MAKERERE UNIVERSITY

ASSESSING THE LEVELS OF PESTICIDE RESIDUES IN LIVESTOCK PRODUCTS AND WATER AROUND LAKE MBURO NATIONAL PARK, SOUTH WESTERN UGANDA

BY

ENID TURYAHIKAYO

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Declaration

I, ENID TURYAHIKAYO hereby declare to the best of my knowledge and belief, except for the literature cited, that this is my original work and has never been submitted to any University or Institute of higher learning for a higher degree award.

Enid Turyahikayo

Date:……………………

APPROVAL

I certify that this dissertation is presented with approval as a supervisor

…………………………

Dr. Ntale Muhammad

Date:…………………..

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DEDICATION

This study is dedicated to my daughters Kyla Ngonzi Tumusiime and Kriska Kengonzi.

LIST OF ACRONYMS

FAO	Food and Agricultural Organization
GC/MSD	Gas Chromatography, Mass Spectrophotometer Detector
GDP	Gross Domestic Product
MAAIF	Ministry of Agriculture, Animal Industry and Fisheries
MRL	Maximum Residue Level
NEMA	National Environment Management Authority
OCPs	Organochlorine Pesticides
OPPs	Organophosphate Pesticides
TBD	Tick-borne disease
WHO	World Health Organization

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ABSTRACT

An assessment on the levels of pesticide residues in livestock products and water around Lake Mburo National Park, South Western Uganda was carried out in the year 2010/2011. This study was necessary in view of the fact that there is inadequate data on the levels of pesticide residues in live stock products and water despite the increasing use of pesticides to control animal disease vectors specially ticks. A structured questionnaire was used to interview the farmers on the types of pesticides used to control animal disease vectors, and their practices, knowledge and attitudes on the use of pesticides. A total of sixty (60) farmers were interviewed and data analysed using Statistical Package for Social Sciences (SPPSS 19). Samples of milk, muscle and water were also collected and analyzed in a laboratory for organochlorine, organophosphate and pyrethroid residues.

From the study, 100% of the farmers controlled animal disease vectors by use of pesticides. The survey revealed a total of ten (10) different pesticides that were being used by the farmers in the study area with synthetic pyrethroids (33.6%) being widely used followed by organophosphates and formamidine (22.1%). There was no organochlorine pesticide being used by the farmers and no organochlorine pesticide residues were detected. However, chlorfenvinphos residues of 0.13 mg/l, 0.11 mg/l, were detected in water sample 1 and 2 from Kanyarweru subcounty while 0.17 mg/l, 0.12 mg/l, 0.41 mg/l, 0.12 mg/l of chlorfenvinphos residues were detected in water sample 1, 2, 3 and 4 from Nyakashara subcounty. In addition, chlorfenvinphos residues of 0.32 mg/l, were detected in milk sample 5 from Sanga Subcounty while 0.28 mg/l and 0.31 mg/l were detected in milk sample 2 and 5 from Nyakashashara subcounty. Also, unquantifiable levels of chlorpyrifos residues, an organophosphate were detected in meat samples 1, 2 and 4 from Nyakasharara Sub County. No pyrethroid residues were detected in the animal products and water samples analyzed.

The presence of pesticide residues in animal products and water could be attributed to the practices, attitudes and knowledge about the use of pesticides. The study revealed that

78% of the farmers disposed their pesticide containers around the spray area. It was also revealed that most of the spray areas were located in the radius of not more than 1km from the water source. This could be the probable cause of water contamination with pesticide residues as a result of runoff. In addition, 88% of the farmers interviewed had no knowledge on the withholding periods of the pesticides.

Despite the fact that some of the pesticide residues detected in the livestock products and water are below the Maximum Residue Levels(MRLs) with increasing pesticide use for tick control in Uganda, there is a possibility of increased pesticide residue levels in livestock products and water above the MRLs. In addition, given that pesticides are toxic compounds their presence in food, even in trace amounts, should be avoided.

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CHAPTER ONE

1.0 Introduction

1.1 Background

Livestock is a strategic export good, a key component of the modern economy and integral to indigenous range land and pastoral culture. In Uganda, livestock contributes 5% of the total Gross Domestic Product (GDP) and 13% of agricultural GDP respectively (UBOS, 2011). Livestock population in Uganda is estimated at 12.8 million cattle, 4 million goats and 3.8 million sheep (UBOS.,2011). Livestock production has continued to grow, at a rate of over 3% per annum, in response to increasing demand for milk and meat in the local market.

Around 60% of the households in the cattle corridor keep livestock, compared to 22% nationally. Uganda's "cattle corridor", stretches from south-west through the central region to the north- east. Livestock productivity in pastoral and agro-pastrol areas in Uganda is constrained by Ticks and tick-borne diseases (TTBDs) (Moran, 1996; Ocaido et al., 1996) and Tsetse flies and trypanosomosis (Magona et al., 1998). Of the diseases, caused by ticks and flies and TBDs, East Coast Fever (ECF, Theileriosis), Anaplasmosis, Babesiosis and Cowdriosis are the most important and wide spread (Otim, 2000).Tsetse flies infest about 41% of the entire landmass of Uganda and 70% of live stock graze under the threat of Trypanosomosis (Nyabahinduka, 1993).

Tick-borne diseases and Trypanosomosis continue to impede livestock productivity in most parts of Uganda due to existence of suitable climate for ticks and inadequate control of both ticks and tsetse flies (Otim, 2000). Farming practices in Uganda have not reached a level where primary health care, through extension services can contain major epizootics. As a result a wide variety of pesticides are used to improve the quality and quantity of crops and livestock. For example, past livestock protection against ticks were dominated by organochlorines (toxaphene and lindane) but these have now given way to organophosphates (Delnav and Supona) and synthetic pyrethroids deltamethrin (Decatix) and cypermethrin (Fendona)(NEMA., 2007a).The use of pesticides to control ectoparasites started with the application of arsenicals from the early years of the last century and the use of pesticides is still the main method for the control of these parasites (Youdeowei, 1983).

According to the FAO.,(2002), a Pesticide is any substance or mixture of substances intended for preventing, destroying, or controlling any pest, including vectors of human or animal disease, unwanted species of plants or animals causing harm during or otherwise interfering with the production, processing, storage, transport, or marketing of food, agricultural commodities, wood and wood products or animal feedstuff, or which may be administered to animals for the control of insects, arachnids or other pests in or on their bodies.

A Pesticide residue means any specified substance in food, agricultural commodities, or animal feed resulting from the use of a pesticide. The term includes any derivatives of a pesticide, such as conversion products, metabolites, reaction product, and impurities considered to be of toxicological significance.

Indiscriminate use of pesticides and their persistence in environment have led to widespread contamination of food sources such as food grains, vegetables, fruits and animal products. Animal derived products, however, are likely to carry a greater load of pesticides due to their higher lipid content (Srivastava et al., 2008). With the exception of occupationally exposed individuals, most exposure to these chemicals occurs via dietary intake (DeVoto et al., 1998; Ahlborg et al., 1995), especially food of animal origin, but also through water, ambient and indoor air, dust and soil (Covaci et al., 2002; Dua et al., 2001; Manirakiza et al, 2002).

Pesticides have been proved to have serious hazards to human health. Organochlorines in particular have high bioaccumulation potential and low degradation rates, have been associated with a number of environmental effects. Due to their persistence pesticides are distributed in air (Strand & Hov, 1996; Wania & Mackay, 1996., Wenzel et al, 1994) water bodies and soil finally making way into the plant and animal biomass. Most pesticides, especially, the organochlorines are very resistant to microbial degradation. They can, therefore, accumulate in human body fats and the environment posing problems to human health (Ejobi et al., 1996).

Pesticide residues in food items have been a concern to environmental and consumer groups of their wide spread use. The most common effects of the wide spread of organochlorine compounds in the environment are birth defects, neurological effects and behavioral effects, reproductive effects and cancer (Windham, 2002). The World Health Organization (WHO) has estimated that each year, there are some 3,000,000 cases of acute pesticide poisonings, with 220,000 deaths. The majority of these incidents occur in developing countries, particularly in Africa, Asia and Central and South America (He, 2000).

Further, the intense use of pesticides in agriculture or disease vector management can lead to the contamination of surface and ground water. Water runoff resulting from heavy rainfall can transport pesticides and their toxic metabolites to distant places located downstream, contaminating lakes, lagoons, reservoirs, ponds, and estuaries, and adversely affecting aquatic organisms. Discarding pesticides, washing spray equipment, or rinsing empty pesticide containers in or near streams and rivers can cause similar damage.

However, livestock development choices and management practices also have impacts on the environment and health. There are concerns about the possible human health impacts of exposures to chemicals and pharmaceuticals used to protect livestock from disease; these may enter food, soil and water supplies, the latter shared by animals and humans. (WHO/UNEP., 2008). In Africa, smallholder farmers and many agriculture officials do not know how to calibrate or use sprayers properly, most farmers do not use safety equipment, recommendations given during safe use pesticide training are not followed, and well-written national regulations are never enforced. Many livestock producers treat their animals themselves. Even if they use the same pesticides as veterinarians, they have little understanding of the conditions and quantities to administer or the waiting periods.

The uncontrolled use of the pesticides can lead to residues in animal products, especially when users fail to respect waiting periods.

Since administration of any chemically active pharmaceutical to food producing animals inevitably leads to presence of residues in foods from such animals, national and international legislation seeks to ensure that consumers of animal food products are not exposed to potentially harmful concentrations. Expert bodies like Codex Alimentarius, Joint FAO/WHO Expert Committee on Food Additives, and the European Union have set a series of maximum residue limits (MRL) on edible tissues. The Maximum Residue Level (MRL) is the maximum amount of the pesticide residue which if found in food substances will not cause any health effect or hazard (Cabtas et al., 1992). The purpose of the MRL is to limit the exposure of consumers to residues of medicines used in food animals, to concentrations that do not pose human health risk.

Surveillance or monitoring on the occurrence of residues in meat and their products was relatively a neglected area until last decade (Horrigan et al., 2002). But with the advancement of technological intervention regarding livestock rearing, disease control and intensive crop production system, the chances of residues in foods of animal origin increased tremendously. This results into a potential risk of various life threatening diseases such as cancer, leukemia, reproductive disorder besides disruption of body's immune, endocrine and nervous system (Horrigan et al., 2002).

Although pesticides play an important role in controlling vectors, the problem of pesticide residues in foods of animal origin has become a major concern due to their effects on human health.

1.2 Problem statement

Although pesticides have been used for the control of animal disease vectors and have been reported to pose several effects on human health, there is inadequate data on the levels of pesticide residues in live stock products and water and their effects on human health in Uganda.

1.3 Objectives

1.3.1 General objective

To assess the levels of pesticide residues in livestock products and water around Lake Mburo National Park, South Western Uganda.

1. 3.1.1 Specific objectives

i. To identify and document the major types of pesticides administered to cattle within the communities around Lake Mburo National Park.

ii. To determine the levels of organophosphate, organochlorine and pyrethroid pesticide residues in livestock products (milk and muscle) and water around Lake Mburo National Park, South Western Uganda.

iii. To determine whether the farmer's perceptions, knowledge and practices contribute to the presence/levels of pesticide residues in livestock products and water around L. Mburo National Park.

1.4 Hypotheses

$H_1$1: The levels of pesticide residues in livestock products (milk, muscle and liver) and water within the communities around Lake Mburo National Park exceed the allowable Maximum Residue Levels.

$H_1$2: The farmer's attitudes, knowledge and practices contribute to the presence of pesticide residues in livestock products and water around L. Mburo National Park.

$H_1$3: The farmer's attitudes, knowledge and practices contribute to the levels of pesticide residues in livestock products and water around L. Mburo National Park.

1. 5 Significance of the study

The data generated will be used by Government Departments involved in monitoring programmes on pesticide residues in livestock products.

Government Agencies, Non Governmental Organizations (NGOs), Civil Society Organizations (CSO) among others can use the data generated to raise awareness among consumers of livestock products about pesticide residues in livestock products.

It is also hoped that data and information generated can contribute to policy dialogue and inform policy change regarding pesticide residues, public health and trade issues in Uganda.

CHAPTER TWO

2.0 Literature review

2.1 Introduction

Uganda's economy relies heavily on the agricultural sector, which accounts for about 43% of the Gross Domestic Product (GDP), and over 80% of the total population derives its livelihood from this sector (NEMA., 2002).

In Uganda, livestock contributes 5% of the total GDP and 13% of agricultural GDP respectively (UBOS., 2011). Livestock population in Uganda is estimated at 12.8 million cattle, 4 million goats and 3.8 million sheep (UBOS., 2011). The table below shows the trends of livestock keeping in Uganda.

Table 1: Trends of livestock keeping in Uganda

	2008	2009	2010	2011	2012	YOY growth
cattle	11,409	11,751	12,104	12,467	12,841	3%
sheep	3,410	3,513	3,621	3,730	3,842	3%
goats	2,450	2,823	3,208	3,604	4,012	3%
pigs	3,184	3,280	3,378	3,479	3,584	3%
poultry	37,437	38,557	39,714	40,905	42,133	3%

*Source: Adopted from UBOS 2011 Statistical abstract. * 2008 are census statistics ; 2009-2012 are based on a 3% growth rate assumed by UBOS.*

The Uganda government's policy on increasing the output of animal products and improving productivity is seriously hampered by several constraints, especially ticks and the tick-borne disease (TBD) complex. The major tick-borne diseases in Uganda are Anaplasmosis, Babesiosis, Cowdriosis and East Coast fever (ECF). Drug treatments of these diseases are expensive and most cases the available drugs are not highly specific. In view of this, the control of these diseases has depended on the use of pesticides and other environmental management strategies (Nyeko & Obuwoya., 1993).

In Uganda, as in other African countries, the principal method employed to control tick-borne diseases is intensive dipping or spraying of cattle with acaricides to free them of tick vectors.

2.2 The use of pesticides in livestock production in Uganda

The use of pesticides to control ectoparasites started with the application of arsenicals from the early years of the last century and the use of pesticides is still the main method for the control of these parasites (Youdeowei, 1983).

In Uganda, the use of pesticides especially in ensuring the sustainability of large quantities of high quality agricultural produce has been steadily increasing over the past half century (Kasozi et al., 2006). Chemical tick control was started in Uganda in the early 1930s and gained momentum after the Second World War (Okello-Onen et al., 1992). After World War II, tick control became very effective. The introduction of exotic cattle in the 1960s led to the construction and use of several dips. The government encouraged farmers to practice tick control by providing a 50% subsidy on costs of construction of dips and purchase of spray pumps and acaricides. However, this subsidy was removed in 1984.

So far, Uganda has gone through a succession of groups of chemicals used to control ticks. In the past, protection against ticks was dominated by organochlorines (Toxaphene and Lindane) but as in other cases, these have largely given way to organophosphates (Delnav and Supona) and synthetic pyrethroids deltermethrin (Decatix) and cypermethrin (Fendona), (NEMA., 2007a). The only proven instances of tick resistance against toxaphene were in *Boophilus decoloratus* at Njeru in East Mengo (now Mukono District) and Nkumba Dairy Farm in West Mengo (now Mpigi District); in *Rhipicephalus evertsi* at Nkumba Dairy Farm and Kigungu at Old Entebbe in West Mengo (Mpigi District) and

Namulonge in East Mengo (Wakiso District); and in *R. appendiculatus* in Mbarara District (Kitaka et al., 1970).

2.3 Categories of pesticides

Pesticides are often referred to according to the type of pest they control. They can also be classified according to their nature e.g chemical pesticides etc.

2.3.1 Organochlorine pesticides (OCPs)

Organochlorine pesticides are insecticides composed primarily of carbon, hydrogen, and chlorine. They break down slowly and can remain in the environment long after application and in organisms long after exposure. OCPs are a class of non polar toxic chemical compounds classified as dichlorodiphenylethane cyclodienes and chlorinated benzenes (Ademoroti, 1996). They may be grouped into three general classes; the dichlorodiphenylethanes (DDT, DDD, dicofol, etc.), the chlorinated cyclodienes (aldrin, dieldrin, heptachlor, etc.) and the hexachlorocyclohexanes (lindane). These compounds differ substantially between and within groups with respect to toxic doses, skin absorption, fat storage, metabolism, and elimination. The signs and symptoms of toxicity in humans, however, are remarkably similar except for DDT (Hayes, 1991). These lipophilic compounds accumulate and even magnify their concentration along the food chain, especially in fatty food (Manirakiza et al., 2002).

Organochlorine Pesticides are widely used by farmers because of their effectiveness and their broad-spectrum activity (Darko & Acquaah, 2007). They are liposoluble compounds and are easily absorbed in lipids of the insect cuticles. In vertebrate animals, variable amounts of chlorinated insecticides are retained in the adispose tissues, from where they are gradually released in circulatory system (Nyeko &Obwoya, 1993). OCPs were commonly used in the past, but many have been removed from the market due to their health and environment effects and their persistence. In Uganda, the use of most OCPs has been banned or restricted under the Rotterdam and Stockholm Convention due to high levels of persistence in the environment and toxicity to non target organisms.

2.3.2 Organophosphate pesticides (OPPs)

Organophosphorus pesticides are widely used in agriculture to protect plants from insects, and hence provide numerous benefits in terms of production and quality. This group of compounds are highly liposoluble but are also soluble in water. Due to their instabilities, the residue levels of organophosphorus pesticides in foods are affected by a number of physical factors applied in food processing, including fermentation, heat treatment and drying. In addition, the chemical nature of organophosphorus pesticides and some environmental factors such as, pH, light, metal ions and ozone, also have impacts on the degradation of pesticide residues (Bogialli et al., 2006). Organophosphate insecticides include chlorpyrifos, diazinon, dimethoate, disulfoton, malathion, methyl parathion, and ethyl parathion among others. The organophosphates are generally categorized as the most toxic of all pesticides to vertebrates (Ware, 2000).

Exposure to organophosphates occurs via inhalation, absorption into the skin, and ingestion (Ecobichon, 1996;Sullivan et al., 1992). Organophosphate insecticides share a common mechanism of toxicity, through inhibitory effects on cholinesterase enzymes in the nervous system .This result in elevated levels of acetylcholine (ACh), which acts on the muscarinic receptors situated at cholinergic junctions in skeletal nerve-muscular junctions, at nicotinic receptors in autonomic ganglia, and receptors in the central nervous system (CNS).

An example of an organophosphate is Chlorpyrifos. These substances poison the nervous system by inhibition of cholinesterase enzyme. Health effects of chlorpyrifos are headache, nausea, dizziness, salivation, excess sweating, blurred vision, chest tightness, muscle weakness, abdominal cramps and diarrhea (WHO., 2002). Chlorpyrifos is moderately toxic following acute oral, dermal and inhalation exposures. Chlorpyrifos affects the nervous system by reversibly inhibiting the activity of cholinesterase (ChE), an enzyme necessary for the proper functioning of the nervous system (Smegal, 2000).

2.3.3 Pyrethroid pesticides

Pyrethroid pesticides were developed as a synthetic version of the naturally occurring pesticide pyrethrin, which is found in chrysanthemums. The US Environmental Protection Agency has classified cypermethrin as a possible human carcinogen though available information on its carcinogenic properties is inconclusive (EXTOXNET., 1996).

This class of insecticides or acaricides includes permethrin, resmethrin and allethrin. Often the formulations contain a synergist (something that enhances the effectiveness of the active ingredient) called piperonyl butoxide, or PBO which itself, is relatively non toxic. The synthetic pyrethroids show properties of low mammalian toxicity but good activity against insects, ticks and mites. They do not appear to be readily absorbed through the skin.

2.3.4 Carbamate pesticides

Carbamates are similar to organophosphates in activity in that they inhibit cholinesterase. Examples of carbamates include Carbaryl, Propoxur, Methomyl among others. These insecticides can cause cholinesterase inhibition poisoning by reversibly inactivating the nzyme acetyl cholinesterase and cause a cholinergic (Hayes& Laws., 1991).

2.4 Impacts of pesticides on human health

Pesticides applied to livestock most frequently include those used to control disease vectors, such as ticks and tsetse flies. Chemical use may therefore have positive impacts on health e.g improving animal productivity and curbing disease transmission to humans. At the same time, chemicals applied to livestock or infiltrating livestock products from the broader environment can impact negatively on health e.g through residues or tainting of food products, pollution of drinking water sources and bioaccumulation in the food chain (WHO/UNEP., 2008).

2.4.1 Acute effects

Injuries may be caused either by a single massive dose being absorbed during one pesticide exposure, or from smaller doses absorbed during repeated exposures over an extended period of time. Illness or damage is referred to as acute when it has a sudden onset and lasts for a short period of time. The type and severity of the symptoms depend on the chemical mode of action and toxicity and the amount of chemical the victim has been exposed to. WHO estimates of 2000 showed that each year three million farmers in the developing world were experiencing severe poisoning from pesticides each year, about 18,000 of whom would die (WHO., 2000). Fifty percent (50%) of the modern pesticides are mutagens, i.e cause heritable changes in the genetic material, DNA. This is of concern since it poses a threat to the gene pool of Uganda's biodiversity, which is quite extensive (NEMA., 2007a).

2.4.2 Chronic effects

Chronic toxicity refers to the effects that occur after exposure over a long period of time, or to symptoms that occur long after exposure and/or persist for a long time. In general, these effects can occur with doses as low as a few micrograms of pesticide per kilogram body weight of the person or animal exposed. Examples of the chronic effects of pesticides on humans are described below.

2.4.3 Carcinogenic effect

Pesticides can exert a carcinogenic effect through a variety of mechanisms, including:

• *Genotoxicity:* Pesticides react with DNA to cause mutations or cancer.

• *Hormonal action:* Pesticide lengthens the oestrous cycle, prolonging exposure to endogenous oestrogen, and can cause mammary and uterine tumours.

• *Immunotoxicity:* Pesticides can alter immune function in a number of ways that can cause cancer.

2.4.4 Neurological effects

According to Poston et al., 2003, there is a growing concern in regard to the developmental neurotoxicity by recent epidemiologic observations that children exposed prenatally or during early postnatal life suffer from various neurological effects. Examples of neurological effects are numbness or weakness of arms, legs, feet or hands; lethargy; memory loss; loss of concentration; and anxiety.

2.5 Pesticide usage and exposure

Human exposure to agricultural pesticides and the subsequent contamination or poisoning may be occupational, non-occupational, intentional or unintentional. Also, exposure may be through ingestion (oral), through the skin (dermal) or through inhalation (respiratory). Occupational contamination or poisoning has been identified as the most serious problem associated with the use of agricultural pesticides, especially in developing countries (Olurominiyi & Monosson, 2007). There is a widespread use of pesticides in the Uganda but these pesticides are handled by persons who are underserved; i.e. not adequately trained, supervised, informed and guided in the proper procedures for pesticide use and handling. Consequently, the pesticides have entered the environment in undesirable levels and caused un quantified deleterious effects (NEMA., 2007b).

Pesticides are the most easily accessible toxic chemicals and most widely stocked in peasant farmer's houses in rural areas and the core problem is poor handling. Handling includes transportation, storage, application, and disposal of pesticides. The victims of pesticide poisoning are not only those who apply the pesticides. Other people working with pesticides, such as storekeepers and farm workers, may also be exposed. Even those who do not work with pesticides may be exposed to them unknowingly, for example, by being sprayed accidentally or eating contaminated food. Children are frequent victims of pesticide poisoning when they eat or drink pesticides that are stored in their reach.

Pesticides may enter the body orally (through the mouth), they may be inhaled as vapours or they may enter through the skin. Oral ingestion may occur by accidentally drinking a pesticide, by splashing spray materials or pesticide dust into the mouth or by eating or drinking contaminated foods or beverages. The ability of a pesticide to be absorbed through the skin depends on the chemical characteristics of the pesticide and its formulation. Respiratory exposure occurs when dust or vapours enter the lungs, or when aerosols are formed when pesticides are sprayed. Pesticides that are more soluble in oil or petroleum solvents penetrate skin more easily than those that are more soluble in water.

2.6 Pesticide residues

When a pesticide product is applied on the field, the chemical is gradually lost as a result of breakdown, leaching and evaporation and the residue is the amount that remains after application (Cox, 1995). While some pesticides have long residual activity and therefore persist in the environment, others have short residual activity and therefore do disappear from the environment or produce low residue concentration. It is therefore not surprising to find or detect residues of pesticides in the environment and food crops after usage. Pesticide residues may enter the food chain causing serious hazards to human and animal lives (Khan et al., 2007). The principal sources of pesticide residues in crops, food animals, soil, water include the following among others;

- Carry-over from insecticide application to soil or to growing crops,
- Leaching of pesticides (herbicides) or insecticides into ground water,
- Drift of the pesticides from adjacent field,
- Translocation of soil applied pesticide into growing crops,
- Disposal of pesticides in streams, rivers lakes occur,
- Effluents of pesticide industry in rivers and streams, and into soil which may be translocated in crops.

CHAPTER THREE

3.0 Materials and Methods

3.1 The study area

3.1.1 Geographical location

This study was carried out in Kiruhura District, South Western Uganda, specifically in the sub-counties of Kanyaryeru, Nyakashashara and Sanga in Nyabushozi County. Nyabushozi county lies in the latitude of -0.25° and longitude of 30.83°. The district borders with other districts of Ibanda and Kamwenge in the North West, Mbarara District in the West, Isingiro District in the South, Rakai District in the South-East, Lyantonde District in the East, Kyenjonjo and Sembabule Districts in the North and North East respectively. It covers an area of 4103.5 Sq Km with an estimated population of 252,648 people by the end of 2007 (MGLD, 2007). It is part of the cattle corridor that extends from the south western, through central to the north eastern region. The map in Figure 2 shows the cattle corridor of Uganda.

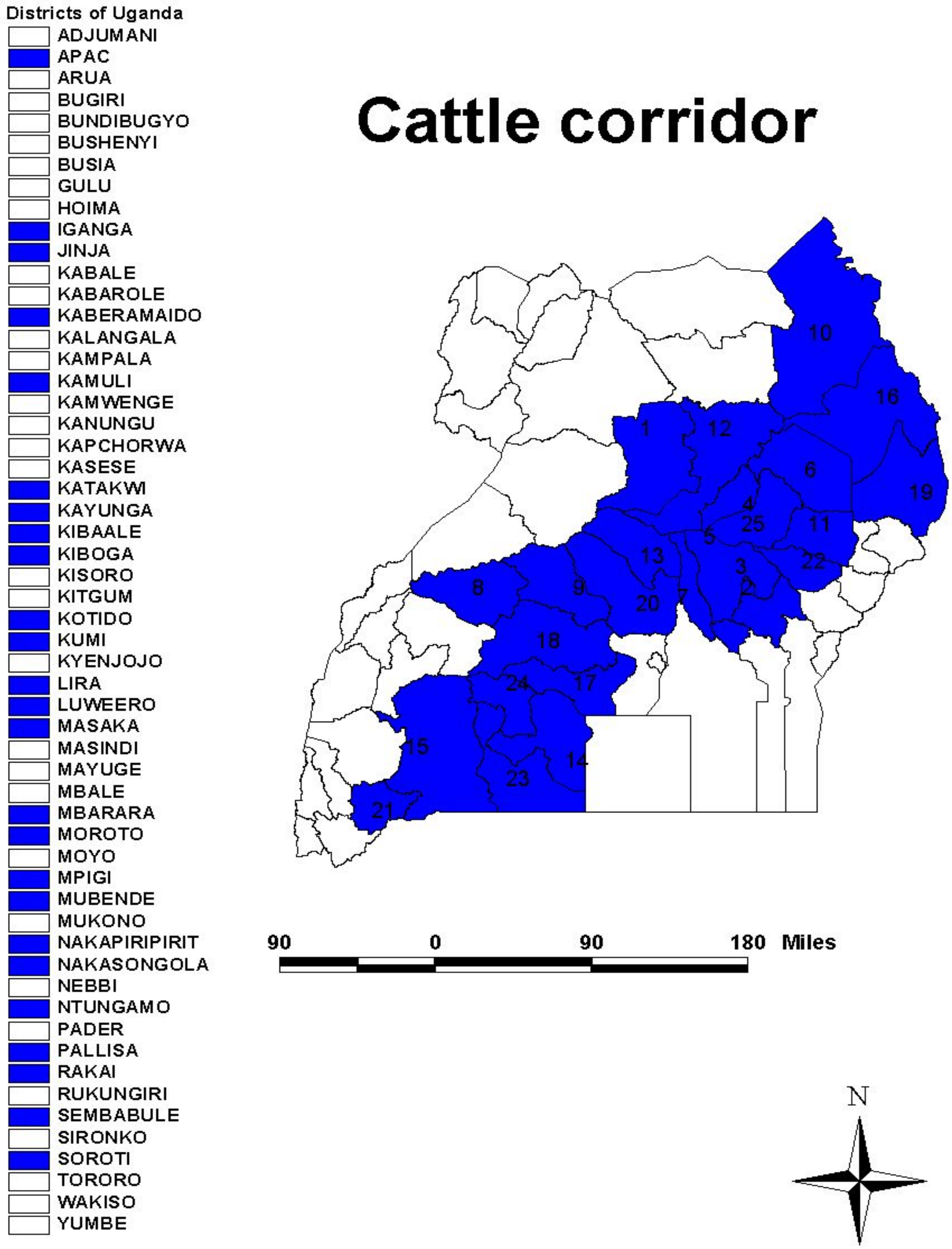

Figure 1: Map showing the cattle corridor of Uganda

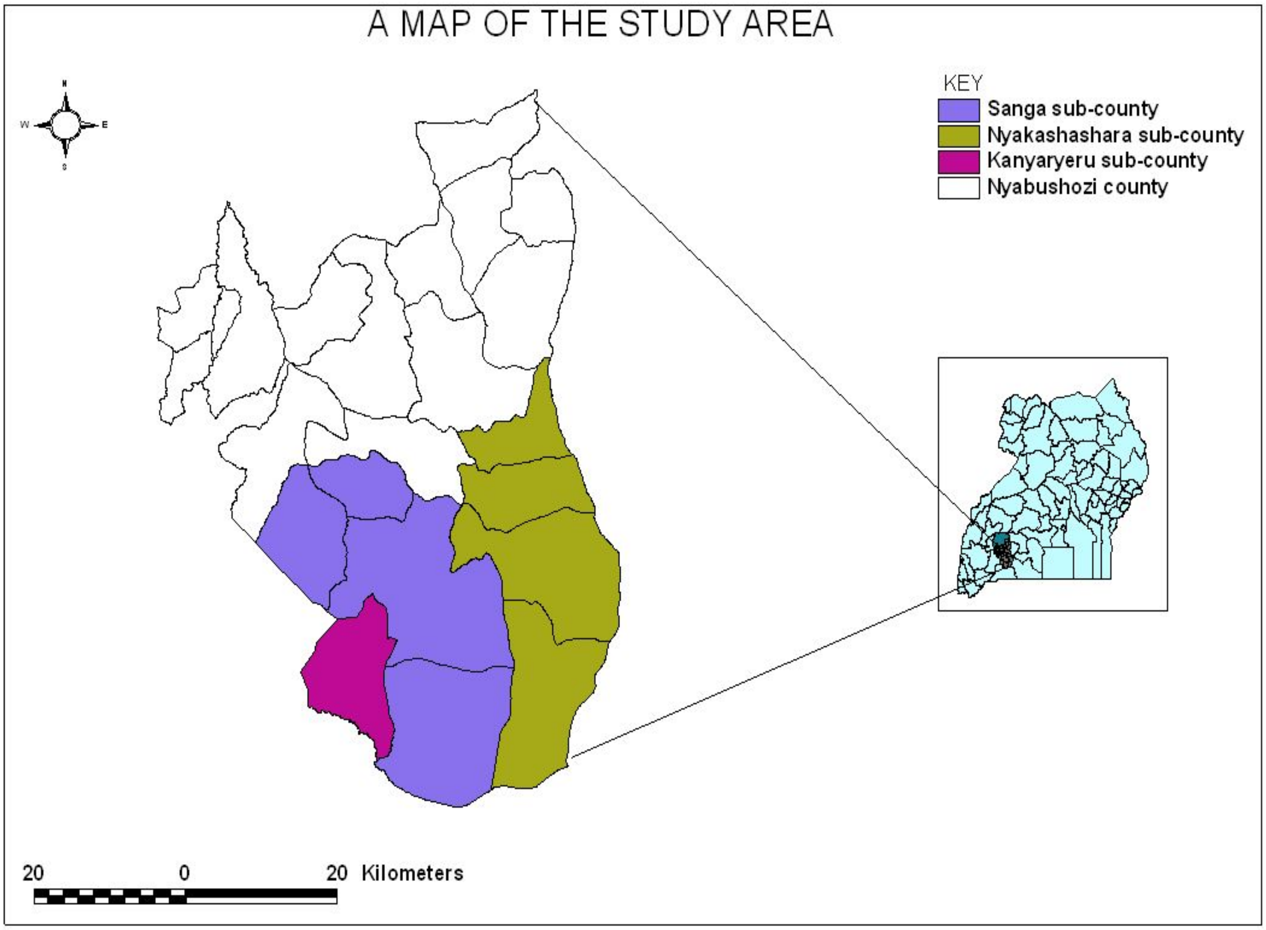

Figure 2: Map showing the location of the study area

3.1.2 Topography and climate

Kiruhura District lies at an altitude ranging between 129 m and 1524 m above sea level with an average rainfall of 882mm per annum. The rainfall pattern is bimodal peaking in the month of April - May (long rain) and September –November (short rains) separated by a long dry spell (June - August) each year. However, due to climatic variation, the rainfall pattern in the area has become unpredictable.

3.1.3 Vegetation

The dominant vegetation type for this study area is described as being dry *Acacia* savanna comprised of an *Acacia / Cymbopogon / Themeda* complex (Langdale - Brown 1964). The dominant vegetation is superimposed on a mixture of grass layer characterized by *Brachiaria decumbens, B. platynota, Themeda triandra,* among others. . There are no reported recent studies undertaken to determine if any significant changes have taken place in species distribution and abundance. With changes in fire regimes (lower frequency) and increased grazing pressure, the likelihood of change in species distribution/composition and abundance would be high.

3.1.4 Land use

Pastoralism has been the main land use activity in this region. However, communal grazing or common property rangeland tenure system is being phased out in favour of individualization (privatization) of land ownership (Kisamba-Mugerwa, 1992) as observed in many other pastoral areas of East Africa. As a result, pastrolism as a land use system is slowly being replaced by agro-pastrolism.

Today most pastoralists have become agro-pastoralists due to the changing social economic conditions in addition to deliberate government policies promoting diversification and increase in income generation. In this region, the cropping aspect is still poorly developed.

3.2 Site selection

Kiruhura District lies in the cattle corridor of Uganda and bordered by Lake Mburo National Park. As a result, there is high infestation of ecto- parasites especially ticks due to interaction of game park animals and livestock. Therefore, there is heavy reliance on pesticides to control ticks and tick borne diseases.

3.4 Sampling method

3.4.1 Objective 1

To identify and document the major types of pesticides administered to cattle within the communities around Lake Mburo National Park.

Data was collected using a detailed structured questionnaire comprising of open and close ended questions. The questionnaires were administered using direct household interviews with selected pastoralists in the month of October 2010.The interviews were conducted in the local language (Runyakore). Heads of the randomly selected households were interviewed. The households were selected according to proximity and approximately 60 households were interviewed.

The questionnaire (appendix 2) focused on;, identification of the commonly used pesticides, the assessment of knowledge, perceptions and practices on the use of pesticides among others.

Figure 3: Pastoralists involved in a household interview

3.4.2 Objective 2

To determine the levels of organophosphate and pyrethroid pesticide residues in livestock products (muscle and milk) and water around Lake Mburo National Park in Uganda.

Five 1kg samples of cow muscle were randomly collected from butcher shops from the three sub counties and frozen until the time of analysis.

Five 2L milk samples were collected at the time of milking from randomly sampled households in the selected villages from each Sub County making a total of 15 milk samples. Sample bottles were rinsed three times before collection of the samples. The samples were frozen until the time of analysis.

Ten 2L water samples were also collected in clean plastic containers from water sources (ponds/dams) within the selected villages. Sample bottles were rinsed three times before collection of the water samples. The samples were refrigerated at -24^0C until the time of analysis. All samples were frozen at -24^0C on the same day of collection. They were then kept in a cool box containing ice packs and then transported to Chemiphar laboratory where chemical analysis was done.

3.4.3 Objective 3

To determine whether the farmer's perceptions, knowledge and practices contribute to the presence/levels of pesticide residues in livestock products and water around L. Mburo National Park. For this objective, a similar procedure as for objective 1 was used.

It is important to note that the sample size (farmers interviewed and samples collected) during the study was limited by funding.

3.5 Analytical laboratory analysis of samples

3.5.1 Method of analysis

Samples of cow muscle, milk and water were analyzed at Chemiphar Laboratory using a Gas Chromatographic system equipped with a Mass Spectrophotometric Detector (GC/MSD) to determine the levels of pesticide residues. The samples were analyzed according to the (A.O.A.C., 1995).This method was applied to determine organochlorine, organophosphate and pyrethroids residues in the muscle, milk and water samples.

3.5.2 Reagents

The following substances/reagents were used; Nitrogen gas, Helium, alphagaz 2, air liguide, petroleum ether, acetonitrite, anhydrous sodium sulphate, hexane, de-mineralized water, acetone, decachlorobiphenyl, 2,4,5,6-tetrachloroxylenechlorofenvinphos standard, organochlorine standards which include; α-HCH, β –HCH, γ –HCH, Heptachlor epoxide, Aldrin, Oxychlordane, endosulfan 1, endosulfan II, endosulfan sulphate, chlorodane, dieldrin, endrin, DDE and DDT. Others include; Organophosphorous pesticide standards: Dichlorvos, Methacrifos, Diazinon, Etrimfos, Phosphamidon, Chloropyrifos methyl, Fenitrothion, Malathion, Chlorpyrifos, Parathionnethyl, Piriphosethyl, chlorfenvinphos. Pyrethroid standards included; Lambda-cyhalothrin, Permethrin cis, Cyperthrin trans, cypermethrin, Tau-Fluvalinate, Deltamethrin

3.5.3 Analytical procedure for muscle samples

3.5.3.1 Sample homogenization

Samples of muscle were divided into smaller portions by cutting them into small pieces and blended into much smaller pieces. Two samples weighing 10 g each were weighed into a stomacher bag and each labeled with an identification number for reference i.e. sample and spike.

3.5.3.2 Extraction

To each 10 g of the muscle sample, 10 g of anhydrous sodium sulphate, 500 ng/mL of a surrogate standard (decachlorobiphenly diluted to a concentration of 10µg/mL in ethyl acetate) was added into each stomacher bag. 100 µL of the working standard 2,4,5,6-

tetrachloroxylene(1µg/ml) was added into the spike. To each stomacher bag, 50 mL of petroleum ether was added and the samples were ground using a stomacher for thirty (30) seconds.

The solution of each sample was poured off into separate 250 mL volumetric flasks through a filter paper. The extraction was repeated a 3^{rd}, 4^{th}, 5^{th} and 6^{th} time with 30 ml of petroleum ether until a 100 mL of the sample and spike was extracted.

3.5.3.3 Sample clean up and concentration

The extracts (50 mL) were put on a rota vapor for approximately five (5) minutes and concentrated to approximately 10 mL. The extracts were further concentrated to 1 mL using a sample concentrator. 1 mL of acetonitrile was then added to the 1 mL extract to remove most of the lipids from the extracts. The extract was further concentrated to 1 mL. 100 µL of an internal standard (diphenly-D10, diluted to a concentration of 10µg/mL in ethyl acetate) was added and rotated vigorously for proper mixing. The extract was then transferred to the freezer at -24^{0}C for one (1) hour. The clear acetonitrile was transferred to a vial and analyzed on a Gas Chromatography-Mass spectrometer detector (GC-MSD)

3.5.3.4 Calculations

$$Cp = \frac{Cpgcms * Vte}{Vex * M * 1000}$$

Cp = Pesticide concentration

Cpgcms = Concentration of the pesticide on GC-MS

Vte = Total extraction volume

Vex = Extraction volume concentrated

Me = Mass extracted

3.5.4 Analytical procedure for water samples

3.5.4.1 Homogenization and extraction

The sample was shaken well and an aliquot of 500 ml was taken for extraction. 500 ml of the water sample was transferred into a separating funnel. To the separating funnel, 16 µl of 2,4,5,6-tetra-chloroxylene (surrogate standard) was added and shaken well to mix. 1 mL of saturated tartaric acid solution was also added and homogenized well by shaking for 30 seconds. To the separating funnel, 20 ml of petroleum ether was added and shaken carefully for 7 minutes and let to settle down for 5 minutes. The water phase was drained off by opening the tap and kept for the second extraction. The extract was collected in a 100 mL volumetric flask. The extraction was repeated the 2^{nd} and 3^{rd} time using 100 mL and 70 mL of petroleum ether respectively until the extract made 100 mL.

3.5.4.2 Concentration

The extract (50 mL) was transferred into a vial and concentrated to 3 mL by evaporation at 40^{0}C in a warm water bath. The 3mL filtrate was transferred into a 4mL vial and concentrated further to dryness using a water bath at 40^{o}C. To the dried concentrate, 1000 µl of hexane was added and mixed well. The concentrate (2 ml) was transferred into a GC Vial for GC-MS analysis results (see appendix) of which were computed using the calculation in 3.5.4.3 below. .

3.5.4.3 Calculations

$$Cp = \frac{Cpgcms * Vte}{Vex * M * 1000}$$

Cp = Pesticide concentration

Cpgcms = Concentration of the pesticide on GC-MS

Vte = Total extraction volume

Vex = Extraction volume concentrated

M = Volume extracted

3.5.5 Analytical procedure for milk samples

A milk sample (500 mL) was transferred into a separating funnel. Petroleum ether (50 ml) was also added and shaken well for mixing. An excess of petroleum ether (200 mL) was added and Sodium Chloride (NaCl) was also added to enhance separation. The sample was rotated and allowed to settle for overnight. The lower layer was run off. In the top layer of the sample, Petroleum ether (200 mL) was added and NaCl(2 g) was also added to enhance separation. The lower layer was run off. This process was repeated three times and the top layer was collected in a 250 mL volumetric flask. The petroleum ether was concentrated to 1 mL. Acetonitrile(10 mL) was added and further concentrated to 5 ml. Petroleum ether (3 mL) was added and the mixture was mixed well and put in the freezer for two hours. The petroleum ether was sacked off to remain with acetonitrile to remove the fats. The extract was further concentrated to 1 mL. Decachlorobiphenly (100 µl) an internal standard was added and rotated vigorously for proper mixing. The concentrate (2 mL) was transferred into a GC Vial for GC-MSD analysis.

3.5.6 Gas chromatographic analysis

An Agilent Technologies GC/MSD model 6890N series was employed in the analysis of samples for organochlorines, organophosphates and pyrethroids residues. Chromatographic separation was achieved on a 30 m x 0.25 mm HP-5ms capillary column with 0.25 µm film thickness. The GC temperature limits ranged from -60^0c to 325^0C while the oven temperature for analysis of organochlorine and organophosphate residues was set at 70^0C while the oven temperature for the analysis of pyrethroids was set at 100^0C. Helium gas was used as a carrier gas at a flow rate of 120 mL/min. The mass spectrometer was used in electron ionization mode (EI) and in Selected Ion Monitoring mode (SIM). Both data acquisition and processing was accomplished by software Chemstation (Agilent Technologies™). The pesticide residue chromatograms for the samples analysed are attached as appendix 5.

3.6 Data Analysis

Data analysis was performed using the Statistical Package for the Social Scientist (SPSS 16.2).

3.6.1 Objective (i)

The identified pesticides administered to cattle within the communities around Lake Mburo National Park were analyzed, and were presented in text, tabular and graphical formats.

3.6.2 Objective (ii)

Results from the laboratory regarding the concentration of organophosphate, organochlorine and pyrethroid residues in milk and water were presented in tabular and graphical formats.

3.6.3 Objective (iii)

The farmers' Perceptions, knowledge and practices towards the presence of pesticide residuals in livestock products and water were tabulated, frequency and percentages were presented in tabular and text formats.

CHAPTER FOUR

4.0 Results and discussions

4.1 Common pesticides administered to cattle within the communities around Lake Mburo National Park.

From the survey, there are ten (10) commonly used pesticides used in the surveyed area. It was reported that 34%, 22%, 22%, 7%, 5% 3%, 3%, 3%,3%, 1% used Baytical, Milibraz, Protaid, Tactic, Supoona, Alphapol, Cooperthion, Decatix, Renegade and Norotraz are used respectively. Out of the pesticides identified, 50% were Synthetic pyrethroids, 30% formamidine and 20% organophosphates. This confirms earlier research by (George, 2000) who indicated that a wide range of acaricides, including arsenical, chlorinated hydrocarbons, organophosphates, carbamates and synthetic pyrethroids are being used for controlling ticks on livestock. From the survey, there were no organoclorine pesticides being used for vector control in the study area. This is explained by (Kunz et al., 1994) that Organochlorine products for treating livestock are now unavailable or have been withdrawn from the market. According to studies carried out by NEMA, 2007a, OCPs were commonly used in the past, but many have been removed from the market due to their health and environmental effects and their persistence (e.g. DDT and chlordane). Organophosphates have gained popularity worldwide in preference to organochlorines, which are persistent and more damaging to the environment. The graph below shows the commonly used pesticides (brand names) in the study area.

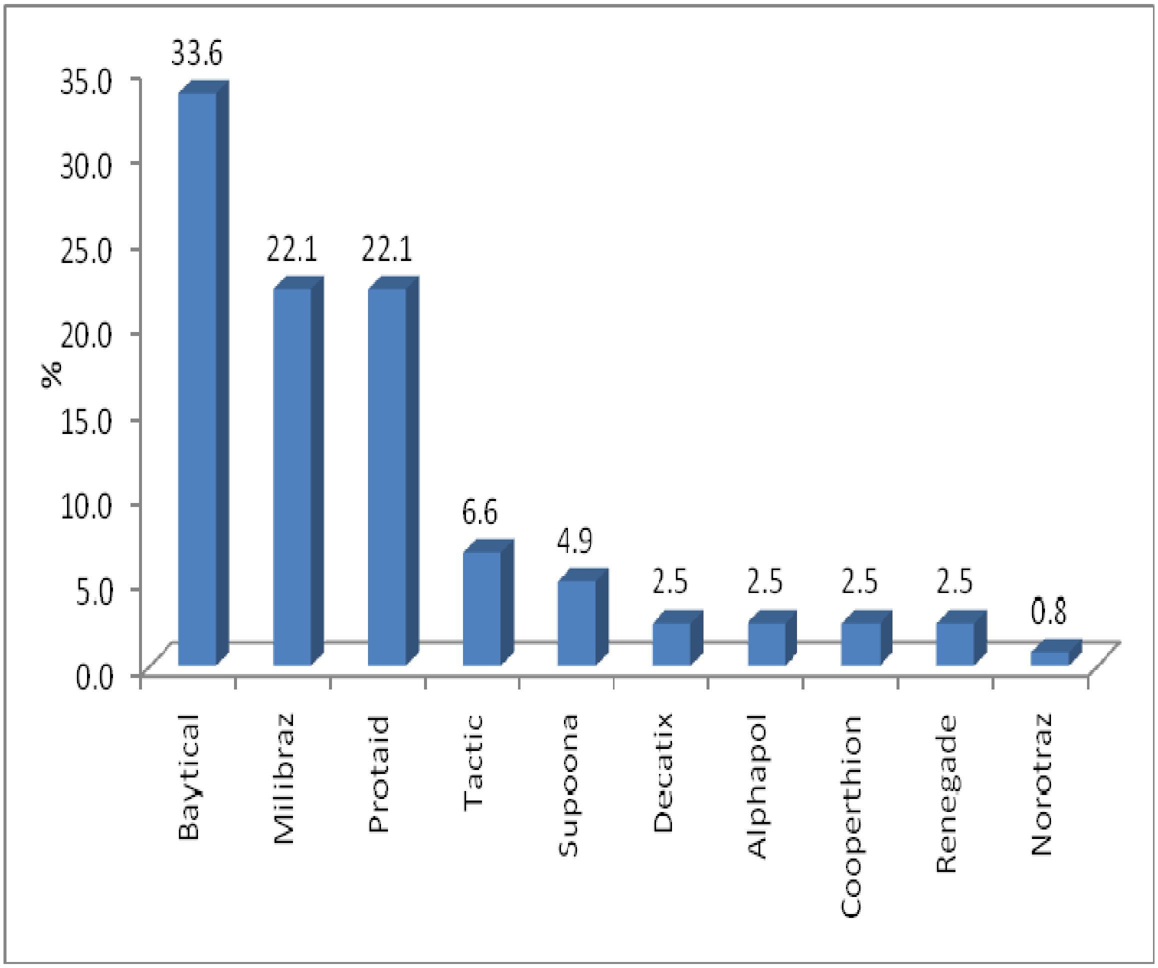

Figure 4: Graph showing commonly used pesticides

The major types of pesticides used were summarized in table 3 indicating the brand name, chemical composition, chemical group and mode of application.

Table 2: Major types of pesticides used

No	Pesticide Brand Name	Chemical composition	Chemical Group	CAS Number	IUPAC Name	Mode of Application
1	Bayticol	Flumethrin 2%m/v	Synthetic Pyrethroid	69770-45-2	Æ-cyano-4-fluoro-3-phenoxybenzyl 3-(ß,4-dichlorostyryl)-2,2-dimethylcyclopropanecarboxylate	Spraying
2	Protaid	Chlorfenvinphos 330g/l Alphacypermethrin 30g/l	Organophosphate	470-90-6	2-chloro-1-(2,4-dichlorophenyl)vinyl diethyl phosphate	Spraying
3	Milibitraz	Amitraz 12.5%m/v	Formamidine	33089-61-1	N'-(2,4-dimethylphenyl)-N-[(2,4-dimethylphenyl)iminomethyl]-N-methylmethanimidamide	Spraying
4	Alfapor	Alpha-cypermethrin 50g/l	Synthetic Pyrethroid	67375-30-8	α-cyano-3-phenoxybenzyl (1*S*)-cis-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate and (*S*)-α-cyano-3-phenoxybenzyl (1*R*)-cis-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate	Spraying and dipping

5	Renegade	Alpha-cypermethrin 50g/l	Synthetic Pyrethroid	As (4) above	As (4) above	Spraying
6	Norotraz	Amitraz 12.5%m/v	Formamidine	As (3) above	As (3) above	Spraying
7	Tacktic®	Amitraz 125g/l	Formamidine	As (3) above	As (3) above	Spraying
8	Supona aerosol	Chlorfenvinphos 0.48%m/m Dichlorvos 0.74%m/m Gentian Violet 0.145%m/m	Organophosphate	As (2) above	As (2) above	Spot treatment
9	Cooperthion	Deltamethrin	Synthetic Pyrethroid	62229-77-0	[(S)-cyano-(3-phenoxyphenyl)methyl] (1R,3R)-3-(2,2-dibromoethenyl)-2,2-dimethylcyclopropane-1-carboxylate	Spraying
10	Decatix	Deltamethrin	Synthetic Pyrethroid	As (9) above	As (9) above	Spraying

4.2 The levels of organochlorine, organophosphate and pyrethroid pesticide residues in livestock products (muscle and milk) and water

4.2.1 Levels of organochlorine pesticide (OCP) residues in meat, milk and water samples

From the survey, there were no organochlorine pesticide residues detected in meat, milk and water samples respectively. This finding could be attributed to the fact that the use of most organochlorine pesticides has been banned or restricted in Uganda under the Stockholm convention due to high levels of persistence in the environment and toxicity to non target organisms (NEMA., 2007a).

Battu et al., (2004) also indicated that organochlorine insecticides are known to be extremely persistent compounds and are either banned or severely restricted from use in most of the developed countries in the world. Similar studies as reported by NEMA, 2007a indicate that in the past, livestock protection against ticks was dominated by organochlorines (toxaphene and lindane) but these have now given way to organophosphates (Delnav and Supona) and synthetic pyrethroids deltamethrin (Decatix) and cypermethrin (Fendona).

The absence of organochlorine pesticide residues in the samples analyzed can also be explained by the fact that the use of most organochlorine pesticides has been banned or restricted in Uganda under the Stockholm Convention due to the high levels of persistence in the environment and toxicity to non target organisms.

4.2.2 Levels of selected organophosphate residues in meat, milk and water samples from the study area

4.2.2.1 Levels of selected organophosphate residues in meat samples

From the study, unquantifiable levels of chlorpyrifos were detected in meat samples 1, 2 and 4 from Nyakasharara subcounty (refer to Appendix 1). The presence of chlorpyrifos residues in muscle can be attributed to the feeding on contaminated fodder with pesticides.

4.2.2.2 Levels of selected organophosphate residues in milk samples

Liquid milk is an essential nutritional food for infants as well as the aged. Contamination of this milk with residues of pesticides is a matter of serious concern. Of the eleven pesticides analyzed, Dichlorvos Methacrifos, Diazinon Etrimfos, Chloropyrifos methyl, Fenitrothion, Malathion, Chlorpyrifos, Parathionnethyl, Piriphosethyl were not detected in any of the milk samples collected from the study area. However, Chlorfenvinphos residues of 0.32mg/l, were detected in sample 5 from Sanga Subcounty while 0.28mg/l and 0.31mg/l were detected in sample 2 and 5 from Nyakashashara subcounty as indicated in appendix 2. The values were above the EU MRL of 0.01mg/l. Nyakasahara recorded the highest mean value(0.12mg/l) of the Chlorfenvinphos residues detected followed by Sanga sub county with a mean value of 0.06mg/l while non of the milk samples collected from Kanyarweru had been detected with any organophate residue as shown in figure 6. A study undertaken by (John et al., 2001) indicates that animals can accumulate these pesticide residues from contaminated feed and water. He also explained that due to the lipophilic nature of pesticides, milk and other fat-rich substances are the key items for their accumulation.

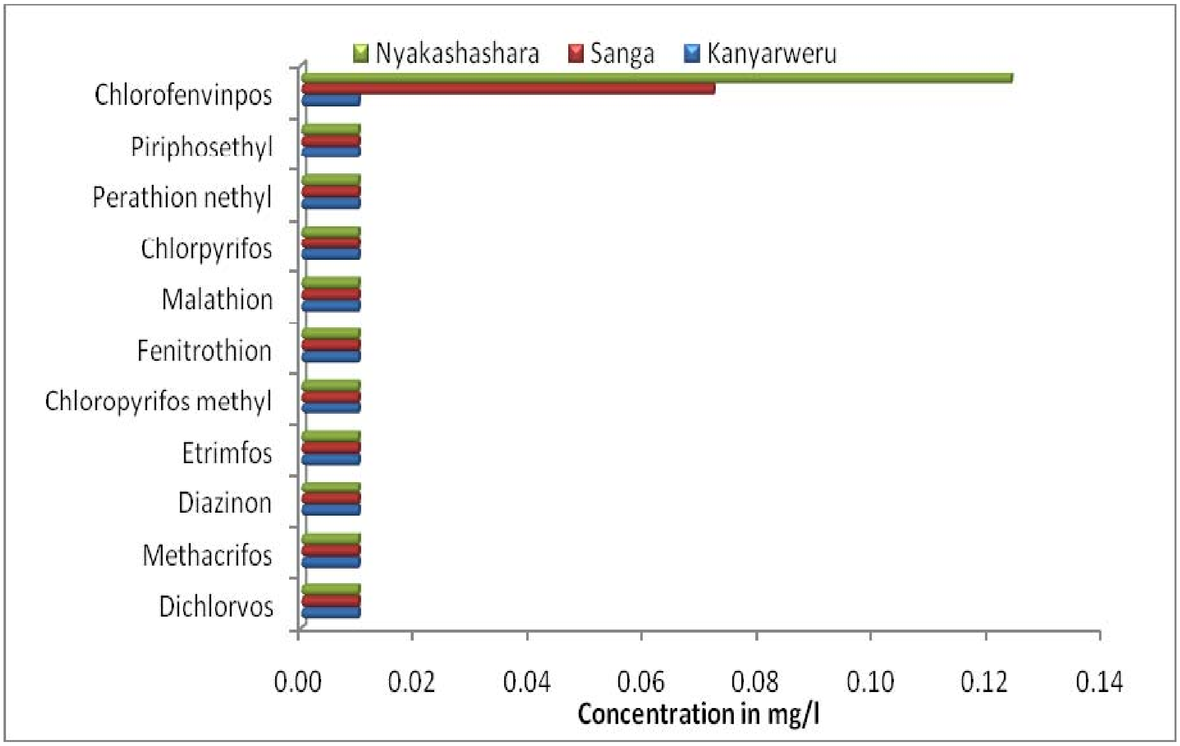

Figure 5: Levels (mg/l) of Pesticide residues in milk samples from the study area
MRL: 0.01 mg/l

4.2.2.3 Levels (mg/l) of selected organophosphate residues in water samples

Pesticide contamination can pose a hazard to both livestock and human health. These pesticides applied in the field or those by accidental spills of pesticides and careless discarding of pesticide containers finally find their way into the aquatic environment either through storm water run off and or as aerosols carried by wind. (Chindah et al., 2004). From the study results, chlorfenvinphos residues of 0.13 µg/l, 0.11 µg/l, were detected in sample 1 and 2 from Kanyarweru subcounty while 0.17 µg/l, 0.12 µg/l, 0.41 µg/l, 0.12 µg/l of chlorfenvinphos residues were detected in sample 1, 2, 3 and 4 from Nyakashara Sub County as indicated in appendix 3. The highest mean value of chlorfenvinphos residues (0.2µg/l) was recorded at Kanyarweru Sub County followed by Nyakashashara subcounty with a mean value of 0.10µg/l. The limit of detection of the instrumenent was 20 ng/mL. Presence of chlorfenvinphos residues may be attributed to the poor disposal of pesticides containers as the study showed that some of the pesticides containers were disposed off near the water sources. However, chlorfenvinphos may also have been released to water via runoff after its application to the cattle.

The study also indicated that most of the water sources used for the spray of the animals are located near the spray areas within 500 m for easy accessibility. As a result, heavy rains can lead to runoff waters therefore carrying pesticides into the water ponds. Studies undertaken by (Beaton, 2010) also indicated that the greatest danger of water contamination exists when spraying is carried out close to wells or surface water.

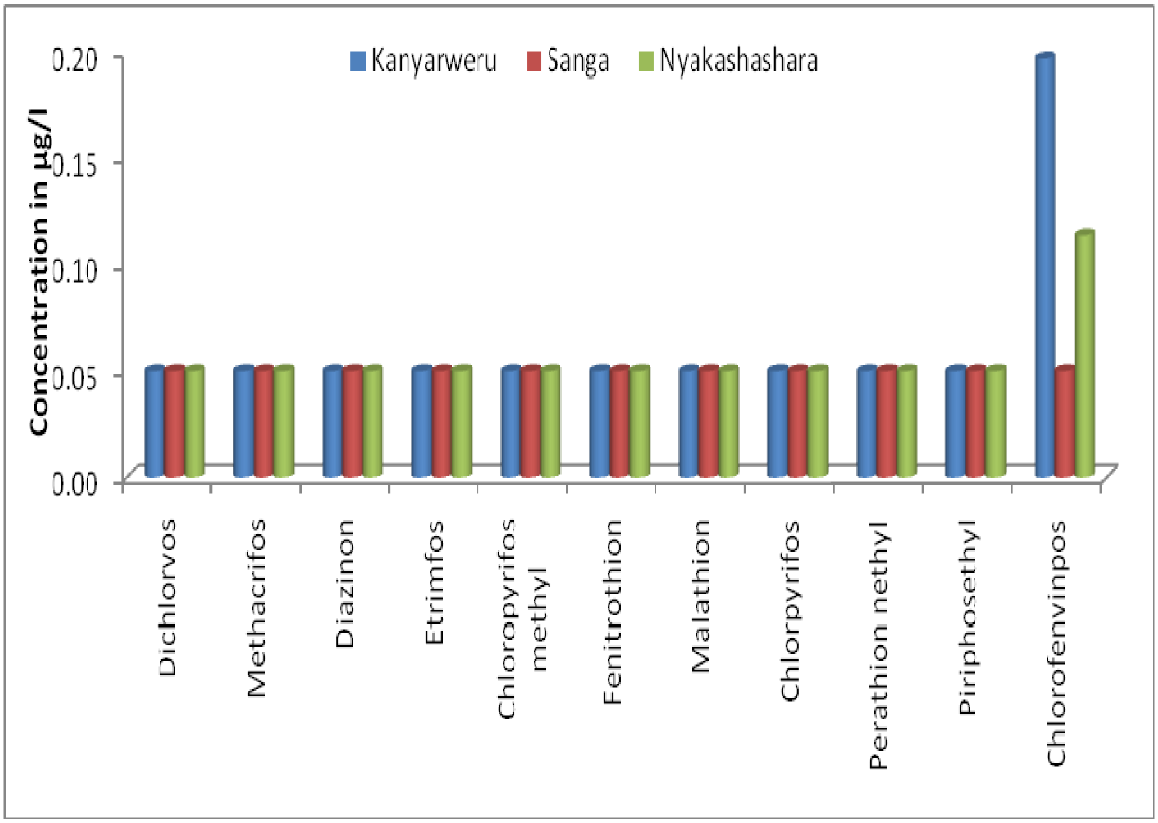

Figure 6: Mean levels of Chlorfenvinphos residues (µg/l) in water samples from the study area
NOTE: MRL for meat =0.05ug/l

4.2.3 Levels of selected pyrethroids residues in muscle, milk and water samples

The study results indicated that there were no pyrethroid residues detected in the animal products and water. These results are similar to (Khay et al., 2009) who analyzed muscle samples and could not find detectable amounts of cypermethrin. In my opinion, this was related to the duration of time between the spray times and sampling time given the fact that synthetic pyrethroids are not accumulated and not retained in the organisms for a long time i.e they do not have an accumulative effect. Also, time lag between sampling and analysis as well as preservation during transportation of samples could have contributed to the absence of pyrethroid residues in the samples analyzed.

4.3 Farmer's perceptions, knowledge and practices that contribute to the presence/levels of pesticide residues in livestock products and water around L. Mburo National Park.

Table 3: Farmers' knowledge on the use of pesticides

		Responses in Percentage (%)
Are animals affected by disease vectors?	Yes	100.0
Vectors affecting animals	Ticks	96.4
	Flies	3.6
Know what withholding time is?	Yes	12.5
	No	87.5
Ideal time between animal treating and slaughter or milking	Anytime hrs after spraying	46.9
	Immediately after spraying	34.7
	Some longer time after spraying	18.4
Know of any effects of consuming milk, eat meat of water with pesticides residues	Yes	72.7
	No	27.3
Effects of consuming milk with pesticides	Sneezing	33.7
	Dizziness	16.9
	Itching	36.1
	Eye irritation	12.0
	General weakness	1.2
Pesticides containers may be used for domestic use	Yes	60.4
	No	39.6

Table 4: Farmers' perceptions towards the use of pesticides

		Responses in Percentage (%)
Not following prescriptions may be dangerous	Agree	70.3
	Disagree	29.7
Think it is good to take milk or eat meat immediately after use of pesticide?	Yes	60.0
	No	40.0
Pesticides can be used to treat human diseases	Agree	39.2
	Disagree	29.4
	Strongly Disagree	31.4

Table 5: Farmers' practices regarding the use of pesticides

		Responses in Percentage (%)
Use Pesticides?	Yes	100.0
Sources of pesticides	Veterinary drug shops	83.6
	Open Markets	10.9
	Veterinary Doctors	5.5
Person who treats the animals	Vet doctor	3.8
	Family Head	77.4
	Anyone available	5.7
	Paid worker	13.2
How pesticides are applied	Spraying	94.4
	Dipping	5.6
Use protective gear while applying pesticides	Yes	1.8
	No	98.2
Follow prescriptions on label of pesticides?	Yes	56.8
	No	43.2
Ever used pesticides to treat human diseases	Yes	33.3
	No	66.7
How pesticide drug containers are disposed off	Anywhere around spray area	78.2
	Collected and burned	5.5
	Disposed into dustbins/pits	10.9
	Used for domestic activities	5.5

4.3.1 Farmers' knowledge on the use of pesticides

4.3.1.1 Vectors affecting animals

From the survey, 96% of the respondents indicated that their livestock are affected by ticks. This is similar to the findings of Soneshine, 1991 who indicated that ticks are the most common external parasites of economic importance in the small holder farming areas of the southern region of Africa . (Keyyu et al., 2003) and (Swai, 2005)also explained that ticks and helminths have a wide host and geographic diversity and hence constitute a major constraint to livestock production in the tropics and subtropics. Analysis of variance (ANOVA) showed that there is no significant difference ($P>0.05$, ANOVA) on the types of vectors affecting the three sub counties. (FAO., 2006) and (CRC-VT., 2001) indicated that ticks have different effects in cattle, including being a vector of tick-borne diseases (theileriosis, babesiosis and ehrlichiosis), suck blood, and cause teat, ear and hide damage. They predispose the animal to abscesses, foot rot and screwworm infection. Ticks also have an indirect effect on cattle in terms of milk production (damage of the teats) and reduced weight gain (L'Hostis & seegers, 2002; Peter et al., 2005). These effects seriously limit livestock production and improvement (Latif & Jongejan, 2002). Therefore, ticks need to be controlled if livestock production is to meet world needs for animal protein.

4.3.1.2 Withholding Period

The withholding period (WHP) is the minimum period which must elapse between last administration or application of a veterinary chemical product, including treated feed, and the slaughter, collection, harvesting or use of the animal commodity for human consumption. WHPs are mandatory for domestic slaughter and on the label of every registered product. Most farmers in developing countries do not often care about withdrawal periods in treated animals (Keyyu et al., 2003) and cows are usually treated and milked at the same time in the morning before being let out to graze (Awumbila et al., 1994).

In this study, farmers were interviewed on the importance and knowledge of withdrawal periods and the public health significance. 88% of the farmers interviewed had no knowledge on the withholding periods of the pesticides while 12% were knowledgeable about the withholding periods of the pesticides. 47% of the farmers milked their cows a few hours (maximum 2 hours) after spraying the animals with the acaricides, 35% indicated that they milked their cattle immediately after spraying. 18% of the farmers indicated that they milked their cattle after some longer time after spraying for example, if the cows were sprayed in the morning, the cows would be milked in the evening. The graph below shows the trends time taken by the farmers to milk their cattle after application of the pesticides.

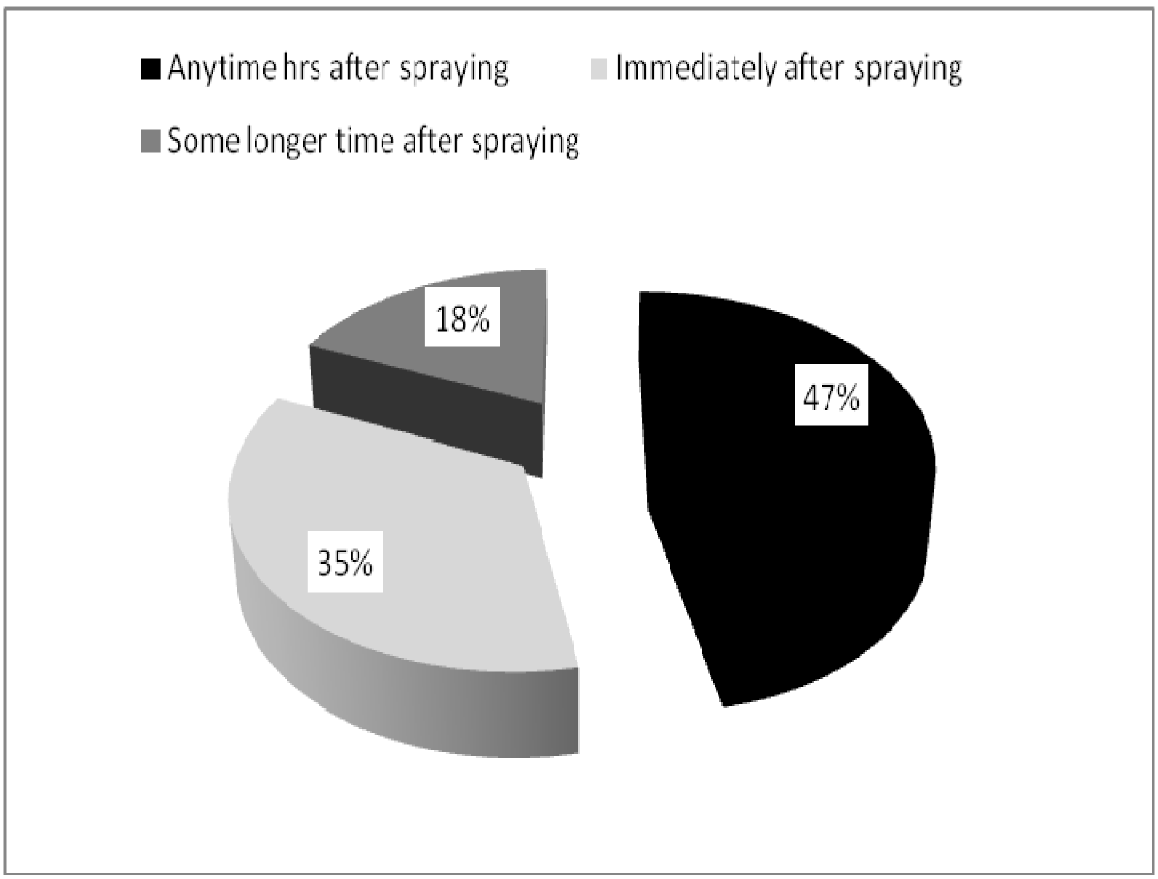

Figure 7: Time taken to milk after application of pesticide

4.3.2 Farmers perceptions regarding the use of pesticides

4.3.2.1 Following pesticide prescriptions

70% of the farmers interviewed agreed that not following prescriptions of the pesticides may be dangerous while 30% disagreed. There is need to raise awareness of the farmers and bring some perceptional change towards their conventional practices of applying pesticides without following the pesticide prescriptions on the labels. Anecdotally, there was a report that even when able to read, some farm workers were reluctant to read manufacturer's labels(Avory & Coggon, 1994).

4.3.2.2 Perception on consuming animal products immediately after using the pesticide

From the study, 60% of the farmers interviewed thought it was not harmful to consume animal products specifically milk and meat immediately after the use of the pesticide while 40% thought otherwise.

4.3.3 Farmers practices regarding the use of pesticides

4.3.3.1 Use of pesticides

All the farmers interviewed indicated that they use pesticides (acaricides) to control ticks on their livestock. Another study by (Martins et al., 1995) revealed that chemical control with acaricides was considered as one of the best methods to control ticks. However these chemicals are toxic and costly. Problems of acaricide resistance, chemical residues in food and the environment and the unsuitability of tick resistant cattle for all production systems make the current situation unsatisfactory, which is why there is debate on the development of an alternate and absolute control method, such as through vaccine (Zahid et al., 2006). A study conducted by (Laffont et al., 2001) indicate that conventional acaricides tend to contaminate the environment and indiscriminately kill beneficial insects (parasitoids), birds (oxpecker) and have harmful effects on the food chain.

4.3.3.2 Sources of pesticides

The survey indicated that 84% of the pastoralists obtained their pesticides from veterinary drug shops while 11% obtained their pesticides from open markets and 5% of the farmers obtained their pesticides from veterinary doctors as shown in figure 9 below. Keyyu et al., (2003) also reported that private drug shops were the primary sources of veterinary drugs for 66.7% of smallholder dairy farmers compared to only 33.3% from official Government extension agents in Tanzania.

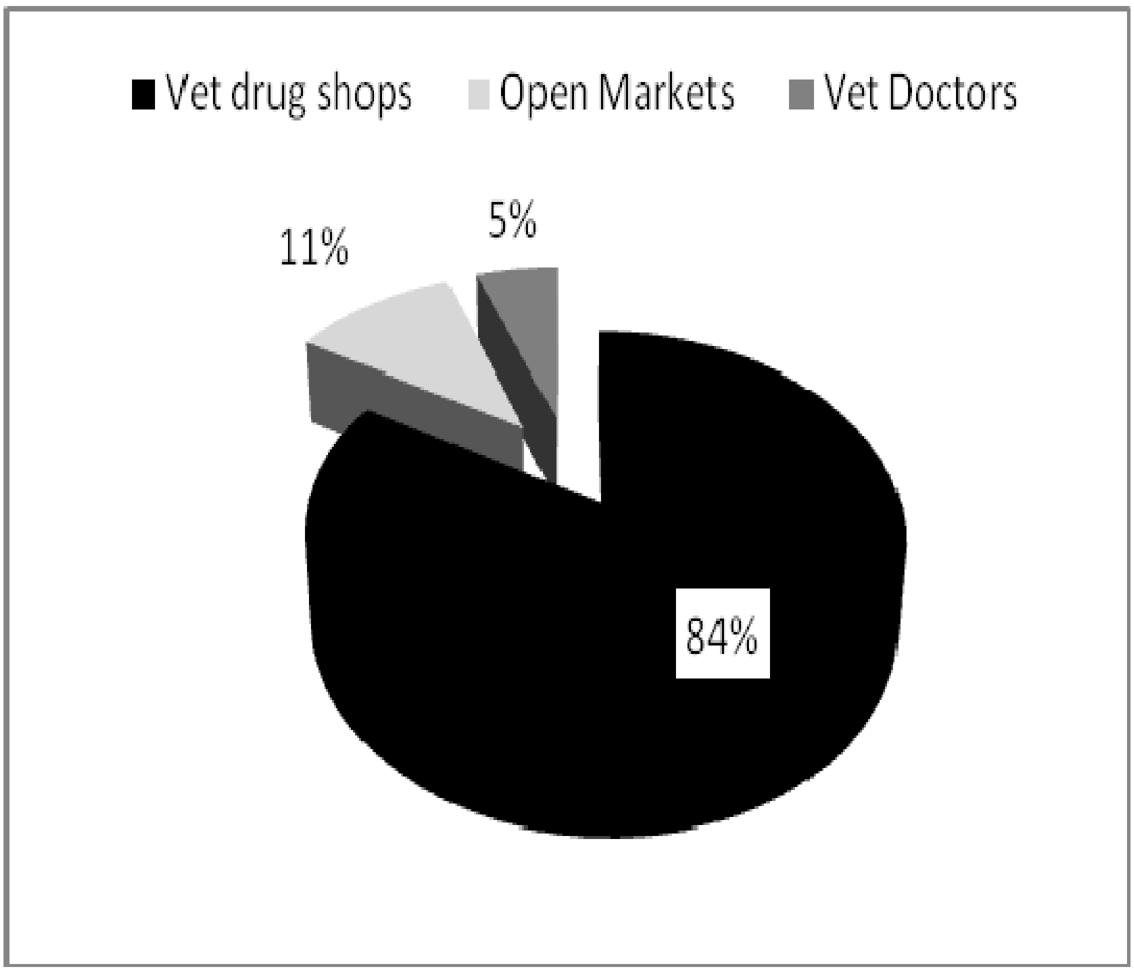

Figure 8: Sources of pesticides

4.3.3.3 Mode of application of pesticides

Ticks are generally controlled with the use of conventional acaricides that are applied by dipping, spraying or pour-on which is considered as one of the best methods(Martins et al., 1995). Their application to cattle has been simple and widely accepted by farming communities, governments and donor agencies (Latif and Jongefan, 2002). From the study, majority of the farmers (94%) applied the pesticides by spraying and 6% by dipping. (Swai et al., 2005) also indicated that hand spraying was the most common method used in application of the acaricides in the pastoral Maasai community-Ngorongoro, Tanzania. The application of fluid acaricides to an animal by means of a spray has many advantages and has been successfully practiced for controlling ticks on most of the animals (Barnett, 1961). Dipping is consequently expensive, incoherent and inconsistent (Kivaria, 2006a). In general, dipping vats provide a highly effective method of treating animals with acaricides for tick control. However, their immobility, high initial cost of construction, and the cost of the acaricides may make vats impractical for many small ranching operations. Also, dipping vats must be managed carefully so that the dips are maintained at the proper concentration and the cattle are dipped properly (Drummond, 1983). The study also revealed that 77% of the family head applied the pesticides on the cattle, 13% indicated that the worker could apply the pesticide and 4% indicated that the spraying was done by the veterinary doctors. While 13% indicated that anyone available can apply the pesticide. Whereas 57.8% of the farmers followed the prescriptions on the labels of the pesticide containers, 43% did not necessarily follow the prescriptions. Whatever the treatment method, adherence to procedures developed by the manufacturer is essential for maximizing the degree of tick control that will occur. Inadequate veterinary personnel and registered drug shops in rural areas have also been associated with improper use of acaricides in Africa. Similar studies among nomadic Massais in Tanzania also showed that 76% of them administered acaricides themselves and did so at the wrong dosage rates (Swai et al., 2005).

4.3.3.4 Safe use of pesticides

In this study only 57% of the farmers followed directions and precautions on the label of the pesticide. Further, the results of field survey regarding the safe use of pesticides indicated that 99% of farmers did not wear protective gear such as eye glasses, goggles or overalls when mixing or applying pesticides. Such unsafe practice was also reported among pesticide sprayers in the Mississippi Delta (Rucker, 1994).

4.3.3.5 Disposal of pesticide containers

Pesticides (herbicides, insecticides, fungicides, etc.) are designed to be toxic. Even in very small quantities, some can have a substantial adverse impact on sensitive organisms. Improper disposal of pesticides or their containers can lead to environmental contamination (Amy, 1999).

From the survey, 78% of the farmers disposed their pesticide containers around the spray area, 11% disposed their pesticide containers into dust bins, 6% used the containers for domestic use while another 5% indicated that the empty pesticide containers were burnt. The contents of the containers can be washed into the environment and water bodies thus contaminating the water quality. It was also observed that the distance from the water source and spray area ranged from 4m to 2km. Figure 8 below shows how the farmers in the study area disposed off their pesticide containers while figure 9 shows the poorly disposed off pesticide containers near the spray area.

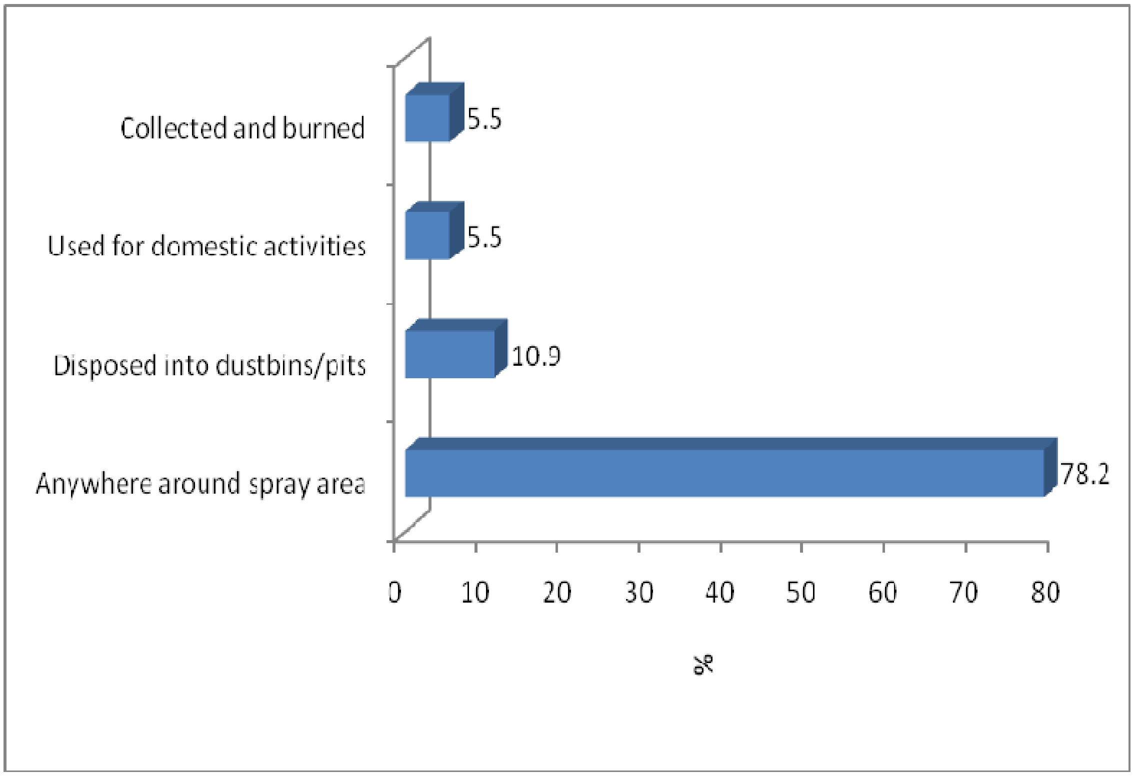

Figure 9: Gragh showing disposal of pesticide containers

Figure 10: Poorly disposed off pesticide containers at the spray area and water source

CHAPTER FIVE

5.0 Conclusions and recommendations

5.1 Conclusions

The results from the study indicate that 100% of the pastoralists use pesticides for control of ecoto-parasites. The survey revealed a total of ten (10) different pesticides that were being used by the farmers in the study area with synthetic pyrethroids (50%) being widely used followed by formamidine(30%) and organophosphates (20%). There was no organochlorine pesticide being used by the farmers.

The study results also revealed that there were no Organochlorine pesticide residues and pyrethroid residues detected in any of the samples analyzed. However, residues of chlorfenvinphos (an organophosphate) of 0.13 mg/l, 0.11 mg/l, were detected in water samples collected from Kanyarweru subcounty, 0.17 mg/l, 0.12 mg/l, 0.41 mg/l, 0.12 mg/l from water samples picked from Nyakashara subcounty. In addition, chlorfenvinphos residues of 0.32 mg/l, were detected in milk sample 5 from Sanga Subcounty while 0.28 mg/l and 0.31 mg/l were detected in milk sample 2 and 5 from Nyakashashara subcounty**.** These values were above the WHO maximum residue levels and therefore, these waters and animal products specifically milk are not safe for human consumption.

The presence of pesticide residues in animal products and water could be attributed to the practices, perceptions and knowledge about the use of pesticides amongst the pastoralists. The study revealed that 78% of the farmers disposed their pesticide containers around the spray area. Most of the spray areas were located in the radius of not more than 1km from the water source. This could be the probable cause of water contamination with pesticide residues as a result of runoff from the spray areas into the water samples. In addition, 88% of the farmers interviewed had no knowledge on the withholding periods of the pesticides.

Although the assessment and containment of public health risks in livestock products is a matter of priority, such commitment was not evident in the study area based on the farmers' attitude regarding the importance of good practices of handling the pesticides. For example, most farmers were concerned with the well being of their cattle and would apply the pesticides without any protective gear despite the health risks associated with the pesticides.

The human population and livestock in Kiruhura District and Uganda at large are at high risk of exposure to pesticide residues in livestock products and water as a result of the knowledge, perceptions and practices of pastoralists on the use of pesticides.

5. 2 Recommendations

In order to control the resultant residue problem in foods and promotion of food trade in international market, a national pesticide residues monitoring program should be implemented.

There should be a coordinating team of public private partnership for close monitoring of appropriate use of pesticides at the field level. In addition, there is need for a massive campaign to all farmers, retailers, distributors of pesticides about the use and safety measures of pesticides.

There is also need to enhance the laboratory capacities and facilities in pesticide residue analysis in order to cope with the newly emerging challenges of pesticide residues.

There is need to establish active surveillance programmes to monitor pesticides residues to prevent international trade barriers associated with the presence of pesticides residues in livestock products.

There is need for comprehensive assessment of pesticide residues in livestock products for a longer period to generate data for more research and guiding policy makers.

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Appendix 1: Levels (mg/Kg) of selected Organophosphate residues in Meat, samples from the study area

Subcounty	Sample	Dichlorvos	Methacrifos	Diazinon	Etrimfos	Chloropyrifos methyl	Fenitrothion	Malathion	Chlorpyrifos	Parathion nethyl	Piriphosethyl	chlorfenvinphos
Kanyarweru	Sample 1	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
	Sample 2	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
	Sample 3	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
	Sample 4	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
	Sample 5	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
Sanga	Sample 1	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
	Sample 2	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
	Sample 3	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
	Sample 4	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
	Sample 5	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
Nyakashashara	Sample 1	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	**Detected**	<0.01	<0.01	<0.01
	Sample 2	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	**Detected**	<0.01	<0.01	<0.01
	Sample 3	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
	Sample 4	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	**Detected**	<0.01	<0.01	<0.01
	Sample 5	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01

Appendix 2: Levels (mg/l) of selected Organophosphate residues in Milk samples from the study area

Subcounty	Sample	Dichlorvos	Methacrifos	Diazinon	Etrimfos	Chloropyrifos methyl	Fenitrothion	Malathion	Chlorpyrifos	Parathion nethyl	Piriphosethyl	chlorfenvinphos
Kanyarweru	Sample 1	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
	Sample 2	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
	Sample 3	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
	Sample 4	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
	Sample 5	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
Sanga	Sample 1	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
	Sample 2	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
	Sample 3	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
	Sample 4	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
	Sample 5	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	**0.32**
Nyakashashara	Sample 1	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
	Sample 2	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	**0.28**
	Sample 3	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
	Sample 4	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
	Sample 5	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	**0.31**

MRL for OPPs in milk is 0.01 mg/l

Appendix 3: Levels (µg/l) of selected Organophosphate residues in water samples from the study area

Subcounty	Sample	Dichlorvos	Methacrifos	Diazinon	Etrimfos	Chloropyrifos methyl	Fenitrothion	Malathion	Chlorpyrifos	Parathionn ethyl	Piriphosethyl	chlorfenvinphos
Kanyarweru	Sample 1	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	**0.13**
	Sample 2	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	**0.41**
	Sample 3	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05
Sanga	Sample 1	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05
	Sample 2	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05
Nyakashashara	Sample 1	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	**0.11**
	Sample 2	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	**0.17**
	Sample 3	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	**0.12**
	Sample 4	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	**0.12**
	Sample 5	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05

MRL for OPPs in water is 0.05 µg/l

Appendix 4: Questionnaire used in the survey

Dear Respondent,

You have been randomly selected in the study titled "assessing the levels of veterinary drugs/chemical residues in livestock derived foods and water around Lake Mburo National park in Uganda" which is being carried out as part of an education research in partial fulfillment of the requirements for the award of the degree of Master of Science in Environment and Natural Resources at Makerere University. Your cooperation in filling this questionnaire will lead to the success of the survey. All responses shall be for academic purposes only and will be treated with confidentiality.

Date:

PART (I) BACKGROUND INFORMATION

Name	
Sex	
Village	
Family Position	
Size of herd	

Part II: Attitude, Knowledge and practices of veterinary drugs and effects on human health

1. Are your animals affected by animal disease vectors? Yes ☐ No ☐

2. If yes, which ones?

Ticks ☐

Fleas ☐

☐

Others

3. Do you use pesticides to treat the vectors? Yes ☐ ☐

4. If yes, what pesticides are used?

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5. Where do you get the pesticides from?

Veterinary drug shops ☐

Open markets ☐

Veterinary doctors ☐

Others ☐

6. Who treats the animals?

Veterinary Doctor ☐

Family head ☐

Anyone available ☐

Other ☐

7. How are the pesticides applied?

Spraying ☐

Dipping ☐

Injection ☐

Others ☐

8. Do you follow the prescriptions on the label of the pesticide when treating the animal?

Yes ☐ No ☐

9. Not following prescriptions of the pesticides may be harmful

Agree ☐ Disagree ☐

☐ ☐

10. Do you know about the withholding time? Yes No

11. If not, how soon do you milk or slaughter your cows after treatment?

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12. Do you think it is good to take milk or eat meat immediately after treating the animals? ☐ ☐

Yes NO

13. Do you know of any effects of consuming milk, meat or water with pesticide residues?

Yes ☐ NO ☐

14. If yes, which ones?

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15. Have you suffered from any diseases associated with Pesticide residues?

Yes ☐ NO ☐

16. If yes, which ones?

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17. Do you use the pesticides else where apart from treating animals?

Yes ☐ No ☐

18. These drugs can be used to treat human diseases.

Agree ☐ Disagree ☐ strongly disagree ☐

19. Have you used them before to treat human diseases?

Yes ☐ No ☐

20. How do you dispose off your drug containers after treating the animals?

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21. The containers for pesticides are used for domestic purposes such as collecting water, storing salt among others

True ☐ False ☐

22. Do you use Protective gear when applying pesticides? (Overalls, Gloves, Masks, etc)

Yes ☐ NO ☐

23. What is the distance between the water source and the spray area?

Appendix 5: Pesticide residue chromatogram for samples analyzed