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EVALUATION OF LIVE *ARTEMISIA ANNUA* L. PLANTS AS MOSQUITO REPELLENTS IN BOARDING SCHOOLS AND HOUSEHOLDS OF MWEA IRRIGATION SCHEME, KIRINYAGA COUNTY, KENYA

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OCTOBE

DECLARATION

This thesis is my original work and has not been submitted for the award of degree or any other award in any other University.

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To my late father, Michael Mbulo Munee and my mother, Veronica Salome Nthambi Mbulo.

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TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF APPENDICES

DEFINITION OF OPERATIONAL TERMS

Anthropophilic: Preferring humans to other animals as a source of food.

Asymptomatic: Exhibiting or producing no symptoms.

Efficacy: The degree to which an intervention accomplishes the desired or projected outcomes.

Endophilic mosquito: A mosquito that tends to inhabit or rest indoors.

Exophilic mosquito: A mosquito that tends to inhabit or rest outdoors

Gonotrophic cycle: A life cycle of alternate feeding and laying of eggs

Haematophagous: Feeding on blood

Mesoendemic: Variable transmission that fluctuates with changes in one or many local conditions, e.g., weather or disturbance to the environment.

Morbidity: The state of being diseased or unhealthy within a population.

Mortality: The proportion of deaths to population

Semiochemicals: A chemical compound that conveys a signal from one organism to another so as to modify the behavior of the recipient organism.

Zoophilic mosquito: A mosquito that prefer to take blood meals on animals.

Zooprophylaxis: The use of animals (such as cattle) to divert disease-carrying insects such as mosquitos from humans.

Zoopotentiation: Increasing malaria transmission.

ABBREVIATIONS AND ACRONYMS

ACT	Artemisinin Combination Therapy
AL	Artemether-Lumefantrine
ANOVA	Analysis of variance
CDC	Centre for Disease Control
CHWs	Community Health Workers
CORPS	Community Owned Resource Persons
CQ	Choloroquine
DDT	Dichloro-diphenyl-trichloroethane
DMP	Dimethyl phthalate
DVBD	Division of Vector Borne Diseases
GMMs	Genetically Modified Microbes
IGRs	Insect Growth Regulators
IRS	Indoor Residual Sprays
ITNs	Insecticide Treated Nets
IVM	Integrated Vector Management
KDHS	Kenya Demographic and Health Survey
KNBS	Kenya National Bureau of Statistics
MoH	Ministry of Health
MRIS	Mwea Rice Irrigation Scheme
NACOSTI	National Commission for Science, Technology and Innovation
RBM	Roll-Back Malaria
SEM	The Standard Error of the Mean

SP	Sulphadoxine-Pyrimethamine
SPSS	Statistical Package for the Social Sciences
SSA	Sub-Saharan Africa
UNICEF	United Nations Children's Emergency Fund
UNDP	United Nations Development Fund
USA	United States of America
ULV	Ultra-low-volume
VOC	Volatile organic compounds
WHO	World Health Organization

ABSTRACT

Mosquitoes have attained public health concern than any other arthropod. The blood-feeding females transmit the protozoa causing malaria, filarial worms causing elephantiasis, and viral diseases. Malaria is a serious public health issue in sub-Saharan Africa, infecting between 300 and 500 million people annually, and it's the leading cause of infant and child morbidity and mortality. Approximately 70 *Anopheles* species have been shown to be competent vectors of human malaria. Methods used for malaria control include killing mosquitoes at larval and adult stages and prevention of biting. Mechanical barriers for example, impregnated mosquito bed nets are commonly used to provide protection from mosquito bites. Despite the use of these conventional methods to prevent mosquito bites, there continues to have cases of malarial deaths. The main objective of this study was to evaluate the mosquito repellence effect of live potted *Artemisia annua* L. plants hung at doors and windows of dormitories in boarding secondary schools of Mwea Rice Irrigation Scheme (MRIS), Kirinyaga County, Kenya. Cross-sectional and experimental ecological study designs were used in this study. Four boarding schools in the MRIS were purposively selected. A total of 340 students were randomly selected from the four schools and a structured questionnaire was administered to the students to gather quantitative data. Also four villages neighbouring the boarding schools were purposively selected and 340 households were systematically sampled for this study. A structured questionnaire was administered to the 340 selected households to gather both qualitative and quantitative data. Baseline mosquito density measurement was done before introducing *Artemisia annua* in the schools by collecting mosquitoes in selected dormitories. Potted live *A. annua* plants were hung at the doors and windows of the selected dormitories. Mosquitoes were collected from the selected dormitories after introducing *A. annua* using suction tube method. The level of knowledge on mosquito control strategy amongst students was high at 97%. Mosquito control methods at household level were use of mosquito bed net (75.8%), use of firewood and mosquito coil (17%), use of insecticide (24%), and environmental management practice (70%). Mean (± SEM) baseline mosquito density in boarding schools were 47.62±2.20, 48.00 ±1.93 for girls treatment and control dormitories respectively, 38.50±2.82, 42.50±2.01 for boys treatment and control dormitories respectively, and 50.37±2.78, 61.50±3.29 for girls and boys blanks dormitories respectively. Live *Artemisia annua* introduction resulted in decreases of mean mosquito catches in both the treatments and controls. Differences in the mean mosquito catches in both the control and treatment dormitories were significant at $p = 0.001$ for the months of March-June respectively. The study concluded that live *A. annua* reduced the mosquito density in dormitories of boarding schools even when they were separated by 50 metres. The study recommends that *A. annua* should be planted near dormitories and other buildings to repel mosquitoes.

CHAPTER ONE: INTRODUCTION

1.1 Background Information

Malaria is a common and life-threatening disease in many tropical and subtropical areas. It is a protozoan infection of the red blood cells, transmitted by the bite of a female anopheles mosquito and is caused by the protozoa of the genus *Plasmodium* (WHO, 2013). Malaria is characterized by paroxysms of fever, the length of which depends on the particular *Plasmodium* species that is the causative organism. Serious consequences, including death, can result, especially after infection with *P. falciparum*. Human malaria is caused by four different species of *Plasmodium*: *P. falciparum*, *P. malariae*, *P. ovale* and *P. vivax* and is transmitted by female *Anopheles* mosquitoes (Antinori *et al*., 2013).

Mosquitoes have attained greater public health concern because of the blood feeding females of different species transmitting the protozoa causing malaria, the filarial worms causing elephantiasis, and the viruses causing dengue fever, yellow fever, Rift Valley fever, West Nile fever, Zika and Chikunguya diseases (Lundström *et al*., 2013). As well as being important vectors of human pathogens, they can attack humans and other mammals causing a nuisance that negatively affects individuals and society, and may have large economic consequences (Justicia and Cianci, 2015).

Mosquito biting causes stress and medical problems in schools. This is a serious health issue requiring effective mosquito control measures (Impoinvil *et al*., 2007). Incessant mosquito biting presents a health hazard that ranges from loss of sleep and severe stress to serious infections (Jamieson, 2006). *Anopheline* transmit malaria, a disease that is the main cause of school absenteeism, high dropout rates, child labour (Burlando, 2012), affecting academic performance (Thuilliez, 2010), class repetition (Thuilliez *et al*., 2010), and even death of school age children (Booker, 2009).

The Mwea Rice Irrigation Scheme (MRIS) economic activities, which are water-related, have aggravated the problem of mosquito-borne malaria by increasing the number of larval habitats and extending the duration of transmission season (Muturi *et al*., 2008). Disease transmission can be interrupted by controlling the vector using various methods including botanical phytochemicals with mosquitocidal potential and repellent activities (Govindarajan *et al*., 2010). *Artemisia annua* plant has an important natural anti-malarial drug efficacious against drug-resistant strains of *Plasmodium*, the malarial parasite (Abdin *et al*., 2003).

Many plant-based products are widely used for their insecticidal/repellent properties for control of mosquitoes and protection from mosquito bites (Bhargava *et al*., 2013). The essential oil of *Artemisia annua* L. has shown toxic repellency and inhibitory activities against two economically important stored product insects. *Tribolium castaneum* (Herbst) and *Callosobruchus maculatus* (Linnaeus) (Tripathi *et al*, 2000); and also a fumigant effect against *Oryzaephilus surinamensis* (Linnaeus) (Coleoptera: Silvanidae) (Lü *et al*, 2011). It has adulticidal activity of oil extracted from the leaves of *L. camara* against *Aedes aegypti* Linnaeus, *Culex*, *quinquefasciatus* Say, *Anopheles culicifacies* Giles, *An. fluviatilis* James and *An. stephensi* Liston mosquitoes (Dua *et al*., 2010).

Essential oils as pesticides lies in the fact that the constituents of all essential oils are moderately toxic or mostly found to be nontoxic to mammals, birds and the aquatic ecosystem (Patel *et al*., 2012).

Lantana camara essential oils, just like *Artemisia annua* essential oils, have shown repulsion effect against haematophagous insect. When live *L. camara* was planted as a hedge around houses significant reduction in mosquitoes *A. gambiae* s.s. and *A. funestus* s.s., was reported (Mng'ong'o *et al*., 2011). Other studies by Seyoum *et al* (2002) on mosquito repellence of essential oil from potted plants demonstrated reduction in *A. gambiae* s.s. and *A. funestus* s.s. mosquitoes. This study investigated whether potted live *Artemisia annua* L. plants used as a hedge around school dormitories would reduce mosquito densities and hence be used as an ideal ecofriendly approach for malaria vector control programmes.

1.2 Statement of the problem

Mosquitoes continue to transmit malaria despite the use of several methods of control such as treated bed nets, aerosols and repellent creams (Cibulskis *et al*., 2011). Students in boarding schools in malaria prone areas suffer most from mosquitos through noise and bites leading to stress and medical problems (Impoinvil *et al*., 2007). Disturbance from mosquito affect the learning environment in boarding schools and this has led to poor performance (UNICEF, 2010; Udowa *et al*., 2010). School attendance is also disrupted with approximately 4 million school days lost annually as a result of malaria (Halliday *et al*., 2014).

While a great deal of research effort has been placed on the conventional methods of malaria control, comparatively little research on the repellency effect of live *Artemisia annua* has been investigated in Mwea Rice Irrigation Scheme (MRIS). The current study aimed at investigating the effect of *Artemisia annua* on mosquito populations MRIS.

1.3 Justification

Despite malaria being in existence not much has been done to prevent it using *A. annua* repellence. No study in Kenya has been done to determine the effect of *A. annua* on mosquito density with view to reduce malaria. There is therefore need for innovative approaches to reduce its burden. Therefore this study endeavored to address this problem. The results of this study may be used by policy makers to improve learning environment in boarding schools and household's areas.

1.4 Research questions

1. What is the level of knowledge amongst students on mosquito control strategies in Mwea Rice Irrigation Scheme, Kirinyaga County?
2. What are the mosquito control methods used by the hosueholds neighboring the boarding schools in Mwea Rice Irrigation Scheme?
3. What is the mosquito density in boarding schools in Mwea Rice Irrigation Scheme, Kirinyaga County?

4. What is the effect of *Artemisia annua* L plant on mosquito density in boarding schools in Mwea Rice Irrigation Scheme, Kirinyaga County?

1.5 Null hypotheses

1. The level of knowledge among students on mosquito control strategies is low in Mwea Rice Irrigation Scheme, Kirinyaga County.
2. The households neighboring the boarding schools in Mwea Rice Irrigation Scheme do not have mosquito control methods.
3. The mosquito density in boarding schools is low in Mwea Rice Irrigation Scheme, Kirinyaga County.
4. *Artemisia annua L.* plants do not reduce mosquito density in boarding schools in Mwea Rice Irrigation Scheme, Kirinyaga County.

1.6 Study objectives

1.6.1 General objective

To evaluate live potted *Artemisia annua* l. plants hung at doors and windows as mosquito repellents in boarding schools of Mwea Rice Irrigation Scheme, Kirinyaga County, Kenya

1.6.2 Specific objectives

1. To determine the level of knowledge amongst students on mosquitoes control tactics in boarding schools Mwea Rice Irrigation Scheme;
2. To establish the mosquito control methods used by the households neighboring the boarding schools in Mwea Rice Irrigation Scheme;
3. To determine the mosquito density in boarding schools in Mwea Rice Irrigation Scheme, Kirinyaga County;
4. To evaluate the effect of *Artemisia annua* L. plants on mosquito density in boarding schools in Mwea Rice Irrigation Scheme, Kirinyaga County.

1.7 Significance and anticipated output

The findings and recommendations of this study will help health and education policy makers in Kenya to make crucial decisions on *Artemisia annua* L. use in complementing other preventive strategies in the control of malaria.

1.8 Conceptual framework

The conceptual framework shows the socio-demographic characteristics, factors affecting mosquito entry in the dormitories, moderating variable (*Artemisia annua* plant) which acts as a barrier between the host and vector; and the mosquito density. Chemical compounds produced by the plant would modify mosquito behaviour by acting as repellent reducing their entry to the dormitories, thereby reduced mosquito density in the dormitories.

The conceptual framework influences the choice of intercession, predicts the expected outcomes of the intervention, suggests factors that may influence the results (factors in *italics* were not investigated), and suggests strategies for improving outcomes.

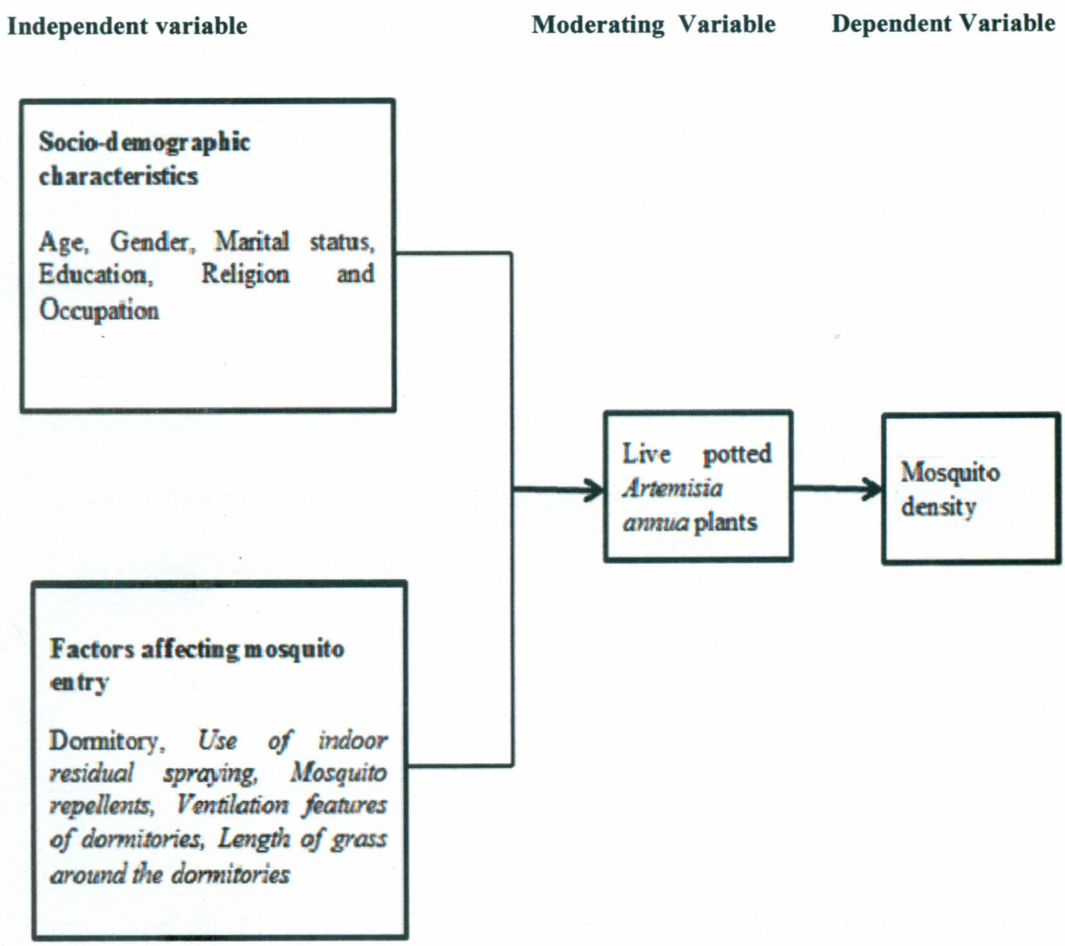


Figure 1.1: Conceptual framework

Source: Modified from Dickens *et al.*, (2013)

CHAPTER TWO: LITERATURE REVIEW

2.1 Global malaria situation

According to WHO (2015) estimates, there were 214 million cases of malaria in 2015 and 438,000 deaths. Malaria incidence fell by 37% globally in 2015; during the same period, malaria mortality rates decreased by 60% between 2000 and 2015. An estimated 6.2 million malaria deaths have been averted globally since 2000. Sub-Saharan Africa continues to carry a disproportionately high share of the global malaria burden. In 2015, the region was home to 89% of malaria cases and 91% of malaria deaths. In 2013, an estimated 437,000 African children died before their fifth birthday due to malaria. Globally, the disease caused an estimated 453,000 under-five deaths in 2013 (WHO, 2014).

Kenya's population is estimated at 43.2 million people, with an estimated population growth of 2.7% per year (World Population Review, 2017). Out of the total population, children under age five account for 16% and children under age 14 account for 42% (KNBS, 2011). Malaria still remains a major public health problem in Kenya and accounts for about 21% of outpatient consultations and 3–5% of hospital admissions (MoH, 2014). Malaria transmission and infection risk in Kenya is determined largely by altitude, rainfall patterns and temperature. About 80% of the Kenyan population is at risk of malaria. Among the at-risk population, 27% (approximately 12 million people) live in areas of epidemic and seasonal malaria transmission where *Plasmodium falciparum* parasite prevalence is usually less than 5%. However, an estimated 28 million people live in endemic areas, and over a quarter (approximately 11 million people) live in areas where parasite prevalence is estimated to be equal to or greater than 20% (Griffin *et al.*, 2013).

2.2 Transmission of malaria

Principal mode of spread of malaria is by the bites of female *Anopheles* mosquitoes which must have been infected through a previous blood meal taken from an infected person (Sinka *et al.*, 2011). Other modes of transmission include: (a) congenital malaria, which involves transfer of parasitized red cells from infected mother to the child either transplacentally or during labor can lead to in malaria in the newborn, congenital malaria is rarely reported and has always been considered to be more frequent in the non-immune population than in the endemic areas (Valencha *et al.*, 2007) and (b) transfusion Malaria, which involves transfusion of blood from infected donors into uninfected recipients. First reported in 1911, transfusion malaria is one of the most common transfusion-transmitted infections today. The risk of acquiring transfusion malaria is very low (1 case per 4 million) in nonendemic countries, whereas in the endemic countries, it is much higher (>50 cases per million donor units) (Chauhan *et al.*, 2009).

Female *Anopheles* mosquitoes transmission intensity depends on factors related to the parasite, the vector, the human host and the environment (Chanda *et al.*, 2012, Lawpoolsri *et al.*, 2010). About 70 different *Anopheles* species are important around the world. All of the important vector species bite at night (Tananchai *et al.*, 2012). *Anopheles* mosquitoes breed in water and each species has its own breeding preference (Chandra *et al.*, 2008); for example some prefer shallow collections of fresh water, such as puddles, rice fields, and hoof prints. Transmission is more intense in places where the mosquito lifespan is longer (so that the parasite has time to complete its development inside the mosquito) and where it prefers to bite humans rather than other animals (Eckhoff, 2011). Transmission also depends on climatic conditions that may affect the number and survival of mosquitoes, such as rainfall patterns, temperature and humidity (Parham *et al.*, 2010).

Studies by Tsefaye *et al,* (2012) suggests that transmission in many places is seasonal, with the peak during and just after the rainy season with malaria epidemics occurring when climate and other conditions favour transmission in areas where have little or no immunity to malaria. Malaria epidemics also occurs when people with low immunity move into areas with intense malaria transmission, for instance to find work, or as refugees (Alli *et al.*, 2010). Human immunity is another important factor, especially among adults in areas of moderate or intense transmission conditions. Partial immunity is developed over years of exposure, and while it never provides complete protection, it does reduce the risk that malaria infection will cause severe disease. For this reason, most malaria deaths in Africa occur in young children, whereas in areas with low transmission and immunity, all age groups are at risk (Doolan *et al.*, 2009).

2.3 Malaria vectors

Although there are about 500 species of Anopheles mosquitoes, 70 are malaria vectors and only about 50 species can transmit malaria through a bite by an infected mosquito female (Hay *et al*, 2010). The *Anopheles gambiae* complex is the dominant malaria vector in Africa. It is comprised of seven species: *A.gambiae sensu stricto* (s.s.), *A. arabiensis*, *A. melas*, *A. merus*, *A. quadriannulatus*, and *A. quadriannulatus* species B (Coluzzi *et al*, 2002). The most important in the complex are *Anopheles gambiae s.s*, and *Anopheles arabiensis*. *Anopheles gambiae s.s.* and *Anopheles arabiensis* are found in 70% of the African continent and *Anopheles funestus* complex is the next most abundant vector in tropical Africa (Weetman *et al*., 2010).

Female *Anopheles* mosquitoes are mammalian ectoparasites that are dependent on a proteinaceous blood meal for egg production. Anopheles mosquitoes also are the exclusive vectors of mammalian malaria (White *et al*., 2011). Four traits are needed to make a highly efficient human malaria vector namely: strong preference for human blood; physiological competence to parasite infection; long life; and a high population density (Sinka *et al*., 2011). Anophelines are found worldwide except in Antarctica. Anophelines that can transmit malaria are found not only in malaria-endemic areas, but also in areas where malaria has been eliminated. The latter areas are thus constantly at risk of re-introduction of the disease (Robert *et al*., 2011).

2.4 Malaria control

Historically, malaria control efforts have led to success in some parts of the world. For example, malaria occurred in the United States (US) and Western Europe until it was eliminated in the US between 1947-1951 due to economic development and public health measures (Cohen *et al.*, 2010). Brazil has made great progress in controlling malaria in most of the country, relying heavily on widespread indoor residual spraying (IRS) and decentralization of finances for malaria control activities were given directly to municipalities, which generated local ownership and facilitated the development of local capacity in malaria control (Griffing *et al.*, 2015). Eritrea made success through scaling up rapid diagnosis and effective treatment of fever cases, environmental management activities, and ITN use. The Eritrean strategy had community involvement in the malaria control activities, including the distribution and re-treatment of ITNs and environmental management (Graves *et al.*, 2008). India put emphasis on full-scale implementation of early diagnosis and prompt treatment of cases at facility and village levels, introduction of ITNs, and alternative vector control methods (including environmental management and use of larvivorous fish) and re-treatment of ITNs (Anvikar *et al.*, 2014). (e) Vietnam success was through the introduction of ITNs and an extensive communications campaign, educating the population on malaria and the importance of using and retreating ITNs (Barat, 2006). Zambia achievements have been attributed to increased advocacy, communication and behaviour changes, efficient partnership coordination including strong community engagement, increased financial resources, and evidence-based deployment of key technical interventions in accordance with the national malaria control programme policy and strategic direction (Chanda *et al.*, 2013).

In 1955 the Global Malaria Eradication Campaign based on indoor and outdoor spraying with Dichloro-diphenyl-trichloroethane (DDT) and use of choloroquine (CQ) medicines was launched focusing on malaria regions of the world but not in most of Sub-Saharan Africa (SSA) countries (Na′ jera *et al.*, 2011). Global eradication efforts were not pursued in SSA countries due to transport problems, water shortages, and diversity in population's habits in Africa, lack of a well-developed public health-oriented infrastructure, the emergence of resistance to DDT and CQ, and lack of political will (Mills *et al.*, 2008).

Since the late 1990s, there has been progress on expanded programmes, funding, technology and advocacy for malaria (Bates *et al.*, 2007). Coordinated malaria control efforts gave rise to initiatives such as Roll Back Malaria (RBM) launched by WHO, the United Nations Children's fund (UNICEF) and the United Nations Development Fund (UNDP) in 1998 (Narasimhan *et al.*, 2003). There has also been increased funding and commitment by governments and other private organizations towards malaria eradication (Snow *et al.*, 2008). Initiatives such as the Multilateral Initiative on Malaria started in 1997 have enhanced scientific and research capacity for malaria (Haque *et al.*, 2015). Other initiatives include the Malaria Vaccine Initiative under the Bill and Melinda Gates Foundation, the World Bank's Malaria Global Strategy and Booster programme (World Bank, 2007), and the US President's Malaria Initiative (President

Malaria Initiative, 2008). Such a wide range of contributors and efforts has led to recent calls for the re-adoption of global eradication of malaria, although this has been presented as a long-term vision rather than a near-term goal (Roberts *et al.*, 2007).

2.5 Malaria control efforts in Kenya

In Kenya, malaria control efforts can be traced back to the colonial period when these were directed from the Central Government. Later policies in the first quarter of the 1900s were a direct response to experience and anticipated epidemics (Abuya, 2008). In 1926, there was an 80% increase in malaria epidemics, which affected the European highlands and accelerated concern for the colonial government to control malaria. Epidemics in the late 1930s and early 1940's intensified the need for a nation-wide policy for malaria control (MoH, 1998). Formal centralized control efforts were established by the creation of the Division of Insect-Borne Diseases in 1947. From the 1970s to the 1990s, vector control was organized through the Division of Vector Borne Diseases (DVBD) and administered at district level under the MoH. As from the 1980s, malaria control began to be linked to community-based health care programmes involving CHWs and Community Owned Resource Persons (CORPS). Programmes linked to primary health care approach were established after the 1978 Alma Ata conference. As a result of this there was significant investment in the Bamako Initiative through a number of NGOs which supported training of CHWs in Kenya (MoH, 1998).

2.6 Mosquito larvae control

Mosquitoes go through four stages in their life cycle: egg, larva, pupa, and imago. The first three stages are aquatic. Adult females lay 50–200 eggs per oviposition. The eggs are quite small ($0.5 \cdot 0.2$ mm^2) and are laid singly and directly on water. Mosquito larvae, commonly called 'wigglers,' live in water for 4–14 days, depending on the water temperature. The pupal stage has is comma-shaped, with the head and thorax merged into a cephalothorax and the abdomen curving around underneath. Pupa is considered as a a resting, non-feeding stage. The imago (adult, sexually mature insect) is the final stage of development. Most of the malaria vector Anopheles species prefer breeding sites that are small, numerous, scattered, and shifting. Each species has its own idiosyncratic preferences (Enayati *et al.*, 2009). Larval control (by chemical and non-chemical means) is relevant and a viable method of vector control only if a high proportion of the breeding sites within the mosquito flight range can be located and are accessible, and the breeding sites are of manageable size.

2.6.1 Biological control

Biological control agents can be used against aquatic mosquito stages, such as larva and pupa, and not for adults. Biological control refers to the introduction or manipulation of organisms to suppress vector populations. A wide range of organisms help to regulate mosquito populations naturally through: predation, parasitism, and competition (Chandra *et al.*, 2008). Larval control can be achieved by environmental management and the use of larvicides or larvivorous fish.

The aim is generally to kill larvae without polluting the environment. Larvivorous fish have been used for over 100 years in mosquito control. For example, *Gambusia affinis* has been widely used to control the immature stages of various vector mosquitoes. Other fish species include Tilapia spp., *Poecilia reticulata*, and *Cyprinidae* (Karunamoorthi, 2011). The benefits of larvivorous fish are that the mosquito larvae cannot build up physiological resistance, and the fish populations are generally self-sustaining and do not depend on the presence of larvae (Walker *et al.*, 2007). Even if some Anopheles larvae survive despite the presence of fish, these emerge as smaller adults (Bond *et al.*, 2005). The fish are relatively inexpensive, and 6 months after stocking the larger fish can be harvested, providing a sustainable source of income and protein for rural farmers (Howard *et al.*, 2007).

Certain types of bacteria, especially *Bacillus thuringiensis* var. *israelensis* and *Bacillus sphaericus*, are highly effective for the control of larvae of mosquitoes (Walker *et al.*, 2007) at very low doses. These bacteria are Gram-positive, soil-dwelling, and commonly used as a biological pesticide; alternatively, the crytoxin may be extracted and used as a pesticide. They also occur naturally in the guts of caterpillars of various types of moth and butterflies, as well as on the dark surfaces of plants. During sporulation, many strains produce crystal proteins (proteinaceous inclusions), called δ-endotoxins, that have insecticidal action (Roh *et al.*, 2007). Upon completion of sporulation, the parent bacterium lyses to release the spore and the inclusions; the toxins exist as inactive protoxins. When the inclusions are ingested by insect larvae, the alkaline pH solubilizes the crystal, and the protoxin is then converted to an active toxin.

The activated toxin binds to insect-specific receptors exposed on the surface of the plasma membrane of mid-gut epithelial cells, and then inserts itself into the membrane to create transmembrane pores that cause cell swelling and lysis, and eventually the death of the insect. The major advantages of biolarvicides are reduced application costs, and safety for the environment, humans, and non-target organisms (Walker *et al.*, 2007).

The application of larvicides may not be an appropriate control strategy in terms of cost-effectiveness, owing to widespread breeding reservoirs; their effectiveness is maintained for only a few days, necessitating frequent and repeated applications at least at the end of every week (Cullon *et al.*, 2009). Finally, biocides are effective against mosquito larvae but cannot control the pupal stage (Marcombe *et al.*, 2011).

Evaluation against larval stages of mosquitoes also have been done using other biological agents including mermithid nematodes such as *Romanomermis culicivorax* (Kamareddine, 2012), microsporidia such as *Nosema algerae* (Raghavendra *et al.*, 2011), and several entomopathogenic fungi (Ruiu *et al.*, 2013). Among these fungi, the oomycete *Lagenidium giganteum* has been proven to be successful for vector control in rice fields (Teng *et al.*, 2005) and has recently been commercialized.

2.6.2 Chemical larviciding

Paris green (cupric acetoarsenite) was used in the successful eradication of *Anopheles gambiae* in Brazil in the 1930s, but it is an arsenical compound and is too toxic to comply with modern standards. Temephos is much safer, but it also kills insect predators of mosquitoes (Forstinus *et al.*, 2015). Recently, numerous insect growth regulators (IGRs) have been synthesized and used for mosquito control. Typically, these substances are mimics of juvenile hormones, and act by binding to juvenile hormone receptors in the immature form of an insect, preventing its survival to the next stage of development. IGRs are target-specific, and almost all IGRs have a good margin of safety for most non-target organisms, including invertebrates, fish, and birds. They are also relatively safe for humans and domestic animals (Nisah *et al.*, 2015).

2.6.3 Source reduction

Anti-larval operations causing the reduction or permanent elimination of mosquito breeding places or sites are recognized as source reduction methods. Source reduction primarily aims to prevent development of aquatic stages of mosquito larvae or reducing breeding source. These methods are atmosphere pleasant, inexpensive in the long run with least conservation and surveillance needs. Source reduction measures undertaken can solve the problem of vector borne diseases in an area but also would bring socio-economic and financial benefits to the communities. Habitat modification includes harbourage alteration and source reduction can be used for mosquito control. Harbourage alteration renders the sites unsuitable for resting of adult mosquitoes and source reduction changes the larval habitat so that mosquito oviposition, hatching and larval development are prevented (Sarwar, 2015).

Source reduction also involves ‘modification’ for permanent or long-lasting physical transformation of land, water, and vegetation, including drainage, filling, land levelling and transformation, and multipurpose reservoir margins. Although these works are usually of a permanent nature, proper operation and adequate maintenance are essential for their effective functioning. Environmental manipulation is process through which recurrent activities are employed in the production of temporary conditions that are unfavourable to the breeding of vectors as a vector control strategy within their ecological zones (Quiñones *et al.*, 2014). Several methods are employed in this procedure, they include: water salinity changes, stream flushing, regulation of the water level in reservoirs, dewatering or flooding of swamps or boggy areas, vegetation removal, shading, and exposure to sunlight.

2.7 Adult mosquito control

Adult vector control helps to reduce the host – vector contact and therefore reducing the transmission. Vector control is a cornerstone in malaria control, owing to the lack of reliable vaccines, the emergence of drug resistance, and unaffordable potent antimalarials. Several methods have been used to control the vector and highlighted below;

2.7.1 Indoor residual spraying (IRS)

IRS is an effective method of vector control, and involves applying a long-lasting insecticide to the inside walls of houses and other structures where people sleep. It is aimed at killing mosquitoes that enter houses when they rest on sprayed surfaces (e.g. walls and ceilings). IRS is widely used in areas of seasonal transmission, including epidemic-prone areas, and increasingly in more malaria-endemic areas. The most common insecticides used are DDT and pyrethroids. IRS is appropriate in epidemiological settings where vectors mainly stay indoors, and in countries where the necessary logistical capabilities can be deployed (Roll Back Malaria, 2011).

In terms of its immediate impact, IRS remains the most powerful vector control intervention for reducing/interrupting malaria transmission. Its use in the last 60 years has played a major role in the elimination of malaria from Southern Europe, Mediterranean, Russia, large parts of Asia, Latin America, and many areas of South Africa (WHO, 2006; Pluess *et al.*, 2010).

Despite its initial widespread use and contribution to the success of malaria eradication and control efforts, the use of IRS has declined in recent years. This is because of the lack of commitment and financing from governments to sustain these efforts over the long term, concerns about insecticide resistance, and fear of its harmful effects on the environment and human health (WHO, 2006). IRS is relatively demanding in terms of planning, logistics, infrastructure, the skills required, and the coverage levels that are needed for a successful operation. Reaching areas with no roads, particularly in the rainy season, may be exceedingly difficult.

The use of IRS is highly effective in achieving rapid reduction in transmission for example during epidemics and other emergencies (Kleinschmidt *et al.*, 2009). Treatment with insecticides greatly improves the effectiveness of the net by acting not only as a barrier but also as a repellence of the contact insecticide.

2.7.2 Ultra-low-volume (ULV) space spraying (fogging)

Space sprays are widely employed for the control of adult mosquito populations worldwide. Space sprays may be applied as thermal fogs, in which kerosene or oil is used as a carrier for insecticides that produce dense fogs of droplets, or as ULV sprays, in which fine droplets of insecticide concentrate are applied. They need to be carefully planned, timed, supervised and evaluated by professional staff if they are to be effective. Equipment should be well maintained (Karunamoorthi, 2011). ULV space spraying is generally not cost-effective as a means of malaria vector control, as the operational costs are high and residual effects are low (Karunamoorthi, 2011). It may, however, be considered for use in exceptional circumstances, such as emergency situations in refugee camps. In this case, if the target mosquito species is exophilic, treatment is applied outdoors wherever the mosquitoes rest. If the vector is endophilic, treatment is applied both indoors and outdoors. Suitable insecticides are applied as cold aerosol sprays or as thermal fogs. Where possible, applications need to coincide with the flying times of the local vector (WHO, 2005).

2.7.3 Impregnated treated nets (ITNs)

ITNs (Impregnated treated nets) have become the most widely used form of vector control. ITNs are more powerful than IRS and are usually less demanding logistically. Moreover, their coverage is easier to sustain. Ordinary ITNs need to be retreated every year or so, but this is not the case with long-lasting insecticide nets, which are designed in such a way that the insecticide lasts for as long as the net. ITNs work in two ways: first, they protect the individual user against biting; and second, they can kill some of the mosquitoes that try to bite. Like IRS, the use of ITNs can produce a community-wide reduction in transmission (Hill *et al.*, 2006). Deltamethrin is the most abundantly used compound, constituting about 60% of global usage, followed by permethrin (22%) (Zaim *et al.*, 2007).

The effectiveness of ITN interventions in reducing the burden of malaria has been amply demonstrated in a variety of epidemiological settings. ITN use by children in several settings has been shown to be very cost-effective (Phillips-Howard *et al.*, 2003). Randomized controlled trials in Kenya, Ghana, The Gambia and Burkina Faso have demonstrated that wide scale use of ITNs can reduce all-cause child mortality by approximately one-fifth; saving an average of six lives for every 1000 children aged 1–59 months protected every year (Karunamoorthi, 2011). In an area of intense perennial transmission in western Kenya, ITN use reduced episodes of clinical malaria and anaemia in infants by >60% (ter Kuller *et al.*, 2003), and reduced by nearly one-third the incidence of sick child visits to peripheral health facilities (Phillips-Howard *et al.*, 2003).

It has been estimated that adequate coverage of malaria in-pregnancy control measures, such as the use of insecticide-treated bed-nets and intermittent preventive treatment in pregnancy, may prevent 3–8% of infant deaths (Guyatt *et al.*, 2001). In the highly malarious western Kenya, studies indicated that women who were protected by ITNs every night in their first four pregnancies delivered approximately 25% fewer babies who were either small for gestational age or born prematurely than women who were not protected by ITNs. Furthermore, the infant who sleeps under the net with the mother will also have marked benefits: reduced malaria exposure, decreased incidence of anaemia, decreased risk of death, and enhanced development (Karunamoorthi, 2011). Where community level ITN coverage is greater than about 60%, a community effect is seen in which non-users receive similar protection to ITN users (Hawley *et al.*, 2003). The use of insecticide-treated bed-nets or curtains substantially reduces the burden of malaria (Karunamoorthi, 2011). The major challenges to the implementation of ITN programmes are Lack of coordination between private and public sectors in the manufacture

and distribution of ITNs, lack of affordable (or free) ITNs for the rural poor. It is a requirement that ITNs are to be re-impregnated every 6-12 months to improve their efficiency pyrethroids resistance, equity and access, seasonal variation of ITN use in the community and low rates of net treatment with insecticides are some of the constraints facing the locals poor resourced communities (WHO, 2005). In malaria endemic areas, the use of ITNS increases child survival substantially in several sites in Africa (Fegan *et al.*, 2007); reduces all-cause mortality among children below five years by approximately 20% (Steketee *et al.*, 2010).

2.7.4 Zooprophylaxis

This strategic placement of livestock sheds or pens is proposed as a component of integrated vector management (IVM) to reduce contact between vectors and human hosts (Donnelly *et al.*, 2015). The purposeful use of livestock as dead-end hosts to divert mosquitoes away from humans is described as active zooprophylaxis. Passive zooprophylaxis occurs where normal presence of livestock draws mosquitoes away from humans (Bogh *et al.*, 2001). Insecticide zooprophylaxis is the treatment of cattle by sponging or dipping them in insecticides in order to pass on a lethal dose of insecticides to the blood-feeding mosquitoes (Mathys, 2010). Zooprophylaxis may be dependent on the relative preference of mosquitoes for hosts. A key example is where the predominant mosquito species prefers human to animal hosts and human hosts are available, and keeping livestock nearby is unlikely to result in zooprophylaxis (Donnelly *et al.*, 2015). Studies have reported zoopotentiation may occur due to proximity or location of livestock relative to humans (Iwashita *et al.*, 2014).

2.7.5 Genetic control of mosquito vector

These are control measures that involve the use of genetic manipulation to modify the ability of the natural vector population to transmit the pathogen (Greenwood *et al.*, 2008). In the last decade, molecular biology has been a source of great hope for the creation of genetically modified microbes (GMMs). Genetic control offers a unique opportunity to control vector-borne diseases, particularly malaria (Amenya *et al.*, 2010). The aim of GMM applications is to suppress or manipulate vector mosquito populations by reducing their ability to transmit diseases (WHO, 2009). Site-specific gene recombination technologies insert the antipathogen effector genes in the integration sites of the genome, making it more effective (Amenya *et al.*, 2010), and as a result, the reared sterile mosquitoes can be released into the environment (WHO, 2009).

Sterile insect technique is a species-specific and environmentally non-polluting methodology that relies mainly on the release of large numbers of sterile insects (Dyck *et al.*, 2005). Mating of released sterile males with native females leads to a decrease in the females' reproductive potential, and ultimately leads to local elimination or suppression of the vector population. Field trials in the 1970s and 1980s demonstrated the effect of sterile insect technique against mosquitoes, even with the technology then available (Klassen *et al.*, 2005). For instance, *Anopheles albimanus* was successfully controlled by the use of chemo-sterilized mosquitoes during a trial in El Salvador (Wilke *et al.*, 2012).

2.7.6 Chemical repellents

Insect repellents are produced in many different chemical forms such as sprays, creams, lotions, aerosols, oils and grease sticks. The type of formulation plays an important role in the efficacy of a substance (Bissinger *et al.*, 2010). Chemical repellents are important in protecting people from blood-feeding insects, ticks, mites, and other arthropods. Consequently, they have been used in the reduction of the transmission of arthropod borne diseases (Abagli *et al.*, 2011). The majority of commercial repellents are prepared by using chemicals such as allethrin, N-N-diethyl-m-toluamide, dimethyl phthalate (DMP), N, N-diethyl phenylacetamide, and N,N-diethyl mendelic acid amide. It is reported that these chemical repellents are not safe for public use (Jayapriya *et al.*, 2015). Synthetic repellents have several limitations, including reduced efficacy owing to sweating, expense, and allergic reactions (Sears *et al.*, 2012). Repellent-treated fabrics might obviate some of these limitations (Karunamoorthi, 2011).

2.7.7 Plant repellents

Most plants contain compounds that they use in preventing attack from phytophagous (plant eating) insects. These chemicals fall into several categories, including repellents, feeding deterrents, toxins, and growth regulators. Although the primary functions of these compounds is to promote defence against phytophagous insects, many are also effective against mosquitoes and other biting Diptera, especially those volatile components released as a consequence of herbivory (Pichersky *et al.*, 2002). Many plant volatiles are deterrent or repellent because they have high vapour toxicity to insects (Lee *et al.*, 2001, Moore *et al.*, 2006).

Repellency of plant material have been exploited for thousands of years by man, most simply by hanging bruised plants in houses, a practice that is still in wide use throughout the developing countries (Moore *et al.*, 2006). Plants have also been used for centuries in the form of crude fumigants where plants were burnt to drive away nuisance mosquitoes and later as oil formulations applied to the skin or clothes. Plant-based repellents are still extensively used in this traditional way by rural communities in the tropics. This is because it is the only means of protection available for the poorest communities (Maia *et al.*, 2011). Most of the communities prefer the natural smelling repellants; they are perceived as safe and a trusted means of protection from mosquito bites (Moore *et al.*, 2007). Many plants have been used as repellents against *Anopheles arabiensis*: *Corymbia citriodora* (Govere *et al.*, 2000; Dugassa *et al.*, 2009), *Eucalyptus camaldulensis* (Dugassa *et al.*, 2009), *Lippia javanica* (Goverre *et al.*, 2000), *Lantana camara* (Seyoum *et al.*, 2003), *Ocimum suave* (Dugassa *et al.*, 2009), *Ocimum* *basilicum* (Dugassa *et al*., 2009), *Cymbopogon citrates* (Karunamoorthi *et al*., 2010), *Pelargonium reniforme* (Goverre et *al*., 2000), and *Azadirachta indica* (Seyoum *et al*., 2002; Maia *et al*., 2011).

Smoke produced by burning of dried leaves of various plants has also been useful in the protection against mosquitoes since the ancient times (Mittal *et al*., 2003). *Suregada zanzibariensis* Verdc. (Angiospermae: Euphobiaceae) repellent volatiles generated by direct burning or by thermal expulsion have been shown to play an important role in protecting households against *Anopheles gambiae* s.s. Giles (Diptera: Culicidae) [Innocent *et al*., 2009]. The major advantages of plant-based traditional repellents are that they are inexpensive, easily available, locally known, and culturally acceptable (Karunamoorthi, 2011).

A large number of plant extracts have been reported to have mosquitocidal or repellent activity against mosquito vectors (Innocent *et al*., 2010; Elumalai *et al*., 2015). Crude extracts from *L. viburnoides* sp *viburnoides* var *kisi* have been demonstrated to have larvicidal effect in various mosquito habitats (Innocent *et al*., 2008). Repellents of plants origin are currently receiving massive attention, owing to their environmental and user-friendly nature (Karunamoorthi *et al*., 2009).

2.8 Public health education on mosquito control

For malaria control, public awareness and health education regarding the habitat and life cycle of the mosquito vector, as well as its physical and cultural control, are important in population management. Health education is proven to be an essential step in any vector control program, which requires that proper information and scientific knowledge to the society on transmitted diseases and their vectors is provided and sustained. The knowledge on the vector life cycle, its ecology and biology should be delivered to help the people live in healthy conditions. The population needs to be sensitized on eco-friendly methods of destruction of vectors breeding sites without the interference of biodiversity (Aziz *et al.*, 2014). Health education strategies should educate the people to break the mosquito life cycle by destroying the possible mosquito breeding sites such as concrete pools, water tanks, aquaria, irrigation ditches and drainages as well as air-conditioners and disposable tires. Moreover, education needs to provide information to the public on matters such as mosquito ecology, vector-borne disease, proper cultural practices for mosquito larva reduction, and mosquito control methods. In addition, health education should be geared towards policy makers in order to gain their support for policy issues on the vector control, provision of information to the public, prevent exposure and lower risk, inform the public of pesticides and benefit; and gaining support for financial resources for mosquito control programme project developments and implementation (Naranjo *et al.*, 2014).

2.9 *Artemisia annua L.* Plant medicinal use

Artemisia annua (Asteraceae) is native to China and commonly referred to as Qinghao (green herb). It has been in use for more than 2,000 years in the management of fever associated with malaria. In antiquity, plants of the genus Artemisia were used to control the pangs of childbirth, to regulate women's menstrual disorders, and as an abortifacient. In 1969, the Chinese screened their medicinal plants in search of an effective antimalarial. A diethyl ether extract of *Artemisia annua* was found to be effective against *Plasmodium sp*., and in 1972 the active ingredient, Artemisinin, was isolated and identified by the Chinese. *Artemisia annua* is currently the main source of artemisinin out of the 400 species of Artemisia. Highest levels of artemisinin are found in the main and branch stems, leaves and flowers of *Artemisia annua* (Ferreira, 2004). Extracts from this plant are currently used for development of the artemisinin based combination therapy ACT (WHO, 2001; Dalrymple, 2010). *Artemisia annua* L. is used for malaria prophylaxis as *Artemisia annua* 'tea' (Meier zu Biesen, 2010; Ogwang *et al*, 2011). *Artemisia annua* 'teas' is also used as a home-based treatment for malaria in situations where conventional treatments are not available (Willcox *et al*., 2011). The hexane extract of leaves of *Artemisia annua* have been used against *Culex quinquefasciatus* and *Aedes aegypti* larvae (Tonk *et al.,* 2006). The essential oil of *Artemisia annua* L. demonstrate a toxic repellent and inhibitory activities against two economically important stored product insects: *Tribolium castaneum* (Herbst) and *Callosobruchus maculatus* (Linnaeus) (Tripathi *et al.,* 2000). They also possess fumigant effect against *Oryzaephilus surinamensis* (Linnaeus) (Coleoptera: Silvanidae), one of the most serious pest insects of stored cereal grains and flour throughout the world (Lü *et al*., 2011). Other studies on repulsive activities of essential oil extract from the leaves of *A. annua* against *Anopheles gambiae* have also been demonstrated (Yimer and Sahu, 2014; Nkuitchou-Chougouo *et al*., 2016).

The age of a plant, and the age of maturation, flowering and senescence, have a significant bearing on its content of secondary metabolites (repellents). The content of artemesinin in Artemisia annua rises in a straight line to reach a peak at 130 days, after which there is a rapid progressive decline to values reached at 50 days. Production of artemesic acid in the same species escalate after 80 days and peaks at 100, well before the peak of artemesinin. The level of arteannuin B shows a sharp rise from day 50 to day 130, reaching a high peak on the same day as artemesinin. Its decline thereafter is not as rapid as the other two compounds (Sangwan *et al.*, 2010, Korkina *et al.*, 2017). Soil, climate, altitude and grower's knowledge can affect the content of artemisinin substantially (Paul *et al.*, 2014).

Based on the findings of its medicinal value and its common practice and use as a hedge in many of the communities, this has created curiosity on the use of the *Artemisia annua* L. as alternative to synthetic insecticide repellents and as a complementary to an ideal ecofriendly approach for malaria vector control programmes. The importance of malaria as a life-threatening disease in many tropical and subtropical areas and the potential medicinal benefits of the *Artemisia annua* against malaria drove the choice of *Artemisia annua* plants for this study.

CHAPTER THREE: MATERIALS AND METHODS

3.1 Research design

The research employed cross-sectional and experimental ecological study designs. Cross-sectional study design was used to determine the level of knowledge amongst students on mosquito control strategies in boarding schools and to establish the mosquito control methods used by the households neighboring the boarding schools. The experimental ecological study design was used to evaluate the effectiveness of two months old potted live *Artemisia annua* plants (from Kenyatta University botanical gardens) in mosquito repellence. The cross-sectional study involved a baseline survey on mosquito density for a month before introduction of *A. annua* followed by three months post planting observations. *A. annua* potted plants were hung around dormitories, and mosquito collection for three months as per WHO, (1975) protocol was done. *A. annua* was introduced around dormitories in one boys and one girls boarding schools to act as treatment dormitories while another one boys and one girls dormitories had no *A. annua* hanged around them to act as controls. Two other schools, one for boys and the other for girls which had no potted *A. annua* hanged around dormitories acted as blanks.

3.2 Variables

For the students and households surveys the independent variables were knowledge and vector control preference, while the dependent variables were age, gender, marital status, education, religion, occupation. For effectiveness of potted *A. annua* plants, the independent variable was the introduction of potted *A. annua* plant, while the dependent variable was mosquito density, while the moderating variable was *A. annua* plant. Confounding variables included size of dormitories, vector control measures practiced in schools, height of grass around the building, gender of students, seasonal changes in mosquito density.

3.3 Location of the study

The study was conducted within Mwea Rice Irrigation Scheme in Kirinyaga County (Figure 3.1). Mwea Rice Irrigation Scheme is approximately 100km North East of Nairobi. It occupies the lower zone of the Kirinyaga County which is wet savannah ecosystem. Mwea Rice Irrigation Scheme has altitude of about 1159 m above sea level and covers an area of approximately 13,640 hectares with more than half of the scheme area under rice irrigation. It has mean annual rainfall of 950mm which occurs in April-May and October-November, with average temperatures being 16°C and 26.5°C; and relative humidity of between 52% to 67%. This climatic condition provides a suitable condition for increased development and survival of mosquitoes in the area.

MRIS has only two girls and two boys boarding secondary schools; Karoti Girls, Wang'uru Girls, Mwea Boys and Tebere Boys.

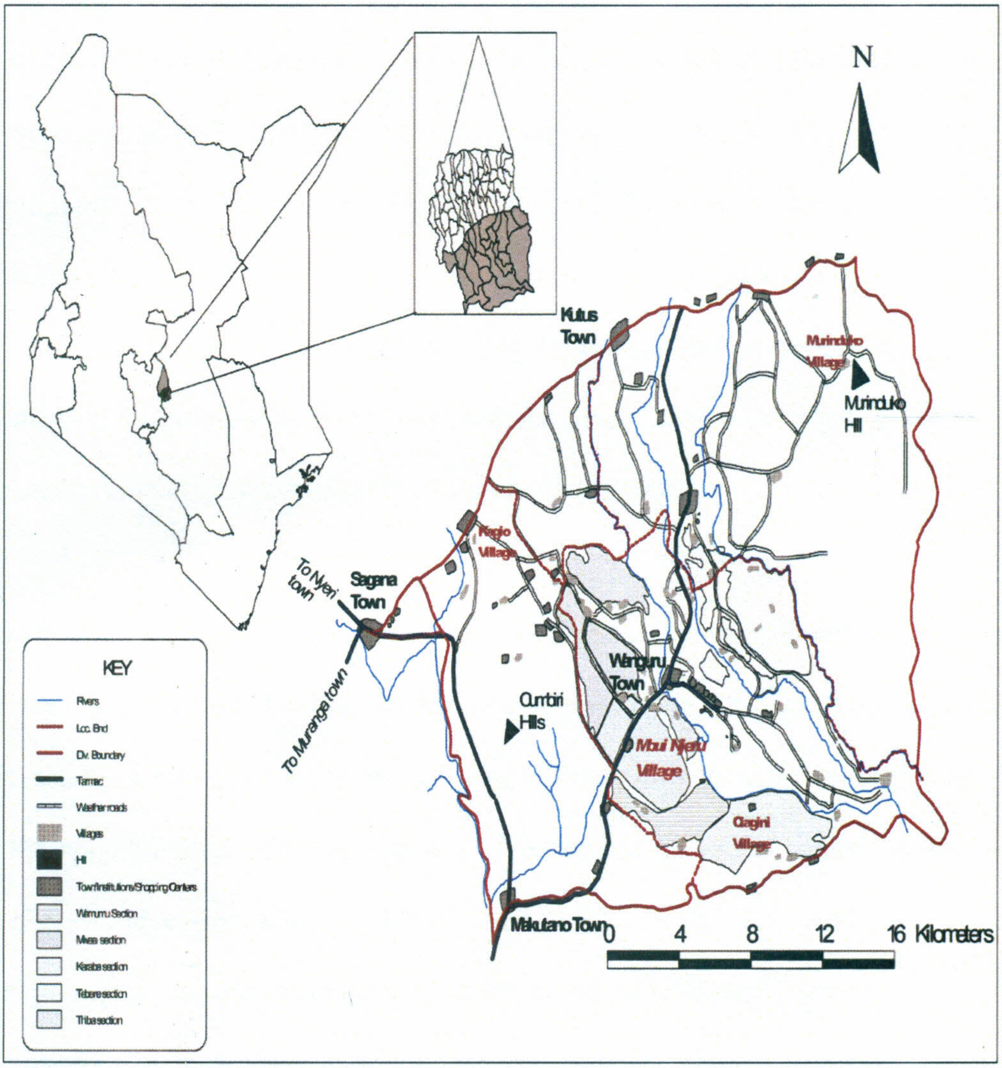


Figure 3.1: Map of Kenya showing Mwea Rice Irrigation Scheme, Kirinyaga County.

3.4 Target and study population

Target population for student survey was high school student while high school students in boarding schools in MRIS were the study population (MRIS has a high student population of 5,910; 54.5% (3,221) and 45.5% (2,689) being boys and girls respectively). The boarding secondary schools have a student population of 3,500; 57.1% (2,000) and 42.9% (1,500) being boys and girls respectively. The target population for community survey was communities residing in irrigation schemes while the study population was communities residing in MRIS.

The knowledge about mosquito control methods and their utilization was conducted among students and the households neighbouring the schools. This was important because the control strategies or their absence may directly affect the mosquito density in the neighborhood boarding schools. Presence of mosquito control strategies in the neighborhood communities would be beneficial for the determination of mosquito densities in an organized institution like the boarding schools.

3.4.3 Inclusion and exclusion criteria

The inclusion criteria for student survey was secondary school students who were boarding at the time of the study, students who gave assent or consent to participate in the study. For the community survey the inclusion criteria was household heads in the community neighbouring schools aged 18 years and above; and household heads who consented to participate in the study. The inclusion criterion for schools was schools with boarding facilities in MRIS. The inclusion criteria villages next to school was villages within 2-5 kilometers. The inclusion criteria for dormitories was dormitories within 50 metres apart. In each school two dormitories were sampled.

The exclusion criteria for student survey were secondary school students who did not assent or consent to participate in the study. For the community neighbouring the schools the exclusion criteria was household heads aged below 18 years and household heads who did not consent to participate in the study. The exclusion criterion for the school was schools in the MRIS without boarding facilities.

3.5.1 Sampling techniques

Convenient sampling was used to select the only four boarding schools in MRIS. Proportionate sampling was based on the student population in each school as shown in Table 3.1. Stratification of classes in each school was done to select the number of students in each class for the study. Simple random sampling was used to select the number of students in each class for the study. Pieces of paper for the number of students to be recruited for the study were written "**Yes**" while the other pieces were blank and put in a box and shuffled. Students in each class who picked "**Yes**" were recruited in the study. Systematic sampling was used to select every **5th** household in the villages for the study.

Table 3.1: Students sampling frame

Secondary School	Student population	Number of students recruited
Karoti Girls SS	1200	116
Wang'uru Girls SS	800	78
Mwea Boys SS	1000	97
Tebere Boys SS	500	49
TOTAL	**3500**	**340**

3.5.2 Sample size determination

The sample size for students was calculated based on Yamane's formula (Yamane, 1967).

$$\mathbf{n} = \frac{\mathbf{N}}{\mathbf{1 + N\,(e)^2}}$$

Where,

n = the sample size

N = the size of population (3,500)

e = the error of 5 percentage points (**0.05**)

$$\mathbf{n} = \frac{\mathbf{3{,}500}}{\mathbf{1 + 3{,}500\,(0.05)^2}} = \mathbf{309}$$

By using Yamane's formula of sample size with an error 5% and with a confidence coefficient of 95% (Yamane, 1967), the calculation from a population of 3,500 students came up with 309 students from all four schools. To account for possible attrition, the number of subjects was increased to 340; hence the study had a sample size of 340 students and 340 household heads.

3.6 Research instrument

Self-administered structured questionnaires (Appendix 1) were used to collect primary data from the students. These questionnaires were preferred for the students because they could express themselves without fear. Interviews using interview schedule (Appendix 2) were used to collect data from systemically selected heads of households of the communities neighbouring the boarding schools.

3.7 Validity, reliability and pre-testing

Validity is the degree to which conclusions on the relationship among variables based on the data are correct. It involves the use of adequate sampling procedures, appropriate statistical tests, and reliable measurement procedures. Reliability the extent to which an experiment yields the same results on repeated trials. Pretesting is testing the effectiveness of a research instrument.

3.7.1 Validity

In order to ensure validity, subjects that were involved in the study had to meet the required characteristics. Proper selection, training and supervision of the research assistants on the research instrument which was used to collect data was done and daily tracking and checking of completed tool. The accuracy was ensured by questions within the questionnaire being precise, language used being understandable and clear to the respondents. All the objectives were addressed by the research instrument.

3.7.2 Reliability

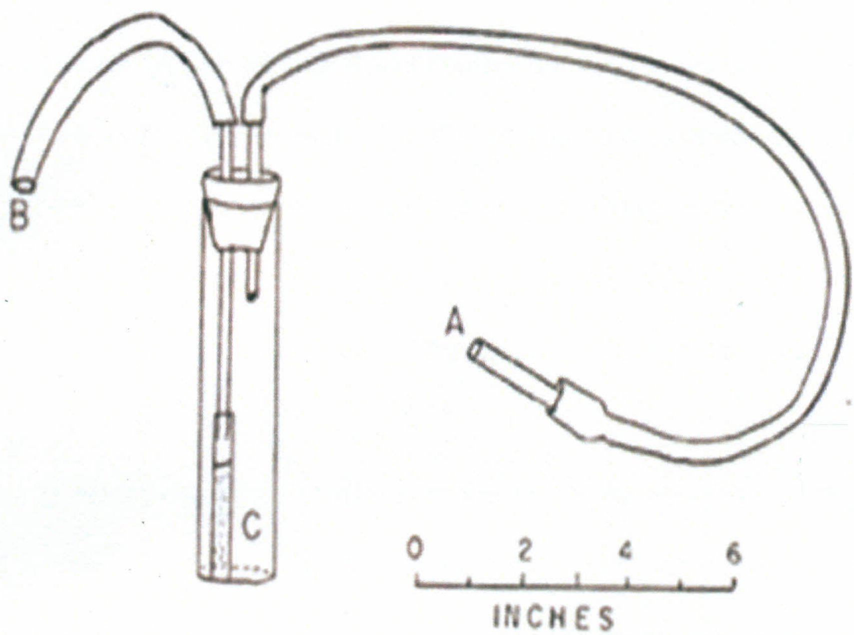
Reliability was done by pre-testing the study instrument before undertaking the study.

3.7.3 Pre-test of the instrument

Pre-testing of the instrument was done at Mutithi secondary school which is 2 ½ km from the study area.

3.8.3 Determination of mosquito density in dormitories

Collections of adult mosquitoes in the (a) baseline treatment and control dormitories and; (b) post planting treatment and control dormitories were conducted biweekly in the study area in the mornings between 8.30 am and 10.30am. The collection of indoor–resting mosquitoes was done using the sucking tube method (Figure 3.2) (World Health Organization, 1975). Baseline survey was done in February 2014 when the *Artemisia annua* L. plant had not been introduced. Mean of mosquitoes resting indoors collected was used to determine the mosquito densities (Masaninga *et al.*, 2012).



A = Mouth piece B = Suction tube C = Collection tube

Figure 3.2: Diagram of sucking tube

3.8.4 Determination of mosquito density after introducing *Artemisia annua* L in dormitories

Seedlings of *Artemisia annua* L. from the Kenyatta University Medicinal Plants Research Garden were obtained and propagated in the selected schools; Mwea Boys Secondary School and Karoti Girls Secondary School. Two months old seedlings were transferred into potting plastic containers (Figure 3.3) and using a wire, the plastic containers were hung next to the doors and windows of the treatment dormitories (Figure 3.4). The control dormitories had plastic containers filled with soil only and hung next to the doors and windows.

Among the dormitories in each school, those were farthest from one another were chosen for this study. The treatment and control dormitories were within a distance of 50 meters from each other (Figure 3.5). The blanks were carried out in another boys (Tebere Secondary School) and girls (Wang'uru Secondary School) boarding schools with the plastic containers filled with soil only hung next to the doors and windows of the selected dormitories. The locations of the schools is as shown in Table 3.2.

The effect of *Artemisia annua* on mosquito density in the dormitories was investigated for three months after being introduced within the doors and windows. Indoor-resting adult mosquito collections in the dormitories were conducted biweekly between March and June on Mondays and Fridays between 8.30 -10.00am in the morning and the number recorded. With the help of flashlight the resting adult mosquitoes in dark places of the dormitories and underneath the beds were spotted and sucked into the tube by squeezing the suction tube. Mean of mosquitoes resting indoors collected was used to determine the mosquito densities (Masaninga *et al*., 2012).

Table 3.2: Aerial distances (Km) between the four schools

Schools	Karoti Girls SS	Mwea Boys SS	Tebere Boys SS	Wang'guru Girls SS
Karoti Girls SS	0.00	5.00	6.00	3.50
Mwea Boys SS	5.00	0.00	7.00	2.50
Tebere Boys SS	6.00	7.00	0.00	4.00
Wang'guru Girls SS	3.50	2.50	4.00	0.00

SS – Secondary school.

Figure 3.3: Some Potted *Artemisia annua* L. plants

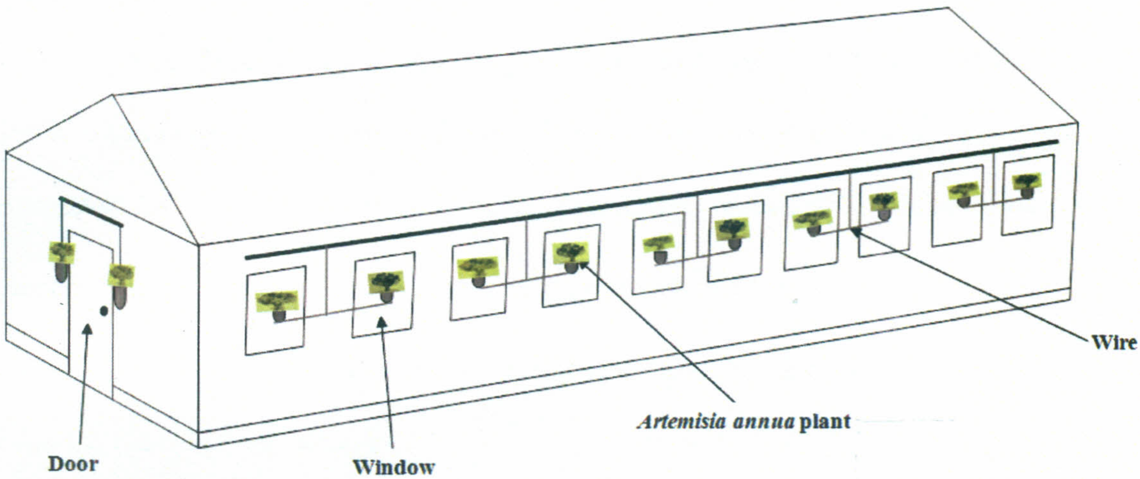


Figure 3.4: Potted *Artemisia annua* L. placed next to the door and windows of treatment dormitory.

Figure 3.5: Treatment and control dormitories in boarding school (distance between the two is approximately 50 metres)

3.9 Data analysis

Data editing was done to check for completeness. Data was coded and entered in statistical computer package, SSPS software. Data cleaning was done to check for error or discrepancies. Data was analyzed using descriptive statistics to obtain means, frequencies and percentages; and Chi square (χ^2) test was done to determine association between the variables. Student unpaired t test was used to test for the difference between the means of the variables.

3.10 Ethical considerations

Permission to conduct the research was granted by Kenyatta University Graduate School, National Commission for Science, Technology and Innovation (NACOSTI) (Appendix 3 and 4) and Ministry of Provincial Administration and Internal Security (Appendix 5) while ethical approval was granted by Kenyatta University Ethics Review Committee (Appendix 6). Informed consent was obtained from the school administrators (Appendix 7) and the selected household members (Appendix 8); and assent from students (Appendix 9) was sought. Participation in the study was fully voluntary. The study participants were not identified by names, but by code numbers to ensure confidentiality.

CHAPTER FOUR: RESULTS

4.1 Socio-demographic profiles of the study population

4.1.1 Socio-demographic profile of the students

Majority of the students selected for this study were in form two to form four. The girls were more than the boys with the boys being 146 (41.2%) and girls being 194 (58.8%). The highest age group of the students was 13 – 15 years (Tables 4.1 and 4.2).

Table 4.1: Number of students per school

Secondary School	Student population	Number of students recruited
Karoti Girls SS	1200	116
Wang'uru Girls SS	800	78
Mwea Boys SS	1000	97
Tebere Boys SS	500	49
TOTAL	**3500**	**340**

Table 4.2: Number and ages of students recruited for the study

Form	Boys (%)	Girls	Age of students	Number (%)
1	26 (7.6%)	24 (7.1%)	< 13yrs	0 (0%)
2	40 (11.8%)	50 (14.7%)	13-15yrs	170 (50%)
3	40 (11.8%)	60 (17.6%)	16-18yrs	136 (40%)
4	40 (11.8%)	60 (17.6%)	>18yrs	34 (10%)

4.1.2 Socio-demographic profile of the household members

Out of a total of 340 household members, 163 (47.9%) were males and 177 (52.1%) were females. Their age range was 20 – 40 years, with the majority 110 (32.4%) were 31 – 40 years. Ninety-eight 98 (28.8%) of the household members were single, 140 (41.2%) were married, while 42 (12.4%) were widowed and 60 (17.6%) were divorced. A majority 172 (50.6%) were Protestants, while 108 (31.8%) were Catholics and 20 (5.8%) were Muslims. Educational level shows that majority of the household members had primary education 108 (31.8%), while 92 (27.1%) had attained secondary school, 80 (23.5%) had attained college education and 60 (17.6%) had attained university education. Majority of the respondents 137 (40.2%) were farmers with 105 (3.9%) being business people and 98 (28.9%) being formally employed (Table 4.3).

Table 4.3: Socio-demographic profile of the household members

Variables	Frequency (n = 340)	Percentage (%)
Gender		
Male	163	47.9
Female	177	52.1
Age in years		
≤ 20	70	20.6
21- 30	85	25.0
31 – 40	110	32.4
≥ 40	75	22.0
Marital status		
Single	98	28.8
Married	140	41.2
Widowed	42	12.4
Divorced	60	17.6
Faith		
Catholic	108	31.8
Protestant	172	50.6
Muslim	20	5.8
Others	40	11.8
Education level		
Primary	108	31.8
Secondary School	92	27.1
College	80	23.5
University	60	17.6
Occupation		
Farming	137	40.2
Business	105	30.9
Employed	98	28.9

The household members were drawn from the villages neighbouring the boarding namely: Karoti, Mwea, Wang'uru and Tebere villages.

4.2 Knowledge amongst students on mosquito control strategies

Out of the 340 students interviewed, 97 % were aware use of mosquito bed net prevents mosquito bite, 59% knew that insecticides use prevents mosquito bite, 40% of the students had knowledge on repellant creams use as prevention method and 24% had knowledge on stagnant water drainage as a control strategy (Table 4.4).

Table 4.4: Level of knowledge amongst students on mosquito control strategies

Control strategy	Response	
	Yes (%)	No (%)
Use of mosquito bed net	330 (97%)	10 (3%)
Use of insecticides	200 (59%)	140 (41%)
Use of repellant creams	136 (40%)	204 (60%)
Draining stagnant water	82 (24%)	258 (76%)

4.3 Mosquito control methods at the household level

4.3.1 Determinants of mosquito bed net use

This study found out that there was association between mosquito bed net use and different socio-demographic profiles. Nearly seventy six per cent (75.8%) of the respondents reported to own at least a bed net during the time of the study. Out of these, 64 % of the nets were treated and the remaining 36 % untreated. Use of treated bed nets was significantly associated with the different socio-demographic profiles; use increases with age (χ^2= 15.94; df = 3 ; p = 0.001), female were more likely to use (χ^2= 7.93; df = 1 ; p = 0.001), married were most likely while divorced were least likely to use (χ^2 = 14.41; df = 3 ; p = 0.001), use was highest amongst college and university graduates (χ^2 = 15.78; df = 3 ; p = 0.001) and use was highest among the employed (χ^2 = 15.73; df = 2; p = 0.001) (Table 4.5).

Table 4.5: Use of mosquito bed net as a control method

Variables		**Mosquito bed net use**	**N = 258 (75.8%)**			
		Untreated	**Treated**	**χ^2 Value Value**	**df**	***p***
Age	≤ 20 yrs	25 (9.7%)	31 (12%)	**15.94**	**3**	**0.001**
	21-30 yrs	27 (10.5%)	35 (13.6%)			
	31-40 yrs	30 (11.6%)	42 (16.3%)			
	≥ 40 yrs	11 (4.3%)	57 (22.1%)			
Gender	Male	111 (43%)	27 (10.5%)	**7.93**	**1**	**< 0.001**
	Female	33 (12.8%)	87 (33.7%)			
Marital Status	Single	51 (19.8%)	22 (8.3%)	**14.41 0.001**	**3**	**<**
	Married	31 (12%)	72 (27.9%)			
	Widowed	11 (4.3%)	27 (10.5%)			
	Divorced	9 (3.5%)	35 (13.6%)			
Education	Primary	47 (18.2%)	19 (7.4%)	**15.78**	**3**	**0.001**
	Secondary School	19 (7.4%)	27 (10.5%)			
	College	35 (13.6%)	43 (16.7%)			
	University	7 (2.7%)	61 (23.6%)			
Faith	Catholic	55 (21.3%)	33 (12.8%)	**7.32**	**3**	**0.067**
	Protestant	81 (31.4%)	69 (26.7%)			
	Muslim	5 (1.9%)	3 (1.2%)			
	Others	11 (4.3%)	1 (0.4%)			
Occupation	Farming	43 (16.7%)	20 (7.8%)	**15.73**	**2**	**< 0.001**
	Business	37 (14.3%)	61 (23.6%)			
	Employed	10 (3.9%)	87 (33.7%)			

4.3.2 Use of firewood and mosquito coils

This study found that there was association between lighting fire wood and mosquito coils as a control method of mosquito and different socio-demographic profiles. Seventeen per cent (17%) of the respondents reported lighting fire wood and use of mosquito coils to control mosquito during the time of the study. Married people were more likely to use firewood and mosquito coils (p = 0.001) (Table 4.6).

Table 4.6: Lighting of firewood and coils as a control method

Variables		Lighting of firewood and coils N = 57 (17%)				
		Yes	**No**	**χ^2Value**	**df**	***p* Value**
Age	≤ 20 yrs	7(12.3%)	3(5.3%)	**0.34**	**3**	**0.952**
	21-30 yrs	9(15.8%)	5(8.8%)			
	31-40 yrs	11(19.3%)	4(7.0%)			
	≥ 40 yrs	13(22.8%)	5(8.8%)			
Gender	Male	15(26.3%)	11(19.3%)	**0.08**	**1**	**0.783**
	Female	19(33.3%)	12(21.1%)			
Marital Status	Single	1(1.8%)	11(19.3%)	**17.56**	**3**	**0.001**
	Married	15(26.3%)	3(5.3%)			
	Widowed	9(15.8%)	4(8.8%)			
	Divorced	8(14.0%)	6(12.3%)			
Education	Primary	3(5.3%)	2(3.5%)	**5.95**	**3**	**0.115**
	Secondary School	9(15.8%)	1(1.8%)			
	College	13(22.8%)	3(5.3%)			
	University	25(43.9%)	1(1.8%)			
Faith	Catholic	7(12.3%)	3(5.3%)	**5.83**	**3**	**0.122**
	Protestant	25(43.9%)	11(19.3%)			
	Muslim	1(1.8%)	4(7.0%)			
	Others	5(8.8%)	1(1.8%)			
Occupation	Farming	19(33.3%)	3(5.3%)	**0.73**	**2**	**0.696**
	Business	15(26.3%)	2(3.5%)			
	Employed	17(29.8%)	1(1.8%)			

4.3.3 Use of insecticides

This study found out that there was association between insecticide use and different socio-demographic profiles. Twenty four per cent (24%) of the respondents reported to have been using insecticide during the time of the study. Use of treated insecticide was significantly associated with the different socio-demographic profiles; age ($\chi^2 = 24.55$; df = 3; p = 0.001), men were more likely to use insecticides ($\chi^2 = 11.76$; df = 1; p = 0.001), married people were more likely to use insecticide ($\chi^2 = 29.13$; df = 3; p = 0.001), as the level of schooling increased use of insecticide increased ($\chi^2 = 10.44$; df = 3; p = 0.016), protestants are more likely to use insecticide ($\chi 2 = 7.80$; df = 3; p = 0.052) and employed were more likely to use insecticide ($\chi^2 = 7.97$; df = 2; p = 0.019) (Table 4.7).

Table 4.7: Use of Insecticide as control method

Variables		Insecticide use: N = 81 (24%)				
		Yes	No	χ^2 Value	df	p Value
Age	≤ 20 yrs	5(6.2%)	9(11.1%)	**24.55**	**3**	**0.001**
	21-30 yrs	3(3.7%)	11(13.5%)			
	31-40 yrs	29(35.8%)	7(8.6%)			
	≥ 40 yrs	15(18.5%)	2(2.5%)			
Gender	Male	13(16.0%)	20(24.7%)	**11.76**	**1**	**0.001**
	Female	37(45.7%)	11(13.6%)			
Marital Status	Single	3(3.7%)	10(12.3%)	**29.13**	**3**	**0.001**
	Married	39(48.1%)	2(2.5%)			
	Widowed	11(13.5%)	3(3.7%)			
	Divorced	8(9.9%)	5(6.2%)			
Education	Primary	5(6.2%)	8(9.9%)	**10.44**	**3**	**0.016**
	Secondary School	11(13.6%)	6(7.4%)			
	College	9(11.1%)	13(16.0%)			
	University	23(28.4%)	6(7.4%)			
Faith	Catholic	19(23.5%)	10(12.3%)	**7.80**	**3**	**0.052**
	Protestant	29(35.8%)	6(7.4%)			
	Muslim	3(3.7%)	5(6.2%)			
	Others	5(6.2%)	4(4.9%)			
Occupation	Farming	11(13.5%)	6(7.4%)	**7.97**	**2**	**0.019**
	Business	25(30.9%)	4(4.9%)			
	Employed	33(40.7%)	2(2.5%)			

4.3.4 Environmental management practices

This study found out that there were several environmental management practices at the household level. Among the reported practices at household level included; disposal of household refuse and waste disposal (13%), draining stagnant water around houses (10%), clearing of vegetations in canals (1%) and clearing of bushes and vegetations around houses (70%) (Figure 4.1).

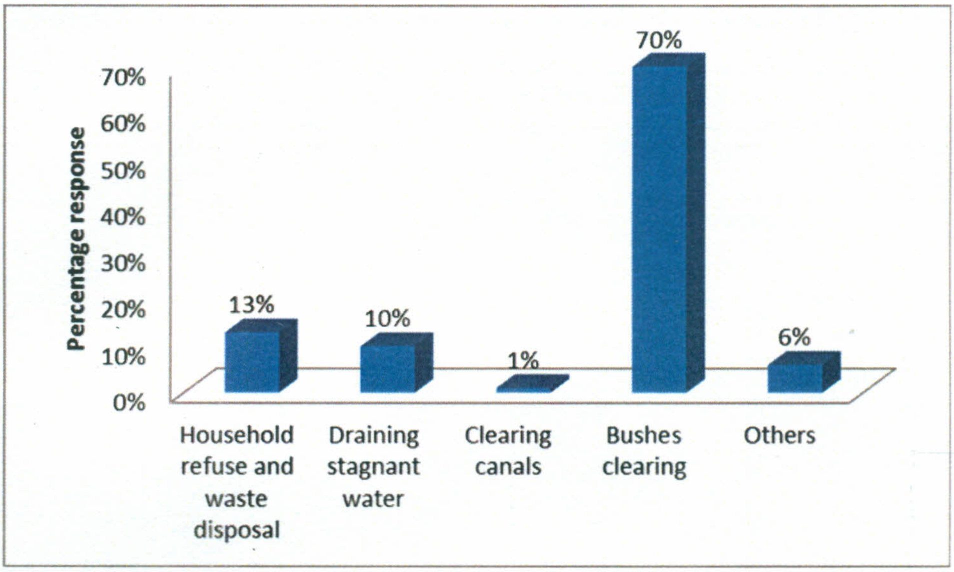


Figure 4.1: Environmental management practices at community level

4.4 Baseline mosquito density in boarding schools in Mwea Rice Irrigation Scheme

This study found out that there were biweekly baseline mosquito densities (expressed in mean ± se) in the month of February in Karoti Girls, Mwea Boys, Wang'uru Girls and Tebere Boys Secondary Schools. In the month of February there was a high mosquito density in the boarding schools: 47.62±2.20, 48.00 ±1.93; (p = 0.904) for Karoti girls treatment and control respectively, 38.50±2.82, 42.50±2.01; (p = 0.268) for Mwea boys treatment and control respectively, and 50.37±2.78, 61.50±3.29; (p = 0.024) for Wang'uru girls and Tebere boys blanks respectively (Table 4.8).

Table 4.8: Baseline mosquito density in dormitories in boarding schools in MRIS

		Number of mosquitoes caught					
		Karoti Girls SS		Mwea Boys SS		Wang'uru Girls SS	Tebere Boys SS
		Dorm. C	Dorm. T	Dorm. C	Dorm. T	Dorm B.	Dorm B.
Week 1	Day 1	35	35	30	44	38	59
	Day 2	43	51	28	52	42	47
Week 2	Day 1	51	52	45	35	56	65
	Day 2	53	50	52	36	50	69
Week 3	Day 1	52	49	35	48	51	60
	Day 2	50	51	36	40	47	73
Week 4	Day 1	45	49	39	43	59	69
	Day 2	52	41	43	42	60	50
	M ± SE	**47.63±2.20**	**48.00±1.93**	**38.50±2.82**	**42.50±2.02**	**50.38±2.78**	**61.50±3.29**
		$p = 0.904$		**$p = 0.268$**		**$p = 0.024$**	

Dorm C- Control, Dorm T - Treatment, Dorm B – Blank, SS- Secondary School, MRIS – Mwea Rice Irrigation Scheme.

The dormitories in blank schools, Wang'uru and Tebere had higher baseline counts than other schools (p-value = 0.024). This could be attributed to the poor environmental conditions around the schools (long grass around the building, water canal short distance from the schools).

4.5 Effect of *Artemisia annua* L. on mosquito density in boarding schools in Mwea Rice Irrigation Scheme.

This study found out that *Artemisia annua* had effect on mosquito density in the boarding schools. In the blank dormitories (Wang'uru Girls SS and Tebere Boys SS), there was a high mean mosquito catches in February to June. Wang'uru girls mean mosquito catches was 50.37±2.78, 56.62±1.74, 63.50±2.22, 58.25±2.53 and 62.12±1.35 for the months of February, March, April, May and June respectively (Figure 4.2, Table 4.14). While Tebere boys mean mosquito catches was 61.50±3.29, 56.25±1.48, 67.37±1.99, 60.12±1.02 and 60.50±1.60 for the months of February, March, April, May and June respectively (Figure 4.3, Table 4.10). In the treatment and control dormitories (Karoti girls secondary school and Mwea boys secondary school) the mean mosquito catches were less compared to the blank dormitories.

The distances between the treatment and cntrol dormitories in Karoti and Mwea were approximately 50 meters. In this study, there was a decrease of mean mosquito catches in both the treatments and controls, Karoti girls control dromitory mean mosquito catches were 47.62±2.20, 36.62±3.88, 38.25±2.63, 37.87±2.53 and 36.00±2.00 for the months of February, March, April, May and June respectively, while the treatment dormitory mean mosquito catches were 48.00±1.93, 19.87±3.28, 15.62±2.25, 20.50±1.47 and 18.12±1.59 for the months of February, March, April, May and June respectively (Figure 4.4, Table 4.11). Mwea boys control dormitory mean mosquito catches were 38.50±2.82, 28.50±2.45, 31.75±2.78, 36.87±2.85 and 44.25±1.11 for the months of February, March, April, May and June respectively, while the treatment dormitory mean mosquito catches were 42.50±2.01, 14.37±1.28, 15.50±1.13, 18.75±1.84 and 21.00±1.33 for the months of February, March, April, May and June respectively (Figure 4.5, Table 4.12).

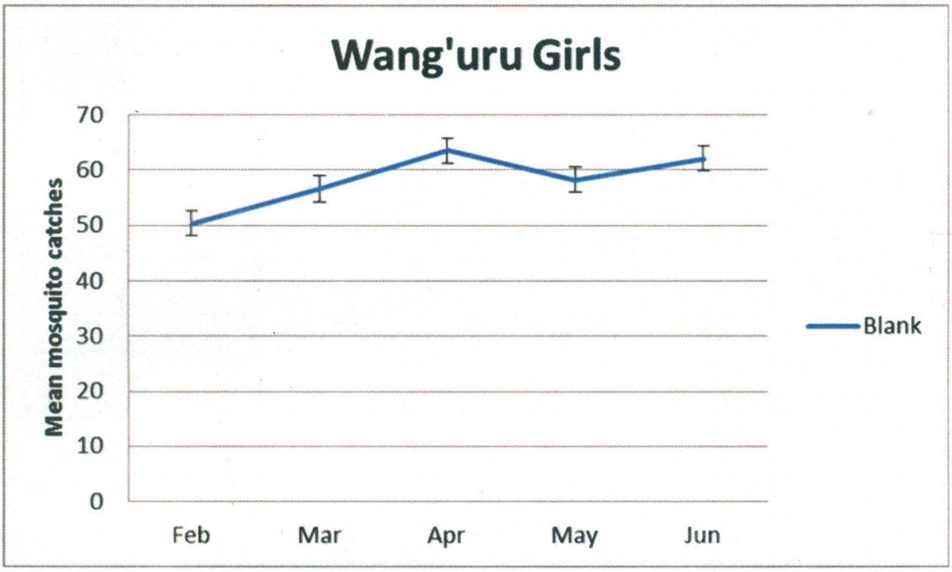


Figure 4.2: Mosquito density (mean ±sem) in a girls blank dormitory

Table 4.9: Mosquito catches in a girls' blank dormitory

WANGURU GIRLS MOSQUITO CATCHES						
		FEBRUARY	MARCH	APRIL	MAY	JUNE
Week 1	**Day 1**	38	55	62	59	69
	Day 2	42	50	65	49	63
Week 2	**Day 1**	56	50	76	54	64
	Day 2	50	59	66	60	58
Week 3	**Day 1**	51	60	64	70	60
	Day 2	47	55	54	65	60
Week 4	**Day 1**	59	61	61	59	58
	Day 2	60	63	60	50	65

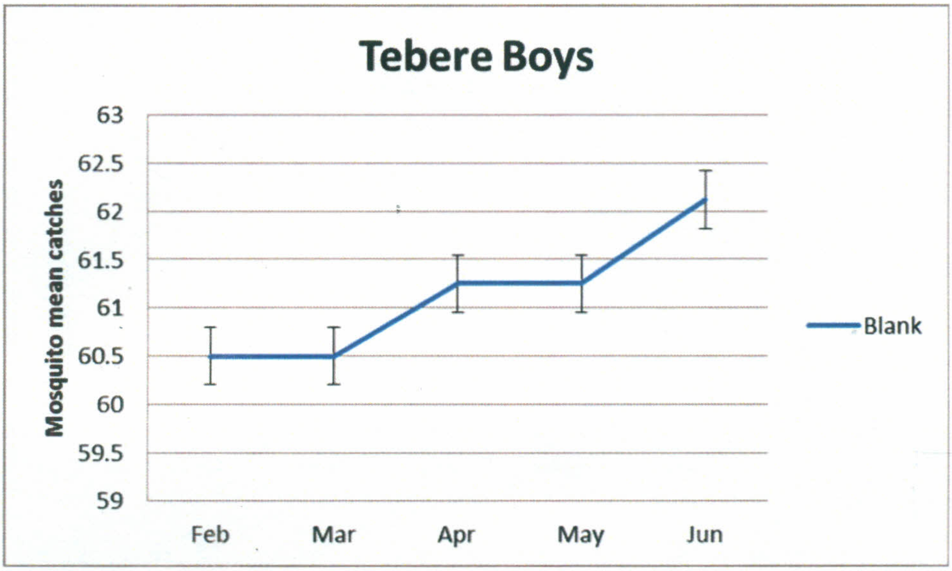


Figure 4.3: Mosquito density (mean ±sem) in a boys blank dormitory

Table 4.10: Mosquito catches in a boys' blank dormitory

		TEBERE BOYS MOSQUITO CATCHES				
		FEBRUARY	**MARCH**	**APRIL**	**MAY**	**JUNE**
Week 1	**Day 1**	59	60	66	58	69
	Day 2	47	57	64	60	59
Week 2	**Day 1**	65	57	63	55	55
	Day 2	69	50	78	62	61
Week 3	**Day 1**	60	54	65	64	57
	Day 2	73	60	74	59	58
Week 4	**Day 1**	69	51	62	60	60
	Day 2	50	61	67	63	65

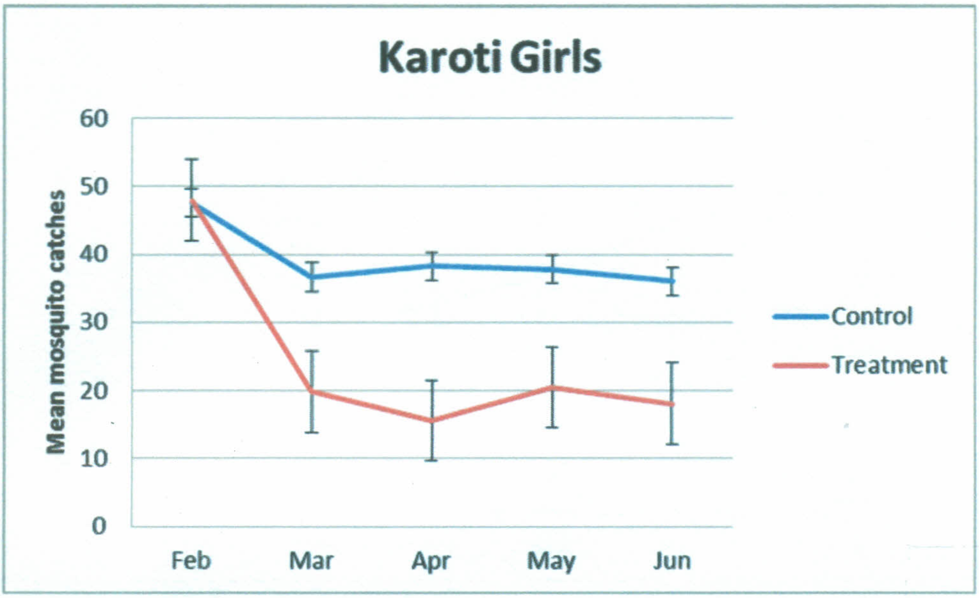


Figure 4.4: Mosquito density (mean ±sem) in a girls control and treatment dormitory

Table 4.11: Mosquito catches after introduction of *Artemisia annua* L. in a girl's control and treatment dormitory

KAROTI GIRLS MOSQUITO CATCHES											
		FEBRUARY		MARCH		APRIL		MAY		JUNE	
		Dorm C	Dorm T	Dorm C	Dorm T	Dorm C	Dorm T	Dorm C	Dorm T	Dorm C	Dorm T
Week 1	Day 1	35	35	24	12	31	13	35	18	38	24
	Day 2	43	51	23	10	51	26	30	19	30	16
Week 2	Day 1	51	52	37	10	30	15	46	21	35	14
	Day 2	53	50	45	26	31	8	35	28	30	17
Week 3	Day 1	52	49	45	30	38	10	30	18	40	22
	Day 2	50	51	36	32	40	11	37	15	30	13
Week 4	Day 1	45	49	54	25	45	20	40	25	40	24
	Day 2	52	47	29	14	40	22	50	20	45	15

Dorm C- Control dormitory, Dorm T – Treatment dormitory

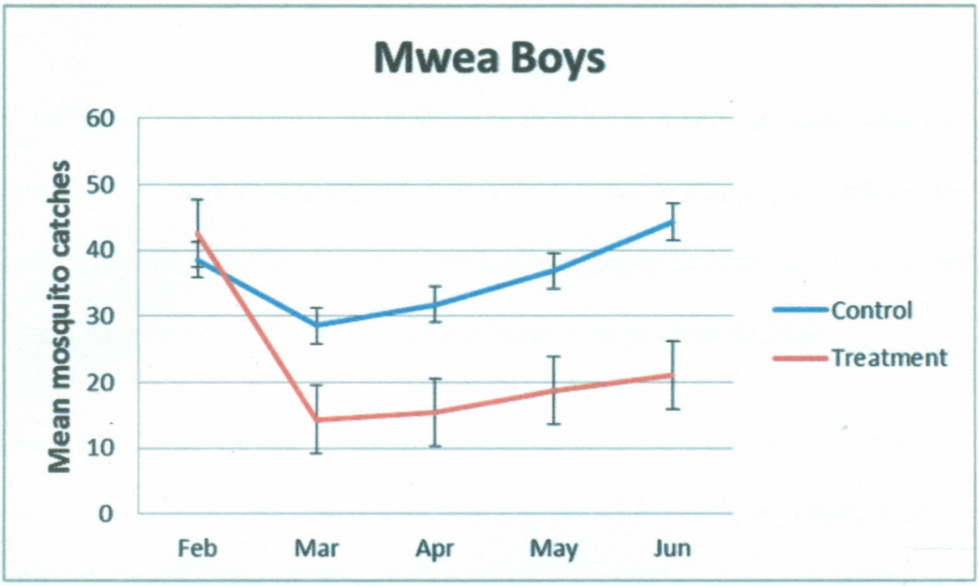


Figure 4.5: Mosquito density (mean ±sem) in a boys control and treatment dormitory

Table 4.12: Mosquito catches after introduction of *Artemisia annua* L. in a boy's control and treatment dormitory

MWEA BOYS MOSQUITO CATCHES											
		FEBRUARY		MARCH		APRIL		MAY		JUNE	
		Dorm C	Dorm T	Dorm C	Dorm T	Dorm C	Dorm T	Dorm C	Dorm T	Dorm C	Dorm T
Week 1	Day 1	30	44	24	15	24	15	40	24	48	22
	Day 2	28	52	21	12	22	18	45	20	43	20
Week 2	Day 1	45	35	21	18	28	13	33	12	40	24
	Day 2	52	36	28	10	32	10	22	16	46	26
Week 3	Day 1	35	48	30	10	38	16	30	12	44	22
	Day 2	36	40	38	16	27	14	39	22	40	18
Week 4	Day 1	39	43	27	14	39	20	46	18	48	20
	Day 2	43	42	39	20	44	18	40	26	45	16

Dorm C- Control dormitory, Dorm T – Treatment dormitory

In the month of February the difference in the the mean mosquito catches in both the control and treatment dormitories was insignificant in Karoti girls with *p*–value (0.776), while the difference in the mean mosquito catches in both the control and treatment dormitories in Mwea boys was insignificant with *p*-value (0.268).

The differences in the mean mosquito catches in both the control and treatment dormitories in Karoti girls were significant with *p*-values 0.005, 0.001, 0.001, and 0.001 for the months of March, April, May and June respectively (Table 4.13), while the differences in the mean mosquito catches in both the control and treatment dormitories in Mwea boys were significant with *p*-values 0.001, 0.001, 0.001, and 0.001 for the months of March, April, May and June respectively (Table 4.14).

Both the decrease in mean mosquito catches and significant differences in both the treatments and controls as compared to the blanks could be attributed to *A.annua* presence.

Table 4.13: Comparison of mean mosquito catches in a girls control and treatment dormitory

KAROTI GIRLS SECONDARY SCHOOL MOSQUITO CATCHES			
Month	**Control**	**Treatment**	**p Value**
	Mean±SEM	**Mean±SEM**	
February	47.62±2.20	48.00±1.93	**0.776**
March	36.62±3.88	19.87±3.28	**0.005**
April	38.25±2.63	15.62±2.25	**0.001**
May	37.87±2.53	20.50±1.47	**0.001**
June	36.00±2.00	18.12±1.59	**0.001**

Table 4.14: Comparison of mean mosquito catches in a boys control and treatment dormitory

MWEA BOYS SECONDARY SCHOOL MOSQUITO CATCHES			
Month	**Control**	**Treatment**	**p Value**
	Mean±SEM	**Mean±SEM**	
February	38.50±2.82	42.50±2.01	**0.268**
March	28.50±2.45	14.37±1.28	**0.001**
April	31.75±2.78	15.50±1.13	**0.001**
May	36.87±2.85	18.75±1.84	**0.001**
June	44.25±1.11	21.00±1.13	**0.001**

4.6 Effect of *Artemisia annua* plant and gender on mean mosquito cathches

This study found out that *Artemisia annua* and gender had effect on mean mosquito cathches. In Wang'uru girls blank dormitory there was a high mean mosquito catches in February to June. Wang'uru girls mean mosquito catches was 50.37±2.78, 56.62±1.74, 63.50±2.22, 58.25±2.53 and 62.12±1.35 for the months of February, March, April, May and June respectively. While Karoti girls treatment dormitory mean mosquito catches were drastically less compared to the blank dormitory in Wang'uru girls February to June. Karoti girls treatment dromitory mean mosquito catches was 48.00±1.93, 19.87±3.28, 15.62±2.25, 20.50±1.47 and 18.12±1.59 for the months of February, March, April, May and June respectively (Figure 4.7, Tables 4.9 and 4.11).

Wang'uru girls blank dormitory mean mosquito catches was high in February to June. Wang'uru girls mean mosquito catches was 50.37±2.78, 56.62±1.74, 63.50±2.22, 58.25±2.53 and 62.12±1.35 for the months of February, March, April, May and June respectively. While Mwea boys treatment dormitory mean mosquito catches were drastically less compared to the blank dormitory in Tebere boys February to June. Mwea boys treatment dormitory mean mosquito catches was 42.50±2.01, 14.37±1.28, 15.50±1.13, 18.75±1.84 and 21.00±1.33 for the months of February, March, April, May and June respectively (Figure 4.9, Tables 4. 9 and 4.12).

In Tebere boys blank dormitory there was a high mean mosquito catches in February to June. Tebere boys mean mosquito catches was 61.50±3.29, 56.25±1.48, 67.37±1.99, 60.12±1.02 and 60.50±1.60 for the months of February, March, April, May and June respectively. While Karoti girls treatment dormitory mean mosquito catches were drastically less compared to the blank dormitory in Tebere boys February to June. Karoti girls mean mosquito catches was 48.00±1.93, 19.87±3.28, 15.62±2.25, 20.50±1.47 and 18.12±1.59 for the months of February, March, April, May and June respectively (Figure 4.6, Tables 4.10 and 4.11).

In Tebere Boys blank dormitory there was a high mean mosquito catches in February to June. Tebere boys mean mosquito catches was 61.50±3.29, 56.25±1.48, 67.37±1.99, 60.12±1.02 and 60.50±1.60 for the months of February, March, April, May and June respectively. While Mwea boys treatment dormitory mean mosquito catches were drastically less compared to the blank dormitory in Tebere boys February to June. Mwea boys treatment dormitory mean mosquito catches was 42.50±2.01, 14.37±1.28, 15.50±1.13, 18.75±1.84 and 21.00±1.33 for the months of February, March, April, May and June respectively (Figure 4.8, Tables 4.10 and 4.12).

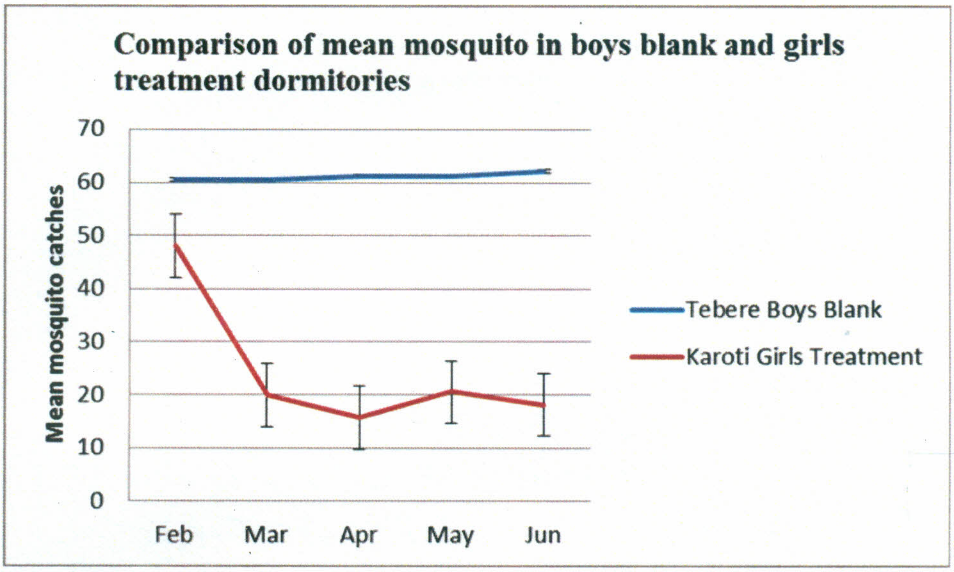


Figure 4.6: Mosquito density (mean ±sem) in girls treatment and boys blank dormitories

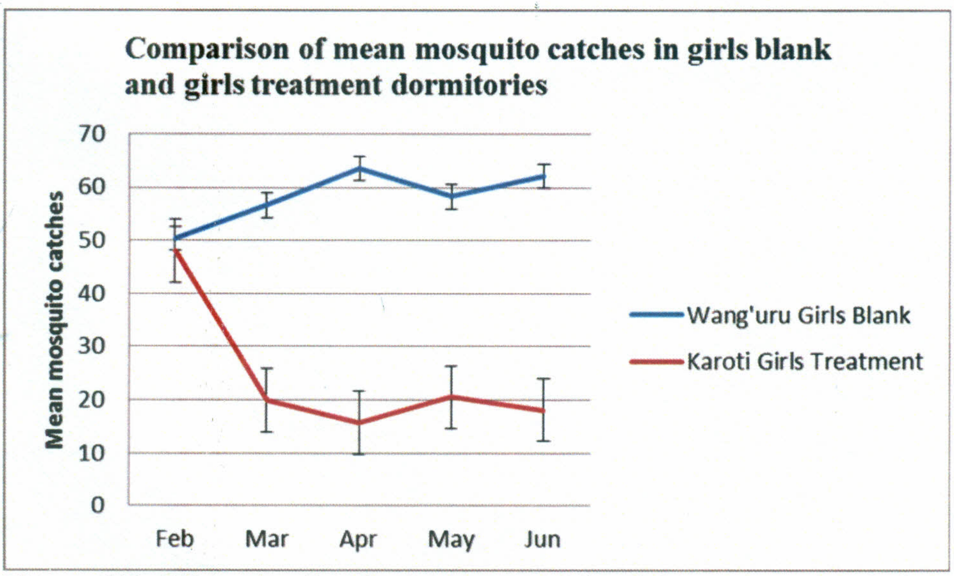


Figure 4.7: Mosquito density (mean ±sem) in girls treatment and girls blank dormitories

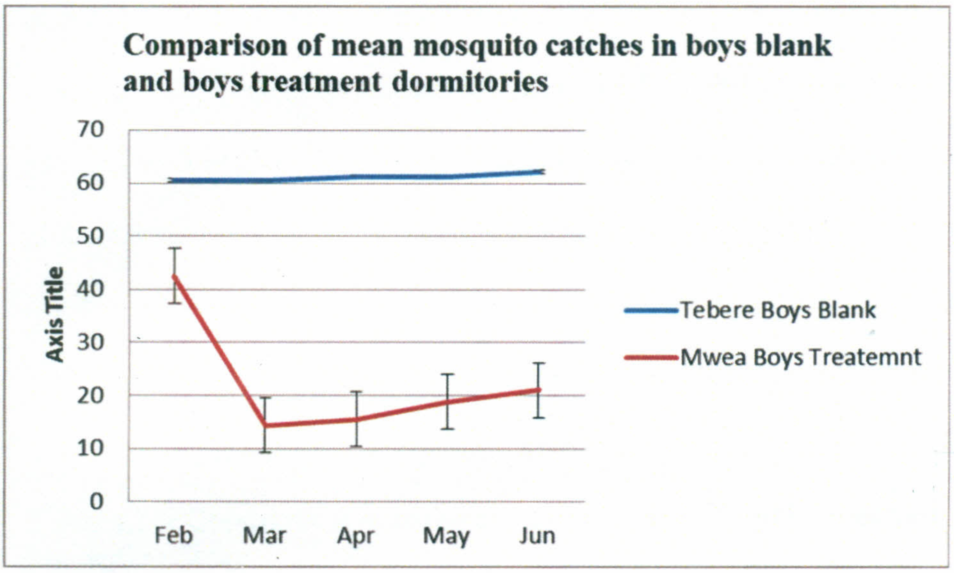


Figure 4.8: Mosquito density (mean ±sem) in boys treatment and boys blank dormitories

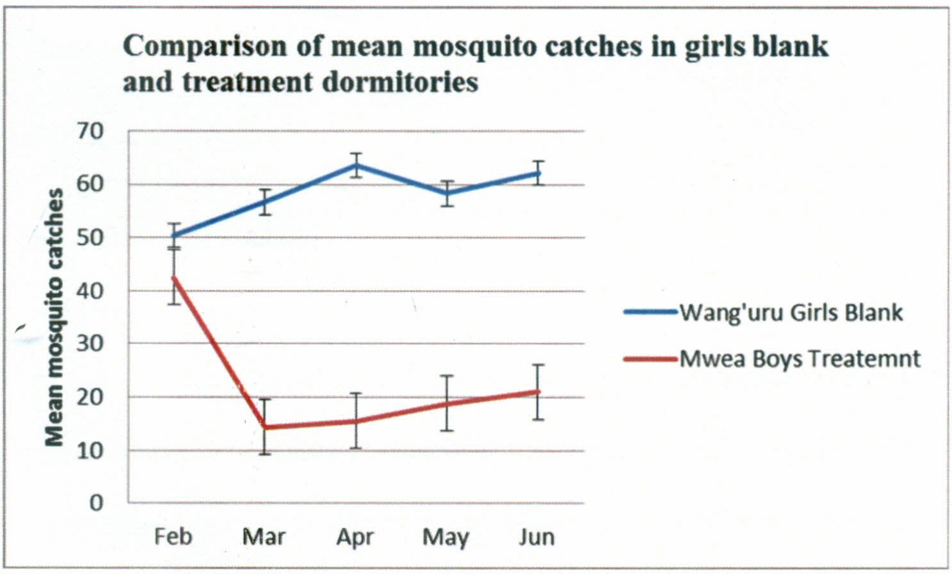


Figure 4.9: Mosquito density (mean ±sem) in boys treatment and girls blank dormitories

In order to find out whether there was any effect on gender of the student dormitories mean mosquito catches comparison was carried out between treatment dormitory of girls school and blank dormitory of girls school. The mean mosquito catches in Karoti girls treatment dormitory were significantly fewer compared to the mean mosquito catches in Wang'uru girls blank dormitory ($p< 0.001$) for the months of February, March, April, May and June respectively (Table 4.15).

Table 4.15: Comparison of mean mosquito catches in girls treatment and girls blank dormitory

Month	Karoti Girls SS treatment dormitory	Wang'uru Girls SS blank dormitory	*p* - Value
	Mean±SEM	Mean±SEM	
February	48.00±1.93	50.37±2.78	**0.001**
March	19.87±3.28	56.62±1.74	**0.001**
April	15.62±2.25	63.50±2.22	**0.001**
May	20.50±1.47	58.25±2.53	**0.001**
June	18.12±1.59	62.12±1.35	**0.001**

SS – Secondary school.

In order to find out whether there was any effect on gender of the student dormitories mean mosquito catches comparison was carried out between treatment dormitory of boys school and blank dormitory of boys school. The mean mosquito catches in Mwea boys treatment dormitory were significantly fewer compared to the mean mosquito catches in Tebere boys blank dormitory ($p< 0.001$) for the months of February, March, April, May and June respectively (Table 4.14).

Table 4.16: Comparison of mean mosquito catches in boys treatment and boys blank dormitory

Month	Mwea Boys SS treatment dormitory	Tebere Boys SS Blank dormitory	*p*-Value
	Mean±SEM	Mean±SEM	
February	42.50±2.01	61.50±3.29	**0.001**
March	14.37±1.28	56.25±1.48	**0.001**
April	15.50±1.13	67.37±1.99	**0.001**
May	18.75±1.84	60.12±1.02	**0.001**
June	21.00±1.13	60.50±1.60	**0.001**

SS – Secondary school.

In order to find out whether there was any effect on gender of the student dormitories mean mosquito catches comparison was carried out between treatment dormitory of boys school and blank dormitory of girls school. The mean mosquito catches in Mwea boys treatment dormitory were significantly fewer compared to the mean mosquito catches in Wang'uru girls blank dormitory ($p < 0.001$) for the months of February, March, April, May and June respectively (Table 4.17).

Table 4.17: Comparison of mean mosquito catches in boys treatment and girls blank dormitory

Month	Mwea Boys SS treatment dormitory	Wang'uru Girls SS blank dormitory	*p* - Value
	Mean±SEM	Mean±SEM	
February	42.50±2.01	50.37±2.78	**0.001**
March	14.37±1.28	56.62±1.74	**0.001**
April	15.50±1.13	63.50±2.22	**0.001**
May	18.75±1.84	58.25±2.53	**0.001**
June	21.00±1.13	62.12±1.35	**0.001**

SS – Secondary school.

In order to find out whether there was any effect on gender of the student dormitories mean mosquito catches comparison was carried out between treatment dormitory of girls school and blank dormitory of boys school. The mean mosquito catches in Karoti girls treatment dormitory were significantly fewer compared to the mean mosquito catches in Tebere boys blank dormitory ($p < 0.001$) for the months of February, March, April, May and June respectively (Table 4.18).

Table 4.18: Comparison of mean mosquito catches in girls treatment and boys blank dormitory

Month	**Karoti Girls SS treatment dormitory**	**Tebere boys SS blank dormitory**	***p* - Value**
	Mean±SEM	**Mean±SEM**	
February	48.00±1.93	61.50±3.29	**0.001**
March	19.87±3.28	56.25±1.48	**0.001**
April	15.62±2.25	67.37±1.99	**0.001**
May	20.50±1.47	60.12±1.02	**0.001**
June	18.12±1.59	60.50±1.60	**0.001**

SS – Secondary school.

The significant fewer mosquitoes in the treatment dormitories as compared to the blank dormitories could be attributed to *A. annua* presence which could have repulsive effect on the mosquito population.

4.7 Effect of *Artemisa annua* plant and distance on mean mosquito catches

This study found out that *Artemisia annua* plant and distance had effect on the mean mosquito catches. Wang'uru girls blank dormitory mean mosquito catches was high in February to June. Wang'uru girls mean mosquito catches was 50.37±2.78, 56.62±1.74, 63.50±2.22, 58.25±2.53 and 62.12±1.35 for the months of February, March, April, May and June respectively. While Karoti girls control dormitory mean mosquito catches were drastically less compared to the blank dormitory in Wang'uru girls February to June. Karoti girls control dromitory mean mosquito catches was 47.62±2.20, 36.62±3.88, 38.25±2.63, 37.87±2.53 and 36.00±2.00 for the months of February, March, April, May and June respectively (Figure 4.11, Tables 4.9 and 4.11).

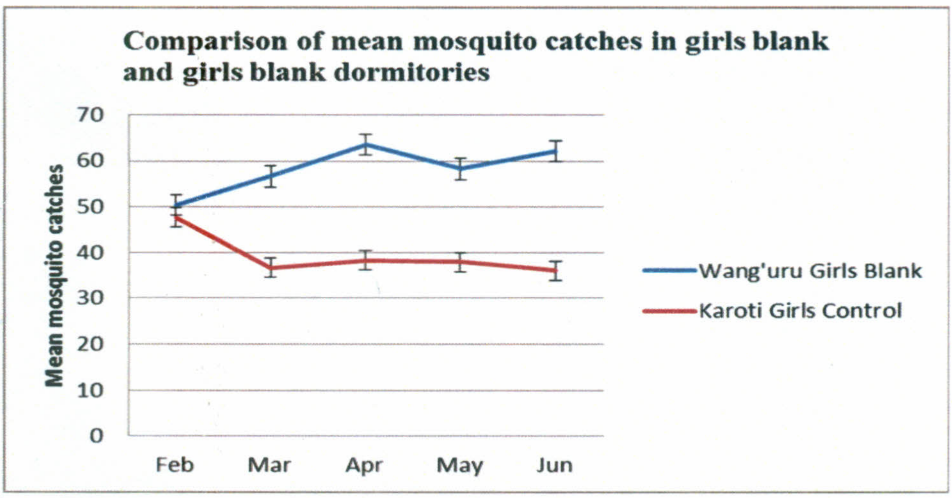


Figure 4.10: Mosquito density (mean ±sem) in girls control and girls blank dormitories

In order to find out whether there was long distance effect on mosquito density, and whether gender was a factor comparison was carried out between control dormitory of girls school and blank dormitory of girls school. The mean mosquito catches in Karoti girls control dormitory were significantly fewer compared to the mean mosquito catches in Wang'uru girls blank dormitory ($p < 0.001$) for the months of February, March, April, May and June respectively (Table 4.19).

Table 4.19: Comparison of mean mosquito catches in girls control and girls blank dormitory

Month	Karoti Girls SS control dormitory	Wang'uru Girls SS blank dormitory	*p* - Value
	Mean±SEM	**Mean±SEM**	
February	47.65±2.20	50.37±2.78	**0.001**
March	36.65±3.88	56.62±1.74	**0.001**
April	38.25±2:63	63.50±2.22	**0.001**
May	37.85±2.53	58.25±2.53	**0.001**
June	36.00±2.00	62.12±1.35	**0.001**

SS = Secondary school.

Wang'uru girls blank dormitory mean mosquito catches was high in February to June. Wang'uru girls mean mosquito catches was 50.37±2.78, 56.62±1.74, 63.50±2.22, 58.25±2.53 and 62.12±1.35 for the months of February, March, April, May and June respectively. While Mwea boys control dormitory mean mosquito catches were drastically less compared to the blank dormitory in Wang'uru girls February to June. Mwea boys control dormitory mean mosquito catches was 38.50±2.82, 28.50±2.45, 31.75±2.78, 36.87±2.85 and 44.25±1.11 for the months of February, March, April, May and June respectively (Figure 13, Tables 4.9 and 4.12).

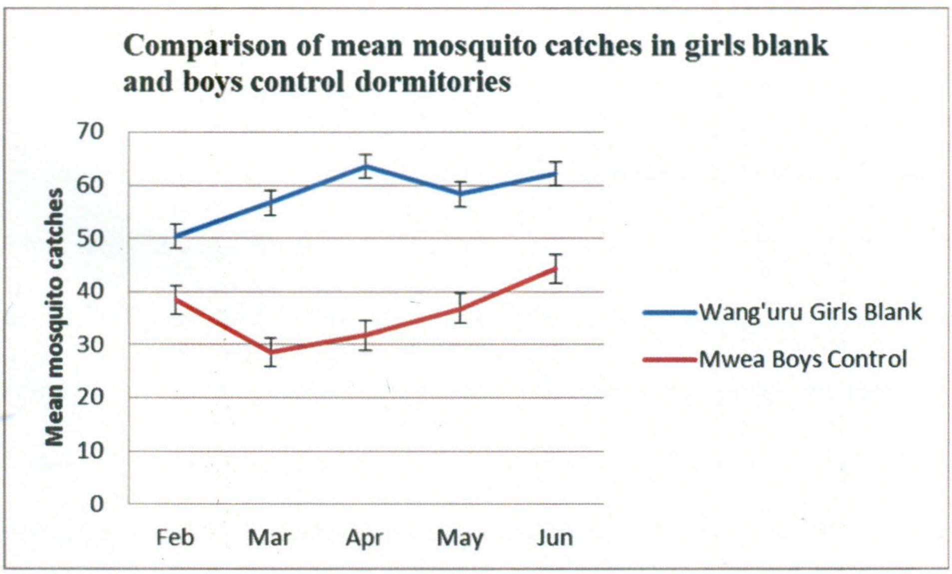


Figure 4.11: Mosquito density (mean ±sem) in boys control and girls blank dormitories

In order to find out whether there was long distance effect on mosquito density, and whether gender was a factor comparison was carried out between control dormitory of boys school and blank dormitory of girls school.

The mean mosquito catches in Mwea boys control dormitory were significantly fewer compared to the mean mosquito catches in Wang'uru girls blank dormitory ($p < 0.001$) for the months of February, March, April, May and June respectively (Table 4.20).

Table 4.20: Comparison of mean mosquito catches in boys control and girls blank dormitory

Month	Mwea Boys SS control dormitory	Wang'uru Girls SS blank dormitory	*p* - Value
	Mean±SEM	**Mean±SEM**	
February	38.50±2.82	50.37±2.78	**0.001**
March	28.50±2.45	56.62±1.74	**0.001**
April	31.75±2.78	63.50±2.22	**0.001**
May	36.87±2.85	58.25±2.53	**0.001**
June	44.25±1.11	62.12±1.35	**0.001**

SS = Secondary school.

In Tebere boys blank dormitory there was a high mean mosquito catches in February to June. Tebere boys mean mosquito catches was 61.50±3.29, 56.25±1.48, 67.37±1.99, 60.12±1.02 and 60.50±1.60 for the months of February, March, April, May and June respectively. While Mwea boys control dormitory mean mosquito catches were drastically less compared to the blank dormitory in Tebere boys February to June. Mwea boys control dormitory mean mosquito catches was 38.50±2.82, 28.50±2.45, 31.75±2.78, 36.87±2.85 and 44.25±1.11 for the months of February, March, April, May and June respectively (Figure 4.12, Tables 4.10 and 4.12).

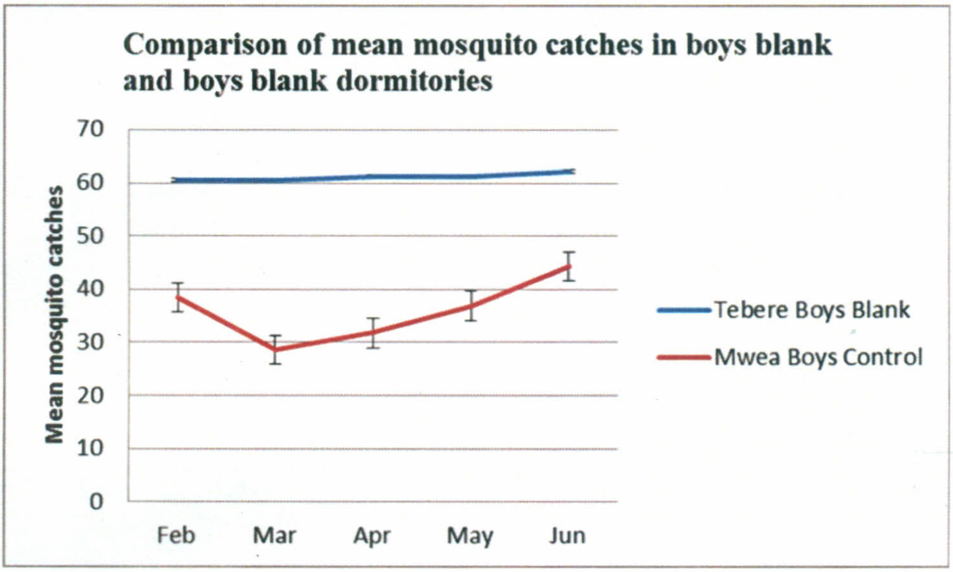


Figure 4.12: Mosquito density (mean ±sem) in boys control and boys blank dormitories

In order to find out whether there was long distance effect on mosquito density, and whether gender was a factor comparison was carried out between control dormitory of boys school and blank dormitory of boys school. The mean mosquito catches in Mwea boys control dormitory were significantly fewer compared to the mean mosquito catches in Tebere boys blank dormitory ($p < 0.001$) for the months of February, March, April, May and June respectively (Table 4.21).

Table 4.21: Comparison of mean mosquito catches in boys control and boys blank dormitory

Month	Mwea Boys SS control dormitory	Tebere Boys SS blank dormitory	*p* - Value
	Mean±SEM	Mean±SEM	
February	38.50±2.82	61.50±3.29	**0.001**
March	28.50±2.45	56.25±1.48	**0.001**
April	31.75±2.78	67.37±1.99	**0.001**
May	36.87±2.85	60.12±1.02	**0.001**
June	44.25±1.11	60.50±1.60	**0.001**

SS = Secondary school.

In Tebere boys blank dormitory there was a high mean mosquito catches in February to June. Tebere boys mean mosquito catches was 61.50±3.29, 56.25±1.48, 67.37±1.99, 60.12±1.02 and 60.50±1.60 for the months of February, March, April, May and June respectively. While Karoti girls control dormitory mean mosquito catches were drastically less compared to the blank dormitory in Tebere boys February to June. Karoti girls control dormitory mean mosquito catches was 47.62±2.20, 36.62±3.88, 38.25±2.63, 37.87±2.53 and 36.00±2.00 for the months of February, March, April, May and June respectively (Figure 4.10, Tables 4.20 and 4.21).

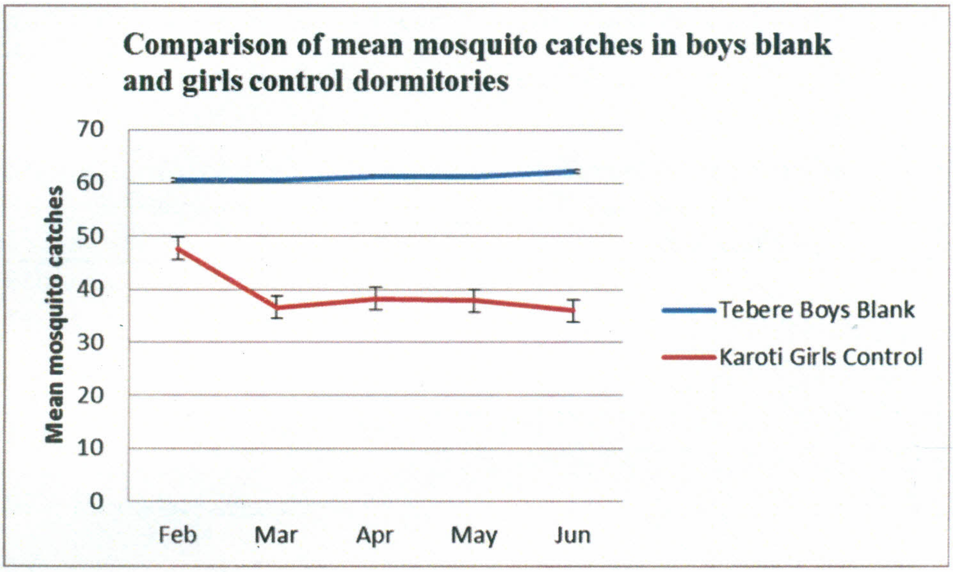


Figure 4.13: Mosquito density (mean ±sem) in girls control and boys blank dormitories

In order to find out whether there was long distance effect on mosquito density, and whether gender was a factor comparison was carried out between control dormitory of girls school and blank dormitory of boys school. The mean mosquito catches in Karoti girls control dormitory were significantly fewer compared to the mean mosquito catches in Tebere boys blank dormitory ($p < 0.001$) for the months of February, March, April, May and June respectively (Table 4.22).

Table 4.22: Comparison of mean mosquito catches in girls control and boys blank dormitory

Month	Karoti Girls SS control dormitory	Tebere Boys SS blank dormitory	*p* - Value
	Mean±SEM	Mean±SEM	
February	47.65±2.20	61.50±3.29	**0.001**
March	36.65±3.88	56.25±1.48	**0.001**
April	38.25±2.63	67.37±1.99	**0.001**
May	37.85±2.53	60.12±1.02	**0.001**
June	36.00±2.00	60.50±1.60	**0.001**

SS = Secondary school

In order to find out whether there was long distance effect on mosquito density, and whether gender was a factor comparison was carried out between blank dormitory of girls school and blank dormitory of boys school. The mean mosquito catches in Wang'uru girls blank dormitory were comparable to the mean mosquito catches in Tebere boys blank dormitory (p = 0.871, 0.213, 0.505 and .0445) for the months of March, April, May and June respectively (Table 4.23).

Table 4.23: Comparison of mean mosquito catches in girls blank and boys blank dormitory

Month	Wang'uru Girls SS blank dormitory	Tebere Boys SS blank dormitory	*p* - Value
	Mean±SEM	Mean±SEM	
February	50.37±2.78	61.50±3.29	**0.024**
March	56.62±1.74	56.25±1.48	**0.871**
April	63.50±2.22	67.37±1.99	**0.213**
May	58.25±2.53	60.12±1.02	**0.505**
June	62.12±1.35	60.50±1.60	**0.445**

SS – Secondary school

The decrease in mean mosquito catches and siginificant differences in both the controls and blanks could be attributed to the presence of *Artemisia annua* semiochemicals in the wide distance locations of the schools. This may be attributed to the long distance the *Artemisia annua* semiochemicals may have travelled in the space between the different schools that may have had repulsive effect on the mosquito population.

CHAPTER FIVE: DISCUSSION, CONCLUSIONS AND RECOMMENDATIONS

5.1 Discussion

5.1.1 Level of knowledge amongst students on mosquito control strategies

The study established that there was high knowledge of some of mosquito control strategies (use of repellant creams and drainage of stagnant water) amongst the students sampled. Students' knowledge on mosquito control strategies is an important determinant of success of a malaria control programs as students can be reached at influential stages of their lives during childhood and adolescence (WHO, 2007), when lifelong behavioural patterns are formed. Students' knowledge can be an effective and efficient way to reach large portions of the population, including young people, families and community members.

High mosquito control strategy knowledge is consistent with the findings of a similar study among high school students in China (Yin *et al.,* 2013) and inconsistent with similar study done among secondary school students in Cape Coast, Ghana (Kudom *et al.*, 2010). Preventing mosquitoes from breeding in standing water could be an effective and relatively inexpensive strategy. Students' knowledge on mosquito breeding sites especially in water could help in controlling the malaria disease by removal of water basins, drain and flush major mosquito breeding sites. This is important in the prevention of malaria by decreasing mosquito breeding sites and prevention of mosquito bites. The results of this study showed that the students had low knowledge on mosquito breeding sites (24%). This finding is consistent with similar study in schools in the municipality of Caxias, State of Maranhão, Brazil (Bezerra, 2011).

This study observed that students were not motivated to use ITN as they claimed that mosquitoes bite even before bed time. A few of them knew different types of mosquitoes though to majority of them all mosquitoes were the same, hence any mosquito bite could result to malaria. Therefore, majority of them did not appreciate how sleeping under ITNs prevented malaria. People could appreciate sleeping under ITNs when they have adequate knowledge on the behaviour and life history of mosquitoes. This finding is in agreement with another study among school children in Kyela District, south western Tanzania (Edson *et al.*, 2007). These findings are also supported by the findings of Kudom *et al.*, (2010) which found lack of motivation to use ITNs. This study was done on secondary school students in Cape Coast, Ghana.

Students' knowledge of vectors' life cycle helps in understanding how the vector transmits malaria. Investigations on vectors control strategies aim at breaking contact between human and vector, hence preventing malaria as well as other mosquito-borne diseases. Understanding the biology and behavior of mosquito, will enable people to be more proactive towards malaria control. Furthermore, the knowledge of the life history of mosquitoes will help understand how various control strategies work against mosquitoes and enable one to choose an appropriate control strategy in specific situations.

5.1.2 Mosquito control methods at household level

The study findings showed that the most frequent occurring diseases at household level were malaria (53%), typhoid fever (29%) and diarrhea (18%). Rice cultivation through irrigation has brought changes in the ecosystem which has affected the farmers' health in addition to creating habitats ideal for the breeding of vectors of diseases such as malaria, typhoid fever and diarrhea (Colwell *et al.*, 2011). This is in addition to changing the epidemiological pattern of malaria from seasonal to perennial, consequently raising the disease incidence in communities with little prior exposure or immunity (Pemberton-Ross *et al.*, 2015). This explains the occurrence of malaria, typhoid fever and diarrhea as the most frequent at household level in MRIS.

The study established that there were several control methods at the community level in Mwea Rice Irrigation Scheme. It was noted that 75.8% of the respondents used mosquito nets, 24% insecticides and 17% light firewood and coils. Environmental management strategies used by the respondents at the house hold level were 70% bush clearing and 10% for draining of the breeding sites. Man-vector contact reduction effectiveness has been reported to be brought by environmental management practices. Braks *et al.*, (2011) study reports that environmental management practices are normally not very effective by themselves and they need to be integrated with other control measures. This is because they do not have immediate effect in reducing the number of biting vectors and may take several days or weeks before reduction in their numbers can be achieved. Environmental management practices covering high proportions of breeding sites within vector flight range and large proportion of community members' active participation have been shown to have significant impact. This finding is consistent with similar studies done in Lhasa and Tibet where 77.2% respondents had applied environmental measures to control mosquito population (Liu *et al.*, 2014). This is inconsistent with similar studies done in Tanzania where community based control strategies did not achieve 70% coverage (Vanek *et al.*, 2006).

The study established that there was significant association between use of treated bed nets and insecticide use; and different socio-demographic profiles including age, gender, marital status, education and occupation indicating regular use of bed nets and insecticides. Affordability of mosquito bed nets in MRIS could be due to distribution of bed nets by the health care facilities to pregnant women and mothers with young children. Also occupation might have led to improved socio-economic enabling the community to afford the bed nets and insecticides. High level of education empowers people knowledge on protective methods against mosquitoes.

Socio-economic based measurement may not be a right measurement of affordability since what appears affordable to outside may not be affordable to many households. Households not well endowed with finances may use other less expensive methods, for example burning wood which eventually may not be effective.

5.1.3 Baseline mosquito densities in boarding schools in Mwea Rice Irrigation Scheme

The objective of this study was to assess the effectiveness of *Artemisia annua* L. on mosquito densities in boarding schools. In order for these to be achieved, *Artemisia annua* L. plants were introduced in school dormitories. Biweekly baseline monitoring for a month assessed the density of mosquito in the dormitories before introducing the plant, from which the effect of *Artemisia annua* L. plant on the mosquito density was compared after introduction for three months. The aim of this study was to provide a time-course basis of quantifying the efficacy of the potted plants in reducing the number of mosquitoes that enter the dormitories.

Effective control of vector-borne parasitic infections through vector management requires information on the abundance of vectors in the targeted areas (Mwangangi *et al.*, 2009). In this study, there were marked weekly variations in the mosquito abundance, with more abundance recorded in Wang'uru and Tebere. Mosquito population density variations are closely linked to rainfall, temperature and environmental factors (Uttah *et al.*, 2009). The pattern observed in this study was similar to those reported in a study done in Dar es Salaam, Tanzania (Fillinger *et al.*, 2008).The rains make more vector breeding sites available, and therefore areas of rain-dependent agriculture precursor ideal aquatic habitats that support high density of diverse mosquito species (Githeko *et al.*, 2001). The other breeding sites such as sunlit pools and forest ponds, and temporary ponds created during the rainy season add to ensure sustenance of higher population density during the rains.

Flood irrigated rice fields in the study area serve as a breeding site for potential vector mosquito species resulting into increased mosquito density and negative impacts on human health. There is evidence for direct relationship between irrigation development and increased malaria transmission (Mwangangi *et al.*, 2010). Studies have demonstrated that rice field and marshland habitats have significant influence on production of mosquito larval populations including vector mosquitoes and hence the disease transmission (Amerasinghe *et al.*, 2014).

5.1.4 Effect of *Artemisia annua* L. on mosquito density in boarding schools in Mwea Rice Irrigation Scheme

The study established that there was decreased mosquito density in the months of March to June when potted live *Artemisia annua* plants were hung on the windows and doors of the dormitories However, the density of the mosquito was higher in the baseline survey (in the month of February when the plant was not hung). Other studies done on other plants have shown similar findings. For instance, findings on use of *Lantana camara* plant in Kagera, Tanzania showed a reduction in mosquito densities when mosquito house entry points were screened by planting the tall and densely foliated repellent plant *Lantana camara* L. around houses (Mng'ong'o *et al.*, 2011).

Mosquitos are sensitive to environmental change; their survival, density and distribution are dramatically influenced by small changes in environmental conditions, such as chemicals produced by plants (Rejmánková *et al.,* 2013; Srivastava *et al.,* 2013). Plant chemicals including repellents, feeding deterrents, toxins, and growth regulators would affect the mosquitos' environmental conditions hence reducing their population density (War *et al.*, 2012).

This study has demonstrated that a potted live intact *Artemisia annua* plant can repel mosquitoes to reduce human –vector contact in a field trial experiment. The results of this study demonstrate *Artemisia annua* has effect on mosquito population density when planted as a whole plant ($p < 0.05$). Similar studies on the plants *Lantana camara, Ocimum americanum* and *Lippia uckambensis* reported they are already being used for hedges around huts in many villages around Lake Victoria because of their citronella-like scent (Seyoum *et al.*, 2002).

Seyoum *et al.*, 2002, reported that in East Africa use of *Ocimum basilicum* (*Lamiaceae*) branches in houses was a traditional way to keep mosquitoes away. In recent years, studies on the plant families *Lamiaceae*, *Nepeta cataria* (catmint oil) as well *Ocimum basilicum* and *Ocimum americanum* extracted oils have provided good protection lasting 2-8 hours depending on the mosquito species and repellent concentration (Tawatsin *et al.*, 2006). *Rosmarinus officinalis* has also exhibited good repellent properties against *Anopheles, Aedes, and Culex* mosquito species (Tawatsin *et al*, 2006). *Vitex trifolia* has excellent efficacy against *Aedes albopictus*, *Anopheles dirus* and *Culex quinquefasciatus*, while it repelled *Aedes aegypti* only for a short interval (Tawatsin *et al.*, 2006).

Several studies conducted on *Myrtaceae*, the plant family of *E. citriodora*. *Eucalyptus*, *Melaleuca leucadendron*, *Melaleuca quinquenervia* and *Psidium guajava* have provided good efficacy against *Culex* mosquitoes and moderate protection against *Anopheles* and *Aedes* (Lupi *et al.,* 2013). Field studies have also shown plant repellent effects on mosquitoes by several plants; *Corymbia citriodora, Eucalyptus spp, E. camaldulensis, L. uckambensis, Lantana camara, Ocimum spp. O. americanum, O. basilicum, Hyptis spp. Hyptis suaveolens, Mentha spp. M. piperata, C. citratus* (Maia *et al.,* 2011).

Mitro *et al*., (2012) study has shown humans, like other mammals may possess individual differences in odors which may convey information on individual identity, sex, age, and motivational state. Studies have shown that individuals possess different degrees of attractiveness to mosquitoes and other blood-sucking insects and these differences have been exploited by haematophagous insects (Rebollar-Téllez, 2005). Wong *et al*., (2011) laboratory studies have shown that *Aedes aegypti* (L.) is preferentially attracted to certain individuals more than others. Also individual differences in sweat production and composition have been shown to play a role in differential attractiveness the anthropophillic mosquitoes to humans. This sweat difference in individuals has been demonstrated to be an important cue for these mosquitoes since the volatile organic compounds (VOCs) emanate from sweat provide an invitation to bite (McMeniman *et al*., 2014).

Men produce more sweat than women during exercise and more sebum is produced per cm^2 male skin, presumably because of differing hormonal levels between the sexes

(Giacomoni *et al.*, 2006). Men have been found to be more attractive to *Aedes aegypti* mosquitoes than women (Lenochova *et al.*, 2008), whereas, *A. gambiae* does not discriminate between human individuals based on gender (Qui *et al.*, 2006). Many haematophagous arthropods use host-characteristic volatile cues to find their animal or human hosts, and some appear to change their behaviour according to the reproductive status of the host (Nordeus *et al.*, 2014). Also *A. aegypti* express varying degrees of attractiveness to women owing to the estrogen content of their urine (Takken *et al.*, 2013). *A. gambiae* are more attracted to pregnant women than to women who were not pregnant (Lindsay *et al.*, 2000).

Evidence for long distance location of semiochemicals by both plants and animals has been well demonstrated. Haematophagous insects using their olfactory mechanisms can locate hidden host within a range of about 23 metres away (Gikonyo *et al.*, 2003; Odalo *et al.*, 2005). Similarly, different species of tsetse flies; *G. morsitans* and *G. pallides*, can positively approach the host by detecting their odour (Gikonyo *et al.*, 2003). Odour produced by monitor lizard attracts *G fuscipes* at a higher frequency compared to the normal trap (Gouteux *et al.*, 1995). Mosquitoes also detect human odour from a distance (Zwiebel *et al.*, 2004). Insect repellency is associated with plant semiochemicals, they can be located within the air around the plant, and can be detected by the strong olfactory senses of insects, such as mosquitoes (Reynolds *et al.*, 2011). Probably, it is possible that mosquitoes detected semiochemicals produced by *A. annua*. This may explain the lower significant ($p < 0.05$) values for the mosquito population density in the treatment groups when compared to the controls and blanks.

5.2 Conclusions

Based on the findings of the study, the following conclusions were reached;

I. Knowledge amongst students on mosquito control strategies by using repellant creams and drainage of stagnant water was low.

II. Mosquito control methods at community level include the use of mosquito nets, mosquito coils, insecticides and environmental management.

III. Baseline mosquito density in boarding schools was moderate.

IV. *Artemisia annua* L. significantly reduced the mosquito population density around the dormitories where it was introduced.

V. Gender of students did not influence *A. annua* mosquito population reduction within the dormitories.

VI. There was long range mosquito density reduction in the control dormitories which were 50 metres away from the treatment dormitories implying long range repellence of mosquitoes.

5.3 Recommendations from this study

I. Public health education to increase knowledge on new mosquito control strategies such as use of repellant creams, draining of stagnant water need to be intensified among the students by education directors

II. Principals of schools should encourage planting of *A.annua* as a hedge in dormitories in their schools in order to reduce mosquito density.

III. Provincial administrators and health care workers should encourage the household members to plant *Artemisia annua* as a hedge so as to reduce mosquito density and thereby reduce malaria.

5.4 Recommendations for further research

The following are areas recommended for further studies;

I. Identification of semiochemicals in *A. annua* L. plant responsible for the reduction of the mosquito densities.

II. Spatial studies on behaviour of mosquitoes with respect to *A. annua* L. plant to establish the effective repellence distance.

III. Laboratory experiments on behaviour of mosquito with respect to *Artemisia annua* L. plant to establish how the plant affects the behaviour.

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APPENDICES

APPENDIX 1: QUESTIONNAIRE FOR STUDENTS ON MALARIA KNOWLEDGE

PART I MALARIA PREVENTION STRATEGIES KNOWLEDGE

Demographic information

1) Sex □ Male □ Female
2) How old are you………………?
3) You are in form?
   - □ Form 1 □ Form 3
   - □ Form 2 □ Form 4

Level of knowledge and malaria prevention strategies

1) Do you know a mosquito is responsible for the transmission of malaria?
   - □Yes
   - □ No
2) Do you know malaria is a life threatening disease?
   - □Yes
   - □ No
3) Have you ever been diagnosed and treated for malaria?
   - □Yes
   - □ No

PART 1I MALARIA PREVENTION STRATEGIES

1)Do you prefer non chemical methods of malaria prevention over chemical methods?
   - □Yes
   - □ No

2)If Yes in question one above which of the following is the reason behind your choice?
   - □ Cost
   - □ Availability
   - □ Convenient to use
   - □ Environmentally friendly

3) Which non chemical methods of malaria prevention do you use?
   - □ Clearing bushes
   - □ Insecticides
   - □ Insecticide treated nets
   - □ Draining stagnant water
   - □ Mosquito repellants creams
   - □ None

   Others specify……………

4) How would you rate your current method of malaria prevention if any?
   - □ Very effective

☐ Effective
☐ Not very effective
☐ Ineffective

PART III MALARIA PREVALENCE

1) Is malaria prevalent in your district?
☐Yes
☐ No

2) Is malaria a significant cause of mortality in your district?
☐ Very significant
☐ Significant
☐ Less than significant ☐ Insignificant

3) Which age group is most affected by malaria?
☐ 0-10 years ☐ Above 51years
☐ 11-21 years
☐ 21-31 years
☐ 41-51 years

APPENDIX 2: QUESTIONNAIRE FOR HOUSEHOLD'S ON MALARIA VECTOR CONTROL

SECTION 1: IDENTIFICATION		
1	Serial number of questionnaire	
2	Name of interviewer	
3	Village code (see below)	
4	Homestead Number	
5	Household number (list provided)	
7	Name of respondent	
8	Date of interview (or Date of visit)	
9	Type of visit (see below)	

Village codes: 1. Mwea 2. Wang'uru 3. Karoti 4. Tebere
Type of Visit: 1. Successful 2. Unavailable 3. Unwilling

SECTION 2: SOCIO-DEMOGRAPIDC PROFILE OF RESPONDENT (Tick Accordingly)

1. Relationship to Head of Household

1	Head	
2	Wife	
3	Husband	
4	Son	
5	Daughter	
6	Relative	
7	Employee	
8	Others (Specify)	

2. Sex of respondent

1	Male	
2	Female	

3. Highest level of education attained

1	Primary school	
2	Secondary school	
3	College	
4	University	

4. Marital status

1	Single	
2	Married	
3	Widowed	
4	Divorced	

5. **Faith**

1	Catholics	

2	Protestants	
3	Muslims	
4	Others (Specify)	

6. Main Occupation

1	Farming	
2	Business	
3	Employed	

SECTION 3: KNOWLEDGE OF MALARIA

7. What are the three most frequently occurring diseases in your household?
   1……………………………………………………..
   2……………………………………………………….
   3……………………………………………………….

8. When was the last time a member in your household had malaria?
   ………………………………………………………………………………………
   ……………………

9. What do you perceive as the causes (s) of malaria in this area? (multiple answers possible)

	Perceived causes	**Tick**
1	Working in the sun	
2	Long rains/Being rained on	
3	Wet and cold condition	
4	Working in rice paddies	
5	Mosquito bite	
6	Eating raw food/mangoes	
7	Evil spirit/demons/witchcraft	
8	Taking dirty /polluted water	
9	From another person with malaria (specify how)	
10	Stagnant water	
11	Dirty home surroundings/Environment	
12	Don't know	
13	Others (Specify)	

10. What are the symptoms and signs of malaria you are aware of? (multiple answers possible)

	Symptoms/ signs	**Tick**
1	Fever	
2	Feeling cold	
3	Headache	
4	Vomiting	
5	Diarrhea	
6	General body weakness	
7	Loss of appetite	
8	Body pain/joint pain	
9	Convulsion	
10	Eyes become yellow	
11	Cough	
12	Don't know	
13	Others (Specify)	

11. Which categories /groups of people do you think are most affected by malaria? (Prompt; multiple answers)

	Category	**Tick**
1	Adults women	
2	Children under five years	
3	All children	
4	Elderly people only	
5	Pregnant women	
6	Adult men	
7	Don't know	
8	Others (Specify)	

SECTION 4: VECTOR BIOLOGY

12. Do mosquitoes cause any trouble to you or your family?

1. Yes ……. 2.No …… (If No go to Question 14)

13. In what ways do they cause you trouble? (Do not prompt)

	Cause you trouble	**Tick**
1	They bite	
2	Their bites are itchy	
3	They carry and cause diseases	
4	They make noise	
5	They finish our blood	
6	Can't sleep at night	

14. In this area where do you think mosquito breeds? (Do not prompt, multiple answers)

	Breeding places	**Tick**
1	In stagnant water (ponds, swarms, hoofprints, wheel ruts etc)	
2	In vegetations outside the house	
3	In rice paddies	
4	In water canals	
5	In animal pens	
6	Rubbish pits/Latrines/Cess pits	
7	In dark places	
8	Don't know	
9	Others (Specify)	

15. Where do mosquitoes' stay/hide before biting? (Multiple responses)

	Places	**Tick**
1	In dark places inside houses	
2	On walls and roofs	
3	In vegetations outside the house	
4	At edges of streams and canals	
5	Don't know	
6	Others (specify)	

SECTION 5: VECTOR CONTROL PRACTICES

16. Are you aware of any method(s) of controlling or protecting you and your family from malaria/mosquito bite?

1. Yes　　2. No

17. What are some of the ways of controlling or preventing your household/family from malarial/Mosquito bite? Indicate how often you apply the methods and rank the order of effectiveness (***Tick accordingly as they are mentioned***).

Codes: **Application**: 1. Daily 2. Weekly 3. Fortnightly 4. Monthly 5. More than monthly
Effectiveness: 1. Effective 2. Not so effective 3. Not effective at all

	Method	**Methods Known to you (*tick*)**	**Methods Currently Applying (*tick*)**	**Application frequency (*prompt*)**	**Effectiveness (*prompt*)**
1	Use of mosquito net-untreated				
2	Use of treated mosquito net				
3	Use of insecticide spray				
4	Taking preventive medicine				
5	Screen windows and doors				
6	Light the fire/ mosquito repellent coils				
7	Apply mosquito repellents to the skin				
8	Use traditional Methods (Specify				
9	**Environmental Management (Household level):**				
	Clearing HH refuse/proper waste disposal				
	Draining/Leveling breeding sites				
	Clearing vegetation in canals				
	Clearing bushes/vegetations around houses				
	Others (Specify)				
10	**Environmental Management (Community level):**				
	Leveling/Draining areas of stagnant water				
	Clearing vegetation in water canals				
	Destroying discarded receptacles				
	Others (Specify)				

18. Please give reasons why you don't apply regularly or at all some of the methods known to you (Mentioned above) for control or self-protection against mosquitoes/malaria.

	Reason for non-use	**Examples of the methods**
1	Not effective	
2	Cannot afford	
3	Don't know how to apply	
4	Don't know how to apply	

5	Hot weather	
6	Denied by my spouse	
7	Not culturally accepted	
8	Side effects e.g. eye irritation, sneezing etc	
9	Don't have time to apply	
10	Others (Specify)	

19. Do you have a Bed net in your household? (Should be asked only if Bed nets have not been mentioned before)

1. Yes 2. No

20. What do you think are the benefits of sleeping under a bed net? (***Multiple answers, don't prompt***)

	Reasons	**Tick**
1	Don't get bitten by Mosquitoes	
2	Don't get malaria	
3	Don't get bothered by other insects	
4	Its warmer	
5	None	
6	Don't know	
7	Others (Specify)	

21. What do you think can be done to improve malaria/mosquito control in this area?

1.
2.
3.
4.
5.
6.

End of the interview

Thank the respondent for his/her participation and spending time with you.

APPENDIX 3: RESEARCH AUTHORIZATION LETTER FROM NACOSTI

REPUBLIC OF KENYA

NATIONAL COUNCIL FOR SCIENCE AND TECHNOLOGY

Telephone: 254-020-2213471, 2241349, 254-020-2673550
Mobile: 0713 788 787 , 0735 404 245
Fax: 254-020-2213215
When replying please quote
secretary@ncst.go.ke

P.O. Box 30623-00100
NAIROBI-KENYA
Website: www.ncst.go.ke

Our Ref: **NCST/RCD/12A/013/35**

Date: **19th April, 2013**

Thomas Mutua Mbulo
Kenyatta University
P.O.Box 43844-00100
Nairobi.

<u>RE: RESEARCH AUTHORIZATION</u>

Following your application dated *2nd April, 2013* for authority to carry out research on "***Effects of Artemisia annua L. As a mosquito repellent in schools in Mwea Rice Schemes, Kirinyaga County, Kenya,***" I am pleased to inform you that you have been authorized to undertake research in **Kirinyaga County** for a period ending **31st December, 2013**.

You are advised to report to **the District Commissioners, the District Education Officers and the District Medical Officers of Health, Kirinyaga County** before embarking on the research project.

On completion of the research, you are expected to submit **two hard copies and one soft copy in pdf** of the research report/thesis to our office.

DR M.K. RUGUTT, PhD, HSC.
<u>DEPUTY COUNCIL SECRETARY</u>

Copy to:

The District Commissioners
The District Education Officers
The District Medical Officers of Health
Kirinyaga County.

APPENDIX 4: RESEARCH PERMIT FROM NACOSTI

PAGE 2

THIS IS TO CERTIFY THAT:
Prof./Dr./Mr./Mrs./Miss/Institution
Thomas Mutua Mbulo
of (Address) Kenyatta University
P.O.Box 43844-00100, Nairobi.
has been permitted to conduct research in

Location
District
Kirinyaga County

on the topic: Effects of Artemisia annua L. As a mosquito repellant in schools in Mwea Rice Schemes, Kirinyaga County, Kenya.

for a period ending: 31st December, 2013.

PAGE 3

Research Permit No. *NCST/RCD/12A/013/35*
Date of issue *19th April, 2013*
Fee received *KSH. 1,000*

Applicant's Signature

Secretary
National Council for Science &Technology

APPENDIX 5: RESEARCH AUTHORIZATION LETTER FROM MINISTRY OF PROVINCIAL ADMINISTRATION AND INTERNAL SECURITY

OFFICE OF THE PRESIDENT

MINISTRY OF PROVINCIAL ADMINISTRATION AND INTERNAL SECURITY.

Telegrams: "DISTRICT" ....
Tel. 0202695022
dcmweaeast@yahoo.com
When replying please quote;

DISTRICT COMMISSIONER
MWEA EAST
P.O. BOX 70
WANGURU

MWEA E. 12/4 VOL.1/36

25th April, 2013

Mr. Thomas Mutua Mbuto
Box 43844
NAIROBI

RE: RESEARCH AUTHORIZATION

Reference is made to your letter ref. NCST/RCD/12A/013/35 of 19th April 2012 from National Council for Science and Technology on the above subject.

This office acknowledge receipt of the same and through this letter you are allowed to carry out your research in the district within the time period set in the letter of research authorization. By copy of this letter, the District Education Officer, District Medical Officer and ass. County commissioner Mwea East Division are request to assist in all your enquiries.

Wish you the very best in your research.

DISTRICT COMMISSIONER
MWEA EAST

S. M. TOWETT
FOR: DISTRICT COMMISSIONER
MWEA EAST

CC
District education Officer
MOH
MWEA EAST

DO – MWEA EAST DIVISION

APPENDIX 6: APPROVAL LETTER FROM KENYATTA UNIVERSITY ETHICS REVIEW COMMITTEE

KENYATTA UNIVERSITY

ETHICS REVIEW COMMITTEE

Fax: 8711242/8711575
Email: kuerc.chairman@ku.ac.ke
kuerc.secretary@ku.ac.ke
Website: www.ku.ac.ke

P. O. Box 43844
Nairobi, 00100
Tel: 8710901/12

Our Ref: KU/R/COMM/51/144

Date: March 19th, 2013

Mbulo Thomas Mutua
School of Public Health
Kenyatta University
Nairobi

Dear Mr. Mbulo,

APPLICATION NUMBER **PKU/096/185** OF 2013 – **'EFFECT OF *Artemisia annua L.* AS A MOSQUITO *REPELLENT IN SCHOOLS IN MWEA RICE SCHEME, KIRINYAGA COUNTY, KENYA'.*** *VERSION 2*

1. **IDENTIFICATION OF PROTOCOL**

The application before the committee is with a research topic, **'Effect of *Artemisia annua L.* as a Mosquito *Repellent in Schools in Mwea Rice Scheme, Kirinyaga County, Kenya'*** *version* 2 dated 6th March 2013.

2. **APPLICANT**

Mbulo Thomas Mutua
School of Public Health
Kenyatta University
Nairobi

3. **SITE**

Mwea Rice Scheme, Kirinyaga County, Kenya

4. **DECISION**

The committee has considered the research protocol in accordance with the Kenyatta University Research Policy (section 7.2.1.3) and the Kenyatta University Ethics Review Committee Guidelines, and is of the view that against the following elements of review,

(i) Scientific design and conduct of study,
(ii) Recruitment of research participant,
(iii) Care and protection of research participants,
(iv) Protection of research participant's confidentiality,
(v) Informed consent process,
(vi) Community considerations.

AND APPROVED that the research may proceed for a period of ONE year from 19th March 2013

5. **ADVICE/CONDITIONS**

i. Progress reports are submitted to the Kenyatta University Ethics Review Committee (KU-ERC) every six months and a full report is submitted at the end of the study.

ii. Serious and unexpected adverse events related to the conduct of the study are reported to this board immediately they occur.

iii. Notify the KU-ERC of any amendments to the protocol.

iv. Submit an electronic copy of the revised proposal to KU-ERC.

When replying, kindly quote the application number above.

If you accept the decision reached and advice and conditions given please sign in the space provided below and return to KU-ERC a copy of the letter.

KENYATTA UNIVERSITY
OFFICE OF THE CHAIRMAN
19 MAR 2013
ETHICS REVIEW COMMITTEE
P.O. BOX 43844-00100 NAIROBI

PROF. NICHOLAS K. GIKONYO
CHAIRMAN: KENYATTA UNIVERSITY – ETHICS REVIEW COMMITTEE

I ....Thomas Mbuto.... accept the advice given and will fulfill the conditions therein.

Signature.................. Dated this day ..26th.. of ....March.... 2013.

cc. Vice-Chancellor
Director: Institute for Research Science and Technology

APPENDIX 7: CONSENT FORM FOR SCHOOL AADMINISTRATORS

Information for Participants

My name is Thomas Mbulo, Box 43844-00100 GPO, Nairobi. I am a student of Kenyatta University. I am conducting a survey on mosquito control strategies entitled **"Effect of *Artemisia annua* L as a Mosquito Repellent in Schools in Mwea Rice Irrigation Scheme, Kirinyaga County"**. The information will be used by the Ministry of Health and Ministry of Education to improve learning environments in boarding schools.

Procedures to be followed

Participation in this study will require your consent as the students' legal tutor to fill in a questionnaire on knowledge of mosquito control strategies and also allow planting of *Artemisia annua* L hedge around dormitories, collection of mosquito species in the dormitories every week.

You have the right to refuse participation in this study. Please remember that participation in the study is voluntary. You may ask questions related to this study at any time. The students may refuse to respond to any questions. You may stop an interview at any time.

Discomfort and Risks

The questions the students will be asked are not on intimate subject and will not be embarrassing or make them uncomfortable.

Benefits

If you participate in this study you will help us learn to improve learning environments in boarding schools. You will also benefit by knowing usage of *Artemisia annua* plant.

Confidentiality

The interviews will be conducted in a private setting within the school. Your name will not be recorded on the questionnaire. The questionnaires will be kept in a locked cabinet for safe keeping at Kenyatta University. Everything will be kept private.

Contacts information

If you have any questions you may contact my supervisors;

Prof. N K Gikonyo, 0722763186; nkgikonyo@gmail.com

Prof. E W Kabiru, 0721998558; ewkabiru@gmail.com

or Chairman, Kenyatta University Ethical Review Committee – email: director-crd@ku.ac.ke

Participants' statement

The above information regarding my participation in this study is clear to me. I have been given a chance to ask questions and my questions have been answered to my satisfaction. My participation in this study is entirely voluntary. I understand that my records will be kept private and that I can leave the study at any time.

Signature or Thumb print of participant ____________________Date__________________

Investigator's statement
I, the undersigned, have explained to the volunteer in a language s/he understands the procedures to be followed in the study and the risks and benefits involved.

Name of Interviewer ____

Interviewers' signature ___________________________ Date _____________

APPENDIX 8: CONSENT FORM FOR HOUSEHOLD HEADS

Information for Participants

My name is Thomas Mbulo, Box 43844-00100 GPO, Nairobi. I am a student of Kenyatta University. I am conducting a survey on mosquito control strategies entitled **"Effect of *Artemisia annua* L as a Mosquito Repellent in Schools in Mwea Rice Irrigation Scheme, Kirinyaga County"**. The information will be used by the Ministry of Health and Ministry of Education to improve leaving environments in the community.

Procedures to be followed

Participation in this study will require your consent to fill in a questionnaire on knowledge of mosquito control methods.

You have the right to refuse participation in this study. Please remember that participation in the study is voluntary. You may ask questions related to this study at any time. The students may refuse to respond to any questions. You may stop an interview at any time.

Discomfort and Risks

The questions the students will be asked are not on intimate subject and will not be embarrassing or make them uncomfortable.

Benefits

If you participate in this study you will help us learn to improve learning environments in boarding schools. You will also benefit by knowing usage of *Artemisia annua* plant.

Confidentiality

The interviews will be conducted in a private setting within the school. Your name will not be recorded on the questionnaire. The questionnaires will be kept in a locked cabinet for safe keeping at Kenyatta University. Everything will be kept private.

Contacts information

If you have any questions you may contact my supervisors;

Prof. N K Gikonyo, 0722763186; nkgikonyo@gmail.com

Prof. E W Kabiru, 0721998558; ewkabiru@gmail.com

or Chairman, Kenyatta University Ethical Review Committee – email: director-crd@ku.ac.ke

Participants' statement

The above information regarding my participation in this study is clear to me. I have been given a chance to ask questions and my questions have been answered to my satisfaction. My participation in this study is entirely voluntary. I understand that my records will be kept private and that I can leave the study at any time.

Signature or Thumb print of participant ________________Date ________________

Investigator's statement

I, the undersigned, have explained to the volunteer in a language s/he understands the procedures to be followed in the study and the risks and benefits involved.

Name of Interviewer ____

Interviewers' signature__________________________ Date ______________

APPENDIX 9: ASSENT FORM FOR STUDENTS

Information for Participants

My name is Thomas Mbulo, Box 43844-00100 GPO, Nairobi. I am a student of Kenyatta University. I am conducting a survey on mosquito control strategies entitled **"Effect of *Artemisia annua* L as a Mosquito Repellent in Schools in Mwea Rice Irrigation Scheme, Kirinyaga County"**. The information will be used by the Ministry of Health and Ministry of Education to improve learning environments in boarding schools.

Procedures to be followed

Participation in this study will require your assent to your Principals' consent as the students' legal tutor to fill in a questionnaire on knowledge of mosquito control strategies

You have the right to refuse participation in this study. Please remember that participation in the study is voluntary. You may ask questions related to this study at any time. The students may refuse to respond to any questions. You may stop an interview at any time.

Discomfort and Risks

The questions the students will be asked are not on intimate subject and will not be embarrassing or make them uncomfortable.

Benefits

If you participate in this study you will help us learn to improve learning environments in boarding schools. You will also benefit by knowing usage of *Artemisia annua* plant.

Confidentiality

The interviews will be conducted in a private setting within the school. Your name will not be recorded on the questionnaire. The questionnaires will be kept in a locked cabinet for safe keeping at Kenyatta University. Everything will be kept private.

Contacts information

If you have any questions you may contact my supervisors;

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Signature or Thumb print of participant ________________Date ________________

Investigator's statement

I, the undersigned, have explained to the volunteer in a language s/he understands the procedures to be followed in the study and the risks and benefits involved.

Name of Interviewer ____

Interviewers' signature ______________________ Date _______________

APPENDIX10: YAMANE FORMULA FOR SAMPLE SIZE DETERMINATION

EXAMPLE: Sample Size Calculation

Yamane's formula:

$$*n = \frac{N}{1 + N(e)^2}$$

Where n = Sample size
N = Population size
e = Level of precision or Sampling of Error
which is ±5%

*Reference: Yamane, Taro. 1967. Statistics, An Introductory Analysis, 2nd Ed. New York: Harper and Row.