) followed by site 33 (Ahero Irrigation Channel) with 8 and site 21 (Mbogo) with 7 (Figure 4). Site 17 had a variety of 6 species (Figure 4). Sites 15, 16, 22, 23 and 26 had 5 to 3 species (Table 3a, Appendix III).

In September 2011, site 18 (Awach Kano at Bridge) showed the highest richness of species (10) followed by site 33 (Ahero Irrigation Channel) with 9 and site 17 (Nyando at Dykes) with 8 (Figure 4). A fewer number of species was found in site 21 (5) compared to the number in July (Figure 4). Sites 15, 16, 22, 23 and 26 had a variety of 5 to 2 species (Figure 4). Site 27 had only one species of fish (Table 3b, Appendix III).

In December 2011, site 33 (Ahero Irrigation Channel) showed the highest richness of species (10) followed by site 18 (Awach Kano at Bridge) with 7 and site 17 (Nyando at Dykes) with 6 (Figure 4). Sites 15, 16, 21, 22, 23 and 26 had a variety of 5 to 2 species (Figure 4). Site 27 had only one species of fish (Table 3c, Appendix III).

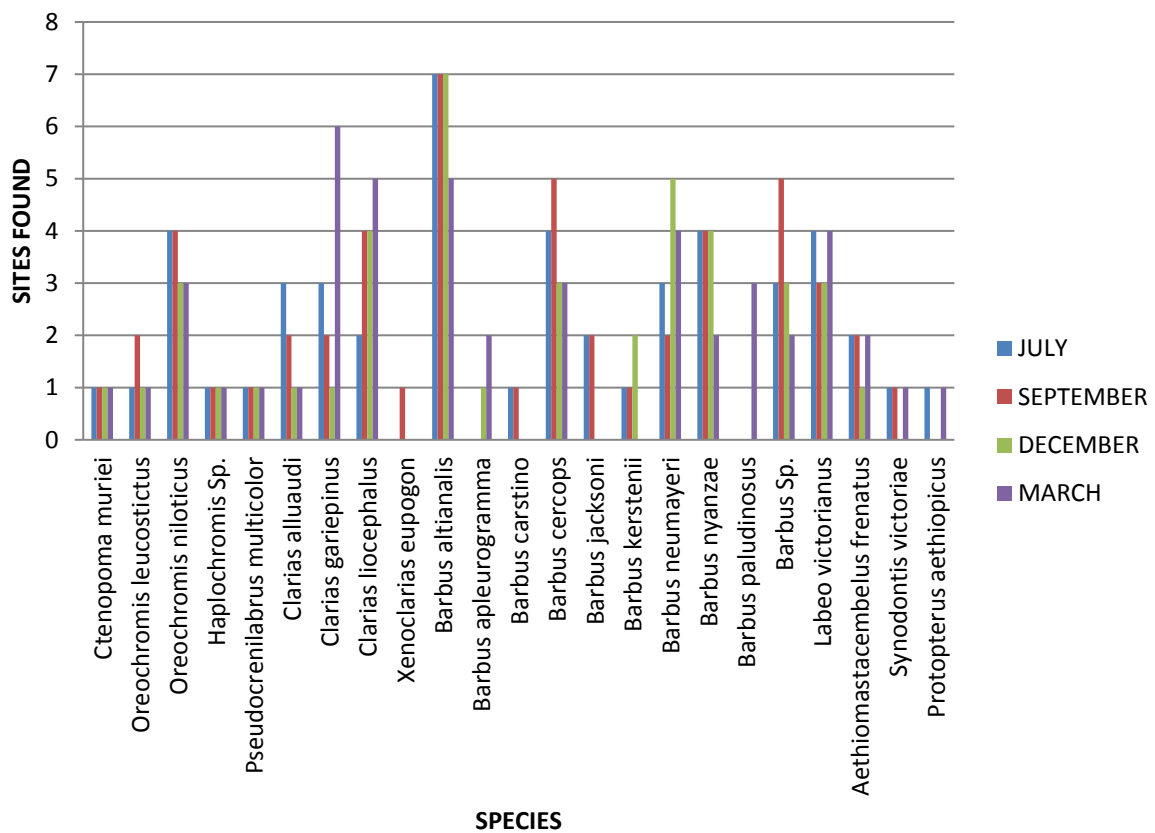
In March, sites 17, 18 and 33 showed the highest richness of species (8). Site 16 had the most number of species (6) compared to the previous sampling periods. Sites 15, 19, 21, 22, 23 and 26 had a variety of 5 to 2 species (Figure 4). Site 27 also had 2 species of fish which was the highest number collected in the four sampling periods (Table 3d, Appendix III).



**Figure 4: Species richness in July, September, December and March from Nandi-Lower Nyando catchment**

Sites 18 and 33 showed the highest variety of species during the study with both recording 10 species in September (Figure 4). In general, these sites are at the lower reaches of the river where there is regular use of pesticides in rice irrigation [Abong'o *et al.*, 2015b]. Sites 26 and 27 had the least variety of species. This distribution of fish species can be explained by variations in physical and chemical parameters such as temperatures, nitrate and phosphate residue levels along the river drainage basin (Tables 3-6).

#### 4.2.3 Density and Distribution of fish species in Nandi-Lower Nyando River sub-catchment



**Figure 5: Density and Distribution of fish species in Nandi-Lower Nyando sub-catchment for the four sampling periods**

None of the sites had all the 23 fish species recorded. The ecologically tolerant *Barbus altianalis* was the most dominant species and was found in 7 of the 13 sampling sites (Figure 5). This was followed by *Oreochromis niloticus* and *Barbus nyanzae* (Figure 5).

Three species, *Oreochromis leucostictus* and *Oreochromis niloticus* which had been introduced into Lake Victoria and *Haplochromis Sp.* which is endemic were found in the Nandi-Lower Nyando, an indication that the river is important in biodiversity conservation since some of the species that have been depleted in Lake Victoria still survive in it [Balirwa, 1998].

Most cyprinids e.g *Barbus cercops*, *Barbus neumayeri*, *Barbus nyanzae*, *Barbus Sp.* and *Labeo victorianus* were present in at least 4 sites (Tables 3a-3d, Appendix III). The highly adaptive and resilient cichlids were dominant in site 33 with some like *Haplochromis Sp.* and *Pseudocrenilabrus multicolor* present in this sampling site only.

A number of rare occurrences were also observed with some species such as *Xenoclaris eupogon*, *Barbus apleurogramma*, *Barbus carstino*, *Barbus paludinosus* and *Protopterus aethiopicus* occurring once and were recorded in small numbers. A past study of benthic macroinvertebrates found the rare order Neuroptera in the Nyando River catchment at Site 21 contributing 0.3% and 0.7% of the total organisms [Abong'o *et al.*, 2015b] in 2005 and 2006 sampling periods respectively.

#### **4.2.4 Diversity indices at sampling sites**

Margalef Index was used to estimate the species richness due to its robustness [Naigaga *et al.*, 2011] whereas Shannon Diversity Index and the Simpson index were used to estimate species diversity

Margalef index [Margalef, 1968] was used to measure species richness by using the following formula:

$$d = (S/1) = \log (N);$$

where; S = total species

N = total individuals.

Shannon Weiner diversity index incorporates the number of species and the corresponding distribution of individuals within species [Shannon and Weaver, 1963]. The Shannon Weiner diversity was calculated by following formula:



$$H = - \sum_{i=1}^n \left( \frac{n_i}{N} \log_2 \left( \frac{n_i}{N} \right) \right)$$

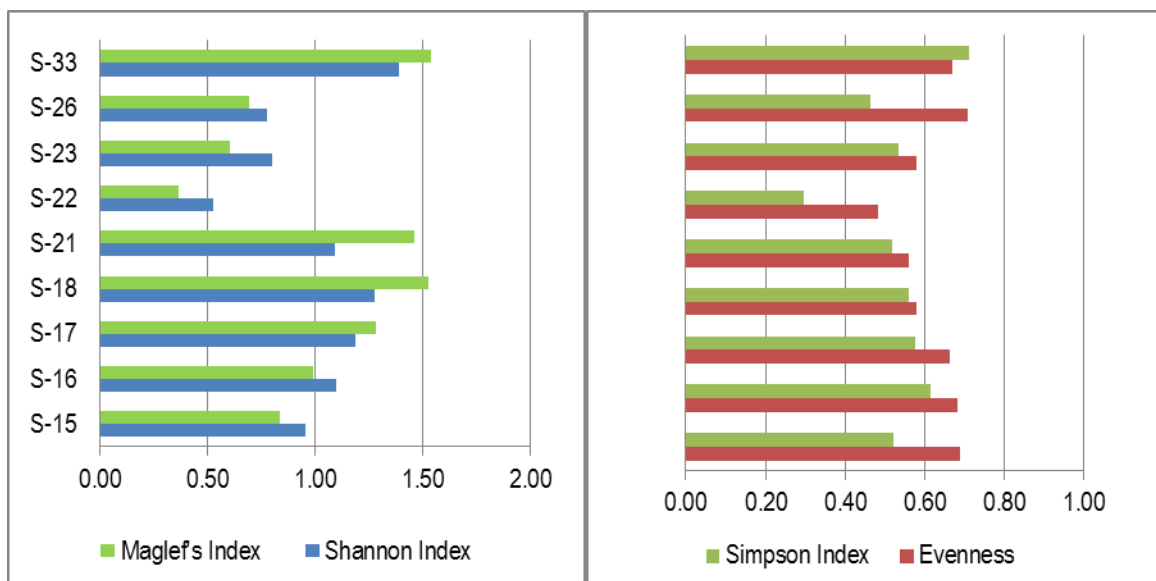
Where: H = Shannon-Wiener index of diversity

$n_i$  = Total No. of individuals of a species

N = Total No. of individuals of all species

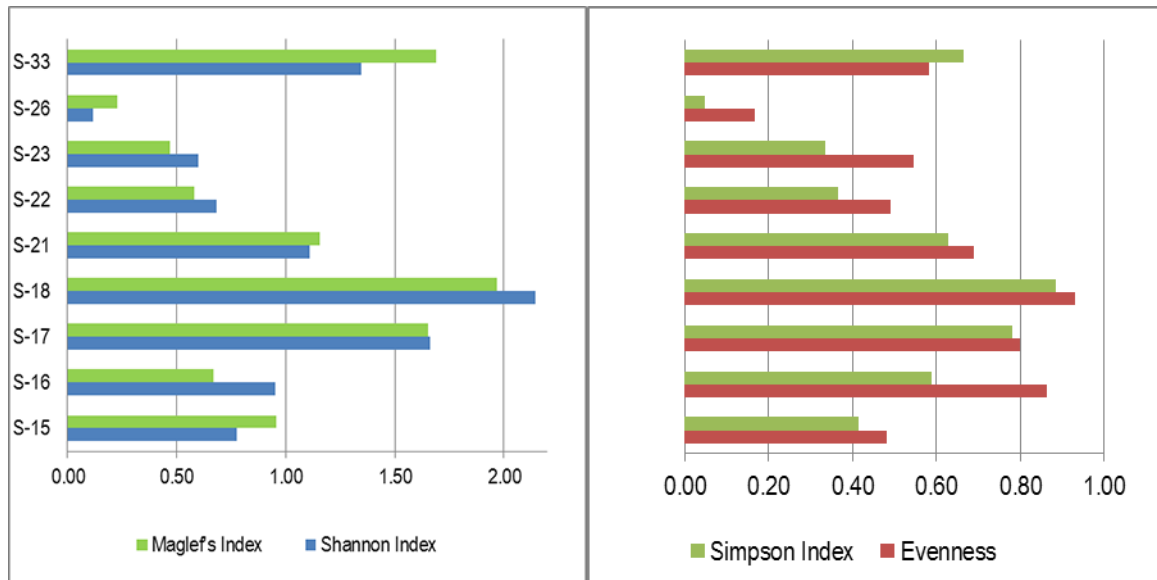
The Simpson's species dominance index measures the probability that two individuals randomly selected from a population will be of the same species [Seaby and Henderson, 2006]. The index is said to be very robust as it depicts variance of species abundance and distribution [Magurran, 2004].

Margalef species richness and Shannon diversity index varied significantly among sampling locations ( $p \leq 0.05$ ) as shown in Table 11. In July, both indices were highest at Ahero Irrigation Channel (Site 33) and lowest at Ainopngetuny (Site 22) (Figure 6). Shannon diversity index was highest at Ahero Irrigation Channel and lowest at Ainopngetuny, while the Margalef richness index was highest at Ahero Irrigation Channel and lowest at Ainopisiwa (Site 22) (Figure 6).



**Figure 6: Diversity Indices in July 2011**

In September, both indices were highest at Awach Kano (Site 18) and lowest at Kapngorium at Bridge (Site 26) (Figure 7). Shannon diversity index was highest at Awach Kano and lowest at Ainopngetuny, while the Margalef richness index was highest at Awach Kano and lowest at Ainopngetuny (Figure 7).



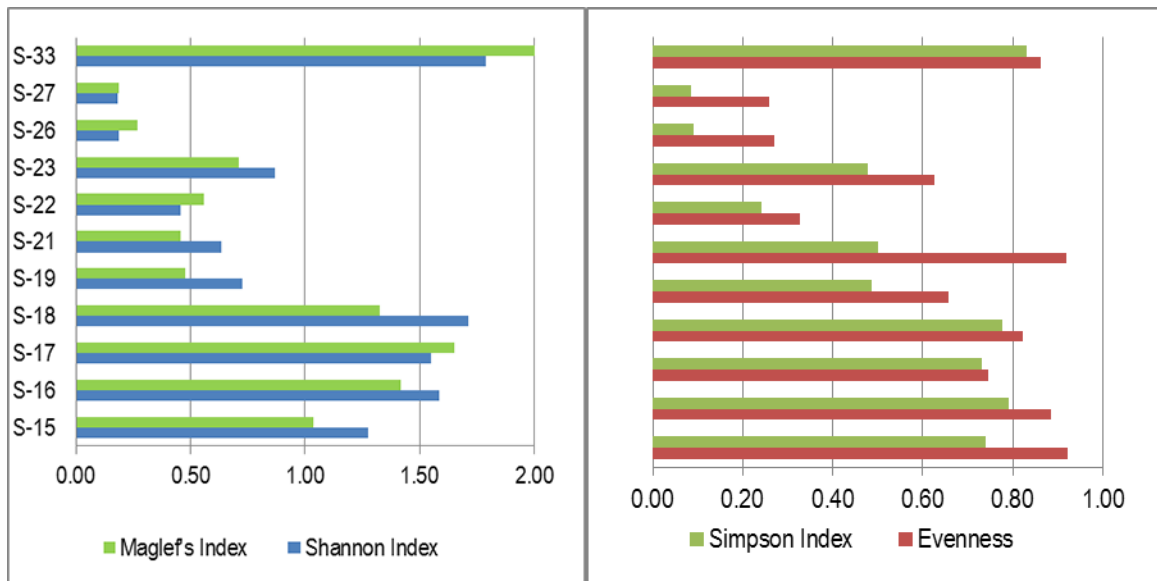
**Figure 7: Diversity Indices in September 2011**

In December, the two indices were highest at Ahero Irrigation Channel and lowest at Kapngorium at Bridge (Figure 8).



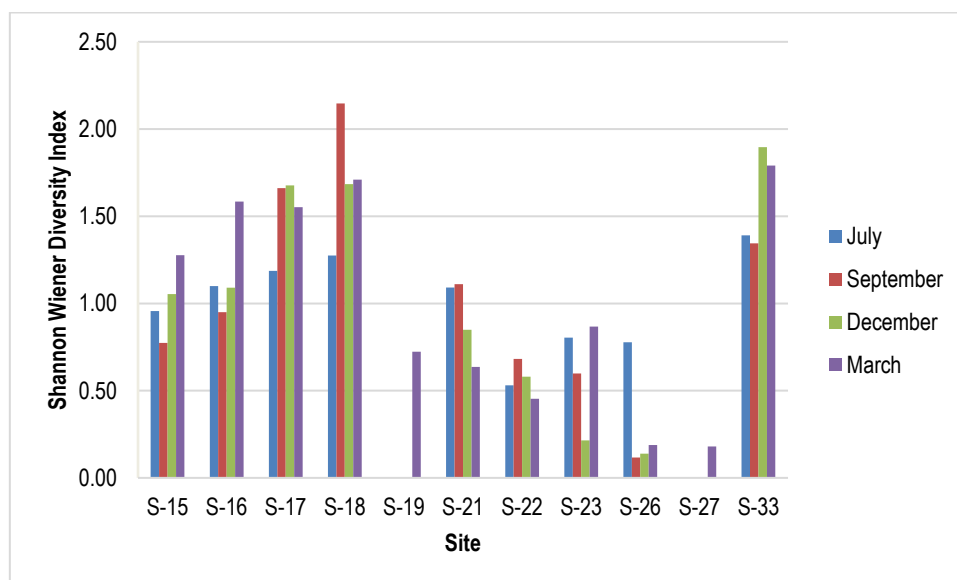
**Figure 8: Diversity Indices in December 2011**

In March, the two indices were highest at Ahero Irrigation Scheme and lowest at Kundos at Bridge (Figure 9).



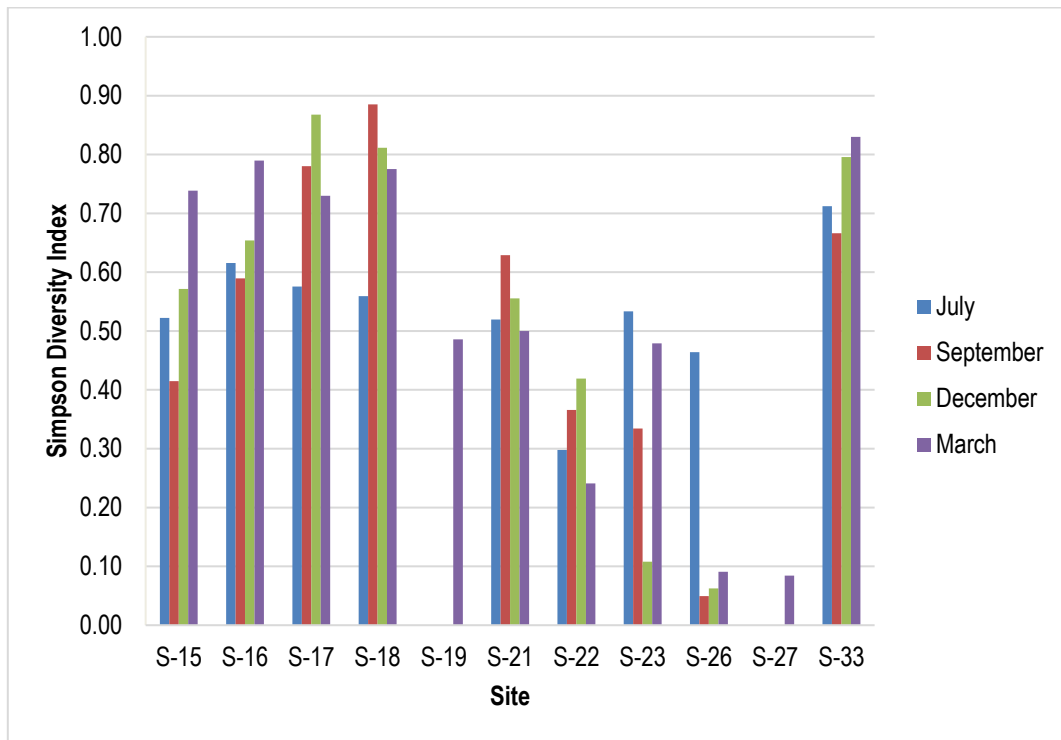
**Figure 9: Diversity Indices in March 2012**

Overall, Shannon diversity index was highest at Awach Kano (Site 18) in September and lowest at Kapngorium (Site 26) in September. Sites further downstream registered relatively higher Shannon indices than sites at higher altitude (Figure 10).



**Figure 10: Variation of Shannon diversity index in the sampling sites**

Simpson diversity index gave similar results with sites further downstream registering higher indices compared to sites at higher altitude (Figure 11). The highest Simpson index (0.89) was registered at Awach Kano (Site 18) in September whereas the lowest (0.05) was registered at Kapngorium (Site 26) in September.



**Figure 11: Variation of Simpson diversity index in the sampling sites**

One-way analysis of variance (ANOVA) was used to analyse the difference in means among the site for the four sampling months.

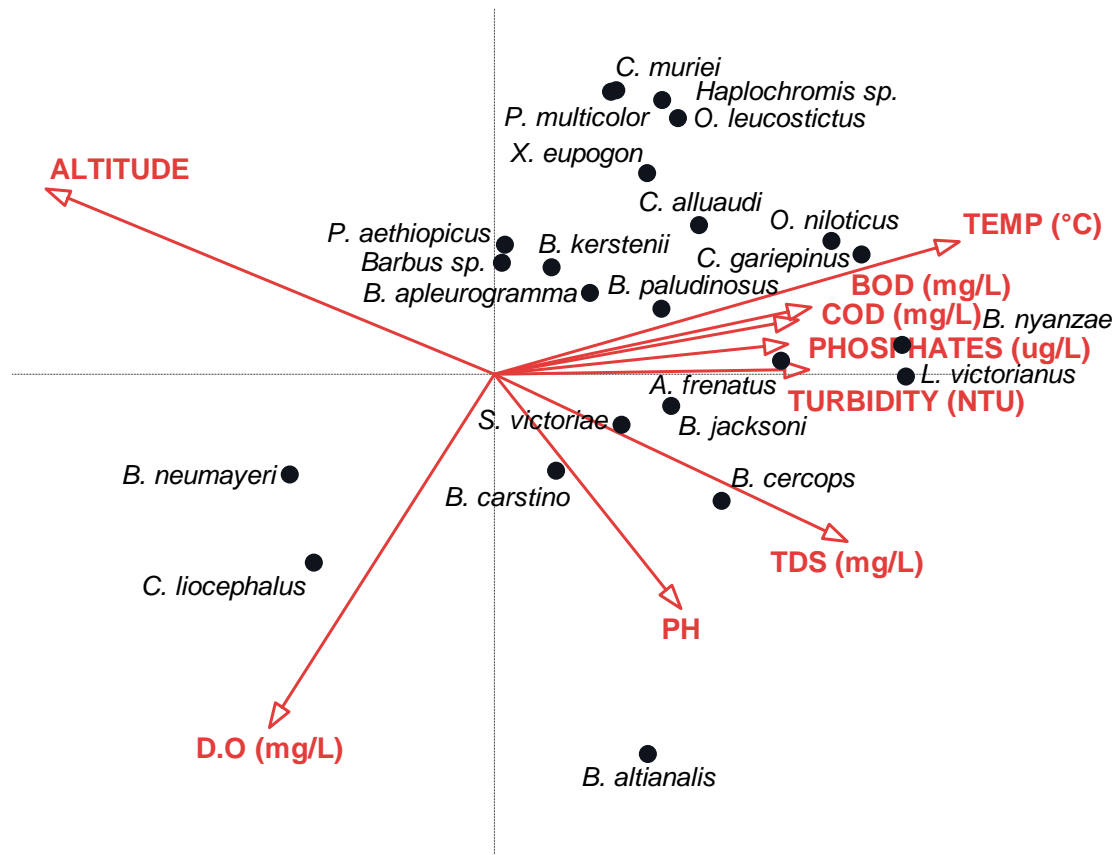
There was significant variation ( $P < 0.05$ ) in diversity indices between sampling sites for the four sampling periods as determined by one-way ANOVA; Shannon Index (ANOVA,  $F_{8,27} =$ ,  $p < 0.001$ ), Evenness (ANOVA,  $F_{8,27} =$ ,  $p = 0.002$ ), Simpson Index (ANOVA,  $F_{8,27} =$ ,  $p < 0.001$ ) and Maglef Index (ANOVA,  $F_{8,27} =$ ,  $p < 0.001$ ) (Table 11).

**Table 11: One-way ANOVA for fish species diversity indices**

**ANOVA**

		Sum of Squares	df	Mean Square	F	Sig.
Log_Shannon Index	Between Groups	.397	8	.050	14.231	.000
	Within Groups	.094	27	.003		
	Total	.491	35			
Log_Evenness	Between Groups	.067	8	.008	4.139	.002
	Within Groups	.055	27	.002		
	Total	.123	35			
Log_Simpson Index	Between Groups	.131	8	.016	10.454	.000
	Within Groups	.042	27	.002		
	Total	.173	35			
Log_Maglef Index	Between Groups	.436	8	.054	17.512	.000
	Within Groups	.084	27	.003		
	Total	.520	35			

### 4.3 Correlating effects of physico-chemical parameters on fish species



**Figure 12: RDA biplot showing the variation in fish community composition in relation to the physico-chemical parameters for the Nandi-Lower Nyando River sub-catchment**

In the variation of fish abundance 4% could be attributed to sampling date and 41% to the physico-chemical parameters (Figure 12). Of the latter variance 43% is displayed on the horizontal axis and another 29% on the vertical one (Figure 12). Only physico-chemical parameters and not the sampling dates explained a significant part of the variation in the fish community composition in the Nandi-Lower Nyando sub-catchment (Figure 12).

All the sampling sites were included in the analysis (Monte Carlo permutation tests,  $p < 0.05$ ). The physico-chemical parameter explain a significant ( $p < 0.05$ ) part of the variance in the community composition of the fish species in the permutation tests. The data sets were therefore analyzed on their changes in fish community composition on the correlation of these changes with the measured physico-chemical parameters (Tables 3-6).

In the RDA biplot in Figure 12 only altitude and dissolve oxygen (DO) explained a significant fraction of the variance in the community composition of the fish composition of the Nandi-Lower River Nyando sub-catchment (Figure 12). Altitude and dissolve oxygen are negatively associated with a higher biodiversity (Figure 12) but positively associate with *Barbus neumayeri* and *Clarias liocephalus* while pH, TDS, turbidity, phosphate, COD, BOD and temperatures are positively correlated, with high biodiversity (Figure 12).

There was significant variation ( $P < 0.05$ ) in most physico-chemical parameters between sampling sites for the four sampling periods as determined by one-way ANOVA (Table 12); temperature (ANOVA,  $F_{12,39} = , p < 0.001$ ), dissolved oxygen (ANOVA,  $F_{12,39} = , p = 0.003$ ), conductivity (ANOVA,  $F_{12,39} = , p < 0.001$ ), turbidity (ANOVA,  $F_{12,39} = , p = 0.001$ ), total dissolved solids (ANOVA,  $F_{12,39} = , p < 0.001$ ), total suspended solids (ANOVA,  $F_{12,39} = , p < 0.001$ ), phosphates (ANOVA,  $F_{12,39} = , p = 0.001$ ), biological oxygen demand (ANOVA,  $F_{12,39} = , p < 0.001$ ), and chemical oxygen demand (ANOVA,  $F_{12,39} = , p < 0.001$ ) (Table 12).

There was no significant variation ( $P > 0.05$ ) in pH and nitrates between sampling sites for the four sampling periods as determined by one-way ANOVA; pH (ANOVA,  $F_{12,39} = , p = 0.254$ ) and nitrates (ANOVA,  $F_{12,39} = , p = 0.837$ ) (Table 12).

**Table 12: One-way ANOVA for physico-chemical parameters**

		ANOVA				
		Sum of Squares	df	Mean Square	F	Sig.
Lg_Temp	Between Groups	.161	12	.013	7.864	.000
	Within Groups	.067	39	.002		
	Total	.228	51			
Lg_D.O	Between Groups	.257	12	.021	3.222	.003
	Within Groups	.259	39	.007		
	Total	.516	51			
Lg_Conductivity	Between Groups	3.280	12	.273	6.861	.000
	Within Groups	1.554	39	.040		
	Total	4.833	51			
Lg_Turbidity	Between Groups	7.226	12	.602	3.522	.001
	Within Groups	6.668	39	.171		
	Total	13.894	51			
Lg_pH	Between Groups	.006	12	.001	1.307	.254
	Within Groups	.016	39	.000		
	Total	.022	51			
Lg_TDS	Between Groups	1.463	12	.122	4.899	.000
	Within Groups	.970	39	.025		
	Total	2.433	51			
Lg_TSS	Between Groups	10.795	12	.900	5.634	.000
	Within Groups	6.227	39	.160		
	Total	17.022	51			
Lg_Nitrates	Between Groups	1.602	12	.133	.589	.837
	Within Groups	8.835	39	.227		
	Total	10.437	51			
Lg_Phosphates	Between Groups	7.661	12	.638	3.600	.001
	Within Groups	6.917	39	.177		
	Total	14.578	51			
Lg_B.O.D	Between Groups	6.413	12	.534	10.765	.000
	Within Groups	1.936	39	.050		
	Total	8.350	51			
Lg_C.O.D	Between Groups	4.358	12	.363	10.870	.000
	Within Groups	1.303	39	.033		
	Total	5.661	51			



## Pesticide Residue Levels in Fish

### 4.4 Quantitative Characteristics

Reference standards of the PCBs and OCPs were used in various steps in the analysis. The analysis was done with GC-ECD. Working reference standard solutions in the range of 2.92-112.28 µg/L were prepared individually for each standard. Quantification was based on calculations from calibration curves for each standard (Figure 13).

Calibration curves for each analyte of interest were produced for quantification of the analytes of interest. The calibration curve of each was a straight line with a correlation factor ( $R^2$ ) above 0.99 indicating a high correlation between instrument response and analyte concentration (Appendix IV).

The noise value was calculated based on the peak height of the blank sample runs around the retention time of each analyte using auto-integrator [Sanagi *et al.*, 2009]. The limit of detection (LOD) was calculated as three times noise value and the limit of quantification (LOQ) was estimated as ten times noise value as shown in Table 13, a method used in cases where baseline noise causes interference [ICH, 1996].

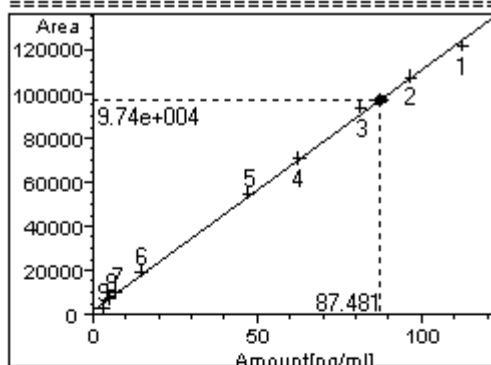
The LOD of the analytes of interest was calculated based on the the corresponding noise signal produced after injecting a low (near baseline) concentration of the calibration standard and using the relationship adapted from the equation below:

$$\text{LOD} = \frac{3 \times \text{Noise peak area} \times \text{concentration of standard injected (ng)}}{\text{Analyte response in the lowest calibration point}}$$

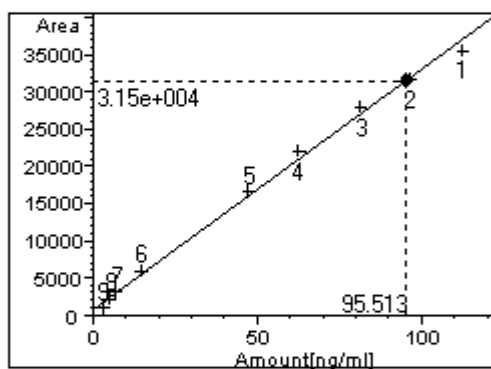
$$\text{LOQ} = 10 \times \text{LOD}$$

*Barbus altianalis* fish species that showed the highest occurrence was used for the percentage recovery test (Table 13). The average percentage recoveries of organochlorine pesticides and PCBs ranged from 76.31±10.87% for PCB 180 to 101.21±8.34% for α-HCH. Table 13 gives percentage recoveries of the pesticides and PCBS in *Barbus altianalis* fish samples. Recovery values (Table 13) of the analytes of interest in this study were within the range described by Hill, 2000 (70-120%) and were therefore not corrected.

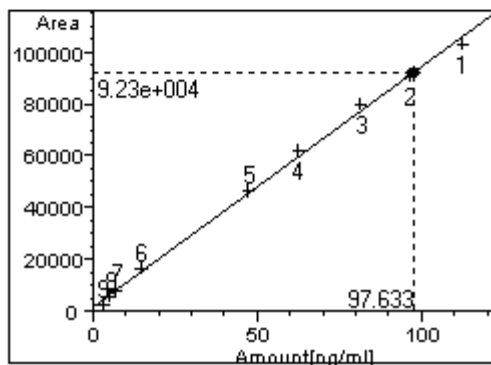
=====  
 Calibration Curves  
 =====



a-HCH at exp. RT: 7.270  
 ECD1 B,  
 Correlation: 0.99925  
 Residual Std. Dev.: 1919.91907  
 Formula:  $y = mx + b$   
 m: 1091.91375  
 b: 1834.89312  
 x: Amount[ng/ml]  
 y: Area



b-HCH at exp. RT: 7.566  
 ECD1 B,  
 Correlation: 0.99826  
 Residual Std. Dev.: 864.15428  
 Formula:  $y = mx + b$   
 m: 321.30788  
 b: 843.71787  
 x: Amount[ng/ml]  
 y: Area



g-HCH at exp. RT: 7.636  
 ECD1 B,  
 Correlation: 0.99891  
 Residual Std. Dev.: 1965.61193  
 Formula:  $y = mx + b$   
 m: 926.25446  
 b: 1826.04014  
 x: Amount[ng/ml]  
 y: Area

Figure 13: Calibration Curves for a, b and g-HCH

**Table 13: Average percentage recoveries tests for polychlorinated biphenyls and organochlorine pesticide in *Barbus altianalis* fish species**

<b>Analyte</b>	<b>% Recovery</b>	<b>LOD (<math>\mu\text{g/L}</math>)</b>	<b>LOQ (<math>\mu\text{g/L}</math>)</b>
a-HCH	101.21 $\pm$ 8.34	0.012 $\pm$ 0.001	0.120 $\pm$ 0.001
b-HCH	94.08 $\pm$ 2.35	0.011 $\pm$ 0.001	0.111 $\pm$ 0.001
g-HCH	96.81 $\pm$ 2.84	0.010 $\pm$ 0.001	0.101 $\pm$ 0.001
d-HCH	92.99 $\pm$ 4.58	0.035 $\pm$ 0.001	0.354 $\pm$ 0.001
Heptachlor	89.22 $\pm$ 7.87	0.019 $\pm$ 0.001	0.193 $\pm$ 0.001
Aldrin	91.71 $\pm$ 4.15	0.023 $\pm$ 0.001	0.225 $\pm$ 0.001
Heptachlor epoxide	86.53 $\pm$ 3.68	0.029 $\pm$ 0.001	0.293 $\pm$ 0.001
Endosulphan I	93.29 $\pm$ 4.35	0.031 $\pm$ 0.001	0.309 $\pm$ 0.001
p,p-DDE	81.72 $\pm$ 9.32	0.027 $\pm$ 0.001	0.268 $\pm$ 0.001
Dieldrin	97.78 $\pm$ 3.18	0.042 $\pm$ 0.001	0.424 $\pm$ 0.001
Endrin	88.36 $\pm$ 3.58	0.059 $\pm$ 0.001	0.595 $\pm$ 0.001
Endosulphan II	80.99 $\pm$ 7.32	0.040 $\pm$ 0.001	0.404 $\pm$ 0.001
p,p'-DDD	92.16 $\pm$ 1.44	0.092 $\pm$ 0.001	0.920 $\pm$ 0.001
Endrin aldehyde	83.14 $\pm$ 5.35	0.043 $\pm$ 0.001	0.431 $\pm$ 0.001
p,p'-DDT	94.08 $\pm$ 6.35	0.045 $\pm$ 0.001	0.446 $\pm$ 0.001
Endosulphan sulphate	77.11 $\pm$ 6.54	0.103 $\pm$ 0.001	1.031 $\pm$ 0.001
Methoxychlor	79.21 $\pm$ 6.56	0.183 $\pm$ 0.001	1.832 $\pm$ 0.001
PCB 28	90.17 $\pm$ 3.15	0.001 $\pm$ 0.001	0.013 $\pm$ 0.001
PCB 52	101.21 $\pm$ 9.34	0.003 $\pm$ 0.001	0.028 $\pm$ 0.001
PCB 101	95.68 $\pm$ 4.13	0.001 $\pm$ 0.001	0.014 $\pm$ 0.001
PCB 138	82.46 $\pm$ 12.35	0.002 $\pm$ 0.001	0.017 $\pm$ 0.001
PCB 153	85.22 $\pm$ 6.84	0.004 $\pm$ 0.001	0.039 $\pm$ 0.001
PCB 180	76.31 $\pm$ 10.87	0.014 $\pm$ 0.001	0.142 $\pm$ 0.001

#### 4.5 Qualitative Characteristics

The standard calibration curves were obtained by running multilevel calibration standards (Level 1 - 9). Chromatograms for the selected OCPs and PCBs were obtained for the individual analytes of interest. Retention time was used for identification whereas peak area was used for quantification. Calibration done on the GC-ECD, model Agilent 6890N (Figure 14 and Appendix V) was confirmed by GC-MS, model Agilent 6890 GC, 5792 MSD and 7683 Autosampler (Figure 15). Confirmation was based on comparison of sample total ion chromatogram retention time and extracted mass spectra against that obtained from reference standards for both OCPs and PCBs.

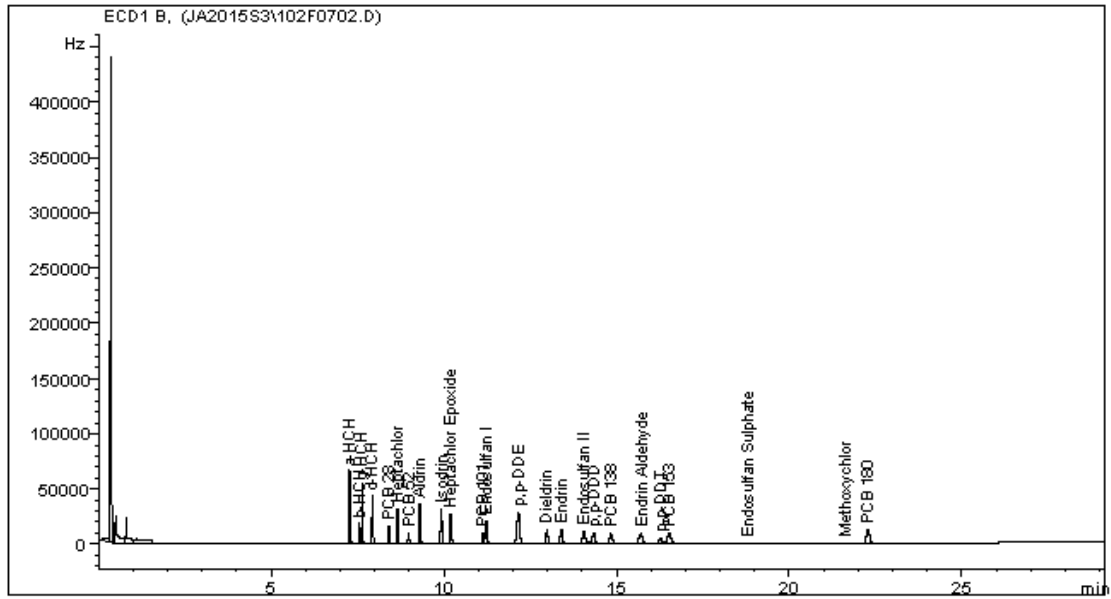


Figure 14: Chromatogram for a Level 3 standard

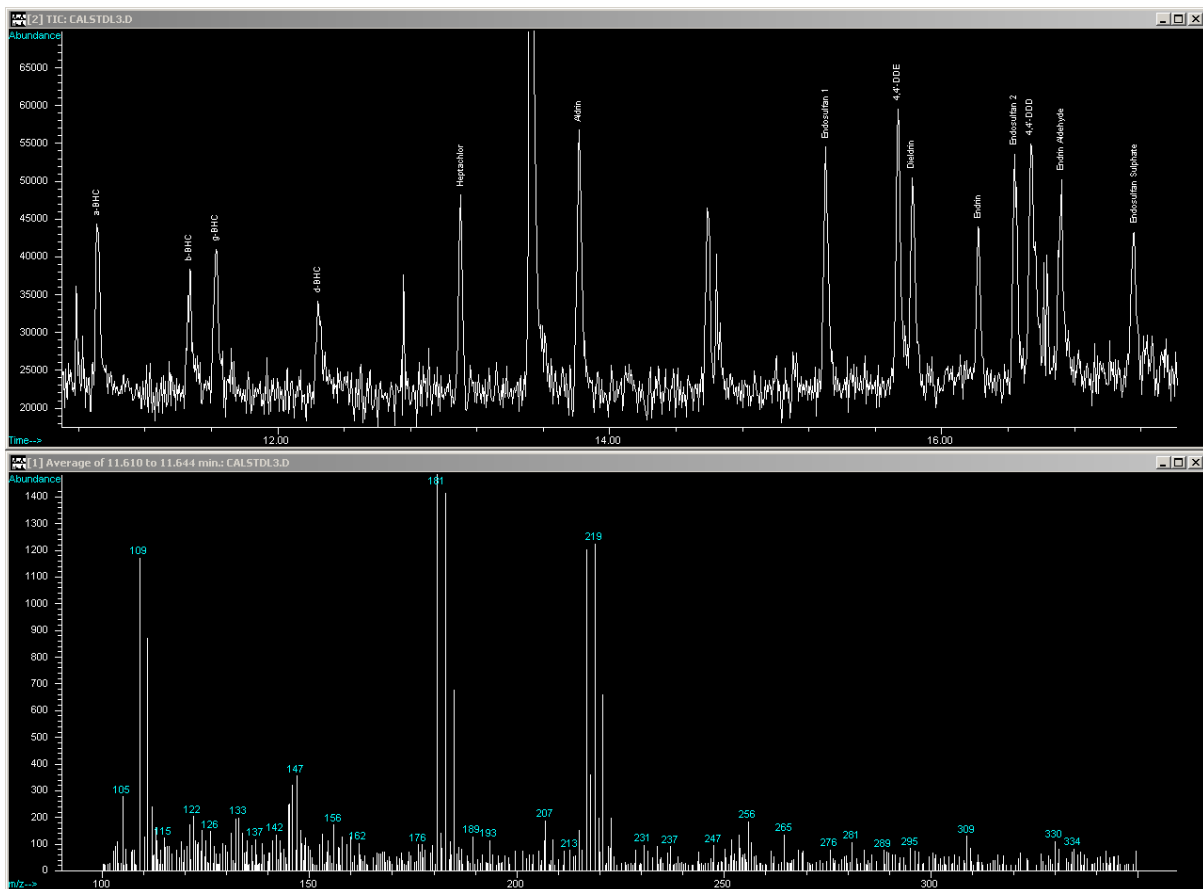


Figure 15: GC-MS Total Ion Chromatogram for a Level 3 standard showing mass spectra for g-HCH

#### 4.6 Concentration levels of organochlorine pesticides in fish muscle tissues

Organochlorine pesticide (OCPs) residue levels in fish tissue were analysed and expressed as concentration per unit of wet tissue weight (ng/g) for the three sampling periods. This is because the sample weight was measured before sample drying. Analysis of fish sample tissue was done in triplicates of 10g each. In cases where the weight of fish tissue was less than that required for analysis (at least 30g), the site was exempted from both OCP and PCB residue level analysis. Therefore results are presented for both OCPs and PCBs as shown below:

- For July sampling period – Site 16, 17, 18, 21, 22, 23 and 33.
- For September sampling period – Site 15, 16, 17, 18, 21, 22, 23 and 33.
- For December sampling period – Site 16, 17, 18, 22, 23, 27 and 33.
- For March 2012 sampling period – Site 15, 16, 17, 18 and 21.

In July, residue levels of  $\alpha$ -HCH were detected in site 18 with mean values ranging from  $2.03 \pm 0.513$  ng/g in *Labeo victorianus* to  $23.38 \pm 1.056$  ng/g of in *Clarius alluaudi*. The highest concentration detected was  $39.03 \pm 0.592$  ng/g of  $\beta$ -HCH in *Clarias liocephalus* at site 22. The lowest concentration detected was  $0.62 \pm 0.051$  ng/g of p,p-DDE in *Clarias gariepinus* at site 21 (Table 14). The variation of total OCPs detected were highest at site 17 (76.88 ng/g) and lowest at site 23 (28.89 ng/g) (Figure 16). Site 17 which is at the lower reaches of the river receives water from domestic sewage discharge from Ahero Township [Abong'o *et al.*, 2014].

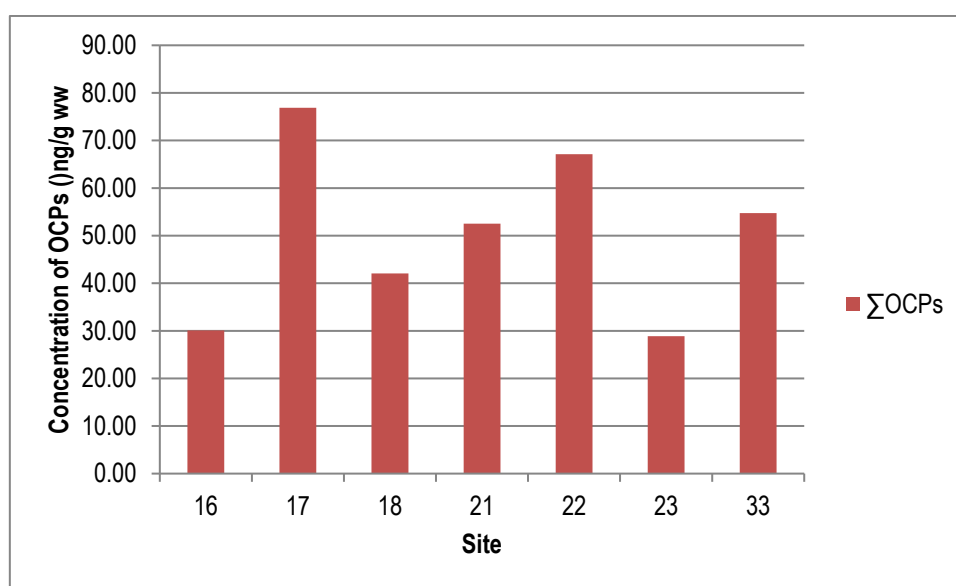


Figure 16: Variation of organochlorine pesticide residue levels in fish species in July 2011

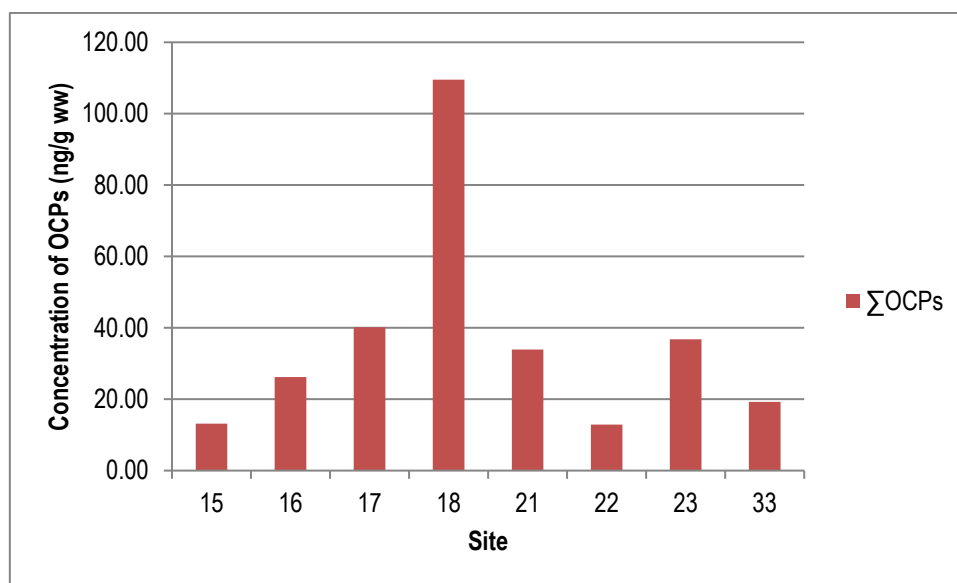
**Table 14: Organochlorine pesticides residue levels in various fish species in July 2011**

SITE	SPECIES NAME	Concentration of Pesticide residue (ng/g)												
		$\alpha$ -HCH	$\gamma$ -HCH	$\beta$ -HCH	$\delta$ -HCH	Hept	Aldrin	Hept. Ep	$\alpha$ -Endo	p,p-DDE	Endrin	$\beta$ -Endo	p,p-DDD	Endr Ald.
16	<i>Labeo victorianus</i>	BDL	3.11 $\pm$ 0.83	BDL	BDL	3.11 $\pm$ 0.45	11.96 $\pm$ 2.528	BDL	BDL	BDL	BDL	11.92 $\pm$ 2.01	BDL	BDL
17	<i>Clarias gariepinus</i>	BDL	BDL	0.70 $\pm$ 0.08	0.90 $\pm$ 0.18	BDL	3.43 $\pm$ 0.364	BDL	BDL	BDL	BDL	0.76 $\pm$ 0.10	BDL	BDL
	<i>Labeo victorianus</i>	5.40 $\pm$ 0.69	BDL	2.90 $\pm$ 0.95	19.17 $\pm$ 0.29	BDL	BDL	BDL	1.05 $\pm$ 0.34	BDL	BDL	BDL	3.68 $\pm$ 0.17	BDL
	<i>Synodontis victoriae</i>	2.48 $\pm$ 0.30	BDL	15.15 $\pm$ 2.13	15.80 $\pm$ 0.94	BDL	BDL	BDL	1.19 $\pm$ 0.35	BDL	0.26 $\pm$ 0.08	BDL	BDL	4.01 $\pm$ 0.36
18	<i>Barbus altianalis</i>	BDL	1.12 $\pm$ 0.25	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL
	<i>Clarius alluaudi</i>	23.38 $\pm$ 1.05	2.43 $\pm$ 0.70	BDL	BDL	BDL	5.16 $\pm$ 0.418	BDL	BDL	BDL	BDL	BDL	BDL	BDL
	<i>Clarias gariepinus</i>	BDL	2.58 $\pm$ 0.45	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL
	<i>Labeo victorianus</i>	2.03 $\pm$ 0.51	2.83 $\pm$ 0.24	BDL	2.54 $\pm$ 0.16	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL
21	<i>Barbus altianalis</i>	BDL	5.20 $\pm$ 0.68	2.55 $\pm$ 0.459	20.78 $\pm$ 0.17	BDL	BDL	BDL	BDL	7.75 $\pm$ 0.28	BDL	BDL	BDL	BDL
	<i>Clarias gariepinus</i>	4.27 $\pm$ 0.75	2.55 $\pm$ 0.46	BDL	4.91 $\pm$ 0.80	1.28 $\pm$ 0.343	BDL	BDL	BDL	0.62 $\pm$ 0.05	2.60 $\pm$ 0.26	BDL	BDL	BDL
22	<i>Barbus altianalis</i>	BDL	15.75 $\pm$ 0.41	BDL	4.25 $\pm$ 0.22	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL
	<i>Clarias liocephalus</i>	BDL	8.09 $\pm$ 0.59	39.03 $\pm$ 0.592	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL
23	<i>Barbus altianalis</i>	BDL	12.62 $\pm$ 1.38	6.17 $\pm$ 0.763	1.06 $\pm$ 0.13	3.36 $\pm$ 0.53	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL
	<i>Clarias liocephalus</i>	BDL	2.44 $\pm$ 0.38	BDL	BDL	3.24 $\pm$ 0.19	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL
33	<i>Oreochromis niloticus</i>	BDL	8.38 $\pm$ 0.26	3.69 $\pm$ 0.205	BDL	BDL	7.05 $\pm$ 0.47	3.66 $\pm$ 0.79	BDL	BDL	BDL	13.14 $\pm$ 0.78	BDL	BDL
	<i>Protopterus aethiopicus</i>	4.66 $\pm$ 0.34	BDL	2.74 $\pm$ 0.431	BDL	BDL	2.93 $\pm$ 0.24	8.50 $\pm$ 0.26	BDL	BDL	BDL	BDL	BDL	BDL

Mean  $\pm$  SD, Wet Weight (ng/g), Below Detection Limit (BDL)

In September, residue levels of  $\alpha$ -HCH were detected with mean values ranging from  $14.54 \pm 0.673$  ng/g in *Barbus altianalis* at Site 21 to  $1.06 \pm 0.165$  ng/g in *Oreochromis leucostictus* at Site 33. Residue levels of  $\delta$ -HCH were detected with mean values ranging from  $12.46 \pm 0.306$  ng/g in *Barbus altianalis* at Site 21 to  $1.92 \pm 0.281$  ng/g in *Oreochromis leucostictus* at Site 33 (Table 15).

The variation of total OCPs detected were highest at site 18 (109.53 ng/g) and lowest at site 22 (12.88 ng/g) (Figure 17).



**Figure 17: Variation of organochlorine pesticide residue levels in fish species in September 2011**

**Table 15: Organochlorine pesticides residue levels in various fish species in September 2011**

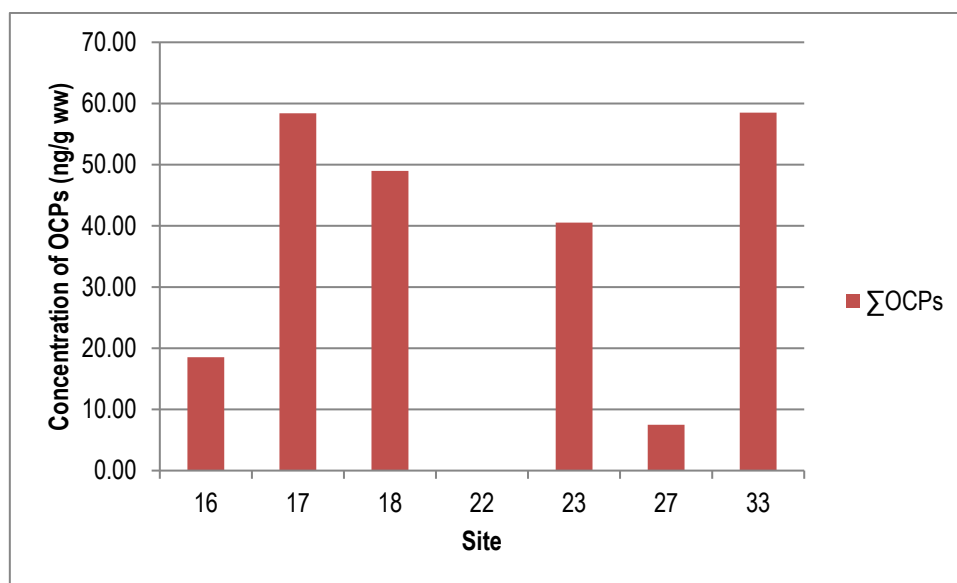
SITE	SPECIES NAME	Concentration of pesticide residue (ng/g)													
		$\alpha$ -HCH	$\gamma$ -HCH	$\beta$ -HCH	$\delta$ -HCH	Hept	Aldrin	Hept. Ep	$\alpha$ -Endo	p,p-DDE	Dieldrin	Endrin	$\beta$ -Endo	p,p-DDD	Endr Ald.
15	<i>Barbus altianalis</i>	4.53 ±0.21	5.11 ±0.58	BDL	3.52 ±0.21	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL
16	<i>Oreochromis niloticus</i>	6.90 ±0.24	11.73 ±0.65	BDL	2.35 ±0.21	BDL	BDL	BDL	BDL	BDL	BDL	BDL	5.21 ±0.03	BDL	BDL
17	<i>Barbus altianalis</i>	3.94 ±0.24	2.71 ±0.30	3.66 ±0.32	5.53 ±0.40	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL
	<i>Oreochromis niloticus</i>	1.15 ±0.09	5.11 ±0.17	BDL	7.42 ±0.59	BDL	BDL	BDL	7.65 ±0.32	BDL	BDL	BDL	BDL	BDL	2.99 ±0.44
18	<i>Barbus altianalis</i>	BDL	5.82 ±0.80	BDL	4.39 ±0.57	1.72 ±0.12	7.86 ±0.60	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL
	<i>Clarias gariepinus</i>	BDL	4.12 ±0.22	BDL	BDL	2.41 ±0.28	BDL	BDL	BDL	BDL	BDL	2.27 ±0.22	BDL	BDL	BDL
	<i>Aethiomastersebelus fretanus</i>	1.95 ±0.18	4.72 ±0.30	BDL	4.32 ±0.30	3.64 ±0.24	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL
	<i>Oreochromis niloticus</i>	1.80 ±0.02	3.01 ±0.41	BDL	11.58 ±0.88	5.93 ±0.12	7.27 ±0.52	17.24 ±0.76	BDL	BDL	BDL	3.96 ±0.49	BDL	15.52 ±0.72	BDL
21	<i>Barbus altianalis</i>	14.54 ±0.67	BDL	4.01 ±0.86	12.46 ±0.30	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL
	<i>Clarias liocephalus</i>	BDL	BDL	2.90 ±0.27	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL
22	<i>Barbus altianalis</i>	BDL	7.71 ±0.22	BDL	5.17 ±0.83	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL
23	<i>Barbus altianalis</i>	6.09 ±0.67	BDL	BDL	8.15 ±0.49	8.37 ±0.245	4.67 ±0.29	BDL	BDL	4.48 ±0.23	1.21 ±0.05	BDL	1.88 ±0.15	1.93 ±0.39	BDL
33	<i>Clarias gariepinus</i>	BDL	1.79 ±0.66	BDL	1.92 ±0.65	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	0.38 ±0.02
	<i>Oreochromis leucostictus</i>	1.06 ±0.16	10.79 ±0.88	BDL	1.92 ±0.28	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	1.38 ±0.02

Mean ± SD, Wet Weight (ng/g), Below Detection Limit (BDL)



In December, residue levels of  $\alpha$ -HCH were detected with mean values ranging from  $7.96\pm 0.382$  ng/g in *Barbus altianalis* at Site 23 to  $3.16\pm 0.619$  ng/g in *Clarias gariepinus* at Site 33. Residue levels of  $\delta$ -HCH were detected with mean values ranging from  $12.82\pm 0.262$  ng/g in *Barbus altianalis* at Site 17 to  $2.31\pm 0.277$  ng/g in *Barbus altianalis* at Site 23 (Table 16).

The variation of total OCPs detected were highest at site 33 (58.50 ng/g) and lowest at site 27 (7.49 ng/g) (Figure 18).



**Figure 18: Variation of organochlorine pesticide residue levels in fish species in December 2011**

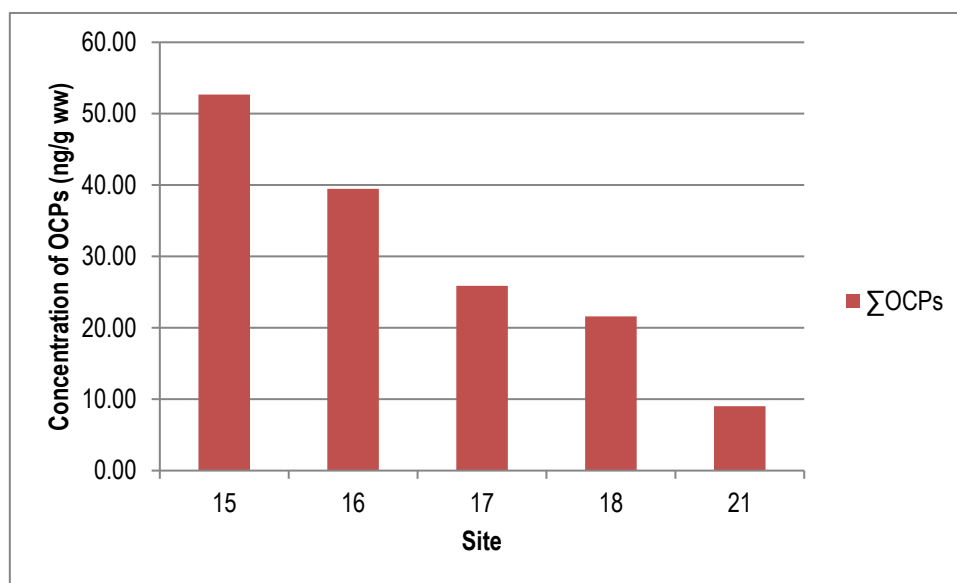
**Table 16: Organochlorine pesticides residue levels in various fish species in December 2011**

SITE	SPECIES NAME	Concentration of pesticide residue (ng/g)												
		$\alpha$ -HCH	$\gamma$ -HCH	$\beta$ -HCH	$\delta$ -HCH	Hept	Aldrin	Hept. Ep	p,p-DDE	Dieldrin	Endrin	$\beta$ -Endo	p,p-DDD	Endr Ald.
16	<i>Oreochromis niloticus</i>	3.46 ±0.22	4.72 ±0.25	BDL	7.96 ±0.89	2.39 ±0.12	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL
17	<i>Barbus altianalis</i>	BDL	16.43 ±0.52	BDL	12.82 ±0.26	BDL	2.44 ±0.15	BDL	BDL	BDL	0.57 ±0.02	BDL	BDL	BDL
	<i>Clarias liocephalus</i>	5.65 ±0.43	6.51 ±0.48	2.43 ±0.27	BDL	7.27 ±0.68	BDL	BDL	BDL	4.38 ±0.37	2.33 ±0.13	BDL	BDL	BDL
18	<i>Labeo victorinus</i>	4.84 ±0.10	12.88 ±0.84	BDL	3.47 ±0.52	BDL	11.36 ±0.41	BDL	3.73 ±0.70	BDL	9.96 ±0.24	BDL	2.74 ±0.16	BDL
22	<i>Barbus altianalis</i>	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL
23	<i>Barbus altianalis</i>	7.96 ±0.38	11.51 ±0.54	BDL	2.31 ±0.27	BDL	BDL	5.04 ±0.35	11.75 ±0.86	BDL	BDL	BDL	1.96 ±0.35	BDL
27	<i>Barbus neumayeri</i>	2.36 ±0.39	BDL	BDL	3.75 ±0.15	BDL	BDL	BDL	1.38 ±0.57	BDL	BDL	BDL	BDL	BDL
33	<i>Clarias gariepinus</i>	3.16 ±0.61	13.41 ±0.61	BDL	BDL	BDL	5.38 ±0.46	BDL	2.86 ±0.78	BDL	BDL	0.32 ±0.07	BDL	1.56 ±0.39
	<i>Oreochromis niloticus</i>	BDL	19.51 ±0.86	BDL	3.37 ±0.41	BDL	BDL	BDL	5.31 ±0.76	BDL	BDL	BDL	3.62 ±0.49	BDL

Mean ± SD, Wet Weight (ng/g), Below Detection Limit (BDL)

In March, residue levels of  $\gamma$ -HCH were detected with mean values ranging from  $14.61 \pm 0.953$  ng/g in *Labeo victorianus* at Site 15 to  $1.59 \pm 0.166$  ng/g in *Clarias gariepinus* at Site 18. Residue levels of  $\delta$ -HCH were detected with mean values ranging from  $3.63 \pm 0.481$  ng/g in *Barbus altianalis* at Site 17 to  $0.31 \pm 0.025$  ng/g in *Labeo victorianus* at Site 16 (Table 17).

The variation of total OCPs detected were highest at site 15 (52.67 ng/g) and lowest at site 21 (9.02 ng/g) (Figure 19).



**Figure 19: Variation of organochlorine pesticide residue levels in fish species in March 2012**

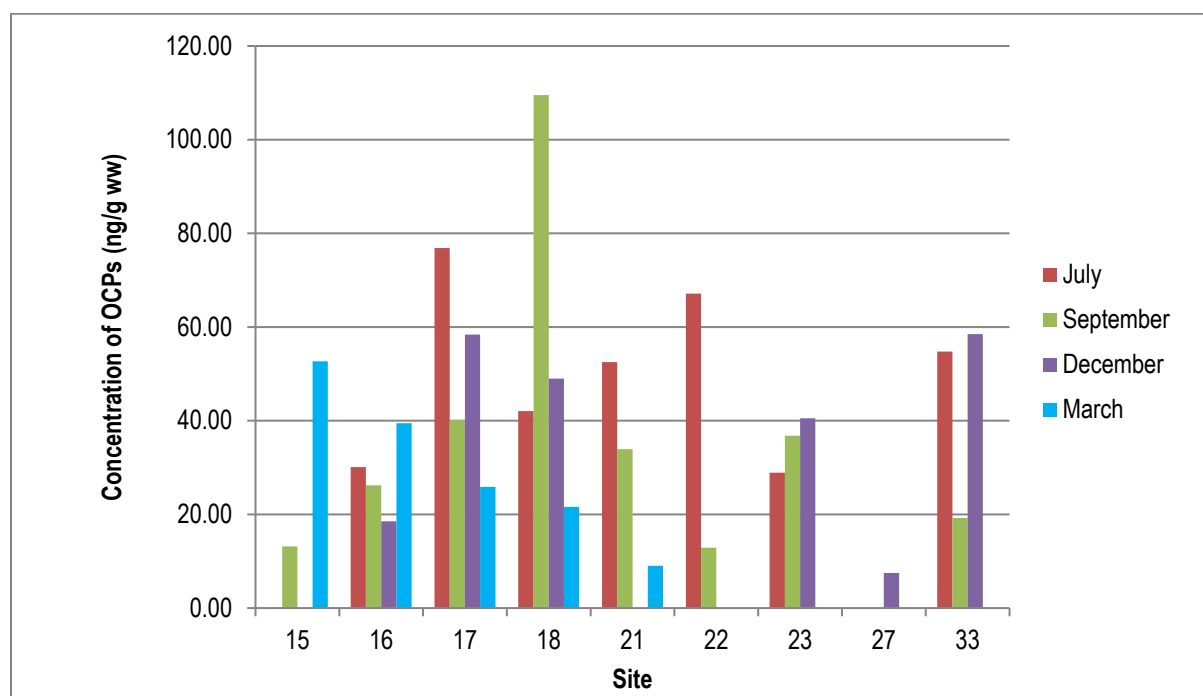
**Table 17: Organochlorine pesticides residue levels in various fish species in March 2012**

SITE	SPECIES NAME	Concentration of pesticide residue (ng/g)											
		$\alpha$ -HCH	$\gamma$ -HCH	$\beta$ -HCH	$\delta$ -HCH	Hept	Aldrin	p,p-DDE	Dieldrin	Endrin	$\beta$ -Endo	p,p-DDD	Endr Ald.
15	<i>Clarias gariepinus</i>	2.31 ±0.27	6.26 ±0.68	BDL	4.19 ±0.23	BDL	2.96 ±0.58	1.52 ±0.34	6.32 ±0.50	BDL	BDL	5.23 ±0.36	BDL
	<i>Labeo victorianus</i>	3.26 ±0.58	14.61 ±0.95	BDL	2.19 ±0.47	2.97 ±0.21	BDL	BDL	BDL	0.85 ±0.05	BDL	BDL	BDL
16	<i>Clarias gariepinus</i>	BDL	10.25 ±0.93	1.01 ±0.42	3.09 ±0.55	BDL	0.20 ±0.01	BDL	BDL	BDL	BDL	0.45 ±0.06	0.54 ±0.01
	<i>Labeo victorianus</i>	BDL	2.83 ±0.55	BDL	0.31 ±0.02	0.11 ±0.09	BDL	BDL	BDL	5.82 ±0.44	BDL	4.59±0.907	BDL
	<i>Oreochromis niloticus</i>	3.79 ±0.48	3.52 ±0.76	1.25 ±0.24	BDL	BDL	BDL	BDL	BDL	0.92 ±0.03	BDL	0.79 ±0.04	BDL
17	<i>Barbus altianalis</i>	BDL	1.75 ±0.313	BDL	3.63 ±0.48	BDL	0.89 ±0.06	0.41 ±0.033	2.09 ±0.16	BDL	BDL	BDL	BDL
	<i>Clarias gariepinus</i>	BDL	6.18 ±0.83	BDL	BDL	BDL	1.60 ±0.57	BDL	1.28 ±0.27	BDL	1.54 ±0.35	BDL	BDL
	<i>Labeo victorianus</i>	BDL	2.29 ±0.44	BDL	1.39 ±0.14	BDL	BDL	2.50 ±0.264	BDL	0.33 ±0.04	BDL	BDL	BDL
18	<i>Barbus neumayeri</i>	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL
	<i>Clarias gariepinus</i>	BDL	1.59 ±0.16	BDL	1.94 ±0.71	BDL	BDL	0.97 ±0.279	5.75 ±0.74	BDL	BDL	BDL	BDL
	<i>Labeo victorianus</i>	BDL	2.28 ±0.49	BDL	1.20 ±0.34	BDL	1.87 ±0.61	BDL	BDL	BDL	1.88 ±0.41	4.11 ±0.30	BDL
21	<i>Clarias liocephalus</i>	2.45 ±0.13	2.92 ±0.49	BDL	BDL	0.71 ±0.05	BDL	1.63 ±0.082	1.31 ±0.14	BDL	BDL	BDL	BDL

Mean ± SD, Wet Weight (ng/g), Below Detection Limit (BDL)

Over the four sampling periods, the variation of total OCPs detected were highest at site 18 (109.53 ng/g) in September and lowest at site 27 (7.49 ng/g) in December (Figure 20). Of the four sampling periods, September had the highest  $\Sigma$ OCP concentrations in 8 of the 9 sites where fish tissue samples were analysed.

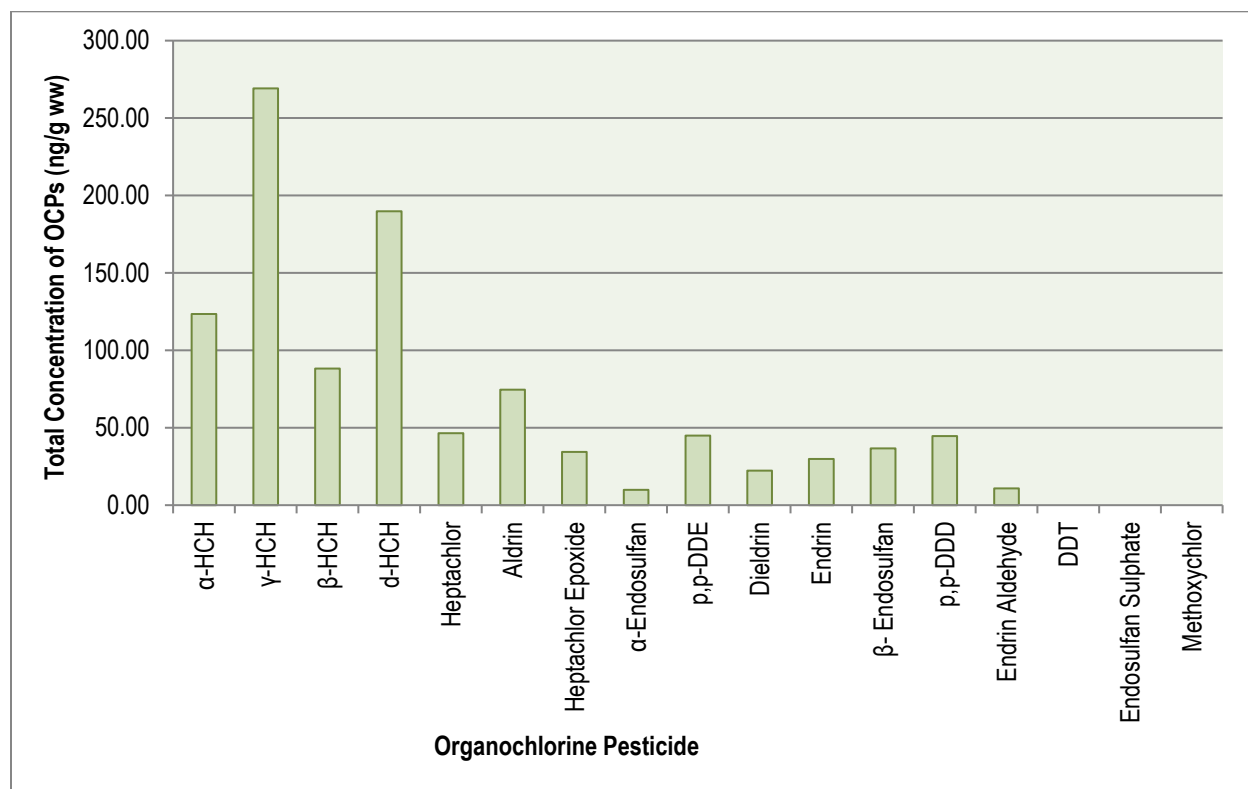
Of the 9 sites, Awach Kano (Site 18) and Nyando at Dykes (Site 17) had consistently high  $\Sigma$ OCP concentrations in the four sampling periods ranging from 109.53 ng/g to 21.59 ng/g while Nyando at Ahero Bridge (Site 16) and Ainopisiwa (Site 23) had low concentrations ranging from 40.53 ng/g to 18.53 ng/g (Figure 20).



**Figure 20: Variation of organochlorine pesticide residue levels in fish species for the four sampling periods**

The concentrations of chlorinated pesticides were high (Table 14-17) in most of the analyzed fish species. All OCPs analysed were detected in fish tissue samples with the exception of p,p'-DDT, endosulfan sulphate and methoxychlor which were below detection limit.

The total concentration of organochlorine pesticide residues over the four sampling periods revealed the following pattern:  $\Sigma\gamma\text{-HCH} > \Sigma\delta\text{-HCH} > \Sigma\alpha\text{-HCH} > \Sigma\beta\text{-HCH} > \Sigma\text{Aldrin} > \Sigma\text{Heptachlor} > \Sigma\text{p,p'-DDE} > \Sigma\text{p,p'-DDD} > \Sigma\beta\text{-Endosulfan} > \Sigma\text{Heptachlor Epoxide} > \Sigma\text{Endrin} > \Sigma\text{Dieldrin} > \Sigma\text{Endrin Aldehyde} > \Sigma\alpha\text{-Endosulfan}$  (Figure 21).



**Figure 21: Total concentration of analysed OCP compounds for the four sampling periods**

#### 4.7 Concentration of polychlorinated biphenyls in fish muscle tissues

Polychlorinated biphenyls (PCBs) residue levels in fish tissue were analysed and expressed as concentration per unit of wet tissue weight (ng/g) during the sampling periods. This is because the sample weight was measured before sample drying.

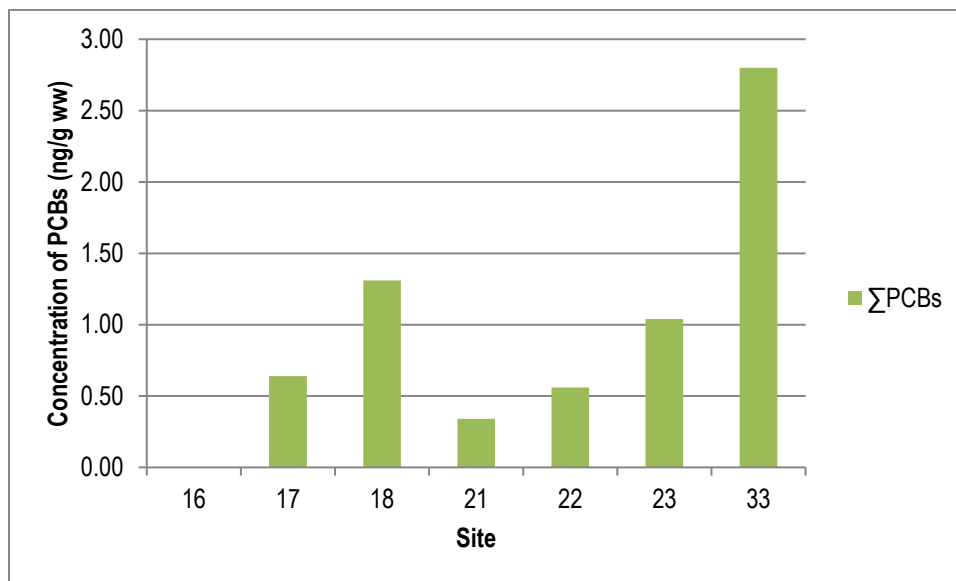
In July, residue levels of polychlorinated biphenyls (PCBs) were investigated. PCB 153 and PCB 180 were below detection limit (BDL). Residue levels of PCB 28 were detected with mean values ranging from  $0.55\pm 0.071$  ng/g in *Barbus altianalis* at Site 18 to  $0.15\pm 0.016$  ng/g in *Barbus altianalis* at Site 21. Residue levels of PCB 101 were detected with mean values ranging from  $0.76\pm 0.047$  ng/g in *Protopterus aethiopicus* at Site 33 to  $0.17\pm 0.009$  ng/g in *Oreochromis niloticus* at Site 33 (Table 18).

**Table 18: Polychlorinated biphenyls residue levels in fish in July 2011**

SITE	SPECIES NAME	PCB 28	PCB 52	PCB 101	PCB 138	PCB 153	PCB 180
16	<i>Labeo victorinus</i>	BDL	BDL	BDL	BDL	BDL	BDL
17	<i>Clarias gariepinus</i>	BDL	BDL	BDL	BDL	BDL	BDL
	<i>Labeo victorinus</i>	$0.25\pm 0.02$	BDL	$0.39\pm 0.05$	BDL	BDL	BDL
	<i>Synodontis victoriae</i>	BDL	BDL	BDL	BDL	BDL	BDL
18	<i>Barbus altianalis</i>	$0.55\pm 0.07$	BDL	BDL	BDL	BDL	BDL
	<i>Clarius alluaudi</i>	$0.03\pm 0.00$	BDL	BDL	BDL	BDL	BDL
	<i>Clarias gariepinus</i>	$0.19\pm 0.01$	BDL	BDL	BDL	BDL	BDL
	<i>Labeo victorinus</i>	BDL	BDL	BDL	$0.54\pm 0.08$	BDL	BDL
21	<i>Barbus altianalis</i>	$0.34\pm 0.04$	BDL	BDL	BDL	BDL	BDL
	<i>Clarias gariepinus</i>	BDL	BDL	BDL	BDL	BDL	BDL
22	<i>Barbus altianalis</i>	$0.15\pm 0.01$	$0.12\pm 0.02$	BDL	BDL	BDL	BDL
	<i>Clarias liocephalus</i>	$0.29\pm 0.04$	BDL	BDL	BDL	BDL	BDL
23	<i>Barbus altianalis</i>	$0.71\pm 0.01$	BDL	BDL	BDL	BDL	BDL
	<i>Clarias liocephalus</i>	$0.33\pm 0.03$	BDL	BDL	BDL	BDL	BDL
33	<i>Oreochromis niloticus</i>	$0.39\pm 0.02$	$0.54\pm 0.04$	$0.17\pm 0.01$	BDL	BDL	BDL
	<i>Protopterus aethiopicus</i>	$0.94\pm 0.08$	BDL	$0.76\pm 0.04$	BDL	BDL	BDL

Mean  $\pm$  SD, Wet Weight (ng/g), Below Detection Limit (BDL)

The variation of total PCBs detected were highest at site 33 (2.80 ng/g) and lowest at site 21 (0.34 ng/g) (Figure 22).



**Figure 22: Variation of polychlorinated biphenyl residues levels in fish species in July 2011**

In September, residue levels of PCB 52 were detected with mean values ranging from  $6.13 \pm 0.427$  ng/g in *Barbus altianalis* at Site 17 to  $0.12 \pm 0.030$  ng/g in *Barbus altianalis* at Site 15. Residue levels of PCB 138 were detected with mean values ranging from  $0.54 \pm 0.026$  ng/g in *Oreochromis niloticus* at Site 16 to  $0.12 \pm 0.015$  ng/g in *Clarias gariepinus* at Site 33 (Table 19).

The variation of total PCBs detected were highest at site 17 (6.13 ng/g) and lowest at site 15 (0.59 ng/g) (Figure 23).



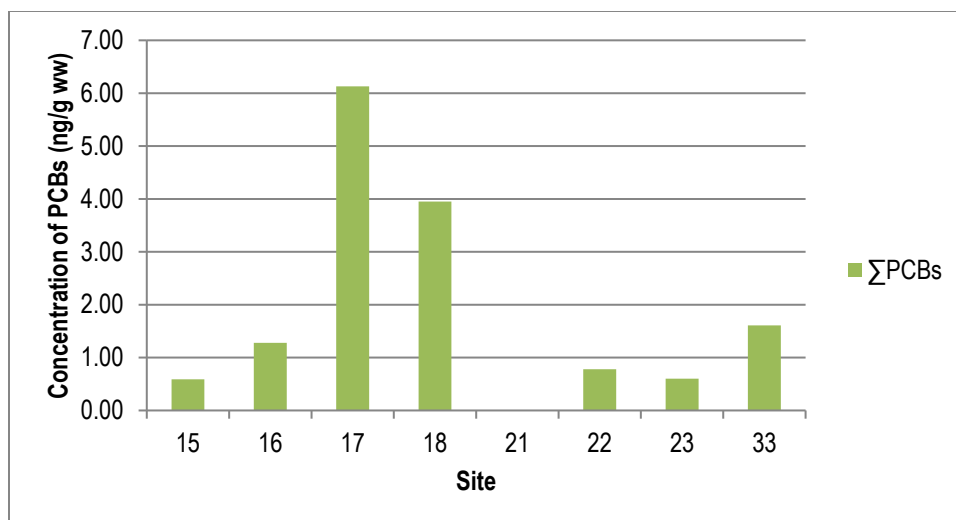


Figure 23: Variation of polychlorinated biphenyl residues levels in fish September 2011

Table 19: Polychlorinated biphenyls residue levels fish in September 2011

SITE	SPECIES NAME	PCB 28	PCB 52	PCB 101	PCB 138	PCB 153	PCB 180
15	<i>Barbus altianalis</i>	BDL	0.12 ±0.03	0.23 ±0.06	0.24 ±0.08	BDL	BDL
16	<i>Oreochromis niloticus</i>	BDL	0.74 ±0.024	BDL	0.54 ±0.02	BDL	BDL
17	<i>Barbus altianalis</i>	BDL	6.13 ±0.427	BDL	BDL	BDL	BDL
	<i>Oreochromis niloticus</i>	BDL	BDL	BDL	BDL	BDL	BDL
18	<i>Barbus altianalis</i>	BDL	0.56 ±0.007	0.85 ±0.07	BDL	BDL	BDL
	<i>Clarias gariepinus</i>	BDL	1.43 ±0.107	BDL	BDL	BDL	BDL
	<i>Aethiomaster sembelus fretanus</i>	BDL	0.86 ±0.034	BDL	BDL	BDL	BDL
	<i>Oreochromis niloticus</i>	0.25 ±0.02	BDL	BDL	BDL	BDL	BDL
21	<i>Barbus altianalis</i>	BDL	BDL	BDL	BDL	BDL	BDL
	<i>Clarias liocephalus</i>	BDL	BDL	BDL	BDL	BDL	BDL
22	<i>Barbus altianalis</i>	BDL	BDL	BDL	BDL	BDL	BDL
23	<i>Barbus altianalis</i>	BDL	0.78 ±0.05	BDL	BDL	BDL	BDL
33	<i>Clarias gariepinus</i>	0.11 ±0.01	BDL	BDL	0.12±0.01 5	0.37 ±0.02	BDL
	<i>Oreochromis leucostictus</i>	BDL	0.17 ±0.03	BDL	0.12 ±0.01	0.72 ±0.06	BDL

Mean ± SD, Wet Weight (ng/g), Below Detection Limit (BDL)

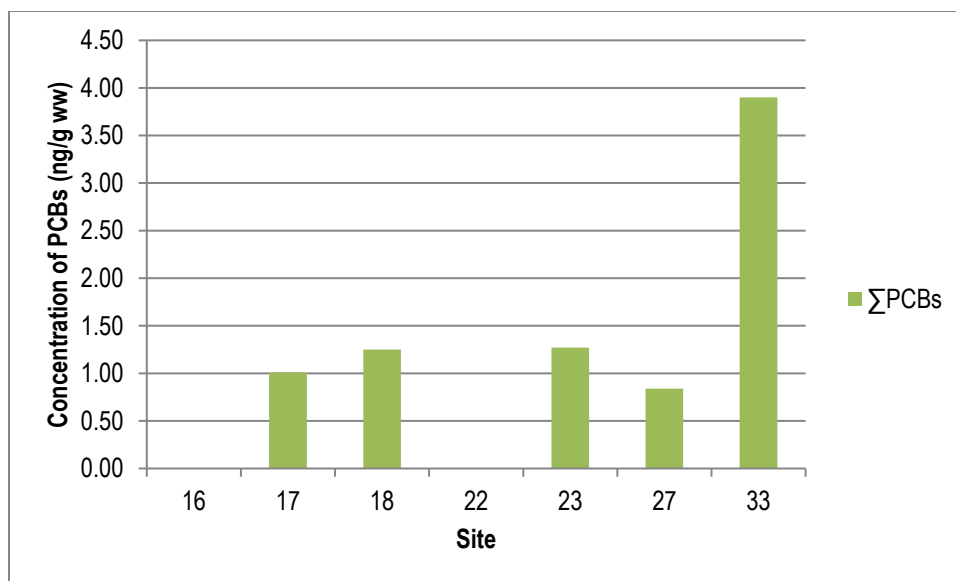
In December, residue levels of PCB 28 were detected with mean values ranging from 0.74±0.076 ng/g in *Labeo victorinus* at Site 18 to 0.25±0.019 ng/g in *Barbus altianalis* at Site 23. Residue levels of PCB 52 were detected with mean values ranging from 0.95±0.023 ng/g in *Clarias gariepinus* at Site 33 to 0.15±0.032 ng/g in *Oreochromis niloticus* at Site 33 (Table 20).

**Table 20: Polychlorinated biphenyls residue levels fish in December 2011**

SITE	SPECIES NAME	PCB 28	PCB 52	PCB 101	PCB 138	PCB 153	PCB 180
16	<i>Oreochromis niloticus</i>	BDL	BDL	BDL	BDL	BDL	BDL
17	<i>Barbus altianalis</i>	0.28±0.01	0.40±0.06	BDL	BDL	BDL	BDL
	<i>Clarias liocephalus</i>	0.33±0.01	BDL	BDL	BDL	BDL	BDL
18	<i>Labeo victorinus</i>	0.74±0.07	0.28±0.01	BDL	0.23±0.02	BDL	BDL
22	<i>Barbus altianalis</i>	BDL	BDL	BDL	BDL	BDL	BDL
23	<i>Barbus altianalis</i>	0.25±0.01	BDL	0.14±0.04	0.88±0.09	BDL	BDL
27	<i>Barbus neumayeri</i>	0.73±0.04	BDL	BDL	0.11±0.06	BDL	BDL
33	<i>Clarias gariepinus</i>	BDL	0.95±0.02	0.86±0.07	BDL	0.19±0.02	BDL
	<i>Oreochromis niloticus</i>	0.69±0.05	0.15±0.03	0.78±0.06	BDL	0.28±0.05	BDL

Mean ± SD, Wet Weight (ng/g), Below Detection Limit (BDL)

The variation of total PCBs detected were highest at site 33 (3.90 ng/g) and lowest at site 27 (0.84 ng/g) (Figure 24).



**Figure 24: Variation of polychlorinated biphenyl residues levels in fish in December 2011**

In March, residue levels of PCB 101 were detected with mean values ranging from 0.64±0.057 ng/g in *Labeo victorinus* at Site 17 to 0.16±0.093 ng/g in *Clarias gariepinus* at Site 18. Residue levels of PCB 153 were detected with mean values ranging from 0.51±0.058 ng/g in *Clarias gariepinus* at Site 18 to 0.47±0.082 ng/g in *Labeo victorinus* at Site 17 (Table 21).

**Table 21: Polychlorinated biphenyls residue levels fish in March 2012**

SITE	SPECIES NAME	PCB 28	PCB 52	PCB 101	PCB 138	PCB 153	PCB 180
15	<i>Clarias gariepinus</i>	BDL	0.86±0.08	0.24 ±0.02	BDL	BDL	BDL
	<i>Labeo victorinus</i>	0.84 ±0.02	BDL	BDL	0.35 ±0.05	BDL	BDL
16	<i>Clarias gariepinus</i>	0.15 ±0.07	BDL	0.53 ±0.03	BDL	BDL	BDL
	<i>Labeo victorinus</i>	0.12 ±0.06	BDL	0.64 ±0.05	BDL	BDL	BDL
	<i>Oreochromis niloticus</i>	BDL	0.42±0.02	0.59 ±0.05	BDL	BDL	BDL
17	<i>Barbus altianalis</i>	BDL	0.23±0.07	BDL	BDL	BDL	BDL
	<i>Clarias gariepinus</i>	0.41 ±0.08	BDL	BDL	0.37 ±0.06	BDL	BDL
	<i>Labeo victorinus</i>	BDL	BDL	BDL	BDL	0.47 ±0.08	BDL
18	<i>Barbus neumayeri</i>	BDL	BDL	BDL	BDL	BDL	BDL
	<i>Clarias gariepinus</i>	0.43 ±0.01	0.27 ±0.097	0.16 ±0.09	BDL	0.51 ±0.05	BDL
	<i>Labeo victorinus</i>	BDL	0.17 ±0.019	0.18 ±0.04	BDL	BDL	BDL
21	<i>Clarias liocephalus</i>	BDL	BDL	BDL	0.44 ±0.09	BDL	BDL

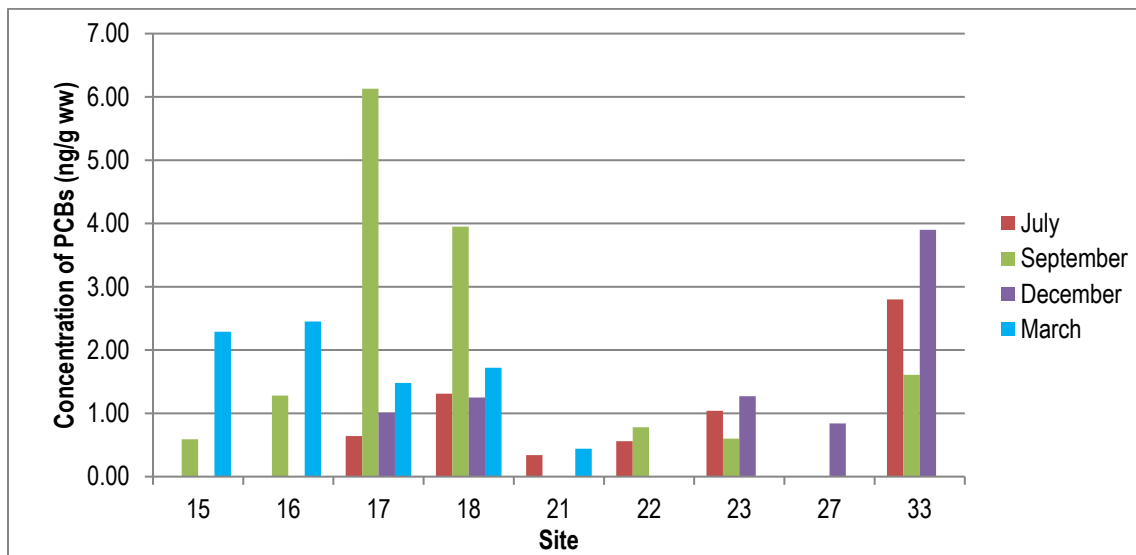
Mean ± SD, Wet Weight (ng/g), Below Detection Limit (BDL)

The variation of total PCBs detected were highest at site 16 (2.45 ng/g) and lowest at site 21 (0.44 ng/g) (Figure 25).



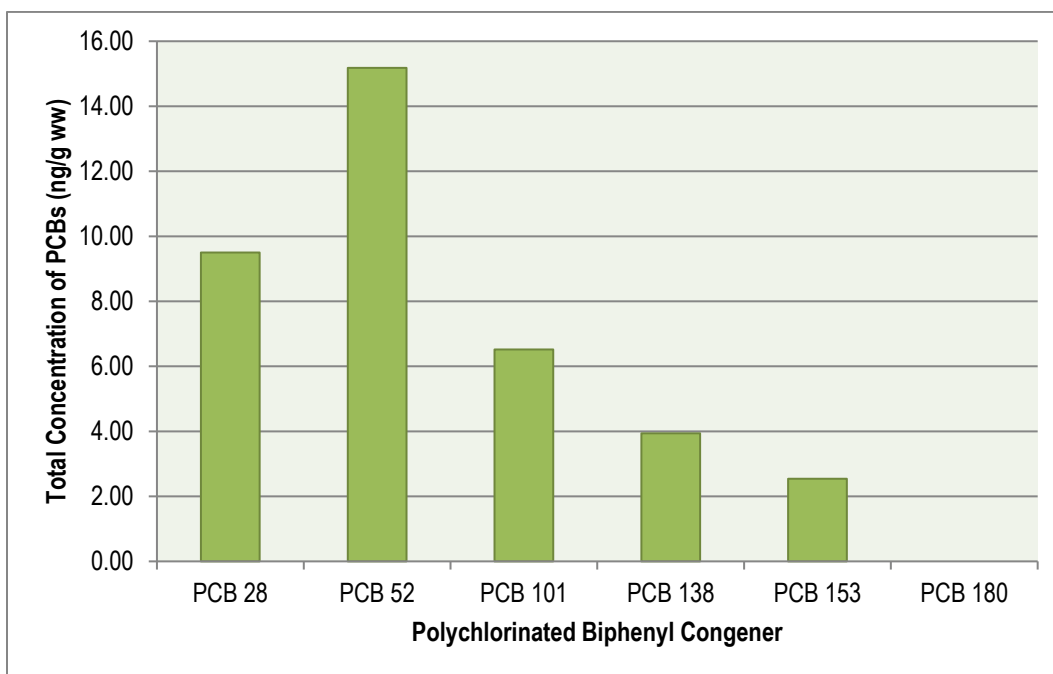
**Figure 25: Variation of polychlorinated biphenyl residues levels in fish in March 2012**

Over the four sampling periods, the variation of total PCBs detected were highest at site 17 (6.13 ng/g) in September and lowest at site 21 (0.34 ng/g) in July (Figure 19). Of the four sampling periods, September had the highest  $\Sigma$ PCB concentrations in 8 of the 9 sites where fish tissue samples were analysed. Of the 9 sites, Nyando at Dykes (Site 17) and Ahero Irrigation (Site 33) had consistently high  $\Sigma$ PCB concentrations in the four sampling periods ranging from 6.13 ng/g to 0.64 ng/g while Mbogo (Site 21) and Ainopngetuny (Site 22) had low concentrations ranging from 0.78 to 0.34 ng/g (Figure 26).



**Figure 26: Variation of polychlorinated biphenyl residues levels in fish species for the four sampling periods**

The concentrations of polychlorinated biphenyls in most of the analyzed fish species (Table 18 - 21) did not exceed the limit for human consumption (125 ng/g ww in muscle meat of caught wild freshwater fish) established by Commission Regulation (EU) 1259/2011 [EC, 2011]. All PCBs analysed were ubiquitous in fish tissue samples with the exception of PCB 180 which was below detection limit. The total concentration of organochlorine pesticide residues over the four sampling periods revealed the following pattern: PCB 52 > PCB 28 > PCB 101 > PCB 138 > PCB 153 (Figure 27).



**Figure 27: Total concentration of analysed PCB compounds for the four sampling periods**

## DISCUSSION

Fish are the most studied species in the aquatic environment [Negi and Mamgain, 2013]. The factors affecting survival of freshwater fish have been identified as modification and loss of aquatic habitat [Hewitt *et al.*, 2008]. 23 fish species from 5 orders and 7 families were recorded in the study sites. The family cyprinidae had the majority of the fish species (48%). The other families were clariidae, cichlidae, mochokidae, protopteridae, mastacembelidae and anabantidae. This is in contrast with studies by Darwall *et al.*, 2005 and Witte *et al.*, 2007 in which cichlids were found to be the most abundant and dominant species in Lake Victoria but is consistent with findings by Raburu, 2013. It can therefore be inferred that Nandi-Lower Nyando River is important in biodiversity conservation.

Sites at the lower reaches of the river (Sites 17, 18 and 33) had the highest richness of species compared to sites in the middle reaches of the river (Sites 22, 23 and 26). Studies on the distribution of fishes along the River Nyando show that more fish species occur in the river mouth wetlands than in the influent rivers upstream [Raburu, 2003]. Twelve major species were collected in the study sites and these included *Barbus altianalis*, *Barbus cercops*, *Barbus jacksoni*, *Barbus kerstenii*, *Barbus neumayeri*, *Barbus nyanzae*, *Clarius alluaudi*, *Clarias gariepinus*, *Clarias leocephalus*, *Labeo victorianus*, *Oreochromis leucostictus* and *Oreochromis niloticus*. Only one of these species (*Oreochromis niloticus*) was introduced, while the rest are indigenous, with *Barbus altianalis* being the most prevalent in the studied sites.

The rare species observed were; *Aethiomastersebelus fretanus*, *Barbus apleurogramma*, *Barbus carstino*, *Barbus paludinosus*, *Barbus Sp.*, *Ctenopoma murei*, *Haps Sp.*, *Protopterus aethiopicus*, *Pseudocrenilabrus multicolour*, *Synodontis victoriae* and *Xenoclaris eupogon*. The dominance and wide distribution of *Barbus Altianalis* can be attributed to its ecological tolerance; inhabiting diverse water habitats where it slowly matures over time to grow to a large size and a diet of various of food items that includes plant material and prey such as other fishes [Robins *et al.*, 1991 and Eccles, 1992]. The two indices used in this study give emphasis to different aspects of diversity. The Shannon index stresses the richness component whereas the Simpson index lays greater emphasis on the evenness component [Nagendra, 2002].

There was significant difference ( $P < 0.05$ ) in diversity indices between sampling sites with sites further downstream (sites 15, 16, 17, 18 and 33) providing higher values than sites in the middle reaches of the river (sites 21, 22, 23, 26 and 27). In general, the highest diversity and richness sites are served with urban discharge and water from rice farms of Ahero Irrigation Scheme where pesticides are regularly applied [Abong'o *et al.*, 2014]. The presence of fish species in the polluted areas may be due to their ability to survive the lasting impacts of pollutants. These species can therefore be termed as adaptive and resistant. For instance, the highly resilient cichlids were dominant at Ahero Irrigation (Site 33) while *Haplochromis Sp.* and *Pseudocrenilabrus multicolor* were only present at this sampling site. The low diversity and richness sites are characterised by large scale coffee and sugarcane farms with a maximum of four different species caught over the four sampling periods.

Herbicides, pesticide residues and fertilizers used in the large scale sugarcane farming in the middle reaches of the river contribute a significant proportion of pollutant loads [Kairu, 2001]. *Barbus altianalis* was predominant in Ainopngetuny (Site 22) with a high number of individuals (192) caught in July.

The redundancy analysis (RDA) biplot (Figure 12) was used to correlate fish species distribution and physico-chemical water quality parameters. The most influential and explanatory environmental variables were altitude, temperature and dissolved oxygen values. pH, TDS, turbidity, COD, BOD, temperature and phosphorus concentration positively influenced the presence of fish species. Altitude and dissolved oxygen negatively influenced fish diversity but were associated with the presence of *Barbus neumayeri* and *Clarias liocephalus* (Figure 12). Electrical conductivity is an important environmental variable to consider for fish community structures [Mondal *et al.*, 2010] since ions in water are an essential source of nutrients that support aquatic life [Galbrand *et al.*, 2008]. Findings of this study show that electrical conductivity had no significant impact on fish species distribution.

None of the 23 fish species found in this study was associated with high nutrient levels. Sampling sites with high phosphorous concentration levels (site 15, 16, 17 and 33) registered low fish species richness indicating that eutrophication reduced fish diversity. These findings are similar to Naigaga *et al.*, 2011 who observed that nutrient load influenced fish diversity in the Nakivubo wetland in Uganda.

*Barbus altianalis* was associated with high pH and total dissolved solids. Overall, the pH values of all sampling sites were within the normal range (pH 6.5 – 8.5). pH decreases can be attributed to the harvesting season when burning of fields releases acid forming ions (SO<sub>x</sub>, NO<sub>x</sub>) which are deposited on the land and enter the river as surface runoff [Bolan *et al.*, 2003].

Hexachlorohexanes (HCHs) exist in eight isomers. In this study four isomers were monitored;  $\alpha$ ,  $\gamma$ ,  $\beta$  and  $\delta$ -isomers. Among these,  $\gamma$  and  $\delta$ -isomers were the most widely detected with total mean concentrations of 269.17 ng/g and 189.76 ng/g respectively over the four sampling periods. The wide distribution of  $\gamma$ -HCH (Lindane) isomer in the fish tissue samples may be explained by its use as an insecticide in coffee and tea farms along lower Nyando drainage basin.  $\gamma$ -HCH can be easily degraded by microorganisms in soil and bottom sediments and photochemically isomerized to  $\alpha$ -isomer (Bhuvaneshwari and Rajendran, 2012).  $\alpha$ -HCH had a mean total concentration of 23.42 ng/g. The  $\beta$ -isomer is highly persistent in the environment and had a mean total concentration of 88.19 ng/g.  $\gamma$ -HCH was found in 8 fish species with mean concentrations ranging from 19.51 ng/g in *Oreochromis niloticus* at Site 33 in December to 1.12 ng/g in *Barbus altianalis* at Site 17 in July (Table 9-12). This finding indicates recent use of the pesticide rather than persistent residues from previous use of the HCH isomers in the lower Nyando river basin.

In this study p,p'-DDT, p,p'-DDD and p,p'-DDE were monitored in fish tissue samples. DDT tends to run off land into water ecosystems. p,p'-DDE is the most persistent metabolite due to its high lipophilicity and lower activity. As a result carnivore fish species accumulate the more degraded form (DDE) while feeding on phytoplankton and macrophyte consuming fish. The highest and lowest mean concentrations of p,p'-DDE were recorded in *Barbus altianalis* and ranged from 11.75 ng/g in at Site 23 in December to 0.41 ng/g in at Site 16 in March. *Barbus altianalis* is an omnivorous fish species, feeding mostly on water plants, and occasionally feeds on other fish [Robins *et al.*, 1991]. Residue levels of p,p'-DDE in analysed fish samples showed the pattern: *Barbus altianalis* > *Clarias gariepinus* > *Labeo victorianus* > *Oreochromis niloticus*. The present study therefore confirms biomagnification of persistent pollutants (e.g DDTs): with higher levels recorded in the carnivorous *Clarias gariepinus* and the lowest levels found in the herbivorous *Oreochromis niloticus*. Rognerud *et al.*, 2002 reported a high rate of biomagnification of p,p'-DDE in freshwater fish.



Endosulfan contains  $\alpha$ -endosulfan and  $\beta$ -endosulfan, two biologically active stereoisomers, mixed in a 7:3 ratio. In this study both isomers were monitored. Endosulfans were identified in 8% of the analysed fish species. The mean concentration of  $\beta$ -endosulfan ranged from not detectable (BDL) to 13.14 ng/g in *Oreochromis niloticus* at Site 33 in July.  $\alpha$ -endosulfan had the least mean total concentration (9.89 ng/g) of all OCPs (Figure 26). Mean concentrations ranged from not detectable (BDL) to 7.65 ng/g in *Oreochromis niloticus* at Site 17 in September. Input of endosulfan in aquatic food webs takes place mainly through plants to herbivore feeders [Fianko *et al.*, 2013]. This explains the high concentrations detected in *Oreochromis niloticus*.

Aldrin and dieldrin are mostly used for pest control [EPA 1980] and exhibit toxic action in insects by contact or ingestion. Aldrin was detected in 32% of fish samples at concentrations ranging from not detectable (BDL) to 11.95 ng/g in *Labeo victorinus* at Site 16 in July. Dieldrin was detected in 14% of fish samples at mean concentrations ranging from not detectable (BDL) to 6.32 ng/g in *Clarias gariepinus* at Site 15 in March. Levels of aldrin in fish were higher than those recorded for its metabolite, dieldrin. The high levels of aldrin in the study thus indicate recent exposure.

Endrin is primarily used as an insecticide [EPA, 1979] since 1951 to control pests in cotton and sugarcane farms. Endrin was detected in 22% of fish samples at concentrations ranging from not detectable (BDL) to 9.96 ng/g in *Labeo victorinus* at Site 18 in December. Endrin aldehyde was detected in 12% of fish samples at mean concentrations ranging from not detectable (BDL) to 4.01 ng/g in *Synodontis victoriae* at Site 17 in July. Levels of endrin in fish were higher than those recorded for its metabolite, endrin aldehyde. The high levels of endrin in this study thus indicate recent exposure.

Heptachlor is primarily used as an insecticide to kill insects in seed grains and on crops [IPCS, 2006]. Heptachlor was detected in 28% of fish samples at concentrations ranging from not detectable (BDL) to 8.37 ng/g in *Barbus altianalis* at Site 23 in September. Heptachlor epoxide was detected in 8% of fish samples at mean concentrations ranging from not detectable (BDL) to 17.24 ng/g in *Oreochromis niloticus* at Site 18 in September. Levels of heptachlor in fish were lower than those recorded for its oxidative metabolite, heptachlor epoxide.

Occurrence of the six indicator non dioxin-like polychlorinated biphenyls (PCB 28, 52, 101, 138, 153, and 180) was monitored in this study. PCBs were detected in 74% of fish samples. PCB congeners 28, 52, 101, 138 and 153) were found, with PCB 52 as the major contributor (40%) to the  $\Sigma$ PCBs in the analysed samples. This is contrary to Oluoch-Otiego *et al.* (2016) who found PCB 153 (27 – 35%) to be the most abundant congener in three fish species (*Oreochromis niloticus*, *Lates niloticus*, and *Rastrineobola argentea*) from Winam Gulf of Lake Victoria. Abundance of congeners with a lower degree of chlorination (e.g PCB 52) is an indication of higher chemical degradation rates along lower Nyando. The highest level of PCBs was recorded in *Barbus altianalis* from Nyando at Dykes (Site 17) with a mean concentration of 6.13 ng/g ww. Considering the low solubility of PCBs in water, most of the PCBs in fish should be derived from food rather than from ambient environment [Brázová *et al.*, 2012].

Consequently, PCB residue levels ranging from 0.15 - 0.78 ng/g were observed in *Oreochromis niloticus*, an herbivorous species that feeds mainly on algae [Deribe *et al.*, 2011]. The mean PCB concentrations in this study are higher than those reported by Ssebugere *et al.* (2014), ranging from 41 to 670 pg g<sup>-1</sup>, in two commercial fish species (*Labeo Niloticus* and *Oreochromis Niloticus*) from Napoleon Gulf on the Ugandan side of Lake Victoria.

Mean concentrations of  $\Sigma$ PCBs in the fishes were as follows: *Barbus altianalis* > *Clarias gariepinus* > *Labeo victorianus* > *Oreochromis niloticus* for the analysed fish species. The results obtained in the present study show higher concentrations of PCBs in carnivorous fish species than in bottom feeders. Similar findings by Ssebugere *et al.* (2014) and Oluoch-Otiego *et al.* (2016) suggest that PCBs can biomagnify to species higher up in the food web.

## CHAPTER FIVE

### CONCLUSION AND RECCOMENDATIONS

#### 5.1 Conclusion

This study achieved its objectives of determining the fish species of lower Nyando, correlating their occurrence to water quality parameters and quantifying the chemical pollution in the basin. In this regard, baseline data that can enable development of a monitoring plan for lower Nyando has been generated. Results obtained in this study can catalyse inception of similar studies in other waterways and the lake.

Various sources of pollution have been identified with severe pollution problems noted closer to the river mouth (from Site 26 heading downstream). However, distribution of fishes along the Nandi-Lower Nyando River has shown that more fish species occur closer to the river mouth wetlands than in the influent rivers upstream. Six major species were collected in the sampling sites with *Barbus altianalis* being the most prevalent. The family cyprinidae had the majority of the fish species (48%).

Water quality parameters strongly influenced fish species diversity as was evident in this study. Human activities and farming practices along the Nandi-Lower Nyando River sub-catchment were shown to have resulted in increased pollutant and nutrient load which in turn affected fish diversity. Low diversity and richness sites were characterised by large scale coffee and sugarcane farming. Adaptive and resilient fish species were predominant in high diversity and richness sites most of which are served with urban discharge and water from rice farms in Ahero Irrigation Scheme. Fish diversity is therefore a useful indicator of ecological integrity in freshwater ecosystems.

The study also analyzed OCPs and PCBs in fish species from the Nandi-Lower Nyando River sub-catchment. The results revealed that aldrin, endrin and heptachlor residue levels were higher than those of their metabolites dieldrin, endrin aldehyde and heptachlor epoxide which may indicate recent exposure to these pollutants.

$\gamma$ -HCH (Lindane) was found to be the most widely distributed hexachlorohexane isomer in the fish tissue samples; a finding attributed to its use as an insecticide in coffee and tea farms along Nandi-Lower Nyando drainage basin. The difference in concentration levels among the sampled species may be due to the different feeding habits and trophic level. Generally the ability of fish to metabolize these pollutants is moderate therefore contaminant loading in fish is reflective of the state of pollution in the surrounding environment.

The tendency of pollutants to bioaccumulate was confirmed in some fish species with higher concentration levels of DDT metabolite p,p'-DDE and PCBs recorded in predator fish *Barbus altianalis* and *Clarias gariepinus*. The data presented is essential in predicting the risk of higher rates of bioaccumulation that would endanger the health of humans along the Nandi-Lower Nyando River sub-catchment.

## **5.2 Recommendations**

### **5.2.1 Research Recommendations**

- Complimentary studies to determine point and non-point sources of POPs along the drainage basin especially around Ahero Irrigation Scheme should be initiated.
- Bioaccumulation studies should be conducted to human beings and animal to determine the degree of absorption of these POPs.

### **5.2.2 Policy Recommendations**

- Increased focus on use of alternative pesticides such as organophosphates that do not persist in the environment.
- Implementation of a monitoring plan for River Nyando by the relevant government organs to keep track of the changing environmental conditions.

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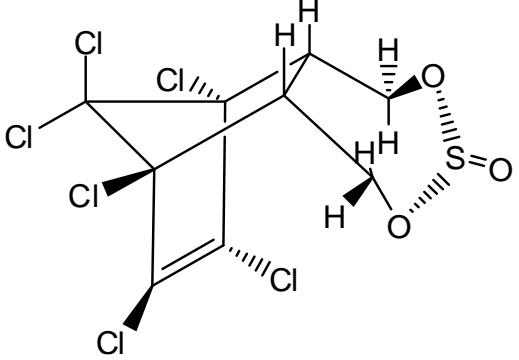
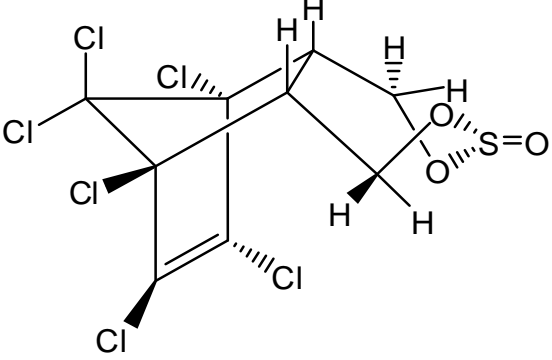
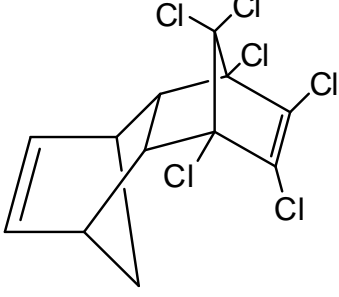
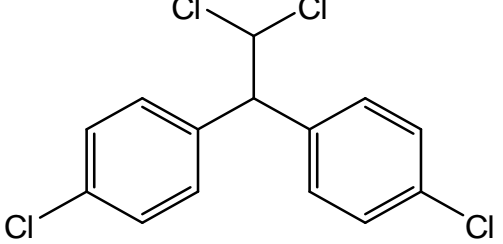
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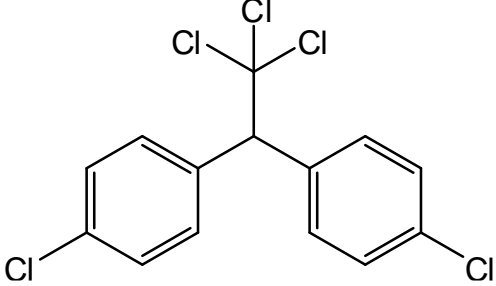
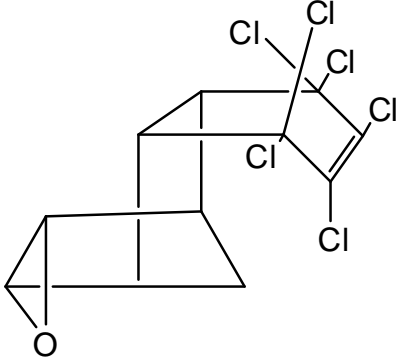
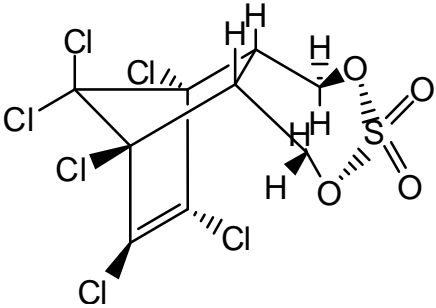
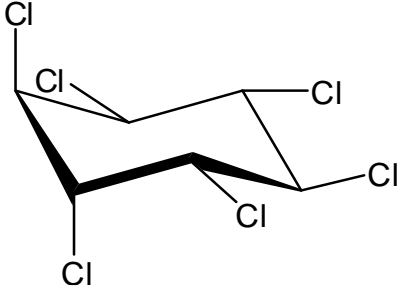
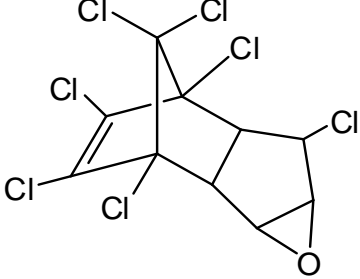
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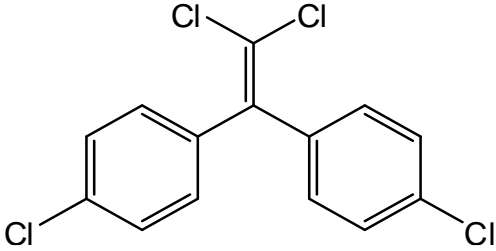
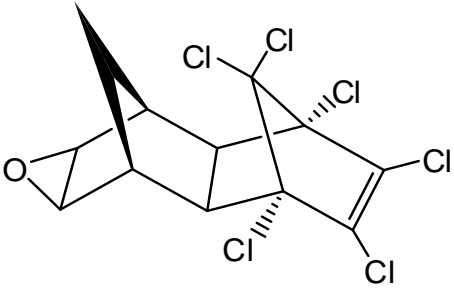
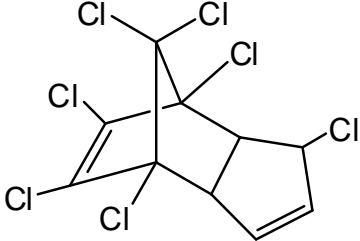
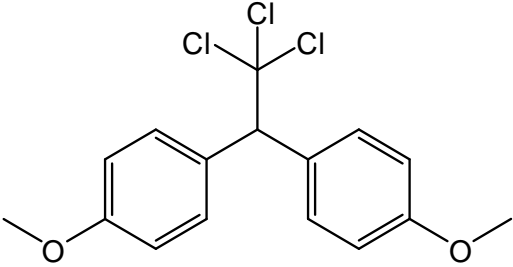


## APPENDICES

### Appendix I: Structures of organochlorine pesticides studied

SN	Name	Structures
	$\alpha$ -Endosulphan	
	$\beta$ -Endosulphan	
	Aldrin	
	(1,1-dichloro-2,2-bis (4-chlorophenyl) ethane) p,p'-DDD	

	<p>dichloro-diphenyl-trichloro-ethane (DDT)</p>	
	<p>Dieldrin</p>	
	<p>EndosulphanSulfate</p>	
	<p>hexachlorocyclohexane (HCH),</p>	
	<p>Heptachlor epoxide</p>	

	(2,2-bis p-chlorophenyl, 1-dichloroethylene) p,p'-DDE	
	Endrin	
	Heptachlor	
	Methoxychlor	

**Figure 1a: Structures of organochlorine pesticides studied**

## Appendix II: Weights and lengths of fish species in Lower Nyando

**Table 2a: Weights and lengths of fish species in Lower Nyando in July 2011**

	15		16		17		18		21		22		23		26		33	
	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)
Aethiomastersebelus Fretanus			15.5	19.1														
Barbus Altianalis	133.8	18.2	61.1	0.0	7.2	9.1			114.0	15.6	122.2	23.5	66.0	19.0				
Barbus Apleurogramma																		
Barbus Carstino					14.0													
Barbus Cercops																		
Barbus Jacksoni																		
Barbus Kerstenii																		
Barbus Neumayeri													26.3	14.3				
Barbus Nyanzae	33.7		60.7		9.7													
Barbus Paludinosus																		
Barbus S.P																18.2	11.9	
Clarius Aluaudi																		
Clarias Gariepinus					600.0	42.6	263.7	27.1	1464.5	56.9								
Clarias Liocephalus											43.7	17.8	14.3	11.4	17.8	14.7		
Ctenopoma Murei																		
Haps S.P																		
Labeo Victorianus	77.6	18.5	75.1	15.5														
Oreochromis Leucostictus	1.4																	
Oreochromis Niloticus			18.6	121.7	13.0		6.7	7.6										
Protopterus Aethiopicus																		
Pseudocrenilabrus Multicolour																		
Synodontis Victoriae																		
Xenoclaris Eupognon																		

**Table 2b: Weights and lengths of fish species in Lower Nyando in September 2011**

	15		16		17		18		21		22		23		26		27		33		
	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)	
Aethiomastersebelus Fretanus					2.1	9.5	25.7	23.3													
Barbus Altianalis	14.6	8.8	9.0	8.8	70.6	17.7	100.3	16.0	53.4	13.7	86.7	16.0	84.8	15.4							
Barbus Apleurogramma																					
Barbus Carstino			0.6	3.9																	
Barbus Cercorps	1.9	5.9			1.8	5.5	1.8	5.6	1.8	5.7	3.1	6.2									
Barbus Jacksoni					11.3	10.4	7.3	8.3													
Barbus Kerstenii																					
Barbus Neumayeri							180.5	23.3			9.2	9.0									
Barbus Nyanzae	3.3	7.0			3.8	7.4	4.4	7.7												4.0	6.9
Barbus Paludinosus																					
Barbus S.P									0.8	4.6			6.7	8.3	14.0	10.3	6.6	8.3	0.6	3.6	
Clarius Alluadi					14.6	11.9															
Clarias Gariepinus							292.5	33.2												37.3	17.7
Clarias Liocephalus													16.1	12.4	28.0	15.5					
Ctenopoma Murei																					
Haps S.P																				2.3	5.3
Labeo Victorianus	14.4	11.4			54.4	17.4	118.3	17.0													
Oreochromis Leocostictus	0.9	3.8																		12.7	7.3
Oreochromis Niloticus			74.2	13.5	57.2	11.0															
Protopterus Aethiopicus																					
Pseudocrenilabrus Multicolour																				2.7	5.0
Synodontis Victoriae							28.9	14.3													
Xenoclarias Eupogon																				1.5	5.7

**Table 2c: Weights and lengths of fish species in Lower Nyando in December 2011**

	15		16		17		18		21		22		23		26		27		33	
	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)
Aethiomastersebelus Fretanus							15.3	20.0												
Barbus Altianalis	6.1	6.7	1.8	6.2	51.7	15.8	6.6	8.1	8.3	8.8	36.1	13.3	41.4	12.7						
Barbus Apleurogramma																				
Barbus Carstino																				
Barbus Cercorps	0.8	4.3			0.8	4.5	0.8	4.3												
Barbus Jacksoni																				
Barbus Kerstenii									2.6	6.6										
Barbus Neumayeri							8.0	9.8			8.1	9.5	8.8	9.1	10.6	10.1	9.6	7.6		
Barbus Nyanzae	5.1	8.2	4.0	7.4	2.9	6.8	2.8	6.5											4.0	6.9
Barbus Paludinosus																				
Barbus S.P					9.2	10.0	4.4	7.6											0.6	3.6
Clarius Alluadi																				
Clarias Gariepinus																			37.3	17.7
Clarias Liocephalus			4.1	7.6	21.7	16.4								13.9	13.2					
Ctenopoma Murei																				
Haps S.P																			2.3	5.3
Labeo Victorianus	1.3	5.1			14.8	10.3	21.1	11.3												
Oreochromis Leocostictus																			12.7	7.3
Oreochromis Niloticus	43.1	13.4	128.6	18.5																
Protopterus Aethiopicus																				
Pseudocrenilabrus Multicolour																			2.7	5.0
Synodontis Victoriae																				
Xenoclarias Eupogon																			1.5	5.7

**Table 2d: Weights and lengths of fish species in Lower Nyando in March 2012**

	15		16		17		18		21		22		23		26		27		33	
	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)	Weight (g)	Length (cm)
Aethiomastersebelus Fretanus							15.3	20.0												
Barbus Altianalis	6.1	6.7	1.8	6.2	51.7	15.8	6.6	8.1	8.3	8.8	36.1	13.3	41.4	12.7						
Barbus Apleurogramma																				
Barbus Carstino																				
Barbus Cercorps	0.8	4.3			0.8	4.5	0.8	4.3												
Barbus Jacksoni																				
Barbus Kerstenii																				
Barbus Neumayeri							8.0	9.8			8.1	9.5	8.8	9.1	10.6	10.1	9.6	7.6		
Barbus Nyanzae	5.1	8.2	4.0	7.4	2.9	6.8	2.8	6.5											4.0	6.9
Barbus Paludinosus							2.6	6.6												
Barbus S.P					9.2	10.0	4.4	7.6											0.6	3.6
Clarius Alluadi																				
Clarias Gariepinus																			37.3	17.7
Clarias Liocephalus			4.1	7.6	21.7	16.4								13.9	13.2					
Ctenopoma Murei																				
Haps S.P																			2.3	5.3
Labeo Victorianus	1.3	5.1			14.8	10.3	21.1	11.3												
Oreochromis Leocostictus	13.1	4.4																	12.7	7.3
Oreochromis Nilotitus			28.6	18.5																
Protopterus Aethiopicus																				
Pseudocrenilabrus Multicolour																			2.7	5.0
Synodontis Victoriae																				
Xenoclarias Eupogon																			1.5	5.7

### Appendix III: Occurrence and distribution of fish species in Lower Nyando

Table 3a: Occurrence and distribution of fish species in Lower Nyando in July 2011

TAXON/SITE	15	16	17	18	19	21	22	23	25	26	27	30	33
<b>Anabantidae</b>													
Ctenopoma muriei	A	A	A	A	A	A	A	A	A	A	A	A	P
<b>Cichlidae</b>													
Oreochromis leucostictus	P	A	A	A	A	A	A	A	A	A	A	A	A
Oreochromis niloticus	A	P	P	P	A	A	A	A	A	A	A	A	P
Haplochromis Sp.	A	A	A	A	A	A	A	A	A	A	A	A	P
Pseudocrenilabrus multicolor	A	A	A	A	A	A	A	A	A	A	A	A	P
<b>Clariidae</b>													
Clarias alluaudi	A	A	A	P	A	P	A	A	A	A	A	A	P
Clarias gariepinus	A	A	P	P	A	P	A	A	A	A	A	A	A
Clarias liocephalus	A	A	A	A	A	A	P	P	A	P	A	A	A
Xenoclarias eupogon	A	A	A	A	A	A	A	A	A	A	A	A	A
<b>Cyprinidae</b>													
Barbus altianalis	P	P	P	P	A	P	P	P	A	A	A	A	A
Barbus apleurogramma	A	A	A	A	A	A	A	A	A	A	A	A	A
Barbus carstino	A	A	A	A	A	P	A	A	A	A	A	A	A
Barbus cercops	A	A	P	P	A	P	A	A	A	A	A	A	P
Barbus jacksoni	A	A	A	P	A	P	A	A	A	A	A	A	A
Barbus kerstenii	A	A	A	A	A	A	A	A	A	A	A	A	P
Barbus neumayeri	A	A	A	A	A	P	A	P	A	P	A	A	A
Barbus nyanzae	P	P	P	P	A	A	A	A	A	A	A	A	A
Barbus paludinosus	A	A	A	A	A	A	A	A	A	A	A	A	A
Barbus Sp.	A	A	A	A	A	A	P	P	A	P	A	A	A
Labeo victorinus	P	P	P	P	A	A	A	A	A	A	A	A	A
<b>Mastacembelidae</b>													
Aethiomastacembelus frenatus	A	P	A	P	A	A	A	A	A	A	A	A	A
<b>Mochokidae</b>													
Synodontis victoriae	A	A	P	A	A	A	A	A	A	A	A	A	A
<b>Protopteridae</b>													
Protopterus aethiopicus	A	A	A	A	A	A	A	A	A	A	A	A	P
<b>TOTAL</b>	<b>4</b>	<b>5</b>	<b>7</b>	<b>9</b>	<b>0</b>	<b>7</b>	<b>3</b>	<b>4</b>	<b>0</b>	<b>3</b>	<b>0</b>	<b>0</b>	<b>8</b>

P = Present, A = Absent



**Table 3b: Occurrence and distribution of fish species in Lower Nyando in September 2011**

<b>TAXON/SITE</b>	<b>15</b>	<b>16</b>	<b>17</b>	<b>18</b>	<b>19</b>	<b>21</b>	<b>22</b>	<b>23</b>	<b>25</b>	<b>26</b>	<b>27</b>	<b>30</b>	<b>33</b>
<b>Anabantidae</b>													
Ctenopoma muriei	A	A	A	A	A	A	A	A	A	A	A	A	P
<b>Cichlidae</b>													
Oreochromis leucostictus	P	A	A	A	A	A	A	A	A	A	A	A	P
Oreochromis niloticus	A	P	P	P	A	A	A	A	A	A	A	A	P
Haplochromis Sp.	A	A	A	A	A	A	A	A	A	A	A	A	P
Pseudocrenilabrus multicolor	A	A	A	A	A	A	A	A	A	A	A	A	P
<b>Clariidae</b>													
Clarias alluaudi	A	A	P	A	A	A	A	A	A	A	A	A	P
Clarias gariepinus	A	A	A	P	A	A	A	A	A	A	A	A	P
Clarias liocephalus	A	A	A	A	A	P	P	P	A	P	A	A	A
Xenoclaris eupogon	A	A	A	A	A	A	A	A	A	A	A	A	P
<b>Cyprinidae</b>													
Barbus altianalis	P	P	P	P	A	P	P	P	A	A	A	A	A
Barbus apleurogramma	A	A	A	A	A	A	A	A	A	A	A	A	A
Barbus carstino	A	P	A	A	A	A	A	A	A	A	A	A	A
Barbus cercops	P	A	P	P	A	P	P	A	A	A	A	A	A
Barbus jacksoni	A	A	P	P	A	A	A	A	A	A	A	A	A
Barbus kerstenii	A	A	A	P	A	A	A	A	A	A	A	A	A
Barbus neumayeri	A	A	A	A	A	P	P	A	A	A	A	A	A
Barbus nyanzae	P	A	P	P	A	A	A	A	A	A	A	A	P
Barbus paludinosus	A	A	A	A	A	A	A	A	A	A	A	A	A
Barbus Sp.	A	A	A	A	A	P	A	P	A	P	P	A	P
Labeo victorianus	P	A	P	P	A	A	A	A	A	A	A	A	A
<b>Mastacembelidae</b>													
Aethiomastacembelus frenatus	A	A	P	P	A	A	A	A	A	A	A	A	A
<b>Mochokidae</b>													
Synodontis victoriae	A	A	A	P	A	A	A	A	A	A	A	A	A
<b>Protopteridae</b>													
Protopterus aethiopicus	A	A	A	A	A	A	A	A	A	A	A	A	A
<b>TOTAL</b>	<b>5</b>	<b>3</b>	<b>8</b>	<b>10</b>	<b>0</b>	<b>5</b>	<b>4</b>	<b>3</b>	<b>0</b>	<b>2</b>	<b>1</b>	<b>0</b>	<b>9</b>

P = Present, A = Absent

**Table 3c: Occurrence and distribution of fish species in Lower Nyando in December 2011**

<b>TAXON/SITE</b>	<b>15</b>	<b>16</b>	<b>17</b>	<b>18</b>	<b>19</b>	<b>21</b>	<b>22</b>	<b>23</b>	<b>25</b>	<b>26</b>	<b>27</b>	<b>30</b>	<b>33</b>
<b>Anabantidae</b>													
Ctenopoma muriei	A	A	A	A	A	A	A	A	A	A	A	A	P
<b>Cichlidae</b>													
Oreochromis leucostictus	A	A	A	A	A	A	A	A	A	A	A	A	P
Oreochromis niloticus	P	P	A	A	A	A	A	A	A	A	A	A	P
Haplochromis Sp.	A	A	A	A	A	A	A	A	A	A	A	A	P
Pseudocrenilabrus multicolor	A	A	A	A	A	A	A	A	A	A	A	A	P
<b>Clariidae</b>													
Clarias alluaudi	A	A	A	A	A	A	A	A	A	A	A	A	P
Clarias gariepinus	A	A	A	A	A	A	A	A	A	A	A	A	P
Clarias liocephalus	A	P	P	A	A	P	A	A	A	P	A	A	A
Xenoclaris eupogon	A	A	A	A	A	A	A	A	A	A	A	A	A
<b>Cyprinidae</b>													
Barbus altianalis	P	P	P	P	A	P	P	P	A	A	A	A	A
Barbus apleurogramma	A	A	A	A	A	A	A	A	A	A	A	A	P
Barbus carstino	A	A	A	A	A	A	A	A	A	A	A	A	A
Barbus cercops	P	A	P	P	A	A	A	A	A	A	A	A	A
Barbus jacksoni	A	A	A	A	A	A	A	A	A	A	A	A	A
Barbus kerstenii	A	A	A	A	A	P	A	A	A	A	A	A	P
Barbus neumayeri	A	A	A	P	A	A	P	P	A	P	P	A	A
Barbus nyanzae	P	P	P	P	A	A	A	A	A	A	A	A	A
Barbus paludinosus	A	A	A	A	A	A	A	A	A	A	A	A	A
Barbus Sp.	A	A	P	P	A	A	A	A	A	A	A	A	P
Labeo victorianus	P	A	P	P	A	A	A	A	A	A	A	A	A
<b>Mastacembelidae</b>													
Aethiomastacembelus frenatus	A	A	A	P	A	A	A	A	A	A	A	A	A
<b>Mochokidae</b>													
Synodontis victoriae	A	A	A	A	A	A	A	A	A	A	A	A	A
<b>Protopteridae</b>													
Protopterus aethiopicus	A	A	A	A	A	A	A	A	A	A	A	A	A
<b>TOTAL</b>	<b>5</b>	<b>4</b>	<b>6</b>	<b>7</b>	<b>0</b>	<b>3</b>	<b>2</b>	<b>2</b>	<b>0</b>	<b>2</b>	<b>1</b>	<b>0</b>	<b>10</b>

P = Present, A = Absent

**Table 3d: Occurrence and distribution of fish species in Lower Nyando in March 2012**

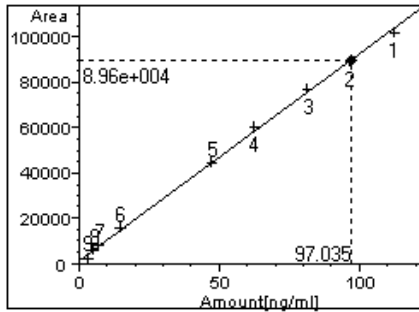
<b>TAXON/SITE</b>	<b>15</b>	<b>16</b>	<b>17</b>	<b>18</b>	<b>19</b>	<b>21</b>	<b>22</b>	<b>23</b>	<b>25</b>	<b>26</b>	<b>27</b>	<b>30</b>	<b>33</b>
<b>Anabantidae</b>													
Ctenopoma muriei	A	A	A	A	A	A	A	A	A	A	A	A	P
<b>Cichlidae</b>													
Oreochromis leucostictus	A	A	A	A	A	A	A	A	A	A	A	A	P
Oreochromis niloticus	P	P	P	A	A	A	A	A	A	A	A	A	A
Haplochromis Sp.	A	A	A	P	A	A	A	A	A	A	A	A	A
Pseudocrenilabrus multicolor	A	A	A	A	A	A	A	A	A	A	A	A	P
<b>Clariidae</b>													
Clarias alluaudi	A	A	A	A	A	A	A	A	A	A	A	A	P
Clarias gariepinus	P	P	P	P	A	A	A	P	A	A	A	A	P
Clarias liocephalus	A	A	A	A	A	P	P	P	A	P	P	A	A
Xenoclaris eupogon	A	A	A	A	A	A	A	A	A	A	A	A	A
<b>Cyprinidae</b>													
Barbus altianalis	A	P	P	P	A	A	P	P	A	A	A	A	A
Barbus apleurogramma	A	A	P	A	A	A	A	A	A	A	A	A	P
Barbus carstino	A	A	A	A	A	A	A	A	A	A	A	A	A
Barbus cercops	A	A	P	P	A	P	A	A	A	A	A	A	A
Barbus jacksoni	A	A	A	A	A	A	A	A	A	A	A	A	A
Barbus kerstenii	A	A	A	A	A	A	A	A	A	A	A	A	A
Barbus neumayeri	A	A	A	A	A	A	P	P	A	P	P	A	A
Barbus nyanzae	A	P	A	P	A	A	A	A	A	A	A	A	A
Barbus paludinosus	A	A	P	P	A	A	A	A	A	A	A	A	P
Barbus Sp.	P	A	A	A	A	A	P	A	A	A	A	A	A
Labeo victorianus	P	P	P	P	A	A	A	A	A	A	A	A	A
<b>Mastacembelidae</b>													
Aethiomastacembelus frenatus	A	P	A	P	A	A	A	A	A	A	A	A	A
<b>Mochokidae</b>													
Synodontis victoriae	A	A	P	A	A	A	A	A	A	A	A	A	A
<b>Protopteridae</b>													
Protopterus aethiopicus	A	A	A	A	A	A	A	A	A	A	A	A	P
<b>TOTAL</b>	<b>4</b>	<b>6</b>	<b>8</b>	<b>8</b>	<b>0</b>	<b>2</b>	<b>4</b>	<b>4</b>	<b>0</b>	<b>2</b>	<b>2</b>	<b>0</b>	<b>8</b>

P = Present, A = Absent

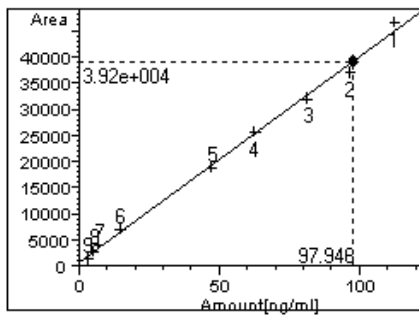
## Appendix IV: Calibration Curves

Data File C:\HPCHEM\1\DATA\JA2015S3\102F0702.D

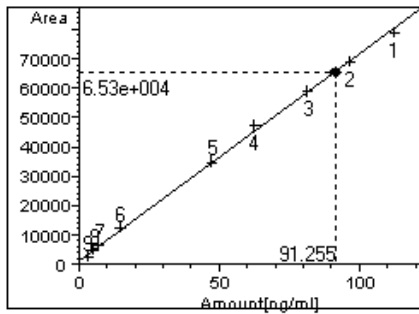
Sample Name: PCB0CPSTD13



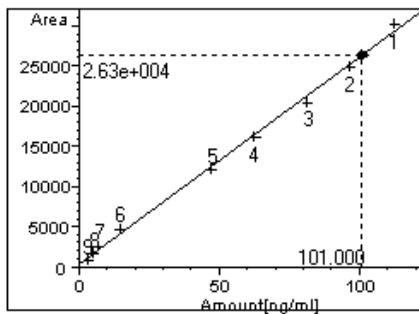
d-HCH at exp. RT: 7.927  
 ECD1 B,  
 Correlation: 0.99944  
 Residual Std. Dev.: 1386.48940  
 Formula:  $y = mx + b$   
 m: 910.70940  
 b: 1262.62438  
 x: Amount[ng/ml]  
 y: Area



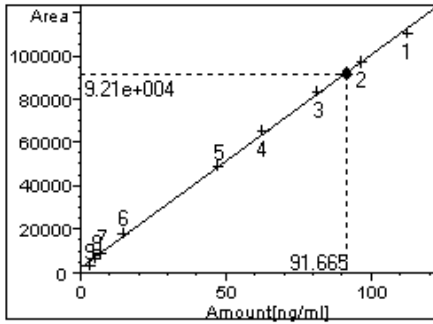
PCB 28 at exp. RT: 8.399  
 ECD1 B,  
 Correlation: 0.99834  
 Residual Std. Dev.: 1029.47756  
 Formula:  $y = mx + b$   
 m: 393.00663  
 b: 694.40971  
 x: Amount[ng/ml]  
 y: Area



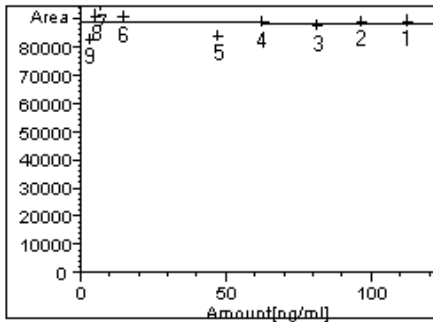
Heptachlor at exp. RT: 8.658  
 ECD1 B,  
 Correlation: 0.99949  
 Residual Std. Dev.: 1021.03778  
 Formula:  $y = mx + b$   
 m: 700.75701  
 b: 1378.52650  
 x: Amount[ng/ml]  
 y: Area



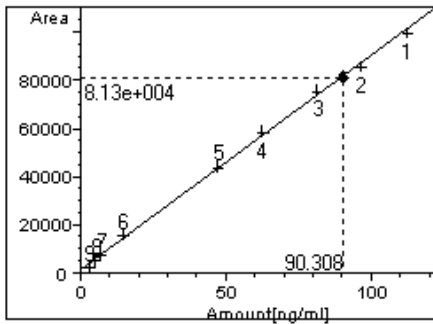
PCB 52 at exp. RT: 8.980  
 ECD1 B,  
 Correlation: 0.99864  
 Residual Std. Dev.: 611.00851  
 Formula:  $y = mx + b$   
 m: 257.10527  
 b: 320.96023  
 x: Amount[ng/ml]  
 y: Area



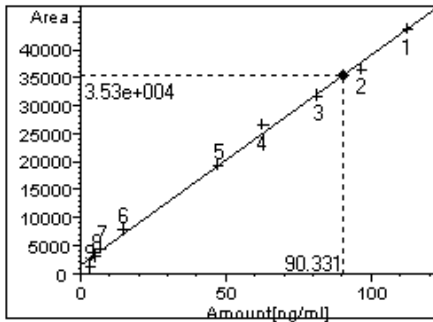
Aldrin at exp. RT: 9.301  
 ECD1 B,  
 Correlation: 0.99933  
 Residual Std. Dev.: 1633.09070  
 Formula:  $y = mx + b$   
 m: 981.02436  
 b: 2128.89116  
 x: Amount[ng/ml]  
 y: Area



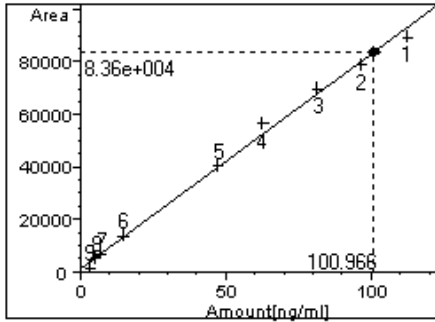
Isodrin at exp. RT: 9.935  
 ECD1 B,  
 Correlation: 0.11143  
 Residual Std. Dev.: 3859.03809  
 Formula:  $y = mx + b$   
 m: -9.51706  
 b: 89009.54837  
 x: Amount[ng/ml]  
 y: Area



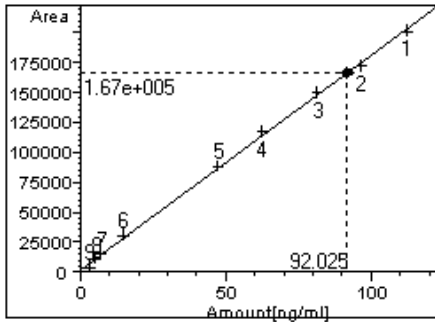
Heptachlor Epoxide at exp. RT: 10.205  
 ECD1 B,  
 Correlation: 0.99930  
 Residual Std. Dev.: 1511.71144  
 Formula:  $y = mx + b$   
 m: 887.47346  
 b: 1124.82469  
 x: Amount[ng/ml]  
 y: Area



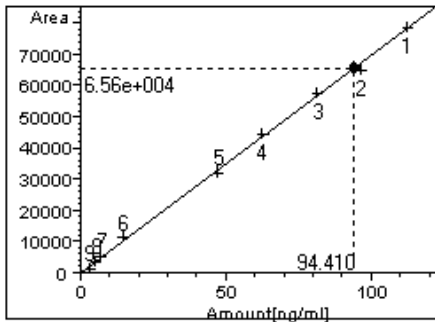
PCB 101 at exp. RT: 11.139  
 ECD1 B,  
 Correlation: 0.99826  
 Residual Std. Dev.: 1008.58055  
 Formula:  $y = mx + b$   
 m: 375.54309  
 b: 1369.51926  
 x: Amount[ng/ml]  
 y: Area



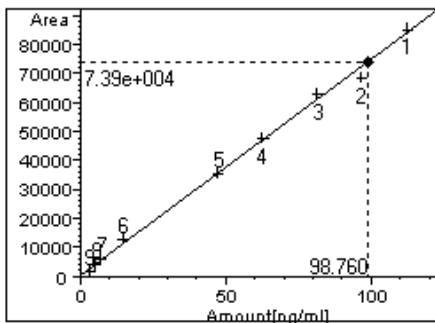
Endosulfan I at exp. RT: 11.236  
 ECD1 B,  
 Correlation: 0.99798  
 Residual Std. Dev.: 2359.86448  
 Formula:  $y = mx + b$   
 m: 815.73116  
 b: 1269.97548  
 x: Amount[ng/ml]  
 y: Area



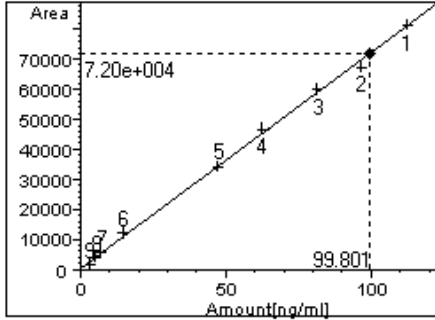
p,p-DDE at exp. RT: 12.160  
 ECD1 B,  
 Correlation: 0.99945  
 Residual Std. Dev.: 2680.41088  
 Formula:  $y = mx + b$   
 m: 1783.97743  
 b: 2461.07495  
 x: Amount[ng/ml]  
 y: Area



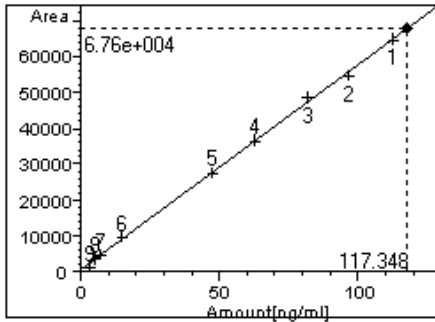
Dieldrin at exp. RT: 12.993  
 ECD1 B,  
 Correlation: 0.99937  
 Residual Std. Dev.: 1120.53243  
 Formula:  $y = mx + b$   
 m: 692.25161  
 b: 196.71217  
 x: Amount[ng/ml]  
 y: Area



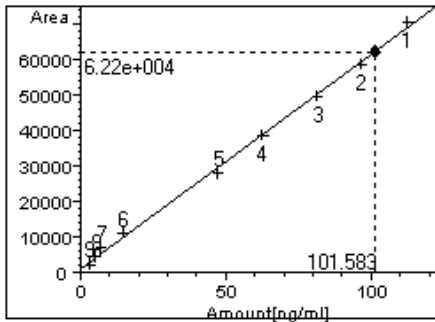
Endrin at exp. RT: 13.413  
 ECD1 B,  
 Correlation: 0.99861  
 Residual Std. Dev.: 1784.50662  
 Formula:  $y = mx + b$   
 m: 743.68001  
 b: 444.07663  
 x: Amount[ng/ml]  
 y: Area



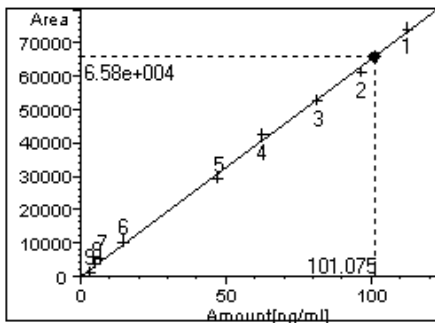
Endosulfan II at exp. RT: 14.069  
 ECD1 B,  
 Correlation: 0.99931  
 Residual Std. Dev.: 1206.97597  
 Formula:  $y = mx + b$   
 m: 715.68594  
 b: 524.42482  
 x: Amount[ng/ml]  
 y: Area



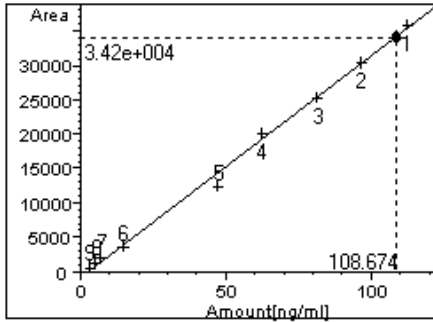
p,p'-DDD at exp. RT: 14.349  
 ECD1 B,  
 Correlation: 0.99952  
 Residual Std. Dev.: 810.43579  
 Formula:  $y = mx + b$   
 m: 572.49729  
 b: 418.87070  
 x: Amount[ng/ml]  
 y: Area



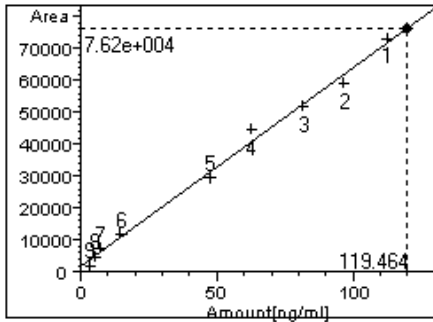
PCB 138 at exp. RT: 14.858  
 ECD1 B,  
 Correlation: 0.99899  
 Residual Std. Dev.: 1228.93425  
 Formula:  $y = mx + b$   
 m: 600.52124  
 b: 1216.97882  
 x: Amount[ng/ml]  
 y: Area



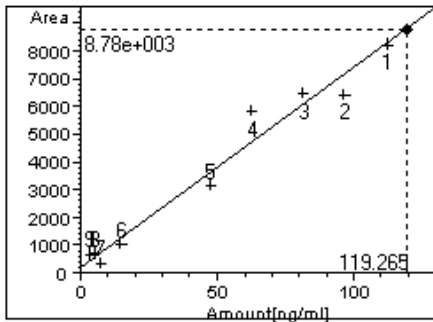
Endrin Aldehyde at exp. RT: 15.719  
 ECD1 B,  
 Correlation: 0.99908  
 Residual Std. Dev.: 1272.50692  
 Formula:  $y = mx + b$   
 m: 650.77621  
 b: 41.51635  
 x: Amount[ng/ml]  
 y: Area



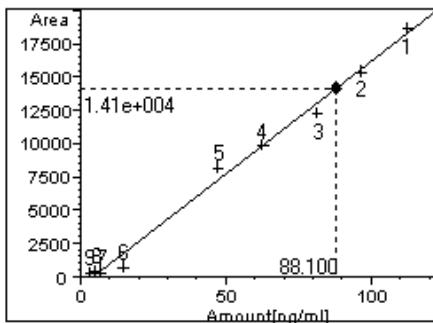
p,p'-DDT at exp. RT: 16.303  
 ECD1 B,  
 Correlation: 0.99805  
 Residual Std. Dev.: 913.12362  
 Formula:  $y = mx + b$   
 m: 320.90469  
 b: -705.87425  
 x: Amount[ng/ml]  
 y: Area



PCB 153 at exp. RT: 16.542  
 ECD1 B,  
 Correlation: 0.99710  
 Residual Std. Dev.: 2161.52107  
 Formula:  $y = mx + b$   
 m: 622.48305  
 b: 1829.31229  
 x: Amount[ng/ml]  
 y: Area



Endosulfan Sulphate at exp. RT: 18.812  
 ECD1 B,  
 Correlation: 0.98380  
 Residual Std. Dev.: 595.52831  
 Formula:  $y = mx + b$   
 m: 71.87420  
 b: 210.27841  
 x: Amount[ng/ml]  
 y: Area

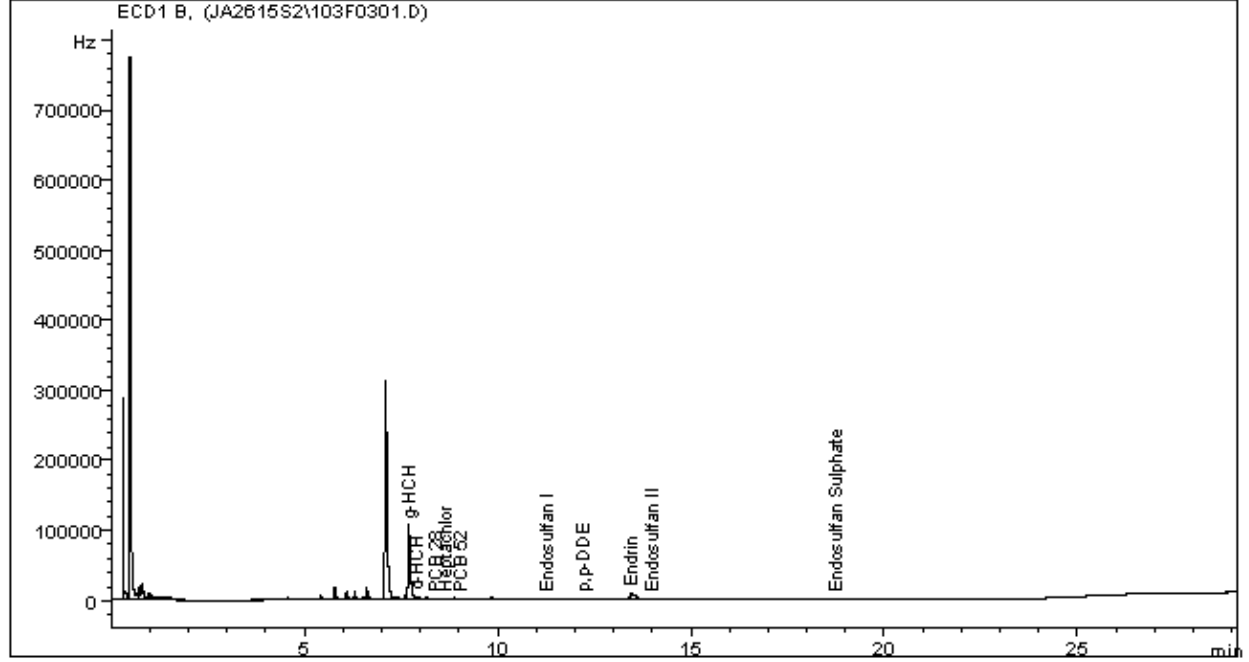


Methoxychlor at exp. RT: 21.472  
 ECD1 B,  
 Correlation: 0.99633  
 Residual Std. Dev.: 659.54482  
 Formula:  $y = mx + b$   
 m: 168.92447  
 b: -735.19771  
 x: Amount[ng/ml]  
 y: Area

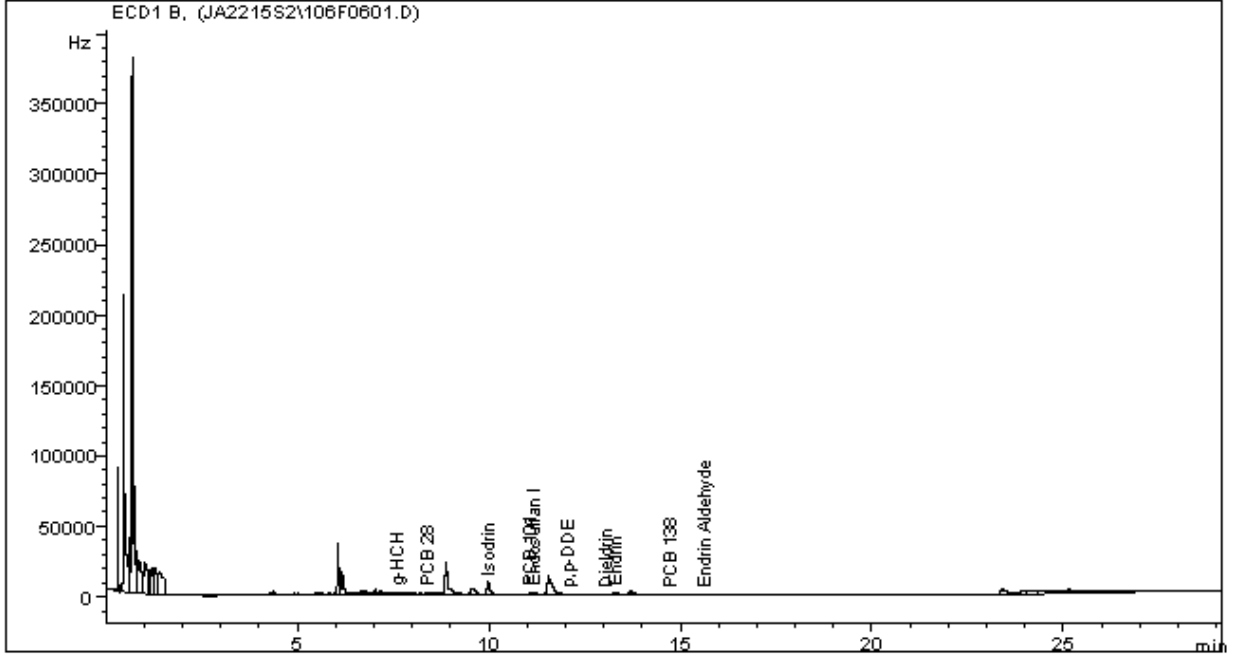


# Appendix V: Chromatograms

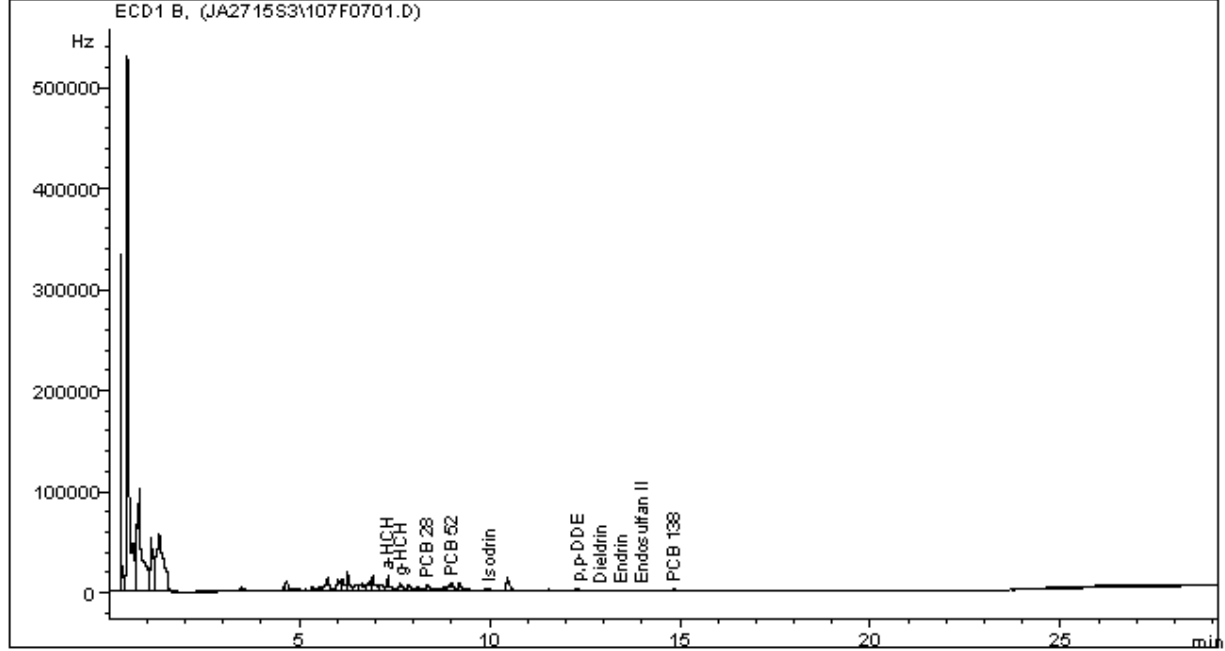
ECD POPs standards test method



ECD POPs standards test method



ECD POPs standards test method



ECD POPs standards test method

