Assessment of pesticide concentrations in environmental and biological parameters from two Kenyan Rift Valley Lakes.

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I dedicate my thesis in loving memory of my mother
Nita Gudka.

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ABSTRACT

In the last two decades Kenyan agriculture has developed rapidly. In particular, horticultural and floricultural activities have intensified on the riparian fringe of Lake Naivasha, a RAMSAR site. The lake supports a large variety of wildlife and avifauna in particular. In the 1980s, the African Fish-Eagle (*Haliaeetus vocifer*) population on the lake was the densest in Africa. As a top predator resident in the Kenyan Rift Valley Lakes the African Fish-Eagle is a good indicator of general ecosystem health but is also highly susceptible to toxic effects of pesticide contamination. Organochlorine pesticide poisoning was a particular threat to birds of prey, during the 1950s and 1960s. Globally, most organochlorine pesticides have been banned, but endosulfan, HCH and methoxychlor are widely used in Kenya, while aldrin, dieldrin and lindane are restricted and DDT, endrin, and heptachlor banned. Studies examining residue levels of these harmful chemicals in African Fish-Eagles are limited to small sample sizes examined in the 1970s and 1980s. This study is the first comprehensive investigation of organochlorine residue contamination in biological and environmental parameters from Lake Naivasha and the control water body at Lake Baringo.

The study aimed to determine the level of organochlorine contamination in both lakes, by evaluating three factors: 1. Differences in organochlorine contamination between lakes, in water, sediment, fish and African Fish-Eagle clotted blood and serum samples 2. Contamination of the Lake Naivasha ecosystem directly resulting from intensive agricultural activities, and 3. Whether contamination is at harmful levels for the African Fish-Eagle and for humans in one or both lake ecosystems. Clotted blood and serum samples were collected from 12 and 8 African Fish-Eagle individuals on Lakes Naivasha and Baringo, respectively, for organochlorine residue analysis. Muscle, fat and liver samples were simultaneously collected from their food base: the Common Carp (*Cyprinus* carpio) on Lake Naivasha, and Nile Tilapia (*Oreochromis niloticus*), Barbus (*Barbus intermedius*), and African Sharp-tooth Catfish (*Clarias gariepinnus*) in Lake Baringo. Water and sediment samples were also collected from five sites on each lake. Lake Naivasha was divided into two regions based on presence of absence of flower farms. The first encompassed the southern and south western riparian edges, which had a high density of flower farms (HFFD) and the second segment encompassed the north and north eastern edges, with a low density of flower farms (LFFD). All 110 samples were analysed for a total of 17 compounds comprising: p,p'-DDT, p,p'-DDD, p,p'-DDE, methoxychlor, aldrin, dieldrin, endrin, endrin aldehyde, endosulfan I, endosulfan II, endosulfan sulphate, heptachlor, heptachlor epoxide, HCH (α, β, δ, γ).

Residues of all organochlorine compounds were detected in both lakes of which, endrin, endrin aldehyde and p,p'-DDE were most frequently detected. Residues detected in African Fish-Eagles were below levels known to cause egg-shell thinning, mortality or depressed productivity. Flower farms were found to be a potential source of endosulfan contamination in Lake Naivasha, as concentrations in African Fish-Eagle blood samples were significantly higher in the high flower farm density (HFFD) area when compared with the low flower farm density (LFFD) area (endosulfan sulphate, LFFD median: 7.1 ng/g, HFFD median 15 ng/g; endosulfan I, LFFD median: 0.0 ng/g, HFFD median 3.1 ng/g, $W = 19$, $p = 0.03$). Median concentrations of the following compounds were all above Threshold Effects Concentrations (TEC) and Environmental Protection Agency (US

EPA) guideline levels: endosulfan (Naivasha: 0.03 ng/l, Baringo: 0.02 ng/l), p,p'-DDT (Naivasha 0.003 ng/l, Baringo: 0.004 ng/l) and methoxychlor (Naivasha: 0.05 ng/l, Baringo: 0.03 ng/l) in water samples, and aldrin (2.11 ng/g), heptachlor (4.85 ng/g) and lindane (6.11 ng/g) in Naivasha and β-HCH (5.51 ng/g) and heptachlor (7.69 ng/g) in Baringo sediment samples. The main hazardous implications of high residue levels are to aquatic organisms, potentially causing mortalities and population declines, leading to trophic cascades higher up the food chain. Impacts at lower trophic levels could adversely affect African Fish-Eagle and fish-eating species impacting ecosystem functions and economic activities.

The additional impact of organochlorine contamination will further perturb the lake ecosystems already faced with, exotic species introductions, habitat loss, eutrophication, siltation, dwindling water supply, and wildlife species declines. Long-term, monitoring and regulation of farming practices in Kenya need to be implemented to maintain aquatic ecosystem integrity.

CHAPTER 1: LITERATURE REVIEW

Persistent Organic Pollutants

Pesticides are chemicals most commonly used in agriculture and horticulture for the control of animal pests, weeds, or plant diseases (US EPA 2011 a). Pesticides include insecticides, herbicides, fungicides, and other pest controlling substances (Wandiga 2001). Between the years 2000 and 2001, the total global usage of pesticides exceeded a weight of 2.27 billion kg of active ingredient, a combined total for both years of over US$ 60 billion spent (Mwanthi 1998). In 2006 and 2007, the total global expenditure on pesticides accounted for a combined total of US$ 75.2 billion (US EPA 2011 b). Africa contributed three percent of total global pesticide imports between 1993 and 1994 and has been experiencing exponential growth since then (Williamson 2003). In Kenya, between 1985 and 1987, the total value of pesticides imported into the country was worth US $69 million (Mwanthi and Kimani 1993), and between 1987 and 1990, a total of 31,234 tons of pesticides was imported (Mwanthi 1998). Agriculture is one of Kenya's main economic activities (IUCN 2005), and pesticide use is highly advocated by African governments to promote development (Williamson 2003).

Insecticides constitute one of the highest quantities of pesticide groups imported into Kenya (Lalah et al. 2003). As such, most focus will be given to insecticides in this study. Organochlorine pesticides are a family of compounds classed as Persistent Organic Pollutants (POPs) (Walker 2001). Organochlorine insecticides are one of the earliest generations of synthetic pesticides (Mineau 2001), and can be divided into three main groups: DDT and its metabolites; the cyclodiene insecticides; and the isomers of HCH (Brooks 1974). These environmentally harmful chemicals are: i) persistent, ii) lipophilic (soluble in fat) and iii) chemically stable (do not metabolise readily) (Mineau 2001, Walker 2001).

The environmental fate of organochlorine compounds is governed by their specific chemical properties (Lalah et al. 2003). Compounds that contain hydrophobic molecules (not easily dissolved) are pushed out of water and bind to organic matter in sediment, and surface water oil films, thus limiting their mobility, and increasing their persistence (Walker 2001). Chemicals also enter the environment more readily if they are highly volatile (Cid et al. 2007). Chemically stable compounds (i.e., p,p'-DDE and dieldrin) are also transported for long distances e.g., atmospheric transport. As a result many organochlorine residues have been detected as far as the polar-regions (Oehme et al. 1994, Loganathan and Kannan 1994). The climate and ecosystem in which organochlorine compounds are released into, determine how they react (Frank et al. 1977), such that, tropical aquatic environments provide the necessary conditions for rapid compound breakdown, whereas temperate terrestrial environments demonstrate slow decomposition and higher build-up of organochlorine compounds (Walker 2001, Wandiga 2001). Chemicals in field conditions are affected by variables such as wind speed, temperature, pH, solar radiation (Wandiga 2001), and precipitation (Walker 2001).

Historically, each organochlorine compound has been used for specific pest controlling functions. In Kenya, heptachlor was used as a seed dressing on maize and bean crops (Wasswa et al. 2011, Lalah et al. 2003), DDT was used in malaria control (Wandiga 2001). Some uses of organochlorine compounds have been maintained

such as endosulfan used in cash crops such as tea, flowers, and vegetables (Cid et al. 2007) and lindane used in tick control and seed dressing (Wandiga 2001). Aldrin continues to be used as a termite control agent and occasionally in malaria control (Lalah et al. 2003).

Pesticides and Biodiversity: Global Impacts on Birds of Prey

Raptors and piscivorous birds are particularly vulnerable to the impact of POPs (Mineau and Tucker 2002, Mineau 2003, Martinez-Lopez et al. 2009, Guitart et al. 2010). organochlorine insecticides are particularly harmful as they bioaccumulate and bioconcentrate increasingly at each trophic level, causing deleterious impacts on top predators (Mora et al. 2002, Martinez Lopez et al. 2009, Mineau 2009, Guitart et al. 2010). Birds of prey, as top predators, are the most acutely impacted, causing mass mortalities and reducing reproductive success (Bowerman 2000). Organochlorines were particularly damaging to wildlife between the period of 1947 and 1985 due to their widespread use (Henny and Elliot 2007).

Exposure of organochlorine compounds to birds of prey can occur even at great distances from application sites through atmospheric transport, e.g., elevated concentrations in the Canadian arctic (Barrie et al. 1992), contaminated migratory prey species, or migratory birds of prey transporting material themselves from distant sources (Henny et al. 1982).

An account of the events and studies conducted since the discovery of insecticidal DDT are extracted from texts in (Walker 2001) and summarised in Table 1. Most studies focusing on organochlorines and their impacts were conducted between the 1950s and 1960s (Anderson et al. 1972, Cooper 1973, Fox and Lock 1978, Newton 1979, Lincer et al. 1980, Fry and Toon 1982, Henny et al. 1987, Kolbe and Hill 1987). DDT and its metabolites are the most widely studied organochlorine insecticides, particularly p,p'-DDE, due to its egg-shell thinning effects in birds (Frank et al. 1977, Fox and Lock 1978, Grier et al. 1982, Jarman et al. 1994). Heptachlor, p,p'-DDE, dieldrin, and others can cause reduced productivity in raptor species (Lockie et al. 1969, Ratcliffe 1970, Henny et al. 1983). The first account of adverse pesticide effects to avifauna was reported 50 years ago in 'Silent Spring' by (Carson 1962). The book documented the dangers of biomagnifications in food chains and egg-shell thinning associated with organochlorine compounds (Fry 1995, Mineau 2003). As a result, the severe implications of DDT use to the sustainability of wildlife, as well as human safety, were beginning to receive more attention. Some of the most severe eggshell thinning effects were found in the Peregrine Falcon (*Falco peregrinus*), Osprey (*Pandion haliaeetus*) and Bald Eagle (*Haliaeetus leucocephalus*) (Snyder *et al.,* 1973; Newton 1979; Cade *et al.,* 1988; Court *et al.,* 1990; Noble *et al.,*1993; Kirt and Hyslop, 1997 Fry 1995, Mineau 2001). The near extinction of *Falco peregrinus* in 1964 from the Eastern United States, and the global population reduction, was attributed to high levels of DDE in the environment (Risebrough 1994). Populations recovered following a DDT ban in 1972 and the species status was delisted from its Endangered Species classification in 1999 (Cade and Burnham 2003, Henny and Elliot 2007) providing significant evidence for organochlorine bans. Other compounds found to inflict deleterious impacts on avifauna included cyclodiene insecticides such as aldrin, dieldrin, and endrin (Mineau 2001), which are among the most toxic organochlorines to birds (Walker 2001). The lipophilic chemicals are most likely to be stored in fat and have

the potential to reach lethal levels in the brain (Beyer et al. 1996). This is particularly harmful during periods of food shortage or when high energy demands force birds to utilise stored fat reserves, releasing the chemicals into the blood stream (WHO 1989). Clear evidence of dieldrin-induced mortalities was found in Eurasian Sparrowhawks (*Accipiter nisus*) in Britain (Newton et al. 1986) causing population declines in some parts of the country (Sibley et al. 2000). Causes of mortality in Bald Eagles found dead in the USA from 1966 to 1983 were reviewed by Peakall (1996). It was discovered that dieldrin-attributed deaths decreased from 13.0% in 1966-70 to 1.7% in 1978-83, following its ban (Cromartie et al. 1975, Peakall 1996). Mineau (2003), argued that governments took notice of the negative pesticide effects not for the wildlife mortalities but primarily due to hazards posed to humans through food chains (Mineau 2003).

As a result of the organochlorine bans, organophosphate and carbamate pesticides (due to their lower environmental persistence) were quickly consented for use by government regulators as replacements of organochlorines, despite their acute toxicity (Mineau 2003). After the ban of DDT and cyclodienes in the United States in 1972, the number of studies examining organochlorine insecticides has decreased. Most current published literature focuses on the synthetic agrochemicals including organophosphates and carbamates (Mineau 2003, Mineau 2005 (a), Mineau 2009, Vyas et al. (1998), Fleischli et al. (2004), Wobeser et al. (2004), Guitart et al. (2010). Yet, several isolated studies are still concerned with identifying levels of DDT in the environment and avifauna (Wyk et al. 2001, Rivera-Rodriguez et al. 2007, Drooge et al. 2008, Mora et al. 2008, Martinez-Lopez et al. 2009 and, Dhananjayan and Muralidharan 2010). It does however, appear that, the general trend in modern toxicology research is moving away from organochlorine studies. They are seen as outdated pesticides, which pose a much less significant threat to wildlife and natural environments than the current newer, highly toxic generation of agrochemicals (Peall, S. pers comm., 11 April 2011). A complete shift in study focus from organochlorines to other pesticides might be relevant to some countries (Mineau 2003), but many developing countries including Kenya, where certain organochlorine compounds are widely used (Wandiga 2001, Lalah et al. 2003) risks of continued natural ecosystem contamination exist (Greichus et al. 1977, Grobler 1994, Evans and Bouwman 2000, Bouwman et al. 2008). As such, organochlorine studies remain integral to detecting those potential hazards.

Even where organochlorine pesticide use has been discontinued, their residues remain in the environment for decades with harmful potential. More recently, organochlorine pesticide toxicity has been realised by their re-emergence in the environment, e.g., bird mortalities as a result of contaminated prey (earthworms and insects bringing organochlorine residues to the surface from soil sinks: Fry 1995), DDE induced eggshell-thinning and reduced nesting success of Ospreys between 1997 – 1998 on the Colombia River, United States (Henny et al. 2004). Two general types of organochlorine studies exist: (1) long-term monitoring of productivity and populations of bird species previously affected by organochlorine contamination, which may include some collecting of eggs or blood plasma, and (2) evaluations of potentially sensitive species based on diet (usually fish or bird-eaters) at locations where few or no studies have been conducted (Henny and Elliot 2007). Not all organochlorine pesticide impacts are as conspicuous as mortalities, some more subtle consequences (causing hormone disruption effects) could be acting on bird species through pesticide contact and may be harmful

nonetheless (Fry 1995). This could require continued organochlorine residue studies determining a wider long-term array of organochlorine-induced problems. The consequences of chemical-induced hormonal imbalances in embryos and foetuses would take years to manifest as they impact birds only at sexual maturity (Colborn et al. 1996).

1874	Synthesis of DDT by Zeidler (Walker 2001).
1939	Discovery of insecticidal property of DDT by Muller.
1940s	Discovery of insecticidal property of HCHs
1946	First signs of Bald Eagle population declines in Florida (Broley 1958). 15 – 19% eggshell thinning was related to diminishing breeding success (Walker 2001). Eggshell thinning in Peregrine Falcons and Sparrowhawks causing population declines only eight years later in Britain.
1950s	Introduction of the cyclodiene insecticides aldrin, dieldrin and heptachlor. (after the effect of eggshell thinning has occurred in bird species).
1956	Reports of large scale predatory bird mortality from farm land in Western Europe.
Late 1950s	Sparrowhawk, Peregrine and Kestrel populations crash in Britain due to cyclodienes.
Early 1960s	Restrictions placed on DDT and cyclodienes due to their environmental persistence and toxicity. Metabolite p,p'-DDE was found to be the most abundant organochlorine compound in the environment and biota.
1961 – 1962	American Robins eliminated from 75 h plot treated with DDT (Bernard 1966). Sandwich Terns on the Netherlands coast poisoned in the field (Koeman et al. 1967).
1962 – 1964	Dieldrin and p,p'-DDE residues in fish-eating birds found 1000-fold higher than prey and lower trophic levels.
1965 – 1966	Buzzard (*Buteo buteo*) in Netherlands near extinction due to Dieldrin poisoning (Fychs 1967).
1969 – 1984	Eggshell thinning of 19 – 30 ppm in Gannets from Quebec, Canada (Elliot et al. 1988). 1975, seed-dressing uses of cyclodienes banned in Britain (Newton 1986, Ratcliffe 1993). Bioaccumulation in fish-eating Shag due to DDE 50-fold higher than prey of Sand Eel (Robinson et al. 1967). Discovery that dietary levels of p,p'-DDT at 3 ppm in American Kestrels could cause egg-shell thinning (Weimeyer and Porter 1970, Peakall 1993). Discovery that soil and sediment are sinks for DDT compounds (Newton 1986).

Table 1: A summary of studies and events related to organochlorine insecticides and their impacts on Wildlife in particular avifauna. Information has been extracted and summarised from Walker (2001)

Birds of Prey and Pesticides in Africa

The field of avian toxicology is primarily informed through studies and research conducted in the northern latitudes, specifically from North America and Western Europe (Henny et al. 1987, Risebrough 1994, Helander

1994, Bowerman et al. 1994, Fry 1995, Shimmel & Snell 1999, Mineau 2001, Mineau and Tucker 2002, Fleishchli et al. 2003, Mineau 2003, Mineau 2005 (a), Drooge et al. 2008, Martinez-Lopez et al. 2008, Mora et al. 2008, Mineau 2009, Guitart et al. 2010). The greater attention devoted to pesticide studies in temperate regions is due in part to the longer persistence of organochlorine compounds under those climatic conditions than in tropical regions (Lalah et al. 2003). As a result of this climate effect, impacts of pesticides on wildlife have been far greater in places like the United States and Europe (Frank et al. 1977).

The few African case studies to have emerged in literature in the last few decades are based on detection of organochlorine insecticides, including: Koeman et al. (1972), Cooper (1973), Lincer et al. (1980), Douthwaite (1992), Kairu (1994), Wyk et al. (2001), Hollamby et al. (2004 a). In Africa, avian toxicology is greatly understudied with few publications (Koeman et al. 1972) in contrast to the prolific pesticide use on the African continent (Ngaio Richards, pers. comm. April 2011).

Koeman et al. (1972) represents one of the first studies conducted on organochlorine contamination in East African birds. The study investigated DDT, endrin, and dieldrin compounds (range: 0.6 – 20 ppb wet weight) in African cormorants (*Phalacrocorax carbo*), Lesser Flamingo (*Phoeniconaias minor)* and White Pelican (*Pelecanus onocrotalus*) tissue samples at Lake Nakuru, Kenya. Frank et al. (1977) was among the pioneer organochlorine related raptor studies in the region. The study surveyed 18 raptor species from agricultural and non-agricultural areas in Kenya. DDT and dieldrin were detected in pectoral muscle tissue in agricultural areas, whereas birds from non-agricultural areas were not contaminated with organochlorine residues. Some of the first in-depth studies of raptor populations and behaviour were focused on the Rift Valley Lakes of Kenya were conducted in the early 1980s by Brown (1980), and Lincer et al. (1981). Both investigated DDT residues on small sample sizes of African Fish-Eagle eggs, finding low levels of organochlorine contamination in birds and African Fish-Eagle in the Naivasha and Baringo lake ecosystems.

Eleven organochlorine compounds were detected in the same species in Lake Nakuru in a follow up study to Koeman et al. (1972) by Kairu (1994). The highest concentration of 20700 ppb was detected of p,p'-DDE as a result of large scale industrial activities around the Lake. More recent studies on organochlorine contamination in birds of prey, particularly African Fish-Eagle, have not been conducted in Kenya since the 1980s. African Fish-Eagle was studied by Brown (1980) and this species is a good indicator of aquatic ecosystem health due to its top predatory position in the food chain (Bowerman 2000). Plasma organochlorine concentrations in African Fish-Eagle adults from Uganda, were investigated by Hollamby et al. (2004 a), who detected low concentrations of p,p'-DDE residues. This study is of significance to Kenyan African Fish-Eagle studies due to the similarities in climate, species and ecosystem in concern.

Recent organochlorine contamination studies, of blood plasma of raptor species, include vulture samples from South Africa (Van Wyk et al. 2001), birds of prey (including Sparrowhawks) from China (Da Chen et al. 2009), and Pariah Kites (*Milvus migrans*), and 12 other bird species from India (Dhanajayan and Muralidharan 2010). Other studies have focused used on organochlorine contamination in non-raptorial species, such as Great Grebe (*Podiceps major*), Neo-tropic Cormorant (*Phalacrocorax brasilianus*), Great Kiskadee (*Pitangus*

sulphuratus) from Argentina (Cid et al. 2007), and numerous migratory species from five Asian countries (India, Vietnam, Japan, Philippines and Russia), (Kunisue et al. 2003). Studies and research concerned with chlorinated hydrocarbon insecticides may still prove necessary in Asia and Africa as production and use of organochlorine pesticides still continue (Wandiga 2001, Droogue et al. 2008, Da Chen et al. 2009, Mineau 2009). In Kenya, organochlorine pesticide use in the 1990s was extensive, when the highest quantity of pesticides were imported into the country (Wandiga 2001, Gitahi et al. 2002) and although restrictions have been placed on certain organochlorine pesticides, endosulfan, methoxychlor and lindane are still in use (Lalah et al. 2003) warranting continual monitoring of their environmental impacts.

Study Area

Lake Naivasha Climate and Physical Attributes

Lake Naivasha, one of the Kenyan Rift Valley lakes, is approximately 160km^2 in area, occurs at an elevation of 1890 m, and lies in a semi-arid region where average annual rainfall is 680 mm (Hickley et al. 2004). The lake has no surface outlet (Becht and Harper 2002). As a wetland of international importance it gained an official title in 1995 as Kenya's second designated RAMSAR site (Becht and Harper et al. 2002). The Malewa River (permanent flow) with a watershed encompassing an area of 1730 km^2, the Gilgil, and the Karati rivers (seasonal flow) are the three main rivers feeding into Lake Naivasha (Adams et al. 2002). The sediments found in Lake Naivasha are introduced by these three rivers and transported to the central, eastern, and southern parts of the lake (Tarras-Wahlberg et al. 2002). A variety of sediment types occur from silt to clay (Hickley et al. 2004), and in some areas include a top layer of humus and water hyacinth (*Eicchornia crassipes)* debris (Pers. obs 2010). Three decades ago, the Lake used to be entirely bordered by *Cyperus papyrus* (Gaudet, 1977; Gouder et al., 1998, Adams et al. 2002, Hickley et al. 2004). The papyrus stands form an important habitat for many, wildlife occurring on the lake, from herbivores (e.g. Hippopotamus and buffalo), to waterfowl, including some threatened species (e.g. Papyrus Yellow Warbler genus species) (IUCN 2003), and the loss of nursing habitats for juvenile fish. Lake Naivasha supports a dense population of African Fish-Eagle important to the economy (tourism) and ecology (trophic control) of the lake.

At the beginning of 2010, Kenya experienced a severe drought of a magnitude that had not been seen for over 100 years causing severe reductions in the lake water levels, dropping by almost 4 vertical meters (M Virani pers. comm. 2009). At the time when the field-work begun, the water levels had risen dramatically by 3 m.

Conservation Problems

Introduced Exotic Species

Since the 1920s, under the British colonial rule, Lake Naivasha has experienced a variety of anthropogenic alterations to its ecology and riparian environment (Harper et al. 2011). Introductions of exotic species began with *Orechromis spilurus niger* and later, in 1927, the introduction of Large-Mouth Bass, *Micropterus salmoides,* for sport fishing (Harper et al. 2011). Additional species were introduced into the lake in 1956: *Tilapia zillii, Orechromis leucostitcus, and Procambarus clarkii* (IUCN 2005). The fish species composition of Lake Naivasha has been subsequently changing throughout the years. The composition of fin-fish caught by local fishermen between 1987 and 2000 provides some insight into what species were dominant in the lake during that period. In order of quantity was (1) *Oreochromis leucostictus,* (2) *Micropterus salmoides,* and (3) *Tilapia zillii* (Hickley et al. 2004). A further introduction of *Cyprinus carpio* in 1999 (Harper et al. 2011) shifted the species composition in the lake so that between the years 2002 and 2006, Common Carp dominated fishermen's catches, while the contribution of the other species fell (Njiru et al. 2008). In 2010, the scenario remained the same with Common Carp dominating and a significant reduction of all other species with possible extermination of Large Mouth Bass (pers. obs.).

Additional exotic species introductions included a floating water-fern, *Salvinia molesta* introduced in 1962 and in 1988, Water Hyacinth (*Eichhornia crassipes)* a floating weed, forming large floating mats on the lake surface, obstructing fishing and boating activities on the lake (IUCN 2005). Water Hyacinth colonised the lake within a few short years (IUCN 2005), its mats fringing the entire perimeter of the lake, the largest of which is found near the Malewa River mouth (Adams et al. 2002). The mats are moved through the lake by prevailing winds, shifting their location frequently (pers. obs.). Salvinia supports very few invertebrate and vertebrate species compared to Water Hyacinth, and more recently the fern has shown marked declines in its extent (Adams et al. 2002). Exotic species introductions and continual fluctuations have altered the ecology of Lake Naivasha, and thus continuously changing the mode for transfer and absorption of organochlorine pesticides through the ecosystem and food chains, as each individual species reacts uniquely to organochlorine chemicals. Salvinia and Water Hyacinth introductions in Lake Naivasha have led to an increased amount of evapotranspiration and a subsequent loss of water from the Lake. A papyrus swamp dominated the northern edge of the lake near the river mouths, and has been gradually reduced to a fringe around the perimeter of the lake (Adams et al. 2002). The reduction in the 'North Swamp' (Kitaka et al., 2002), has led to a loss of natural pollution distillation by the papyrus (Gaudet and Muthuri 1981).

Agricultural Developments

In the 1980s, Lake Naivasha experienced a surge of riparian agricultural activities (Boar et al. 1999), of which horticulture and floriculture are the most economically significant (Hickley et al. 2004) and most dense (Njuguna 2007) with approximately 30 flower farms (IUCN 2005) throughout Kenya. The farms have aided Kenya build its identity as a 'Newly Agriculturalising Country' (NAC), contributing 75% of the total 32 200 tonnes of flowers exported from Kenya in 1995, valued at US$ 80 million (IUCN 2005). Flower exports from 1999 to 2005 grew by 20% each year (IUCN 2005). Although the bulk of agriculture in the area is dominated by flower and vegetable farming, the area is also comprised of cotton and small-scale subsistence farming (Kitaka, 2000). The growth of the riparian floriculture industry has been implicated in the lakes current eutrophic state (Kitaka, 2000), as a result of chemical pollution (Everard and Harper 2002) drainage and accelerated riparian vegetation clearance (*C. papyrus* and *Acacia xanthophloea*) with a consequent loss in the potential to absorb effluent and run off from settlement and agriculture(Boar et al. 1999). Papyrus acted as an important filter of incoming suspended sediment from rivers (Tarras-Wahlberg et al. 2002), and helped to intercept eroded terrestrial topsoil run-off (Hickley et al. 2004) entering the lake, thereby improving water quality (Tarras-Wahlberg et al. 2002).

Pesticide use (of which some is banned in other countries) is regarded as one of the prime hazards of horticultural farms (Everard and Harper 2002). Water extraction for irrigation, drinking water supplies and hydroelectric power generation purposes have also been implicated as a threat to the ecosystem integrity (Becht and Harper et al. 2002). Agrochemicals (i.e., insecticides, fungicides, herbicides, fertilizers) are used intensively at the flower farms around the periphery of the lake. Some of the flower farms have illegally encroached within 50 m of the lake edge, increasing the potential for untreated run-off directly from the green

houses to enter the lake (pers. obs.). As a result of increased job opportunities in horticulture/floriculture farms, ad-hoc settlements have expanded, depositing a large amount of untreated and unregulated run-off, including sewage, heavy metals, and chemicals into the lake at multiple sites (Adams et al. 2002, Kapila, S. pers. comm. 2012). Pesticides can enter fresh water systems through a number of channels (e.g.. through land run-off, washing into natural water bodies). Eye-witness accounts have been reported by the Food and Water Watch 2008 - "We saw pipes pumping water from the lake to the flower green-houses and a ditch where waste water drained back into the lake." Evidence of organochlorine residues (p,p'-DDT, p,p'-DDE, aldrin, dieldrin and endosulfan II) in samples of various fish species occurring at the lake were found (Gitahi et al. 2002). These levels were reportedly higher than levels found in a study conducted by Mugachia et al. (1992), on the same fish species in Lake Naivasha, suggesting there is a build-up of chemical residues in the lake.

Farms are recommended to be more responsible with their use of pesticides, ensuring that contamination of the lake and its natural environs does not occur, rather than limiting pesticide usage altogether (Everard and Harper 2002). This has potential for success, but the nature of certain chemicals makes it difficult to control their contamination (Walker 2001). For example, endosulfan is highly volatile and easily transported through the air, contaminating not only nearby environments, but those further afield as well (Loganathan and Kannan 1994).

Lake Baringo Climate and Physical Attributes

Lake Baringo is a shallow lake, approximately 140 km^2 and lies in a semi-arid region (Hickley et al. 2004). It is also one of the six sites in Kenya designated as a RAMSAR wetland of international importance (Omambia et al. 2009). Lake Baringo has been experiencing dramatic land cover changes primarily due to increasing human population around the Lake catchment area, and in part due to climate change impacts (Kiage et al. 2007). The main land use in the region is pastoralism as 46% of the land is unsuitable for agriculture (Hickley et al. 2004). Forest cover has been the most affected around the lake, in some sections reaching cover losses of over 40% over a 14-year period, over-grazing blamed for much of the problem. Deforestation and subsequent land degradation have increased the sediment yield in the lake resulting in reduction in lake surface area by over 10% (Kiage et al. 2007) and also leadaing to highly turbid waters, primarily due to deforestation of the surrounding hills (Aloo 2004). The high siltation of the lake has led to significantly reduced primary productivity (Lincer et al. 1981), dramatically reducing the invertebrate richness and diversity (Aloo 2004). Turbidity has led to the near extinction of submerged macrophytes and a lake bed virtually devoid of benthic fauna. Fishing pressure has added to the environmental stresses endured by the fish populations and commercial catches have been detrimentally affected. Lake Baringo supports a wide variety of wildlife including waterbirds, hippos and crocodiles, and a resident Fish Eagle population. This population has suffered previously from the effects of chemical pollutant – in 2006 poisoned fish bait intended for crocodiles led to the mortalities of 13 African Fish-Eagle individuals (Kapila, S. Pers. Comm. 2012). Fives fish species historically occurred in Lake Baringo, of which *Barbus intermedius* are rare in the fisherman's catches, while *Labeo cylindricus* has almost disappeared from the lake since the inflowing rivers were dammed, affecting its breeding habits (Aloo 2004).

CHAPTER 2: INTRODUCTION

The African Fish-Eagle is a highly territorial, avian apex predator occurring extensively across sub-Saharan Africa on permanent water bodies. It is widespread throughout its range and often occurs locally in dense populations, preferring tall riparian trees as habitats for nesting and perching (Brown 1980, Simmons 2005). Its main prey are fish and aquatic birds (Stewart et al. 1997, Harper et al. 2002), with preferences differing slightly from one locality to the next (Brown 1980, Simmons 2005). Leslie Brown reported the Lake Naivasha population as the densest population in Africa between the 1960s and early 1970s (Harper et al. 2002). Between the 1960s and 1980s, 144 - 172 individual fish eagles were recorded on Lake Naivasha (Smart 1991). At this time, a total of 350 bird species were also reported at Lake Naivasha (Lincer et al. 1981) and it was subsequently designated a RAMSAR site in 1995, a wetland of international importance (Everard and Harper 2002). In Lake Baringo, a smaller Fish Eagle population was recorded of approximately 35 pairs in the 1980s (Brown 1980). At this time 300 bird species were reported to occur at Lake Baringo (Lincer et al. 1981). Of the Kenyan Rift Valley lakes, Lake Baringo and Lake Naivasha are largest freshwater lakes (Gitahi et al. 2002, Campbell et al. 2003, Hickley et al. 2004).

African Fish-Eagles from Lake Naivasha experienced population fluctuations since the 1980s, decreasing from 80 pairs (Lincer et al. 1981) to only 119 individuals in 2010 (S. Kapila, unpublished data 2009-2011). This coincided with the increased economic activities in the area (mainly floriculture). The African Fish-Eagle population in Lake Baringo has similarly declined from 70 individuals in the 1980s to 20 individuals in 2010 (S. Kapila, unpublished data 2009-2011). The impact of dwindling populations is two-fold: biological and socio-economic, and at Lake Naivasha, this is potentially related to contaminated effluent and run-off from the fringing farms.

The impacts of persistent organic pollutants are frequently observed in birds of prey (Mineau 2003) and as such these birds have been used as indicator species to quantify the health of a variety of ecosystems for over three decades (Harper et al. 2002). Eagles in particular are important indicator species as they often occupy top predatory status, feeding over extensive but defined areas (Peakall, 1993, Bowerman 2000). African Fish-Eagle's 'top predator' status, its sensitivity to the effects of organic pollutants (Douthwaite 1992), and its conspicuous nature as a research subject (Virani 2010, M. Virani pers. comm. 2010), have made it a useful environmental sentinel (Mineau 2003). Their predominantly fish diet, make them effective indicators of residue concentrations and aquatic ecosystem health in general (Bowerman 2000). Bird tissues are often used as a means to quantify the pollutants found in these ecosystems (Martinez-Lopez et al. 2009). The type of tissue used varied between studies. Mineau 2003 reports that three main factors can influence the impacts of pesticides on wildlife. The first two factors relate to the ecology of the species (i.e., diet, behaviour) as well as the chemical characteristics (i.e., persistence, toxicology, bioaccumulation properties), and finally the intended use of the chemicals in question. Birds can be exposed to chemical pesticides through their food, skin, feet, or inhalation of vapours or droplets.

Persistent organic pollutants (POPs) (e.g., DDTs, PCBs, dieldrin) are resistant to degradation and may remain in the environment for decades (Martinez-Lopez et al. 2009). The organochlorine pesticides are infamous for their long environmental persistence (Hollamby et al. 2004 a), due to their slow metabolism and enduring breakdown products (Mineau 2003). Accordingly, they were recognised as one of the major environmental pollutant groups (Van Wyk et al. 2001). During the 1950s and 1960s organochlorines were used intensively as insecticides and herbicides in agricultural practices for pest control, contaminating both terrestrial and aquatic habitats (Koeman 1972, Martinez-Lopez et al. 2009). In many parts of the world, they exhibited long-range transport (i.e., atmospheric), bioaccumulation in both humans and animals due to high fat solubility (Mineau 2003), biomagnifications in food chains, and toxic effects (Koeman 1972, UNEP 2001, Dhananjayan and Muralidharan 2010). The global transport of pesticide compounds such as p,p'-DDT has been well documented and in Lake Victoria, Uganda, air samples were reported to contain chlorinated compounds, including DDT (Hollamby et al. 2004 a).

Organochlorines are readily soluble in lipids, storing in the body fat and metabolizing very slowly in birds with a half-life of one year (Dhananjayan and Muralidharan 2010). The chemical compounds can be mobilized from the adipose tissue compartment, causing an increase in blood level resulting in toxic manifestations (WHO 1989). The toxicity of organochlorines can vary widely between compounds (Walker 2001). Cyclodienes (i.e., aldrin, dieldrin, endrin,) are almost 100 times more toxic to quail than any DDT compounds (De Witt, 1956), and dieldrin was reported to be between 35-70 times more toxic than DDE in Peregrine Falcons *(Falco peregrinus)* (Fox and Lock 1978).

Endosulfan is an insecticide and acaricide (i,.e. killing arachnids) of the cyclodiene subgroup, which acts as a poison against a wide variety of insects and mites on contact. It is used primarily for a wide variety of food crops like citrus, fruit, vegetables and cereals (Martinez-Lopez et al. 2009). Lindane, on the other hand, was used to treat the soil against plagues of insects in crops such as onion, potato, kale, and cereals. It was also used to treat seeds (Martinez-Lopez et al. 2009). Some non-POP pesticides (e.g., hexachlorocyclohexane (HCH)) also exhibit substantial bioaccumulation and toxic effects, and are no longer produced in many countries (Da Chen et al. 2011). However, α-HCH and γ-HCH isomers are still legal in Kenya (Lalah et al. 2003).

The import of pesticides (20% insecticides and 20% herbicides) into Kenya in 1995 had increased by 1095.6 tonnes since 1986 (Wandiga 2001). In 2005, 8370 tonnes of pesticides valued at Kenya shillings 4.68 billion were imported into the country of which insecticides made the bulk (Njuguna 2007). However, DDT was banned in Kenya in 1985, the year of the last import. Aldrin and dieldrin were banned in 1992 (Lalah et al. 2001). Before the ban, as much as 70 tonnes of DDT were used annually for agricultural pest control on maize and cotton, and for mosquito control. Lindane, aldrin, and dieldrin were used for seed dressing (Lalah et al. 2001). Despite the official ban of these pesticides, they are still available in the market and detectable in the environment (Lalah et al. 2001). Other cyclodiene and hexachlorocyclohexane compounds were also used in

Kenya (Wandiga 2001). Endosulfan is one of the cyclodiene compounds used in agricultural practices and especially in horticulture and floriculture (Gitahi et al. 2001). The compound has been banned in many parts of the world including other developing nations (e.g. India) due to its toxicity and persistence.

In the early 1960s drastic declines in some bird populations were observed, coinciding with high residues of organochlorines in the tissues and eggs of birds in North America and Europe (Risebrough 1985). Koeman et al. (1972), emphasised the existence of numerous well documented cases of unwarranted mortalities that occurred in fish and bird species due to industrial effluent and the use of organochlorine fungicides in seed dressing activities.

Well documented cases of species declines resulting from pesticide contamination include DDE-induced egg shell thinning in the Bald Eagle (*Haliaeetus leucocephalus*) in North America (Bowerman et al, 2003) and the White-tailed Sea Eagle (*Haliaeetus albicella*) in Sweden (Helander et al, 1982). Both of these species share the same niche requirements as the African Fish-Eagle (Hollamby et al. 2004 a). Other cases include dieldrin-induced mortalities of *Falco peregrines* and Sparrowhawks (*Accipiter nisus*) in Britain, and the population decline of Osprey (*Pandion haliaetus*) from North America (Risebrough 1994).

Organochlorine exposures were correlated with declines in specifically fish-eating bird populations (Bowerman 2000). Exposure to p,p'-DDE has been associated with reduced reproductive success, eggshell thinning and population declines in birds such as *Haliaeetus leucocephalus* and *Pandion haliaetus* (Wiemeyer et al. 1988, Hollamby et al. 2004a, Dhananjayan and Muralidharan 2010). In some cases several compounds were discovered in complex mixtures (i.e., heavy metals and organochlorines), which confound results making it difficult to pinpoint a single factor (Douglas et al. 1999). Autopsies of 145 Bald Eagles between 1966 and 1972 reported 12.4% dieldrin-induced mortalities (Cromartie et al. 1975, Kaiser et al. 1980, Risebrough 1994).

During the 1960s and 1970s the sentiment among scientists was that there was a limited amount of information available on the effects of persistent organic pollutants in the developing world, and indeed in Africa as a whole (Koeman et al. 1972). Most of the studies on organochlorine pesticide contamination in African Fish-Eagle were conducted between the 1970s and 1990s (Frank et al. 1977, Brown 1980, Lincer et al. 1981, Davies and Randall, 1989; Douthwaite, 1994). Sixty percent of the African fauna affected by organochlorine residues of mainly DDT and its metabolites were birds (Douthwaite 1994, Wiktelius and Edwards 1997). African Fish-Eagles in the 1970s and 1980s were investigated for DDT residues and found concentrations of less than 0.01 ppm (Frank et al. 1977, Brown et al. 1980). More recently, two studies were conducted, on the organochlorine contamination in African raptors (Van Wyk et al. 2001, Hollamby et al. 2004 a).

Blood plasma has been used by a variety of researchers to demonstrate the chemical contamination burden in birds of prey (Dhanajayan and Muralidharan 2010): migrating raptors (Elliott and Shutt 1993), nestling Bald

Eagles (Dykstra et al. 1998; Elliott and Norstrom 1998), Peregrine Falcons and nestling Gyrfalcons (Jarman et al. 1994), African White-backed Vultures (Van Wyk. Et al. 2001), Egyptian Vultures (Gomara et al. 2004), and African Fish-Eagle (Hollamby et al. 2004 a).The method has been hailed as a favourable, non-destructive indicator specifically useful in studying species of conservation importance (Donaldson et al., 1999, Rivera-Rodriguez et al. 2007,Martinez-Lopez et al. 2009). The use of blood plasma has been effective in demonstrating the residues of organochlorine compounds in avifauna (Donaldson et al., 1999).

For the past two decades, Kenyan scientists have considered the 'closed system' Rift Valley Lakes of their country as possible sites for contamination. Due to the increase in urban settlements, industrial and agricultural activities, there is a growing danger of aquatic pollution build-up in these lake catchment basins as they have no outlet for the biocides to escape (Koeman et al. 1972). These areas, as 'closed basins', are thought to make good sites for pesticide studies (Lincer et al. 1981). They would appear important since avian mortalities, due to heavy chemical contamination, have still continued to occur in some developing countries today under particular pest-control operations (Harper et al. 2002).

As top predators in the Lake Naivasha ecosystem food chain, African Fish-Eagles could be adversely impacted by compounded effects of pesticide accumulation. Pesticide exposure reduces the breeding success of birds of prey and subsequently results in population declines. African Fish-Eagles are not only important to the biological integrity of the ecosystem, but also to the local economy as a tourist attraction. Pesticide contamination in edaphic and biotic components of Lake Naivasha has been previously investigated. However, organochlorine pesticide levels in African Fish-Eagles in relation to other biotic and abiotic factors from this study will be the first comprehensive analysis since 1979. A range of organochlorine compounds have been analysed to determine if 'old' generation chemicals still impact the Lake Naivasha and Lake Baringo ecosystems, creating a database on African Fish-Eagle toxicology, to then build future research involving the 'new' generation pesticides. As such, this study will provide important baseline residue data on contamination levels in lake water, sediment, fish, and African Fish-Eagles from two similar Rift Valley Lakes.

The specific study focus of this thesis is on differences in organochlorine compound concentrations between Lake Naivasha and Lake Baringo, Kenya. I hypothesize that there will be higher organochlorine concentrations at Lake Naivasha as a result of the use of pesticides on the flower farms around the southern perimeter of the lake, relative to Lake Baringo which lacks such farms. Secondly, this study aims to determine whether sites adjacent to the high density of flower farms were more contaminated than sites with low densities of flower farms within Lake Naivasha. I hypothesize that if flower farms were the source of most pesticides detected, one would expect higher levels of contamination near flower farms relative to areas further from flower farms in all sample types. Lastly, I evaluated if pesticide chemical residues observed at Lake Naivasha and Lake Baringo were at harmful or toxic levels relative to existing international standards and WHO guidelines.

METHODS

Study Sites

The study was conducted at Lake Naivasha, Nakuru District, Rift Valley Province (0° 46' 6.7" S, 36° 21' 2.32" E) (Fig. 2) and Lake Baringo, Baringo District, Rift Valley Province (0° 38' 0" N, 36° 5' 0" E), Kenya (Fig. 3). Lake Naivasha is a shallow freshwater lake, at an altitude of approximately 1890 m creating, warm and semi-arid climatic conditions (Hubble & Harper 2002). The major inflow into the lake is the Malewa River (Adams et al. 2002), while the Gilgil and Karati rivers provide seasonal inflows. Irregular precipitation causes the lake surface area to fluctuate between 100 and 150 km^2 (Adams et al. 2002). Work was conducted on Lake Naivasha between 6 October and 25 November 2010. Lake Baringo is a fresh water lake approximately 168 km^2 at an altitude of 1050 m. Its major inflows include the Ol-Arabel and El-Molo rivers. Research was conducted at Lake Baringo from 16 to 26 October 2010. All work on African Fish-Eagle was performed through the auspices of the National Museums of Kenya and the permission of the Chief Warden of Baringo.

The study methods are divided into two separate sections: 1. Methods related to the field work and sample collections (which were then transported to the Nairobi University Laboratory) and 2. The organochlorine pesticide analyses in the laboratory.

Section 1: Field Sampling Protocol

All field sampling on Lakes Naivasha and Baringo was carried out by boat. Four types of samples were taken: (i) water (ii) lake sediments (iii) fish and, (iv) fish eagle blood and serum from territorial African Fish-Eagles living around the lake perimeter.

Water and Sediment Samples

Site Selection: Water and sediment samples were collected from five sites at both lakes. Sampling sites at both lakes were selected to represent as large a proportion of the entire water body as possible. Sites in Lake Naivasha were chosen based on their proximity to peripheral flower farms. At Lake Naivasha, sites near flower farm out-flows and a river mouth were selected as sampling points for suspected contamination direct from the flower growers. At Lake Baringo, river mouths were sampled to determine pesticide contamination from potential agricultural industries, upstream of the rivers flowing into Lake Baringo. Each site was sampled once.

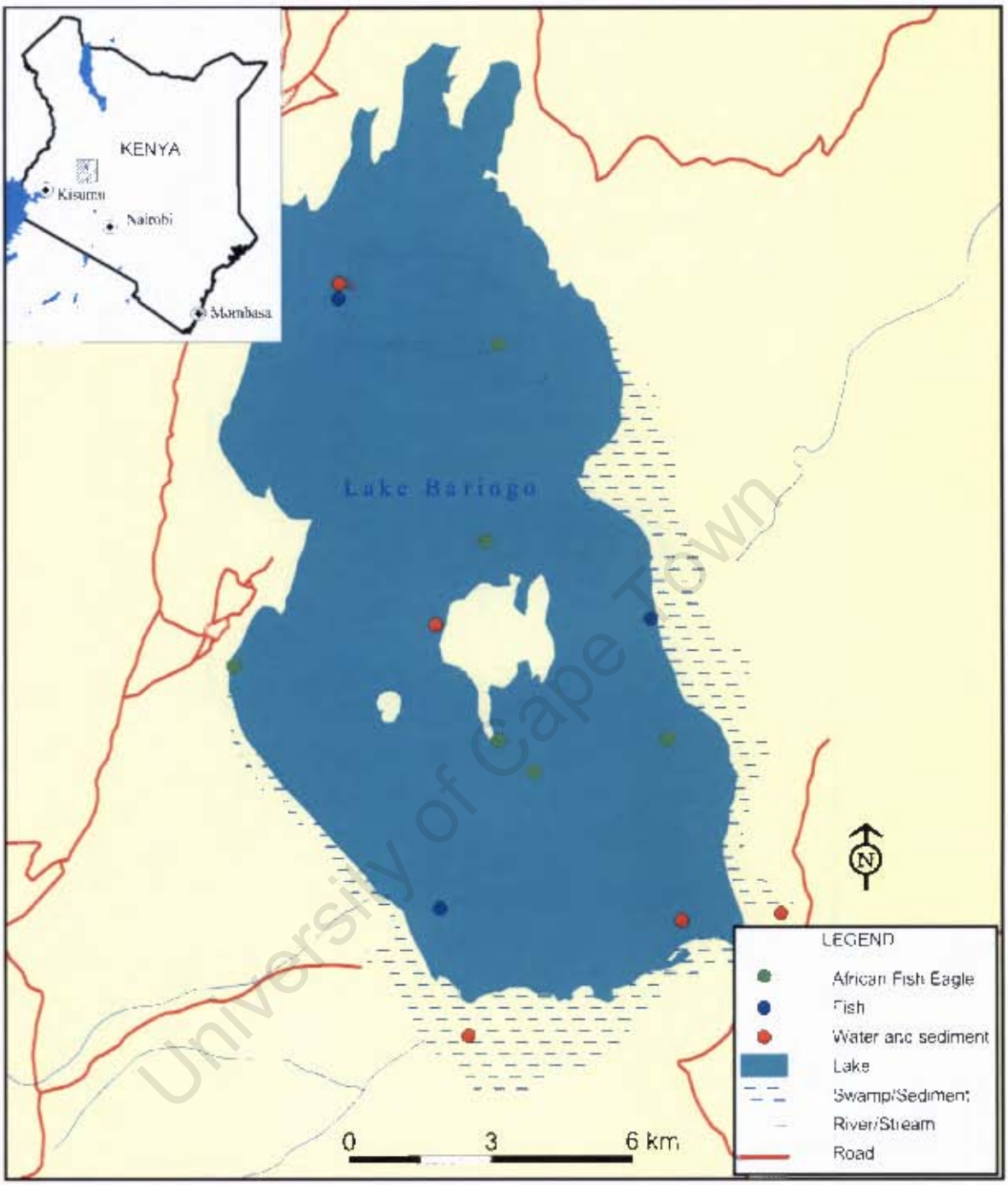

Figure2: Sampling sites for African Fish-Eagle, water and sediment, and fish samples collected from the control lake, Lake Baringo, Kenya, 2010.

At Lake Baringo, river mouths that were still flowing (carrying effluent and waste from upstream) and islands with the highest human population densities were selected for sampling sites. These included the Ol-Kokwe (0° 37' 07"N, 36° 03' 46"E) and Rongena (0° 41' 0.0"N, 36° 02' 40"E) Islands, Ol Arabel (0° 33' 48"N, 36° 07' 42"E)

and El Molo River mouths (0° 33' 4.4"N, 36° 06' 34"E) and the Salabani region (0° 32' 2.5"N, 36° 04' 07"E) between two small river mouths of the Meisori and Ndau (Fig. 3).

Bottle Preparation: Water samples were collected in 1L amber-glass bottles fitted with a glass stopper. Sediment samples were collected in 180ml amber-glass bottles with fitted screw caps lined with clean aluminium foil. Amber-glass bottles were used to prevent sample degradation through UV rays. Each bottle prior to sample collection was washed using clean, hot, water and detergent. Bottles were air dried overnight in a sheltered environment and then rinsed with acetone, to remove any traces of chemical contamination prior to field sampling. Bottles were rinsed in lake water from each specific site before the sample was collected.

Sample Collection: Both samples were collected between 6:30 and 10:30am from each site on the same day. At each sample site two litres of water were collected by grab sampling in individual 1L bottles and were collected 30 cm below the water surface, keeping the stopper on until the appropriate depth was reached. This was done to avoid getting oily residues from surface waters. Sediment samples were collected simultaneously to water samples using an Eckman grab (Cox 2002). All sediment collected in the grab was poured directly into the collection bottle, removing all excess water. Samples were taken from only the top five cm of undisturbed sediment from the lake bed. Multiple Eckman grab samples (two to four) per site were combined to make one sample to obtain a large enough quantity for each sample to be analysed in the lab. No attempt was made in the field to separate meio-fauna and macro-fauna from the samples, as this was conducted at the lab. Each bottle was labelled and stored in a cool box at an ambient temperature (four to six degrees centigrade) maintained by ice packs and chillers. Each sample was labeled with the date, sample type, collectors name and sample code. All bottles were allowed to dry for just over 60 seconds before storing. This allowed any residues on the surface of bottles to evaporate. Gloves were worn when handling bottles to minimise potential external contamination.

At each sample site the GPS co-ordinates were recorded.

***Sample Storage*:** Samples were stored at 4°C both in a cooler after collection as well as in the lab before analysis to prevent rapid biodegradation in the field all sample bottles were stored in a cool box with ice at a temperature of 4°C, and later stored in a refrigerator at 4°C to prevent rapid biodegradation of the sample before laboratory extraction 48 hours after collection.

Fish Samples

Sampling protocol for fish differed between Lake Naivasha and Lake Baringo. This was due to the variation in fishing techniques on each lake and differences in fish species composition. At Lake Naivasha the predominant species during the survey period was Common Carp and therefore the selected species for sampling in Lake Naivasha. Identification of the species was determined by the size, shape and pattern of the scales.

The fish specimens were separated into two groups; those fish collected at Crescent Island and Malewa River Mouth were designated as a low density flower farm area, and specimens obtained from Sher Karuturi, Oserian and HomeGrown sites were designated as a high density flower farm area. Direct observation showed that Lake Naivasha was naturally divided into approximately two segments, related to flower farms. The southern side of Lake Naivasha was found to have a high density of flower farms whereas, the northern side of the lake would have been considered to have a low or sparse density. Oserian, HomeGrown and Sher Karuturi farm sites were from the southern section and Crescent Island and the mouth of Malewa River sites were on the northern section of the lake (Fig. 2).

Three specimens were collected from each of the three most abundant freshwater fish species found in Lake Baringo, at the time of the study, which included: Nile Tilapia (*Oreochromis niloticus*), African Sharptooth Catfish (*Clarias gariepinnus*), Barbus (*Barbus intermedius*) (Aloo 2004). Fresh specimens were purchased from local fishermen while fishing on the lake, from various sites. Specimens were collected from sites at random owing to the nature of the fish distribution. Fishermen were not successful at catching fish at some sites where water and sediment samples were collected (pers. obs.).

Sample Collection: All fish were purchased from local fishermen at their fishing sites. The Common Carp specimens collected during the study weighed between 400g and 850g and were approximately 18 cm in length. The primary method of fishing was carried out using purse seine nets or occasionally by the use of line-fishing. Fish collected for this study were caught by fishermen using seine-nets or fishing lines. For each specimen of fish collected; (i) species (ii) geographical position of where fish were purchased (iii) location of the fish caught and purchased (iv) quantity of fish purchased (v) weight (vi) Identification marking of each specimen to differentiate from one-another, were recorded. Each fish was marked by cutting a small notch in the fin to identify the location at which it had been purchased.

From Lake Naivasha, two fish specimens were purchased at each of five sampling sites in close proximity to the water and sediment sites (10 to 100 m). Live and dead fish were stored in buckets of water for the duration on the boat. All fish were purchased within a few hours of being retrieved by fishermen from their fishing nets/lines. Obtaining fish specimens was opportunistic and dependent on encounters with fishermen at designated sampling sites.

Fish Sample Preparation: Fish specimens had to be dissected to remove specific tissues and organs necessary for this study. The dissection table was prepared using sterilized surgical equipment (scalpels, sutures, surgical gloves and tweezers). Fish specimens were weighed prior to dissection and the weight was recorded to the nearest gram. The first incision was made from the anus to the throat on the under belly of the fish. The muscle tissue (from left and right fillet), liver and fatty tissue and were removed from the fish and packed into separate, sterilized aluminium foil.

Sample Storage: Each aluminium wrapped sample was placed into zip-lock bags and labelled. These were stored in plastic containers and frozen immediately to prevent any sample deterioration. All samples were maintained in a frozen state for transportation to the laboratory. For analysis purposes at the laboratory, samples from Lake Naivasha, collected from the low density flower farm area were combined to make one sample and those from the high density flower farm area were combined to make a second sample. All muscle, liver and fatty tissues were combined during analysis for each of the two samples. Species specific tissue and organ samples from Lake Baringo were combined to create one sample per species, resulting in three fish samples from the Lake. Pooling fish samples from both lakes was necessary to meet the high costs of the sample analysis, if any fish samples were to be included in this study.

Fish Eagle Clotted Blood and Serum Samples

Sample Collection *- Baiting Fish Eagles*

The selection of African Fish-Eagles on both lakes was opportunistic and based on the success of baiting and trapping. African Fish eagles were not considered for baiting if they were perched at a distance greater than 200 m from the water's edge, or those that were too close to another territorial pair or they were rearing chicks. Sampled African Fish-Eagles included both males and females that weighed between 1.95 – 3.23 kg.

Baited fish was required to capture individual African Fish-Eagle for blood sample collection. Common Carp were used as bait for African Fish-Eagle in Lake Naivasha and both barbus (80%) and tilapia (20%) were used as bait for fish eagles in Lake Baringo. The smaller fish (14 cm) had a greater success rate for capturing African Fish-Eagles and were therefore preferred to fish larger than 17 cm, (pers. obs.). The fish entrails were removed and a piece of block-shaped Styrofoam was inserted and stitched in place (using a needle and dental floss) to float the fish belly up.

Four or five monofilament lines approximately 30cm long were pushed into the fish through the center of its back, exiting through the belly (i.e. pointing upwards). A slipknot was tied at the end of each line to create a noose. The nooses were approximately 8 cm in diameter with the knots lying on the fish. The nooses were tied together at the hump located on the back of the fish (Common Carp) and attached to a parachute cord which was in turn attached to a line and drift wood float (heavy enough to prevent Fish Eagles from pulling away). The nooses are thin and black, lying flat on the surface of the water all aiding in their camouflage (Hollamby et al. 2004 b) (Fig. 4).

Figure 3: Noosed Common Carp bait to capture African Fish-Eagles for sample collection from Lake Naivasha (October 2010).

Once a target fish eagle was found, the noosed fish was set afloat onto the water surface from one side of the boat. The boat backed away a safe distance (50 m) to minimise intimidation. When the fish eagle attacked the baited fish and attempted to carry it, one or both nooses closed around the bird's toes and it fell, floating on the water surface. The drift wood float was then quickly approached by boat and the line reeled in first to bring the eagle on board.

Once caught and lowered onto the boat, to subdue the bird a custom-made falconer's leather hood was placed over its head. The eagle was held upright, supported by its tarsus with its wing spread out to enable access to the brachial vein (Bowerman et al. 1994). Before puncturing the vein, the area was disinfected with surgical wipes. Five milliliter syringes were fitted with either a 23 gauge needle (for the smaller sized birds to avoid causing hematomas) or a 22 gauge needle (for larger birds or large veins). Blood collected in the syringe was transferred directly into seven milliliter red-top plastic vacutainer tubes. The tubes were shaken 5 times by hand to mix the blood and then labelled appropriately. From the time of collection until centrifuging, blood was stored upright in cool boxes at a temperature of 4 °C.

The mass of each eagle were taken to the nearest gram using a spring scale in kilograms. To assess the physical health of individuals the tongue, keel and tarsus were observed for abnormalities. Birds were metal-ringed on their lower tarsus and the numbers from any individuals previously metal-ringed were recorded. Fish eagles were kept for no longer than 15 minutes and released. Birds were observed for a few minutes to ensure there were no complications to individuals after release.

Sample Preparation

Blood samples were left to stand for a couple of hours then centrifuged in a hand-held centrifuge for 15 to 20 minutes until the serum separated to the top. The serum was siphoned into a glass transporter vial using glass pipettes and labelled.

Sample Storage

Serum and clotted blood samples were labelled and stored in separate containers. All serum samples in the glass transporter vials and the remnants of whole blood in the vacutainer tubes were stored upright, at - 20^{0}C, until the extraction phase of analysis.

Data Analyses

To compare variables between the two Lakes, Mann-Whitney U tests were performed on the non-normally distributed data. To compare concentrations of organochlorine compounds within the groups of total DDT, total endosulfan and total HCH relative to one another, Kruskal-Wallis ANOVA tests were used. Comparing the variables within each group of compounds to one another a Multiple Comparisons test was used. When comparing metabolites with parent compounds of endrin, aldrin and heptachlor a Wilcoxon Matched Pairs test was used. Differences in African Fish-Eagle compound contamination between high density and low density flower farm areas on Lake Naivasha were compared using a Mann-Whitney U test.

Statistical programs used were Statistica 10.0 and Minitab 13.0. The threshold for statistical significance was set at <0.05.

Pesticide Data Presentation

Some individual organochlorine compounds analysed in this study were associated to a parent compound as either an isomer or a metabolite. The sum of these compounds was calculated and each given its own category, named 'Total'. Compounds with 'totals' calculated include: endosulfan, endrin, DDT, HCH, and heptachlor. Every metabolite and isomer was associated with a 'parent' compound but not all 'parent' compounds were investigated for metabolites or isomers. Technical HCH was divided into its four isomers (α, β, δ, γ), endosulfan was split into its two isomers (I and II) and its metabolite, endosulfan sulphate, technical DDT was separated into its two isomers (p,p'-DDT and p,p'-DDD) and its metabolite p,p'-DDE, however, methoxychlor had no associated compounds. The rest of the organochlorine 'parent' compounds were investigated for their metabolites. The median concentrations of isomers and metabolites associated with their 'parent' compounds were summed, creating an additional organochlorine category and concentration referred to as 'total-suite' of compounds.

Section 2: Laboratory Analysis

All samples of water, sediment, fish and Fish Eagle blood were transported to the Nairobi University laboratory and analysed by Vincent Madadi using gas chromatographic analyses. Samples were analysed for 17 organochlorine compounds: Individual compounds and metabolites, a-HCH, b-HCH, d-HCH, g-HCH(lindane - hexachlorocyclohexane), Heptachlor, heptachlor epoxide, endrin aldehyde, endosulfan I, endosulfan II, endosulfan sulphate, p,p'-DDT, p,p'-DDD and p,p' -DDE. All pesticide standards were available from the Nairobi University, chemistry department. Water, fish and blood samples were analysed on a wet weight basis and sediments were analysed on a dry weight basis. Gas chromatographic analyses were carried out using Varian 3400 series GLC, fitted with Agilent 6890N gas chromatography equipped with electron capture detector. Two, BPX 5 capillary column with dimensions of 60m x 0.25 mm x 0.25 µm and BPX 50 capillary column with dimensions of 60 m x 0.25 mm x 0.25 µm were used. The carrier gas was Helium and nitrogen gas was the mark-up. For further details of the analytical process see Appendix A.

RESULTS

In this section of the thesis and throughout the remainder, the organochlorine compounds are divided into three main 'groups' according to their chemical relatedness.

1. The first group refers to the cyclodiene compounds including, endosulfan (isomers: endosulfan I and II and metabolite: endosulfan sulphate), endrin and endrin aldehyde, aldrin and dieldrin, heptachlor and heptachlor epoxide.

2. The second group are hexachlorocyclohexane (HCH) compounds including (isomers: α-HCH, β-HCH, δ-HCH and γ-HCH also known as lindane), and

3. DDT compounds make up the final group including, isomer: p,p'-DDT, metabolites: p,p'-DDD and p,p'-DDE and sub compound: methoxychlor.

Detection frequencies of organochlorine compounds

All 17 organochlorine compounds (aldrin, dieldrin, endrin, endrin aldehyde, heptachlor, heptachlor epoxide, endosulfan I, endosulfan II, endosulfan sulphate, p,p'-DDT, p,p'-DDD, p,p'-DDE, methoxychlor, lindane, α, β, and δ-HCH) investigated in this study were detected in both lakes. Endrin, endrin aldehyde and p,p'-DDE were the most frequently detected organochlorine compounds, found in 100% of samples from all 5 sample types (Table 2). Dieldrin (LOD, 0.001 ng/ml) and endosulfan I (LOD, 0.007 ng/ml) were detected with the lowest frequency in majority of sample types.

Table 2: Percentage detection frequency of selected Organochlorine compounds in environmental and biological sample types from Lake Naivasha and Lake Baringo, Kenya, 2010.

Organochlorine Compound	DETECTION FREQUENCY (%)									
	Water		Sediment		Fish		AFE Clotted Blood		AFE Serum	
	Naivasha	Baringo	Naivasha	Baringo	Naivasha	Baringo	Naivasha	Baringo	Naivasha	Baringo
	(*n* = 10)	(n = 10)	(n = 5)	(n = 5)	(*n* = 2)	(n = 3)	(*n* = 12)	(n = 8)	(*n* = 12)	(n = 7)
α-HCH	80	30	100	100	100	100	25	50	92	86
β-HCH	100	100	100	100	100	67	100	100	67	100
γ-HCH	10	100	100	100	100	67	100	75	100	86
δ-HCH	70	90	100	100	100	67	92	100	100	86
Heptachlor	20	10	100	100	50	33	25	38	100	29
Heptachlor epoxide	100	100	100	100	50	100	100	63	100	86
Aldrin	90	80	100	100	100	100	92	100	50	100
Dieldrin	90	40	80	60	50	67	42	75	100	57
Endrin	100	100	100	100	100	100	100	100	100	100
Endrin Aldehyde	100	100	100	100	100	100	100	100	100	100
Endosulfan I	100	90	80	40	50	67	67	38	100	100
Endosulfan II	100	100	100	100	50	67	92	88	100	100
Endosulfan sulphate	100	100	100	100	50	100	100	100	100	100
p,p'-DDT	100	100	100	100	100	100	92	100	100	100
p,p'-DDD	100	100	100	100	100	100	92	100	100	100
p,p-DDE	100	100	100	100	100	100	100	100	100	100
Methoxychlor	100	100	100	100	50	50	100	100	100	100

The extent of dieldrin contamination in water samples was higher in Lake Naivasha than Lake Baringo. Sediment samples were the most extensively contaminated, but did not contain the highest concentrations of organochlorine compounds. Fish and African Fish-Eagle samples in Lake Naivasha showed more extensive contamination than samples from Lake Baringo. Serum samples show wider and higher frequency detection of organochlorine compounds, especially endosulfan compounds, than clotted blood samples (Table 2).

Table 3: Comparison of selected organochlorine compound median concentrations in parts per billion measured in water and sediment samples between Lake Naivasha and Lake Baringo, Kenya, 2010. Results that differed significantly between sites are shown in bold.

Organochlorine Compound	Water Median Naivasha (n = 10)	Baringo (n = 10)	Test Statistic	P	Sediment Median Naivasha (n = 5)	Baringo (n = 5)	Test Statistic	P
α-HCH	0.0041	0.0005	149	**0.001**	1.19	0.49	35	0.144
β-HCH	0.0034	0.0196	65	**0.003**	3.15	5.51	20	0.144
δ-HCH	0.0003	0.0004	82.5	0.096	1.36	1.9	29	0.835
γ-HCH	0.0000	0.0017	65	**0.002**	6.11	4.92	29	0.835
Total HCH	0.0080	0.0216	72	**0.014**	11.33	12.08	28	1.000
Heptachlor	0.0000	0.0000	105	1.000	4.85	7.69	21	0.210
Heptachlor Epoxide	0.0016	0.0018	98	0.623	1.13	0.86	34	0.210
Total Heptachlor	0.0016	0.0018	96	0.521	7.23	8.62	22	0.296
Aldrin	0.0007	0.0006	111	0.677	2.11	1.36	27	1.000
Dieldrin	0.0009	0.0000	132	**0.040**	0.21	0.08	26	0.834
Endrin	0.0036	0.0015	136	**0.021**	0.89	1.16	26	0.835
Endrin Aldehyde	0.0097	0.0070	114	0.521	3.38	6.81	17	**0.037**
Total Endrin	0.0131	0.0093	119	0.308	5.93	7.30	22	0.296
Endosulfan I	0.0025	0.0014	145	**0.003**	0.35	0.04	30	0.672
Endosulfan II	0.0062	0.0036	122	0.212	1.67	0.39	30	0.676
Endosulfan Sulphate	0.0202	0.0106	136	**0.021**	5.07	8.65	20	0.144
Total Endosulfan	0.0284	0.0161	142	**0.006**	7.08	8.99	23	0.403
p,p'-DDT	0.0034	0.0035	107	0.910	0.89	0.78	28	1.000
p,p'-DDD	0.0177	0.0043	142	**0.006**	0.29	1.05	25	0.676
p,p'-DDE	0.0028	0.0048	59	**0.001**	0.84	1.25	22	0.296
Total DDT	0.0263	0.0131	134	**0.031**	3.59	2.50	25	0.676
Methoxychlor	0.0479	0.0317	117	0.385	8.95	9.11	24	0.540

Cyclodienes and Their Metabolites

Environmental (water and sediment) Samples

Comparing the cyclodiene compounds between water samples taken from Lake Naivasha and Lake Baringo, dieldrin, endrin, and total-endosulfan (endosulfan I & II and endosulfan sulphate) concentrations were higher in Lake Naivasha. Concentrations of endosulfan sulphate (metabolite) (Table 4) were significantly higher than both endosulfan I and II (isomers) in Lake Naivasha water samples (Multiple Comparisons Kruskal Wallis Test; n = 30, H = 21.30, endosulfan I: p = 0.000 and endosulfan II: p = 0.04).

No significant differences in sediment samples were found for any of the compounds with one exception; endrin aldehyde had a higher concentration in Lake Naivasha (Table 3).

Table 4: Differences in median concentrations of metabolites and isomers for selected organochlorine, parent-compounds in water samples, from Lake Naivasha and Lake Baringo, Kenya, 2010. Significant differences are highlighted in bold.

Lake (n=10)	Metabolites/Isomers				Test Statistic	P
		p,p'-DDT	*p,p'*-DDD	*p,p'*-DDE		
Naivasha		0.003	0.017	0.003	12.65	**0.002***
Baringo		0.004	0.004	0.005	1.86	0.394*
		Endosulfan	Endosulfan II	E. Sulphate		
Naivasha		0.003	0.006	0.020	21.3	**< 0.001***
Baringo		0.001	0.004	0.010	19.78	**< 0.001***
	α-HCH	β-HCH	δ-HCH	γ-HCH		
Naivasha	0.0054	0.0034	0.0003	ND	29.07	**< 0.001***
Baringo	ND	0.0200	0.0004	0.0017	30.61	**< 0.001***
			Heptachlor	H.Epoxide		
Naivasha			ND	0.0016	1.784	0.070•
Baringo			ND	0.0018	2.701	0.006•
			Endrin	E.Aldehyde		
Naivasha			0.0036	0.0097	2.599	**0.009•**
Baringo			0.0015	0.0069	2.803	**0.005•**
			Aldrin	Dieldrin		
Naivasha			0.0007	0.0008	0.663	0.507•
Baringo			0.0006	ND	1.68	0.093•

En Alde= endrin Aldehyde; Hept Epox= heptachlor epoxide; ND= not detectable below 0.040 ng/ml (heptachlor); 0.001 ng/ml (dieldrin), 0.009 ng/ml (lindane); • = Wilcoxon Matched Pairs test; * = Kruskal Wallis Anova test.

Table 5: Comparison of selected organochlorine compound median concentrations measured in fish samples between Lake Naivasha and Lake Baringo, Kenya, 2010.

Pesticide Compound	Median Naivasha (n = 2)	Median Baringo (n = 3)	Test Statistic	P
Total HCH	29.25	38.89	6	1.000
Total Heptachlor	1.21	4.60	4	0.390
Aldrin	7.50	3.36	7	0.773
Dieldrin	0.37	0.01	6.5	1.000
Total Endrin	10.57	6.77	7	0.773
Total Endosulfan	2.73	7.53	3	0.149
Total DDT	5.97	5.16	6	1.000
Methoxychlor	2.96	8.76	3	0.149

Table 6: Comparison of selected organochlorine compound median concentrations measured in African Fish-Eagle clotted blood and serum samples between Lake Naivasha and Lake Baringo, Kenya, 2010. Significant differences are highlighted in bold.

	Clotted Blood				Serum Median			
Pesticide Compound	Naivasha (n = 12)	Baringo (n = 8)	Test Statistic	P	Naivasha (n = 12)	Baringo (n = 7)	Test Statistic	P
α-HCH	0.00	0.53	114	0.300	3.88	1.28	120	1.000
β-HCH	7.04	3.39	147	0.114	5.98	3.19	128	0.526
δ-HCH	12.69	8.50	146	0.133	9.47	8.79	136	0.190
γ-HCH	13.14	7.18	139	0.335	3.50	4.03	119	0.966
Total HCH	37.61	28.47	144	0.177	27.34	18.45	143	0.060
Heptachlor	0.00	0.00	118.5	0.505	0.00	0.00	128.5	0.499
Heptachlor Epoxide	3.95	2.16	156	**0.023**	3.14	2.73	125	0.704
Total Heptachlor	4.97	3.14	138	0.375	8.13	2.73	130	0.422
Aldrin	2.94	1.19	137	0.418	7.85	1.63	150	**0.013**
Dieldrin	0.00	1.16	101	0.203	0.02	1.51	115	0.735
Endrin	1.71	2.31	129	0.847	6.22	3.23	141	0.083
Endrin Aldehyde	8.05	5.45	127	0.969	13.95	5.68	146	**0.031**
Total Endrin	9.66	10.17	120	0.671	21.05	9.65	145	**0.038**
Endosulfan I	0.72	0.00	148	0.082	2.08	2.09	118	0.899
Endosulfan II	1.62	1.13	134	0.563	4.16	0.57	150	**0.013**
Endosulfan Sulphate	10.14	6.74	147	0.114	10.99	7.84	138	0.139
Total Endosulfan	15.09	8.60	149	0.083	15.00	12.86	138	0.139
DDT	2.76	3.08	132	0.671	7.33	3.20	143	0.060
DDD	4.87	2.63	150	0.070	5.81	3.90	130	0.422
DDE	3.74	3.97	133	0.616	3.10	4.49	108	0.331
Total DDT	12.66	8.98	139	0.335	14.93	14.98	125	0.704
Methoxychlor	21.49	24.01	120	0.671	25.5	17.68	126	0.642

Biological (fish and African Fish-Eagle) Samples

In Lake Naivasha during the study, the African Fish-Eagle main fish prey consisted of Common Carp, an invasive species first seen in Lake Naivasha in 2000 (Shiv Kapila pers. comm. 2010 and Harper et al. 2011). No differences were found in organochlorine compound concentrations from fish samples, but aldrin, endosulfan II and total-endrin (endrin + endrin aldehyde) were detected at higher concentrations in Lake Naivasha African Fish-Eagle samples (Tables 5 and 6).

In both lakes, serum samples contained significantly higher concentrations of endosulfan sulphate than endosulfan I and II (Multiple Comparisons Kruskal-Wallis test; Lake Naivasha: n = 35, Z = 12.90, group 1 and 3: p = .004, group 2 and 3 p = .008; Lake Baringo: n = 24, Z = 17.2, group 1 and 3 p = .000, group 2 and 3 p = 0.04).

Table 7: Differences in median concentrations of metabolites and isomers for selected organochlorine parent-compounds in African Fish-Eagle clotted blood samples from Lake Naivasha and Lake Baringo, Kenya, 2010. Significant differences are highlighted in bold.

Sample	Lake					Test Statistic	P
			p,p'-DDT	p,p'-DDD	p,p'-DDE		
Clotted	N		2.76	4.87	3.74	1.38	0.502*
	B		3.08	2.63	6.74	1.42	0.493*
Serum	N		7.33	5.81	3.10	3.08	0.214*
	B		3.20	3.90	4.49	2.23	0.327*
			Endosulfan	Endosulfan	E. Sulphate		
Clotted	N		0.72	1.62	10.14	12.90	**0.002***
	B		ND	1.33	6.74	17.21	**0.002***
Serum	N		2.08	4.16	10.99	16.07	**0.003***
	B		2.09	0.57	7.84	11.72	**0.003***
		α-HCH	β-HCH	δ-HCH	γ-HCH		
Clotted	N	ND	7.04	12.69	13.14	20.4	**0.001***
	B	0.53	3.39	8.50	7.18	9.40	**0.024***
Serum	N	3.88	5.98	9.47	3.50	6.33	0.966*
	B	1.28	3.19	8.79	4.03	9.94	**0.019***
				Heptachlor	H. Epoxide		
Clotted	N			ND	3.94	2.12	**0.034•**
	B			ND	2.16	0.56	0.575•
Serum	N			ND	3.14	1.26	0.209•
	B			ND	2.73	2.03	**0.043•**
				Endrin	E. Aldehyde		
Clotted	N			1.72	8.05	2.67	**0.007•**
	B			2.31	5.45	1.54	0.123•
Serum	N			6.22	13.95	3.10	**0.002•**
	B			3.93	5.68	2.37	**0.017•**
				Aldrin	Dieldrin		
Clotted	N			2.94	ND	2.43	**0.015•**
	B			1.19	1.16	0.56	0.575•
Serum	N			7.85	0.02	2.82	**0.004•**
	B			1.63	1.51	0.00	1.000•

Naivasha = (n = 12); Baringo = (n = 7); En Alde= endrin Aldehyde; Hept Epox= heptachlor epoxide; ND= not detectable below 0.001 ng/ml (dieldrin); 0.040 ng/ml (heptachlor); 0.037 ng/ml (α-HCH), 0.0007 ng/ml (endosulfan I); • = Wilcoxon Matched Pairs test; * = Kruskal Wallis Anova test.

DDT compounds and Methoxychlor

Environmental (water and sediment) Samples

Water samples from Lake Naivasha, contained higher concentrations of total-DDT (p,p'-DDT + p,p'-DDD + p,p'-DDE)when compared to Lake Baringo (Table 3). Concentrations of p,p'-DDD (metabolite of DDT) were detected at the highest concentration, relative to p,p'-DDT and p,p'-DDE in water samples from Lake Naivasha (Multiple comparisons Kruskal-Wallis test, n = 30, H = 12.6, group 1 & 2: p = 0.02, group 2 and 3: p = 0.003).

No differences in concentrations of DDT compounds were found in sediment samples between lakes (Table 3).

Biological (fish and African Fish-Eagle*) samples*

Fish and African Fish-Eagle samples from Lake Naivasha, when compared to Lake Baringo, showed no differences in concentrations of DDT compounds (Table 5 and 6, respectively).

Concentrations of all three DDT compounds showed no significant differences when compared to one another in African Fish-Eagle samples from both lakes (Table 7) i.e., p,p'-DDT, p,p'-DDD and p,p'-DDE concentrations were equal in relation to each other from African Fish-Eagle samples.

HCH compounds

Environmental (water and sediment) samples

Total-HCH (α, β, δ, and γ-HCH) concentrations in water samples from Lake Baringo were higher than in Lake Naivasha (Table 3), and concentrations of β-HCH were significantly higher than δ-HCH in both lakes (Multiple comparison Kruskal Wallis test; n = 40, H = 30.6, Baringo: group 2 & 3: p = 0.000, Naivasha: group 2 & 3: p =0.03).

No differences were found of HCH concentrations in sediment samples, when compared between lakes (Table 2).

Biological (fish and African Fish-Eagle*) samples*

No differences were found of any organochlorine concentrations in African Fish-Eagle or fish samples between lakes (Table 6). However, within each lake, δ, γ isomers showed the highest concentrations in clotted blood samples (Table 7).

High and Low Density Farming Areas

Finally, comparing the organochlorine compounds in water samples taken from within Lake Naivasha in areas with high flower farm density (HFFD) and low flower farm density (LFFD), concentrations of total-HCH were significantly higher in the LFFD area (Table 8).

Methoxychlor in sediment samples was higher in the LFFD area of Lake Naivasha, no differences were found for other compounds (Fig. 4). However, total-HCH in fish samples was higher in the LFFD area of Lake Naivasha (Fig. 5).

Endosulfan I, Endosulfan sulphate, δ-HCH and heptachlor epoxide were significantly higher in the African Fish-Eagle samples from birds with territories in the HFFD area of Lake Naivasha (Table 9).

Table 8: Comparison of median concentrations of selected organochlorine compounds in water samples from a low-density flower-farm area (LFFD) and from a high-density flower-farm area (HFFD) within Lake Naivasha, Kenya, 2010. Significant differences are highlighted in bold.

Organochlorine Compound	Water Median LFFD (n = 4)	HFFD (n = 6)	Test Statistic	P
Total HCH	0.3628	0.0080	32	**0.004**
Total Heptachlor	0.0019	0.0014	28	0.241
Aldrin	0.0006	0.0007	21	0.915
Dieldrin	0.0002	0.0014	10	**0.014**
Total Endrin	0.0143	0.0097	25	0.594
Total Endosulfan	0.0259	0.0337	19	0.594
Total DDT	0.0250	0.0357	20	0.749
Methoxychlor	0.0203	0.0602	10	**0.014**

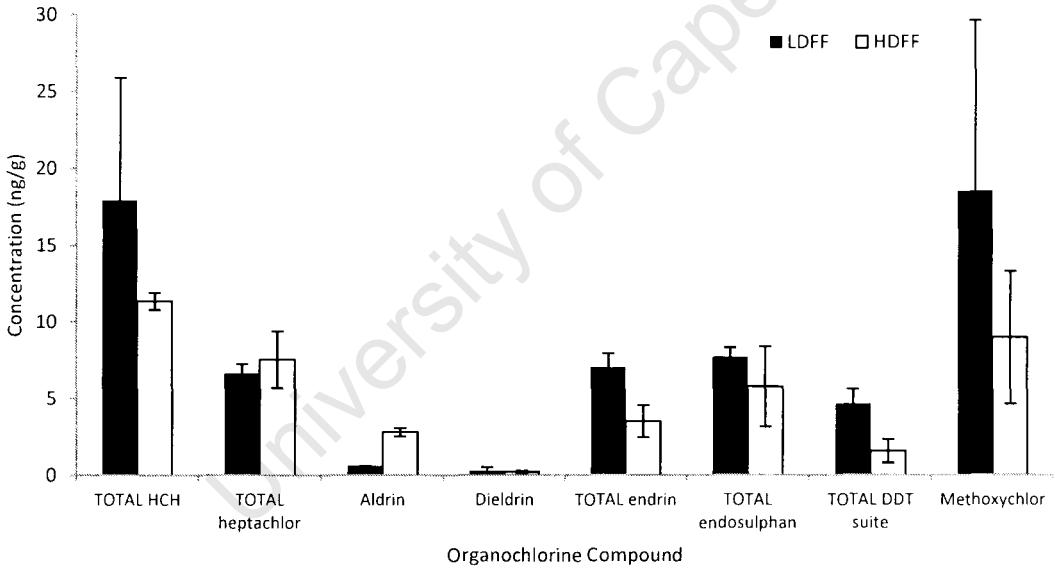

Figure 4: Median concentrations and standard error (SE) of organochlorine compounds in sediment samples collected from a low flower farm density (LFFD) area and high flower farm density (HFFD) area on Lake Naivasha, Kenya, 2010.

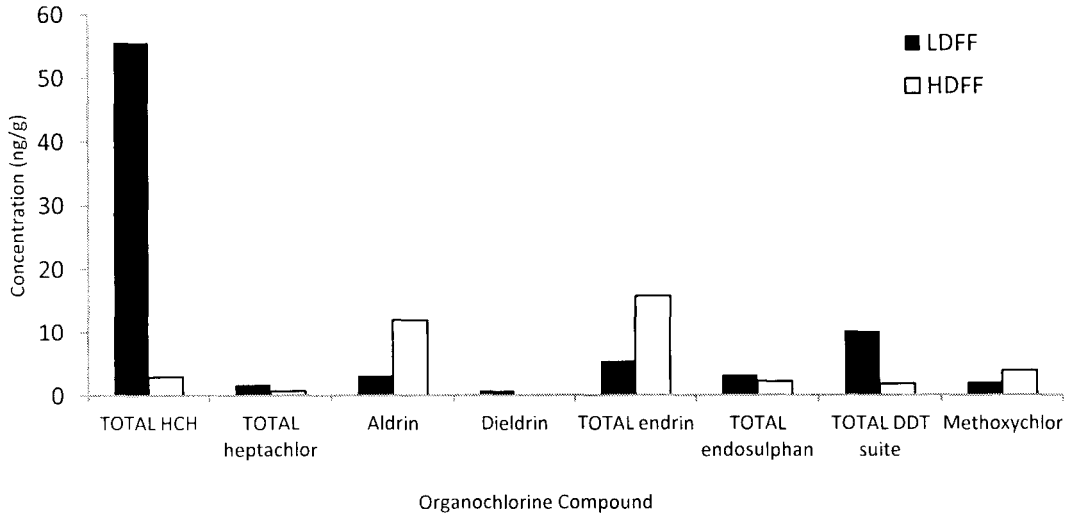

Figure 5: Concentrations of organochlorine compounds in Common Carp collected from a low density flower farm area and a high density flower farm area on Lake Naivasha, Kenya, 2010.

Table 9: Comparison of median concentrations of organochlorine compounds in African Fish-Eagle Serum and clotted blood samples from a low-density flower-farm area (LFFD) and from a high-density flower-farm area (HFFD) within Lake Naivasha, Kenya, 2010. Significant differences are highlighted in bold.

	Serum Median				Clotted Blood Median			
Organochlorine Compound	LFFD (n = 5)	HFFD (n = 7)	Test Statistic	P	LFFD (n = 5)	HFFD (n = 7)	Test Statistic	P
α-HCH	7.2	0.0	37	0.52	0.0	0.0	36	0.63
β-HCH	5.7	6.3	29	0.63	3.9	7.7	25	0.26
δ-HCH	8.7	15.7	18	**0.02**	12.3	18.5	28	0.52
γ-HCH	0.0	10.3	25.5	0.28	10.6	17.3	26	0.33
Total HCH	25.3	48.1	30	0.74	23.0	65.6	22	0.10
Heptachlor	1.2	0.0	37	0.52	0.0	0.0	37	0.52
Heptachlor	3.2	3.1	32	1.00	2.6	5.8	20	**0.05**
Total Heptachlor	7.3	9.0	31	0.87	3.0	5.8	27	0.42
Aldrin	16.5	6.0	36	0.63	2.8	5.9	29	0.63
Dieldrin	0.0	0.1	33	1.00	0.0	0.0	33	1.00
Endrin	6.2	6.2	30	0.74	1.2	2.4	29	0.63
Endrin Aldehyde	12.1	14.2	31	0.87	6.8	10.3	25	0.26
Total Endrin	18.4	21.2	32	1.00	7.8	12.7	27	0.42
Endosulfan I	0.0	3.1	19	**0.03**	0.6	0.8	30	0.75
Endosulfan II	3.6	4.8	32	1.00	3.8	0.8	40	0.26
Endosulfan	10.1	11.2	29	0.62	7.1	15.0	19	**0.03**
Total Endosulfan	15.2	14.8	28	0.52	10.3	16.5	26	0.33
p,p'-DDT	4.8	9.9	32	1.00	2.7	6.5	26	0.33
p,p'-DDD	5.0	6.6	32	1.00	4.8	5.9	30	0.75
p,p'-DDE	4.6	2.3	32	1.00	4.1	3.3	33	1.00
Total DDT	13.0	15.4	30	0.74	11.1	20.5	30	0.75
Methoxychlor	26.7	24.9	37	0.52	11.9	25.0	26	0.33

DISCUSSION

This short study set out to determine the level of pesticide contamination in two lakes that differ in the land use and history. Both lakes Naivasha and Baringo are fresh-water lakes that are used by the top predator, the African Fish-Eagle. This species was used to determine if the high concentration of flower farms on the shores of Lake Naivasha was adding to the pesticide burden of the lake and compromising the lake's ability to function as a healthy eco-system. It is important to remember that several pesticides have been banned in Kenya and this is important in interpreting some of the concentrations detected in this study. In Kenya, DDT was banned in 1997, endrin and heptachlor are banned, lindane, aldrin and dieldrin are restricted in their use, while only endosulfan and methoxychlor have no legal restrictions (Lalah et al. 2003).

Patterns of Pesticide Accumulation

The studies conducted by Mugachia et al. (1992) and Gitahi et al. (2002) are among the few Kenyan studies previously conducted to examine organochlorine impacts at Lake Naivasha. They have created some of the baseline data for environmental and piscivorous samples in the area, and in an attempt to gain an insight into long-term trends of pesticide pollution occurring at the comparisons will be made to the results found in this current study.

Organochlorine concentrations detected in Common Carp in this study were higher than concentrations in fish samples from Mugachia et al. (1992), since no traces of p,p'-DDT, p,p'-DDE, lindane and α-HCH were found. However, Common Carp samples from this study were detected with lower concentrations of organochlorine compounds than Large-Mouth Bass (*Micropterus salmoides*) from Gitahi et al. (2002). Whereas, Common Carp concentrations from this study were similar to Louisiana Crayfish (*Procambarus clarkia)* (i.e., p,p'-DDT: 4.6 ng/g) from Gitahi et al. (2002).

Both this present study, and Gitahi et al. 2002, shows similarly low organochlorine levels of contamination due to detections in the parts per billion concentration range as oppose to in parts per million ranges (Lalah et al. 2003). Small differences in concentrations between fish samples can be attributed to the amount of fat present in them (Munga 1990, from Gitahi et al. 2002) as species with higher fat contents store more pesticide residues (Munga 1990). Common Carp samples were comprised mostly of muscle tissue, as the species have small amounts of fat (pers. obs.) compared to higher fat contents in Large Mouth Bass (Gitahi et al. 2002). Large Mouth Bass are also the more predatory of the two species therefore influencing the contamination potential in the species over Common Carp Ros Boar pers. Comm. Common Carp tissue from this study was contaminated with aldrin, HCH, dieldrin, endrin, heptachlor and their metabolites, whereas *Oreochromis niloticus* samples from Hollamby et al. (2004 a), showed no contamination of these compounds. This suggests that detection of cyclodiene and HCH compounds could be attributed to more extensive, intensive and recent uses in Kenyan, rather than Ugandan Lakes.

The absence of organochlorine compounds (endosulfan II, dieldrin, aldrin, p,p'-DDT and p,p'-DDE) in water and sediment samples from Gitahi et al. (2002), appears to indicate an increase in the amount of contamination in Lake Naivasha between the years 2002 and 2010. This coincides with the increased intensity of agricultural development around the Naivasha region (Harper et al. 2011), and studies have shown an increase in intensive agriculture to negatively impact avifauna (Donald et al. 2001).

Frank et al. (1977), Brown (1980), and Lincer et al. (1981) were the first to conduct studies that surveyed organochlorine contamination in birds of prey associated with Lake Naivasha. Although the studies were not specific to African Fish-Eagles, there were several samples incorporated into the studies, providing a general indication of contamination levels from the 1970s and 1980s period. African Fish-Eagle clotted blood and serum samples from both lakes in this study were contaminated with p,p'-DDE and dieldrin below 10 ppb (no risks of toxicity below this level) as found in Frank et al. (1977). Both previous studies therefore, found similar, low level contamination in the species. In contrast, far lower concentrations of p,p'-DDE were found from African Fish-Eagle clotted blood and serum samples in this study, compared to a contaminated African Fish-Eagle egg sample (1 500 ppb) from Lincer et al. (1981). A comparison between blood and eggs of the same species is possible because organochlorine compounds in eggs are closely matched with those from blood (Iseki et al. 2001). Concentration differences between these studies may be attributed to variations in organochlorine insecticide uses over decades (Wandiga 2001) but generally comparisons between this study and previous ones are hard to make, due to their small, un-representative sample sizes and different sampling techniques.

Comparisons between other similar studies (e.g. Hollamby et al. 2004 a), with larger sample sizes may provide an enhanced indication of the contamination status of Lake Naivasha than comparisons with the older studies (e.g. Frank et al. 1977). Concentrations of Aldrin, HCH, dieldrin, endrin, heptachlor and their metabolites were detected in African Fish-Eagle samples from this study, whereas, none were detected (1.0 ppb detection limit) in adult African Fish-Eagle plasma samples from Lake Victoria and Lake Mburo in Uganda from Hollamby et al. (2004 a). Detections in Kenyan samples could suggest more contamination and/or greater previous and current uses of these organochlorine compounds in Kenyan than Ugandan lakes. Similar levels of p,p'-DDE were detected in African Fish-Eagle samples from this study and Hollamby et al. (2004 a) who detected mean concentrations of 2.6 ppb wet weight perhaps indicating similar extent and quantities of DDT used in both countries. The main use of DDT in Uganda (Hollamby et al. 2004 a) and Kenya (Wandiga 2001) was predominantly used to control malaria epidemics.

The blood organochlorine concentrations from this study were more similar to the plasma concentrations detected in Cape Griffon (*Gyps coprotheres*), Lappet-faced (*Trogos tracheliotos*) and African White-backed Vultures (*Pseudogyps africanus*)(Organochlorine range: 0.0 – 24.3 ug/l wet weight) from four sites in South Africa (Van Wyk et al. 2001). However, Dhananjayan and Muralidharan (2010), found slightly higher

concentrations in Besra Sparrowhawk (*Accipiter virgatus*) (1.1 – 166 ng/ml) and Barn Owl (*Tyto* alba) (0.0 – 79.3 ng/ml) plasma samples from Ahmedabad, India, than concentrations from this study, although still within the parts per billion (ppb) range.

Status of Contamination in Rift Valley Lakes

Few organochlorine compounds in this study showed large concentration differences in sample types between Lake Naivasha and Lake Baringo with the exception of Endosulfan, endrin and aldrin, which had higher concentrations in water and African Fish-Eagle samples from Lake Naivasha. The detection of endosulfan residues in Lake Naivasha is a direct result of its use (Wandiga 2001) in horticulture and floriculture activities around the lake (Gitahi et al. 2001). The presence of aldrin indicates recent use or exposure, albeit in small quantities, since it is known to breakdown rapidly into dieldrin in the environment (Walker 2001, Mineau 2003, Rivera-Rodriguez et al. 2007) and in vertebrates, once in the food chain (Van Wyk et al. 2001, Walker 2001, Rivera-Rodriguz et al. 2007). No difference was found in the sediment and fish sample concentration of lindane, heptachlor and endrin aldehyde potentially indicating that the sample size was too small to detect any differences.

Organochlorine compounds investigated in this study were detected in all sample types from both lakes, indicating their high frequency and wide distribution. The high detection frequencies in the majority of organochlorine compounds (except endosulfan I, and dieldrin), was unexpected due to their rapid decomposition in tropical (WHO 1989, Wandiga 2001) aquatic systems (Koeman et al. 1972, Frank et al. 1977). Metabolite p,p'-DDE was one of the most frequently detected organochlorine compounds and is arguably the most persistent of the organochlorine residues (Walker 2001), as it has been found at the highest frequency in other similar studies (Van Wyk et al. 2001, Cid et al. 2007, Dhananjayan and Muralidharan 2010).Endosulfan sulphate, a metabolite of endosulfan which is commonly used in Kenyan agricultural activities, was frequently detected in both lakes. The higher frequency of endosulfan sulphate (metabolite) detected than endosulfan isomers (insecticidal endosulfan) would appear to be a product of old endosulfan application but misleading due to insecticidal endosulfans rapid breakdown in warm blooded (endothermic) organisms (Gorbach 1988, Walker 2001, Martinez-Lopez et al. 2008) and the environment (Lalah et al. 2003) within four weeks (Cox 2002) so its application is in-fact relatively recent.

The detection frequencies of endrin and endrin aldehyde (metabolite) as equal to endosulfan sulphate but greater than dieldrin, in this study is unexpected. Endrin is considerably less persistent than dieldrin in vertebrates (Walker 2001), yet in this study, it has a higher detection frequency than dieldrin. Banned in the 1980s (Wandiga 2001) with a half-life in water of four years in temperate regions (WHO 1989), the detection of endrin compounds are potentially due to illegal use or long-range atmospheric transport from other regions (Lalah et al. 2003, Cid et al. 2007), or both. Lalah et al. (2003), detected high levels of endrin in water samples

from Tana River in Kenya, attributing its concentrations to illegal use. Its low concentration potentially suggests this is atmospheric contamination.

Aldrin and dieldrin were among the least frequently detected organochlorine compounds, particularly in African Fish-Eagle samples, from both study lakes. Cid et al. (2007), also found low detection frequencies of the two compounds in bird tissue samples from Argentina, corroborating findings in this study. Dieldrin was also not detected in African Fish-Eagle samples from Uganda, despite being highly persistent in the environment (Blus et al. 1996), but it was registered for restricted use and has a history of being used around Kampala (near the study site), (Hollamby et al. 2004 a). Perhaps the compound is more readily excreted in the African Fish-Eagle species than other *Haliaeetus* species, found in temperate areas.

The most conspicuous differences between the two lakes were the significantly higher concentrations in water samples, of lindane from Lake Baringo, and endosulfan from Lake Naivasha. The role of small sample size in sediment and fish samples has already been explored as a possible reason for the lack of differences. However, the African Fish-Eagle samples also showed little variation. This could suggest that the prime source of contamination into the food chain is from sediment, where the bulk of the organochlorine residues in the environment are stored (LeBlanc et al. 2004).

Risk Assessment and Toxicity

Generally, concentrations of organochlorine compounds detected in the various sample types from both study lakes were not considered hazardous to humans or wildlife. Lalah et al. (2003), reports that water, sediment, or fish samples are considered to be less polluted if their organochlorine contamination concentrations are in the range of ng/g (parts per billion), in comparison to mg/kg (parts per million), which are considered more polluted. Sample types in this study are all between the ng/l (parts per trillion) and ng/g (parts per billion) range, thus both study lakes may be considered relatively un-polluted. From the literature , organochlorine contamination in birds of prey, eggshell thinning, avian mortalities, and population declines all occur in the mg/kg (parts per million) range, approximately 1000 times higher than concentrations detected in African Fish-Eagle samples in this study.

Some concerns are highlighted with regards to organochlorine concentrations in water and sediment samples from the study lakes. The United States Environmental Protection Agency (US EPA) has established guidelines related to organochlorine chronic exposure to aquatic organisms (Hamilton et al. 2003) (Table 11). Total-endosulfan, p,p'-DDT and methoxychlor levels detected in water samples from both lakes exceed the chronic-exposure guideline levels recommended (Table 10). The concentrations of these three compounds, if maintained over a period of three or more years, could cause negative impacts (i.e., mortalities, population declines) on the aquatic organisms, impacting the food chain in both study lakes concerned (De Lorenzo et al. 2001). Sediments from both Lakes Naivasha and Baringo have been identified in this study to carry the main pesticide burden of all the sample types analysed. Sediments act as sinks for organochlorine compounds, as a

result of slower decomposition than both, water and vertebrates, as such their concentrations have the potential to reach significant levels (Walker 2001). In sediments, heptachlor, aldrin, and lindane concentrations in Lake Naivasha, and β-HCH and heptachlor in Lake Baringo, are equal to, or above, recommended TEC (Threshold Effects Concentrations) guidelines (Table 10). Concentrations above these TEC levels could lead to potential threats to aquatic organisms (Wasswa et al. 2011), especially benthic dwellers (Table 11).

In bird species, Mineau et al. (2009), suggests that the average toxicity associated with specific organochlorine compounds occurs at concentrations in the mg/kg range (parts per million) (i.e., aldrin: 19.8 mg/kg, dieldrin: 35.1 mg/kg, lindane: 90.8 mg/kg, and DDT: 1334 mg/kg). The four compounds listed above were detected at far lower concentrations in African Fish-Eagle blood samples from this study and as such posed no toxic threats to the birds. Although useful for bench marking toxicity, toxicity levels are often species specific. Henny et al. (1983), reported heptachlor epoxide levels of 1 500 ppb, caused, reduced productivity in American Kestrels. Endrin is known to be one of the most acutely toxic organochlorine compounds (Fox and Lock, 1978), around 100 times more toxic to quail than any DDT compounds (De Witt 1956). Whereas, p,p'-DDE residues in eggs of Bald Eagles and Sea Eagles induce reproductive failure at 15 000 ppb (Wiemeyer et al. 1984) and 26 000 ppb (Helander et al. 1982), respectively. Organochlorine residues in African Fish-Eagle in this study have been detected at similar levels in various raptor species from Bowerman et al. (2000) and Hollamby et al. (2004 a). Both authors concluded that the raptors were under no immediate danger from organochlorine residues at levels reported.

Table 10: WHO Guidelines (GV), Threshold Effects Concentrations (TEC) and FDA regulations for selected organochlorine compounds in sediment, water and fish.

Pesticide Compound	Sediment TEC (ng/g)	Water GV (ug/l)	Fish (ng/g)
Aldrin	2.00*	0.03†	300•
Dieldrin	1.90*	0.03†	300•
α-HCH	4.50*	1.00†	300•
β-HCH	6.00*	1.00†	300•
δ-HCH	5.00*	1.00†	300•
γ-HCH	2.37*	2.00†	300•
Heptachlor	1.00*	0.03†	300•
Heptachlor Epoxide	2.47*	0.03†	300•
p,p'-DDT	4.20*	2.00†	5000•†
p,p'-DDD	3.16*	0.06‡	5000•†
p,p'-DDE	3.54*	0.06‡	_
Endrin	_	0.60†	_
Endrin Aldehyde	_	0.60†	_
Methoxychlor	_	20.0†	_
Endosulfan	NA*	0.05‡	_

*Wasswa, J. (2011), • U.S. Food and Drug Administration (1994), † WHO (2006).
† WHO. (1993), ‡ Hamilton et al. (2003).

Table 11: US EPA water-quality criteria for aquatic organisms of selected organochlorine compounds, acute and Chronic data are summarised from Hamilton et al. 2003.

Organochlorine Compound	Acute (ug/l)	Chronic ug/l)
γ-HCH	2.00	0.0800
TOTAL HCH	6.00	3.6800
Heptachlor	0.52	0.0038
Heptachlor epoxide	0.52	0.0038
TOTAL heptachlor	0.52	0.0038
Dieldrin	0.36	0.0651
Endrin	0.19	0.0610
TOTAL Endosulfan	0.22	0.0056
p,p'-DDT	1.10	0.0010
Methoxychlor	-	0.0300

Illegal Pesticide Uses: Distinguishing Current versus Previous Use

The majority of the organochlorine compounds showed overall low concentrations, but higher concentrations of their metabolites, indicating exposure as a result of old organochlorine applications (before the ban of some organochlorine compounds) (Wandiga 2001, Walker 2001, Lalah et al. 2003). The higher concentration of aldrin than dieldrin, in water and African Fish-Eagle samples from Lake Naivasha, however, indicates relatively recent contamination by aldrin, as it is readily broken-down into dieldrin within 8 weeks in water (Eichelberg and Lichtenberg 1971 *in* Lalah et al. 2003). In vertebrates dieldrin becomes the dominant compound (Van Wyk et al. 2001). Naivasha is a malaria-free area and is therefore unlikely to have been treated with aldrin (substituted for DDT for mosquito control). Termite control is the only other legally accepted, albeit restricted, use for aldrin in Kenya (Lalah et al. 2003), hence this appears to be a source of the contamination. Alternatively, atmospheric deposition is also a plausible source, due to the volatile nature of organochlorine compounds (Walker 2001). The intense solar radiation in tropical environments (Wandiga 2001) accelerates the volatility of compounds and results in their atmospheric transport (Lalah et al. 2003).

It is also worth mentioning the higher concentration of p,p'-DDD than other DDT compounds in Lake Naivasha water samples. Rothane (p,p'-DDD) was also marketed as an insecticide on its own (Walker 2001), and its high levels detected could be a result of direct Rothane use, independent of DDT. In this instance, contamination appears to be relatively recent, because p,p'-DDD is not as persistent as p,p'-DDE (Cid et al. 2007).

The Role of Agriculture and Floriculture

Legally, the only organochlorine compound accepted for use by the horticulture and floriculture farms around Lake Naivasha, and in Kenya as a whole, is endosulfan (Lalah et al. 2003, PCPB 2008). Therefore, I expected to find differences in endosulfan compounds between areas of high and low flower farm densities. However, in

neither water, fish, nor sediment samples are there any indications of higher endosulfan concentrations from high flower farm density (HFFD) areas compared to low density (LFFD) areas. Never the less , African Fish-Eagle individuals with territories in the HFFD area showed significantly higher concentrations of endosulfan I (isomer) and endosulfan sulphate (metabolite). This suggests that African Fish-Eagles on the HFFD side of the lake are impacted by the chemical compounds entering the system from flower farms. The absence of differences in water, sediment samples is attributed to continuous mixing of lake water and sediment by wind induced wave action (Tarras-Wahlberg et al. 2002, Cope 1966), and not only due to differences in chemical quantity used. Other organochlorine compounds demonstrating higher concentrations in African Fish-Eagle samples, from the HFFD area, are: δ-HCH and heptachlor epoxide. Heptachlor is banned from use in Kenya. Therefore, the detection of its metabolite (heptachlor epoxide) in African Fish-Eagle blood could be attributed to bioaccumulation, through diet (Common Carp contaminated by the sediment) or as a result of atmospheric transport (Hollamby et al. 2004 a).

In Common Carp samples, total-HCH showed the most striking difference in organochlorine concentrations between HFFD and LFFD areas, considerably higher in the LFFD area of Lake Naivasha. Fish samples representing the LFFD area of the lake were collected predominantly from locations close to Malewa River mouth. Water samples also contained a significantly higher concentration of total-HCH in the LFFD area, than the HFFD area. Based on these findings, farming activities upstream on the Malewa River are the likely source of lindane and technical HCH contamination in Lake Naivasha. Harper et al. (2011), reported increases in wide-scale agriculture along river banks in the Naivasha basin, increasing soil erosion and sedimentation. As a result, some organochlorine residues previously locked in the soil may be released and washed into the lake, as well as chemicals currently used. This may also be the main source of organochlorine contamination in Lake Baringo, due to the large amounts of soil erosion from deforested highlands and agricultural areas (Hickley et al. 2004) , leaching into the lake. The main farming activities upstream are subsistence cultivation, animal husbandry (Kitaka et al. 2002), maize, vegetables, and fodder production (Becht 2007). Animal husbandry activities could be responsible for illegal lindane (γ-HCH) usage (tick control: Wandiga 2001) or maize cultivation, as lindane is restricted to use in only seed dressing and termite control (Lalah et al. 2003).

Ecosystem Level Impacts and African Fish-Eagle

Direct Impacts

The concentrations differences detected in organochlorine compounds between water and sediment samples were vast. However, the concentrations detected in fish and in African Fish-Eagles in this study showed small differences in organochlorine compounds. This could suggest that the hydrophobicity of organochlorine compounds is very high, leading to residues binding with sediment particles and debris. It would suggest that some organochlorine compounds entering the system are coming through soil and river sediment. In the food chain, the similarities in concentrations between prey and predator indicate a weak biomagnification process. However, the similarities also show evidence of bioaccumulation. Avifauna are also exposed to pesticides in a

variety of ways, independent of their diet (Mineau 2003). Highly volatile chemicals like organochlorine compounds are transported through the air and depending on the type of pesticide application methods used in the surrounds farms, contact varies. If spraying is the method of choice, fine mist can be transported in the air and inhaled or absorbed through the skin of birds, potentially causing toxic manifestations.

Indirect Impacts

Low breeding success and population instability in African Fish-Eagle has been linked to food scarcity, particularly during drought periods (Harper et al. 2002, Harper et al. 2011). In this section the possibilities of organochlorine contamination contributing to the problem of food shortages in Lake Naivasha will be explored. Evidence of potential threats to aquatic organisms constituting the lower trophic and food chain levels with regard to organochlorine residues (De Lorenzo et al. 2001), has already been highlighted in this study. Lake Naivasha is a particularly complex web of interactions as a result of introduced species, confounding the dynamics of the lake ecology and food chains. As a general rule, disturbances induced in the lower trophic levels (i.e. aquatic organisms) have an indirect effect on the higher trophic levels (i.e. African Fish-Eagle). The predominant diet of African Fish-Eagle in both study lakes is comprised of fish, and at Lake Naivasha in particular, *C.carpio*. Aquatic organisms (phytoplankton, diatoms, aquatic-insects) and detritus make up approximately 30% of the Common Carp diet and submerged macrophytes make up 40% of the diet (Njiru et al. 2008) and Common Carp fry feed on phytoplankton (USGS 2005). The alternative, but significant, dietary source for African Fish-Eagle are water birds i.e., Red-Knobbed Coot (*Fulica cristata*), but they too depend upon submerged macrophytes and aquatic insects for their food.

Harper et al. (2011), reports that the current situation at Lake Naivasha is a reduction in submerged macrophytes, as a result of water hyacinth blooms. However macrophytes are also a major food source for the Louisiana crayfish that have in the past been responsible for the collapse of benthic plant communities within a short span of time (Harper et al. 1995, Hickley and Harper 2002, Harper et al. 2011). The reduction of macrophytes, the main food base of both Common Carp and Red-Knobbed Coot would potentially induce a heavier pressure on their alternative food, e.g. aquatic organisms. This is where the organochlorine contamination levels in water and sediment could become a concern, by causing harmful impacts to the aquatic organisms at the toxic levels detected in this study (Table 2), leading to mortalities or behaviour disorders, affecting their abundance. A reduction in food availability for Common Carp and Red-Knobbed Coot provides few alternatives for African Fish-Eagle's diet, despite being a robust opportunistic scavenger. As a consequence of limited food options, due to trophic cascading effects (Frank et al. 2007), African Fish-Eagle productivity and populations could decline or fall far below carrying capacity.

It is possible that the Lake Baringo ecosystem is also affected by the organochlorine compounds in a similar way to Lake Naivasha, directly affecting the lower trophic levels and eventually impacting the top predators. Although the organochlorine contamination may not be the primary cause of reduced fish and African Fish-

Eagle populations (Hickley et al. 2004) it could compound the problems already affecting the lake productivity e.g. siltation.

Additionally, during periods of high water levels in Lake Naivasha (e.g. 2010, during this study), the water hyacinth could act as a medium for rapid atmospheric release of organochlorine residues from water and sediments. Studies have shown that water hyacinth increases the amount of surface water loss, through evapo-transpiration (Adams et al. 2002), this process coupled with the high volatility of organochlorine compounds would enable rapid evaporation of organochlorine residues through water hyacinth into the atmosphere. Organochlorine residues from sediment would be absorbed through water hyacinth roots. The result would be an increase in the atmospheric content of organochlorines, a potential decrease in water and sediment concentrations and a reduction in harmful effects to the aquatic organisms. The displacement of the organochlorine burden from within the lake into the atmosphere will perpetuate their long-range atmospheric transport (Cid et al. 2007). This whole process is subject to water levels in the lake as drought periods reduce the water levels considerably, decreasing the extent and quantity of water hyacinth (Harper et al. 2011). However, during periods of heavy rainfall the water hyacinth blooms and spreads, covering vast areas of the lake water surface (Harper et al. 2011).

The Wider Ecosystem Implications

Harper et al. (2011), reports that some bird species are seen much less frequently in Lake Naivasha today than in the past, including the African Darter (*Anhinga rufa)*, Great Crested Grebe (*Podiceps cristatus*), Great Egret (*Casmerodius albus*), Saddle-billed Stork (*Ephippiorhynchus senegalensis*), White-backed Duck (*Thalassornis leuconotos*), Baillon's Crake (*Porzana pusilla*), African Skimmer (*Rynchops flavirostris*), Red-knobbed Coot (*Fulica cristata*) and Yellow-billed Duck (*Anas undulata*). The reasons for declines in waterbirds have also been associated with food-shortages, as found with African Fish-Eagle at Lake Naivasha (Harper et al. 2002). Although waterbird species occupy various niches, piscivorous waterbirds and African Fish-Eagle species are part of the same food chain and would be impacted by similar food-limiting factors, where organochlorine contamination could have an influence, particularly on aquatic organisms at the lower levels of the food web(as described in paragraph two of ecosystem level impacts). This further emphasises the benefits of using apex predators: African Fish-Eagle to determine wider ecosystem implications of pesticide usage, especially in Lake Naivasha, an area of conservation importance as an Important Bird Area (IBA), and RAMSAR site, that not only support a high diversity and richness of fauna but is also an incredibly important resource to the local community of Naivasha.

When conservation goals are concerned with pesticide toxicology, attention needs to be focused on organochlorine compounds, as well as widely used Organophosphates and Carbamates in respect to the growing intensive agricultural practices throughout Kenya, if an understanding is to be gained of their long-term impacts to the ecosystems in which they are deposited. Organophosphates and carbamates are more predominant in agricultural practices around Lake Naivasha (Gitahi et al. 2002) and demonstrate high toxicity and short environmental persistence. Their toxic effects, which are related more to acute cases of poisoning (Mineau 2005 a) than chronic long-term effects, have been globally documented (Mineau 2003) as well as in Kenya (Paula Kahumbu, Pers. Comm. 2009). Detecting organophosphates and carbamates are more difficult as a result of their chemical properties, but it has been done in the past (Mineau 2003). This study has shown that a connection exists between organochlorine compounds used in farming practices adjacent to Lake Naivasha and their contamination in the ecosystem and its food chains. It would therefore appear that a similar connection is likely to exist for organophosphates and carbamates insecticides. Reported mortalities of 13 African Fish-Eagle adults from Lake Baringo in 2006, were the result of acute poisoning by a CBM, commonly known as Furadan (Kapila, S. Pers Comm. 2012). The implications of organophosphate and carbamate uses, with regard to wildlife, in the Lakes are worrying, to say the least.

Study Limitations and Areas for Improvement

Laboratory Analysis

1. The type of pesticide contaminants investigated in field research is sometimes limited by their availability in local laboratories. The availability of organophosphate and carbamate pesticides in Kenya are limited due to their short life expectancy (retention time) and difficulty associated with

importing hazardous substances into the country. Usually, orders must be placed by specific chemical importers to outsource standards (i.e., from Germany), a time consuming and cumbersome process, taking several months.

2. In Kenya, the field of toxicology is still developing, and as a result, few internationally-certified, independent laboratories with the necessary equipment to conduct pesticide analyses are located here. Research projects requiring specialised equipment may be restricted due to high costs and unreliable equipment.
3. This study was therefore forced to use a cheaper and more readily available organochlorine combination standard (more than one pesticide standard), as well as limit the number of samples analysed for organochlorine pesticides.

Field Sampling

4. Collecting blood samples from African Fish-Eagle was a time consuming, costly, and logistically difficult task requiring the use of a boat and limited by the site population size. Lake Naivasha had a larger population than Lake Baringo hence fewer African Fish-Eagles were caught from Lake Baringo.
5. The choice of a 'control Lake' was governed by two factors: the lake had to be fresh-water and it had to have a resident African Fish-Eagle population, making Lake Baringo the only option in Kenya. Logistical and time constraints prevented the study from extending into other countries in search of an undisturbed fresh-water lake with no or limited peripheral agricultural activities.
6. The number of samples and sample sites were restricted by cost factors, resulting in a reduced sample size for fish tissue samples.
7. Water samples were included in the study to gauge the pesticide compounds being introduced into the lake from high and low density flower farm areas on Lake Naivasha as a substitute for actual data from the farms themselves. The study was limited by the lack of information from flower farms related to pesticide application frequencies, times, and quantities. Obtaining information directly from farms would have aided in differentiating between recent and old contamination but data was not available due to uncooperative farm representatives.

CHAPTER 3: CONCLUSIONS AND SYNTHESIS

The major conclusions of my study are: 1. Pesticides currently used in agricultural practices around Lake Naivasha have caused contamination of the lake. 2. Concentrations of organochlorine compounds in African Fish-Eagle are not at levels where egg-shell thinning, mortalities and low reproductive success will be induced. 3. Organochlorine compounds occur at levels of concern in sediment and water samples with possible negative impacts on aquatic organisms, causing trophic cascades affecting the food chain. My predictions (Introduction) were partly borne out that 1. Sites with a high density of flower farms are more contaminated with endosulfan than sites with low densities of flower farms within Lake Naivasha.

Overview, Future Studies and Recommendations

This study highlights some of the major concerns with developing intensive agriculture and its potential impacts on Kenyan aquatic ecosystems.

The agricultural practices around Lake Naivasha have caused contamination of the lake ecosystem and its wildlife by HCH, methoxychlor and endosulfan compounds. The presence of some compounds i.e., p,p'- DDD indicate possible illegal use in the Naivasha region. There appears to be two distinct sources of certain organochlorine contaminants at Lake Naivasha. The farms along the immediate, southern riparian edge of the lake appear to be the source of endosulfan contamination in African Fish-Eagle samples, and the farms upstream of the Malewa River appear to be responsible for the contamination of Common Carp and water and sediment samples by HCH compounds and methoxychlor.

African Fish-Eagle organochlorine blood concentrations such as p,p'-DDT, p,p'-DDE, dieldrin, and heptachlor were detected at similarly low concentrations as those during and after the 1970s as reported by, Frank et al. (1972), Brown (1980), Lincer et al. (1981), indicating that eagles from both Rift Valley Lakes were not exposed to high or harmful concentrations of these compounds individually. However, potential threats could manifest themselves due to the combined effect of all 17 organochlorine compounds, heavy metals and other pollutants (Donaldson and Hunter 1999) present in these two ecosystems. Pesticides entering the lakes during droughts, in particular, could add to the existing difficulties faced by the wildlife in both study lakes (i.e., from habitat loss, low food availability, poisoning). The organochlorine concentrations detected in African Fish-Eagle in this study are highly unlikely to cause acute lethal poisoning, mass mortalities, and reduced reproductive success, as have been observed with Peregrine Falcons and Bald Eagles in parts of Europe and North America, respectively.

Depressed productivity and breeding success experienced by African Fish-Eagle on both Lakes, specifically Lake Naivasha, may more likely be due to food shortages, rather than direct organochlorine contamination. However, pesticides can potentially influence the abundance of food items, thereby creating food shortages. In both lakes, aquatic organisms appear to be the most likely to experience direct toxic impacts from endosulfan, methoxychlor, and DDT compounds in water, and lindane, aldrin, and heptachlor in sediments, as these compounds exceed guideline levels for these substrates. As a result, long-term harmful effects could impact lower trophic levels with consequences filtering up to the apex predators.

The largely exotic species-dominated lake, particularly by water hyacinth, could be masking the true organochlorine contamination in Lake Naivasha, through the release of organochlorine residues from water and sediment by evapo-transpiration. A process similar to the filtration mechanism by dense papyrus stands (Sue Peall, pers. comm. March 2011, Harper et al. 2011).The facilitation of organochlorine residue loss by water hyacinth and the remaining papyrus in Lake Naivasha is perhaps creating a false scenario, of Lake Baringo being subjected to a similar level of contamination, as the latter is devoid of an extensive 'natural'

filtering mechanism. Analysis of the air samples immediately around the surface of the lake and water hyacinth mats may indicate the contribution of the weed to removal of pesticide residues from the lake.

Owing to the substitution of organochlorines by organophosphates, more emphasis needs to be placed on modern and currently used pesticides for future studies. In Lake Baringo, there has been a reduction in African Fish-Eagle populations since the 1980s when Brown (1980), reported four times the number of individuals than in a 2011 census. The decline in African Fish-Eagle populations could be attributed to food shortages and deliberate poisoning. However, concentrations of other pesticides may be compounding the problems already being faced. Future studies and conservation actions should aim to: (i) determine local perceptions towards the lake and its wildlife, (ii) conduct a survey of pesticides available in "agro-vets" (agriculture and veterinary supply shops) and (iii) analyse the type of farming practices in the region that add most contaminants to the aquatic systems down-stream. Agro-vets are seldom registered businesses (Martin Odino, pers. comm. 2008) and occur extensively throughout rural Kenya supplying easily available and cheap chemicals to local farmers (pers. obs.). This makes it more difficult to regulate how pesticides are used.

Frequent, long-term, monitoring of pesticide contamination in natural systems needs to be implemented (Mineau 2005 b), instead of short, time-limited studies. Goals can be achieved through collaboration and transparency between agriculturalists and other interest groups, and are especially important in areas of economic and environmental importance to both national and international communities. Horticulture and floriculture businesses must work towards open information dissemination related to pesticide products, application methods, and frequencies and quantities used. This dissemination will facilitate the accurate identification of any potential hazards caused to the surrounding ecosystems related to pesticide use and help to mitigate the environmental impacts of chemical use. A potentially useful product of collaboration might be an up-to-date guide for environmentally safe farming practices, which may reduce contamination of air, water, soil or sediment, and would build on the existing Lake Naivasha Growers Group (LNGG) manual. Environmental-contamination monitoring in conjunction with papyrus fringe restoration, as proposed in Harper et al. (2011), could improve the ecological integrity of this RAMSAR site.

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APPENDICES:

APPENDIX A: METHODS: Section 2- Laboratory Analysis

All samples of water, sediment, fish and Fish Eagle blood were transported to the Nairobi University lab and analysed by Vincent Madadi using a GCMS procedure.

Extraction and analysis of water for pesticide residues

Water samples were extracted following solvent-solvent extraction method using dichloromethane. 1 L of water was transferred into 2 L separatory funnel followed by 50 ml phosphate buffer of pH 7. Drops of 0.1 N hydrochloric acid or sodium hydroxide were added to adjust the pH to 7 before adding 100 g of analytical grade sodium chloride to salt out the pesticides from the aqueous layer. The sample was vigorously shaken to dissolve the salt before adding the extracting solvent. Methylene chloride (60 ml) was added to the original glass bottle to rinse the interior parts of the bottle plus the cap and the mixture transferred to the sample in the separatory funnel. The sample was shaken for two minutes while venting in the fume chamber to release excess pressure, and allowed to settle for 10 minutes to allow the layers to separate. The lower (organic) layer was drained into 250 ml conical flask and covered with aluminium foil. The remaining aqueous layer was re-extracted twice with 60 ml portions of methylene chloride and the extracts combined. The combined extract was filtered through glass wool and anhydrous sodium sulphate into 500 ml round bottomed flask. 2 ml of isooctane was added to the sample as a keeper and concentrated to 1 ml using rotary evaporator and a water bath at 40°C. The extract was transferred into a 20 ml glass vial and the round bottomed flask rinsed 3 times with 1 ml portions of hexane. The sample was evaporated under a gentle stream of nitrogen to 1 ml before cleanup. The method was repeated for all water samples and recovery standards.

Extraction of sediments samples

Soil and sediment samples were extracted by Soxhlet method. 20 g of homogenized soil or sediment sample was treated with 60 grams of baked out anhydrous sodium sulphate and ground to powder in a mortar. The sample was allowed to stay overnight to dry further before extraction. 200 ml of 3:1 hexane-acetone mixture was transferred into 500 ml round bottomed flask and placed onto the heating mantle. Two boiling cubes were added to the flask to facilitate smooth boiling. The sample was placed into Soxhlet extractor and fitted onto the round bottomed flask containing the extracting solvent mixture. A Soxhlet condenser was fitted on top of the extractor and extraction initiated for 16 hours.

After extraction, 2 ml of isooctane was added to the extract as a keeper before evaporating to 1 ml using a rotary evaporator. The extract was transferred into 20 ml glass vial and the round bottomed flask rinsed 3 times with 1 ml portions of n-hexane. The rinsates (i.e., the solvent containing compounds of interest that is recovered after cleaning the sample by passing though a chromatography column) were combined with the sample in the glass tube before evaporating to 1 ml under a gentle stream of white spot nitrogen.

Extraction of fish samples

Homogenized fish sample (10 g) was treated with 30 grams of baked out (heated at 200 ^{o}C overnight) anhydrous sodium sulphate and ground to powder in a mortar. The sample was allowed to stay overnight to dry further before extraction. 200 ml of 3:1 hexane-acetone mixture was transferred into 250 ml round bottomed flask and placed onto a heating mantle. Two boiling cubes were added to the flask to facilitate smooth boiling. The sample was transferred into a Soxhlet thimble and placed into an extractor then fitted onto the round bottomed flask containing the extracting solvent mixture. A Soxhlet condenser was fitted on top of the extractor and extraction initiated for 16 hours.

After extraction, 2 ml of isooctane was added to the extract as a keeper before evaporating to 1 ml using a rotary evaporator. The extract was transferred into 20 ml glass vial and the round bottomed flask rinsed 3 times with 1 ml portions of n-hexane. The rinsates were combined with the sample in the glass tube before evaporating to 1 ml under a gentle stream of white spot nitrogen.

Extraction of fish Eagle blood and Serum samples

Homogenized blood samples (1-5 g) were treated with 30 grams of baked out anhydrous sodium sulphate and ground to powder in a mortar. The sample was allowed to stay overnight to dry further before extraction. 200 ml of 3:1 hexane-acetone mixture was transferred into 250 ml round bottomed flask and placed onto the heating mantle. Two boiling cubes were added to the flask to facilitate smooth boiling. The sample was transferred into a Soxhlet thimble and placed into an extractor then fitted onto the round bottomed flask containing the extracting solvent mixture. A Soxhlet condenser was fitted on top of the extractor and extraction initiated for 16 hours.

After extraction, 2 ml of isooctane was added to the extract as a keeper before evaporating to 1 ml using a rotary evaporator. The extract was transferred into 20 ml glass vial and the round bottomed flask rinsed 3 times with 1 ml portions of n-hexane. The rinsates were combined with the sample in the glass tube before evaporating to 1 ml under a gentle stream of white spot nitrogen.

Clean up of water, sediments, fish and Fish Eagle blood samples

The concentrated sample extracts were cleaned by eluting each one through a 15 g deactivated neutral alumina column and eluting with 165 ml of n-Hexane. A 25 cm glass column with sintered glass bottom was packed with 1 cm anhydrous sodium sulphate, followed by 15 g deactivated neutral alumina then topped with 1 cm anhydrous sodium sulphate. The column was conditioned with 15 ml n-hexane and discarded. The sample was gently transferred on top of the anhydrous sodium sulphate and allowed to elute through the stationary phase. The sample vial was rinsed three times with 1 ml portions of n-hexane and transferred into the column. The sample was then eluted further with 165 ml of n-hexane and collected in a 500 ml round bottomed flask. Isooctane (2 ml) was added to the cleaned sample and concentrated to 1 ml using rotary evaporator.

Preparation of anhydrous sodium sulphate for drying samples

Analytical grade anhydrous sodium sulphate (Na_2SO_4) was prepared by baking in an oven at 200°C for 16 hours to remove contaminants. The reagent was cooled and stored in desiccators ready for use.

Preparation of alumina for cleanup of samples

Analytical grade neutral aluminium oxide (Al_2O_3) for sample cleanup was prepared by baking in a Memmert oven at 200°C overnight and cooled. The reagent was deactivated by treating with 8% (w/w) distilled water and allowed to stay overnight before use.

Preparation of the Calibration Curve

The calibration curve was prepared from the stock solutions of PCBs and OCP standards. Nine level calibration curves were prepared from different dilutions of the stock solutions directly into auto-sampler vials. The concentrations were calculated by measuring the appropriate volumes and weighing the exact masses of all stock solutions and solvents added.

Analysis of organochlorine pesticide residues

Analysis of the samples was done using two gas chromatographic systems: Agilent 6890N loaded with Chem-station version 10.4. The performance of the equipments were tested using level 1 of the calibration standards and the responses compared for all compounds to the response of the same level injected in the multi-level calibration of the previous sequences.

The samples injection sequence followed first a solvent blank run (isooctane) followed by the most diluted standard from the calibration curve and the mid-calibration curve standard. Then the blank sample, the reference sample, the real samples and the other calibration standards injected at random. The mid-calibration curve standard was injected as the last sample at the end of the daily sample sequence to monitor the instrument performance. Samples with compounds in levels higher than the calibration window were re-injected after dilution.

Gas Chromatography conditions

Analyses of all samples were conducted in pulsed split-less mode at 4.50 bars. The detector and injector were set at 300° C and 250° C respectively.

For Agilent 6890N, analyses were conducted using BPX 5 capillary column of dimensions 60 m x 250 µm x 0.25 µm and a temperature program with initial temperature of 90 °C for 3 minutes, ramped to 200 °C at 30 °C/min with a hold time of 15 minutes, then ramped to 265 °C at 5 °C/min with a hold time of 5 minutes, then ramped to 275 °C at 3 °C/min with a hold time of 35 minutes. Helium (99.999%) and white spot nitrogen were used as both the carrier and makeup gas respectively. A carrier gas constant flow rate of 1 ml/min was maintained throughout all the analyses, whereas the makeup gas was maintained at a constant flow of 30 ml/min.

Confirmatory analyses were conducted using BPX50 capillary column of dimensions 60 m x 250 µm x 0.25 µm and a temperature program with initial temperature of 90°C for 3 minutes, ramped to 200°C at 30°C/min with a hold time of 15 minutes, then ramped to 265°C at 5°C/min with a hold time of 5 minutes and finally ramped to 275°C at 3°C/min with a hold time of 15 minutes. High purity helium gas (99.999%) was used as a carrier gas whereas white spot nitrogen was use as a makeup gas. A carrier gas constant flow rate of 1 ml/min was maintained throughout all the analyses, whereas the makeup gas was maintained at a constant flow of 30 ml/min.

Integration and Interpretation of Chromatograms

The system stability over the entire analysis was tested by doing an overlay of the middle point of the calibration curve and all the re-injections of this point during the sequence. All chromatograms with the same responses for all peaks within a margin of 5% confirmed the system stability. The system linearity based on r^2 was checked for each compound before consideration of the calibration curve.

Quality Assurance and Quality Control (QA&QC)

Analysis of all samples was done in triplicate to verify the presence of the analytes in the samples. Sample blanks, reference materials, and spiked samples were included in the sample extraction, cleanup and fractionation to verify the method performance.

The method detection limits were calculated by reviewing the noise in the chromatograms next to the peak of interest. The detection limit was set as three times the noise divided by the response of the compound in the lowest calibration point multiplied by the concentration of that point (in nano-grams injected). All compounds found with concentrations below the detection limit were reported as less than the detection limit (<dl) or not detected (ND).

APPENDIX B: Pesticides most commonly used in intensive horticulture/floriculture around Lake Naivasha (Gitahi et al. 2001).

Intensive horticulture/ Floriculture		
Trade Name	Common Name	Family
Lannate	Methomyl	Carbamoyloxime (I)
Apollo	Clofentezine	Tetrazine (A)
Brigade	Bifenthrin	Pyrethroid (I)
Mitac	Amitraz	Amidine (I,A)
Nemacur	Fenamiphos	Organophosphate (N)
Orthene	Acephate	Organophosphate (A,I)
Pentac	Dienochlor	Organochlorine (I)
Thiodan	Endosulfan	Organochlorine (I)
Cascade	Flufenoxuron	Benzoylurea (I)
Dursban	Chlorpyrifos	Organophosphate (A,I)
Folimat	Omethoate	Organophosphate (A,I)
Furadan	Carbofuran	Carbamate (I)
Hostathion	Triazophos	Organophosphate (A,I,N)
Mesurol	Methiocarb	Carbamate (A,I,M)
Omite	Propargite	Sulfite (A)
Tedion	Tetradifon	Bridged-diphenyl (A)
Temik	Alicarb	Carbamoyloxime (I)
Vydate	Oxamyl	Carbamoyloxime (I)
Kelthane	Pirimiphos-methyl	Organophosphate (I)

I- Insecticide; A-Acaricide; N-Nematicide; M-Molluscicide

APPENDIX C: Instrument Limit of Detection (LOD)

Pesticide Name	ng/ml
a-HCH	0.037
b-HCH	0.209
g-HCH	0.009
d-HCH	0.004
Heptachlor	0.04
Aldrin	0.022
Heptachlor epoxide	0.001
Endosulfan I	0.007
p,p-DDE	0.008
Dieldrin	0.001
Endrin	0.09
Endosulfan II	0.014
p,p'-DDD	0.068
Endrin aldehyde	0.012
p,p'-DDT	0.165
Endosulfan sulphate	0.01
Methoxychlor	0.271

APPENDIX D: Elution sequence

Pesticide Name	Retention time (minutes)
a-HCH	23.591
b-HCH	25.516
g-HCH	25.809
d-HCH	27.124
Heptachlor	27.784
Aldrin	34.122
Heptachlor epoxide	37.483
Endosulphan I	40.409
p,p-DDE	42.558
Dieldrin	42.615
Endrin	44.292
Endosulfan II	45.145
p,p'-DDD	46.285
Endrin aldehyde	46.752
p,p'-DDT	50.332
Endosulfan sulphate	51.532
Methoxychlor	52.316
PCB 198	57.823

Appendix E: Comparison of median OC concentrations measured in fish samples between Lake Naivasha and Lake Baringo, Kenya, 2010.

Pesticide Compound	Median Naivasha (n = 2)	Median Baringo (n = 3)	Test Statistic	P
α-HCH	5.76	1.17	6	1.000
β-HCH	13.71	19.61	6	1.000
δ-HCH	5.76	3.39	7	0.773
γ-HCH	4.02	8.07	5	0.773
Heptachlor	0.34	0.00	6	1.000
Heptachlor Epoxide	0.87	1.42	5	0.773
Aldrin	7.50	3.36	7	0.773
Dieldrin	0.37	0.01	6.5	1.000
Endrin	1.17	0.46	8	0.390
Endrin Aldehyde	9.40	6.30	7	0.773
Endosulfan I	1.82	1.41	7	0.773
Endosulfan II	0.42	1.38	4.5	0.564
Endosulfan Sulphate	0.49	4.74	3	0.149
DDT	1.33	1.25	6	1.000
DDD	3.42	0.90	7	0.773
DDE	1.23	2.09	5	0.773
Methoxychlor	2.96	8.76	3	0.149

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Mbatha,Hlengiwe Patience
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