OCCURRENCE OF *SCHISTOSOMA MANSONI* AND ITS TRANSMISSION RISKS IN SCHOOL CHILDREN IN SCHISTOSOME NON-ENDEMIC KAGIO AREA IN KIRINYAGA COUNTY, KENYA

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DECLARATION

This is my original work and has not been presented for a degree in any other university or for any other award.

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To the glory of God and,

To my daughter Melody Marcel Mukami and my friend C. K. Njine.

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ABBREVIATIONS AND ACRONYMS

ELISA	Enzyme-Linked Imunnosorbent Assay.
Epg	eggs per gram.
CDC	Center for Disease Control and Prevention.
RIA	Radio Immuno Assay.
WHO	World Health Organization.
CNS	Central Nervous System.
MPHS	Ministry of Public Health and Santation

DEFINITIONS OF OPERATIONAL TERMS

Ascites: Accumulation of fluid in the peritoneal cavity.

Cercarial dermatitis: Short term immune reaction occurring in the skin of humans who have been infected with water-borne Schistosomatidae. Symptoms include itching papules and occur within hours of infections and do not last for more than a week.

Eosinophilia: A condition in which the eosinophil count in the peripheral blood exceed $0.45x10^9$ per litre of blood or 450/µl.

Fibrosis: The formation of excess fibrous connective tissue in an organ or a tissue in reparative or reactive process.

Granuloma: Tiny collection of immune cells known as histiocytes and forms when the immune system attempts to wall off substances it perceives as foreign, for example infectious organism, which the body is unable to eliminate.

Hematuria: Presence of red blood cell (erythrocytes) in urine.

Hepatomegally: Enlargement of the liver due to infections, heart failure, blockage of blood vessels from the liver, chronic liver disease and viral diseases.

Hydronephrorosis: Distension or dilation of the renal pelvis and calyces usually caused by obstruction of free flow of urine in the kidney.

Katayama fever: Acute schistosomiasis that occurs weeks after the initial infection especially by *Schistosoma mansoni* and it is manifested by abdominal pain, cough, diarrhea, eosinophilia and fatigue.

Lymphadenopathy: Disease of the lymph nodes where they become swollen enlarged due to infection, auto-immune disease or malignancy.

Splenomegally: Enlargement of the spleen caused by infectious mononucleosis, portal hypertension and some bacterial infections.

ABSTRACT

Schistosomiasis is a chronic parasitic disease caused by a blood fluke of the genus *Schistosoma*. An estimated 249 million people are infected worldwide in about 78 countries with 85% infections occurring in sub-Saharan Africa. In Kenya, an estimated 5 million people are infected with about 12 million people at risk of infection. Schistosomiasis is a major public health concern due to the morbidities caused. Kagio area borders Mwea irrigation scheme which is an endemic area with a prevalence of 47%. The close proximity of Kagio area to the irrigation scheme offers labour opportunities to the population including school children who seek menial jobs and this poses a threat of transmission of schistosomiasis which is common in the irrigation scheme. The study sort to establish the occurrence of schistosomiasis in school children of ages 8-15 years in the non-endemic Kagio area and the endemic Mwea irrigation scheme and compare the levels of the disease in the two areas. Two schools in Kagio area; Kagio primary and Kang'aru primary schools which are at close proximity to the irrigation scheme and one school, Kandongu primary in the irrigation scheme where the pupils from non-endemic area of Kagio go to work were sampled. The pupils provided early morning stool collected in Elkay specimen cups that were checked for schistosome eggs and other helminthes. Macroscopically, stool was checked for presence of mucus or blood, nature and colour. Microscopically, direct saline and iodine wet mounts were done. Eggs were counted systematically from all fields of the slide and tabulated. On the same stool samples, Kato katz was done and eggs present counted in order to check the intensity of infection. Eggs counted were multiplied by 24 to convert the counts to eggs per gram (epg). The eggs present were then classified as light (1-100 epg), moderate (101-400 epg) or heavy (>400 epg) as an indicator of the intensity of infection. Questionnaires were administered to the participating pupils to collect data on their interaction with the endemic Mwea irrigation scheme. The data collected was analyzed using chi-square statistics at 95% confidence level to establish the relationship between occurrence of shistosomiasis in the non-endemic Kagio area and labour migration to the endemic Mwea irrigation scheme and t-test statistics was done to check the difference in occurrence of schistosomiasis in non-endemic area compared to the endemic area and Pearson correlation was done to check for the relationship between visiting in the irrigation scheme and occurrence of schistosomiasis. From microscopic examination of the stool samples, 7.2% of pupils from non-endemic area of Kagio had *Schistosoma mansoni* eggs in the stool while 22% of pupils sampled from Mwea irrigation scheme had *Schistosoma mansoni* eggs in their stool. The mean number of eggs for the pupils who had light infection was 56 epg and 104 epg for those who had moderate infection. There were no pupils with heavy infestation. Pupils who worked in the farms were more prone to infection than those who did not with 7.9% of those who had the infection working in the rice paddies compared to 4% of infected pupils who did not. Analysis of results indicated that there was a significant relationship between labour migration and occurrence of *Schistosoma mansoni* infection among school children in Kagio area (χ^2=0.2604; df = 1; P = 0.01). There was no significant difference in infection rates between Kagio and Mwea areas (t=5.33, cl=95%, df=1, P=0.118). The findings from this study conclude that *Schistosoma mansoni* was present in both endemic and non-endemic areas and that labour migration is a transmission risk. It is recommended that the policy makers institute programmes that are designed to eliminate or minimize child labour migration from non-endemic to endemic areas as a way of preventing spread of schistosomiasis.

CHAPTER ONE: INTRODUCTION

1.1 Background information

Schistosomiasis is a parasitic acute and chronic disease caused by blood flukes of the genus *Schistosoma.* Three major species of schistosome are associated with human schistosomiasis namely: *Schistosoma mansoni* and *Schistosoma japonicum* that causes intestinal schistosomiasis and *Schistosoma haematobium* causes urinary schistosomiasis. The parasite lives in the mesenteric vessels for *Schistosoma mansoni* and the vesicle plexus of the urinary bladder for *Schisosoma haematobium* in humans. Shistosomiasis is second to malaria as the most devastating tropical disease in the world (WHO, 2010) and remains the most prevalent parasitic infection with significant economic and public health consequences (Chitsulo *et al*., 2000). The two forms of schistosomiasis, intestinal and urinary schistosomiasis, occur in Africa. Worldwide, intestinal schistosomiasis occurs in 52 countries, most of which are in Africa. Species causing Asian intestinal schistosomiasis include *Schistosoma mekongi* that occurs in several districts of Cambodia and the Lao People's Democratic Republic and *Schistosoma japonicum* that occurs in China, Indonesia and the Philippines. Urinary schistomiasis is caused by *Schistosoma haematobium* and occurs in Africa and the Middle East (John, 2008).

The prevalence and intensity of *Schistosoma* infections in humans follow a characteristic pattern of variation with age, being generally low in young children up to 10 years of age but rises to a peak during the second decade of life (10-14 years) due to complex immune mechanisms that lead to slow acquisition of immune resistance. The prevalence declines to low

levels among the older individuals though re-infection can occur at the same rate among individuals of different age groups (WHO, 2010).

Schistosomiasis occurs worldwide and is responsible for morbity and mortality. An estimated 249 million people are infected worldwide with both urinary and intestinal schistosomiasis in about 78 countries, with 85% infections occurring in sub-Saharan Africa (WHO, 2014). Over 120 million people show symptoms to schistosomiasis and 700 million people are at risk of infection annually (WHO, 2011). Mortality rates vary with different schistosome infections but it is generally low, for example, 2.4 of 100,000 (0.0024%) die each year from infection with *Schistosoma mansoni* (WHO, 2010). However, acute schistosomiasis caused by egg migration through tissue and the host immune responses to the eggs is associated with mortality rate of up to 25% in some cases (WHO, 2010).

Both intestinal and urinary schistossomiasis are prevalent in tropical and sub-tropical areas, especially in communities with poor access to safe drinking water and poor sanitation; all races appear to be equally susceptible if exposed to infested fresh water (Sturrock, 1986; WHO, 2014).

In Kenya, an estimated 5 million people are infected and about 12 million are at risk of infection (Kariuki, 2011). Schistosomiasis can be controlled through targeted treatment of high risk groups using praziquantel combined with public education (WHO, 2012). Chemotherapy is considered a cost effective way of schistosomiasis control as it dramatically reduces prevalence, intensity of infection and morbidity and reduces the volume of schistome eggs

available for environmental contamination. The drug is effective against all forms of schistosomiasis and has successfully been used for the past 20 years to control schistosomiasis in Brazil, China, Egypt, Morocco and Saudi Arabia (WHO, 2012). The target groups for control include school children and adults in endemic areas whose activities put them at risk. Persons at risk activities includes fishermen, farmers in irrigation farming and sugarcane farming, irrigation workers and persons conducting water related activities like drawing water, laundry, bathing and closing rivers (CDC, 2013).

However, available data show that only 14% of people requiring treatment were treated by the year 2011 (WHO, 2012). WHO implemented treatment based control programme for schistosomiasis covering 33.5 million in 2010. This was an increase from 12.4 million treated in 2006 (WHO, 2012). The extent of spread of schistosomiasis to populations in non-endemic areas has not been studied despite the potential transmission risks from interactive activities in endemic areas. The present study was intended to establish occurrence of schistosomiasis in school children aged 8-15 years in non-endemic area of Kagio, which borders an endemic area, Mwea irrigation scheme located in Kirinyaga County, Kenya. Inadequate hygiene and contact with infected water make children more vulnerable to infection hence the age group of 8-15 years who are pupils of class 4 to class 8 was selected for the study.

1.2 Statement of the problem

Schistosomiasis is a major public health concern due to the morbidity caused by the disease especially in developing nations, mainly in sub-Sahara Africa where 85% of infections occurs. Tropical climate and water bodies in Kenya especially Mwea irrigation scheme offer a conducive environment for snail intermediate hosts and egg development. Kagio area borders Mwea rice irrigation scheme; where prevalence level of 47% was reported in Mwea (Kihara and Muhoho, 2007). The close proximity to the endemic area poses a threat of transmission of the disease to persons coming into contact with the contaminated water in Mwea irrigation scheme. Mwea irrigation scheme covers a wide settlement area and it is associated with a wide spread of *Schistosoma mansoni* infection.

Kagio has no rice farming activities due to the regional topography therefore labour migration is common to Mwea irrigation paddies and this may potentially introduce the disease to new foci that were previously non-endemic. Persons below the age of 19 years are at a greater risk of contracting the infection (Muthami, 1995**)** due to slow acquisition of immunity to *Schistosoma* infection (WHO, 2014). Data on the influence of labour migration from non-endemic area to endemic area is lacking therefore, the purpose of this study was therefore to establish the occurrence of schistosomiasis in school children in non-endemic Kagio area and compare it with that of Mwea irrigation scheme and also establish the source of infection among the school children in Kagio.

1.3 Justification

With reference to previous work done on schistosomiasis in Kirinyaga County, a lot has been done in Mwea irrigation scheme excluding the neighbouring areas. A lot of emphasis has been on the snail intermediate hosts and their control using molluscicides (Katsivo, 1993). Some studies focused on co-infections with other diseases, the role of chemotherapy using praziquantel in reducing morbidity and mortality (Kihara and Muhoho 2007) and also the relationship between schistosomiasis and anaemia in pregnant women (Kariuki, 2011). Kagio area does not favour the establishment of snails but cases of *Schistosoma mansoni* infection are known. Identification of factors affecting transmission of the disease is key in controlling the disease, especially in people living near the transmission sites and who have a relationship with the endemic area. This will help understand how people not living in the irrigation scheme may acquire the infection. The data on prevalence and transmission risks of schistosomiasis in non-endemic area due to labour migration will be available to policy makers involved in control to target people living adjacent to the irrigation scheme to avoid them being source of infection to the population.

1.4 Research questions

(i) What are the occurrence levels of *Schistosoma mansoni* infections among school children in Kagio area and Mwea irrigation scheme in Kirinyaga County, Kenya?

(ii) What is the influence of labour migration from non-endemic Kagio area to the endemic Mwea irrigation scheme on transmission of *Schistosoma mansoni* among school children in Kagio area?

1.5 Hypotheses

(i) Children from Kagio area and Mwea irrigation scheme of Kirinyaga County do not suffer from *Schistosoma mansoni* infections.

(ii) Labour migration from Kagio area to Mwea irrigation scheme does not influence transmission of *Schistosoma mansoni* among school children in Kagio area.

1.6 Objectives

1.6.1 General objective

To determine the occurence of *Schistosoma mansoni* infections among school children in the non-endemic Kagio area and Mwea irrigation scheme of Kirinyaga County in Kenya and establish effects of labour migration as a transmission risk of the infection with *Schistosoma mansoni* among school children.

1.6.2 Specific objectives

(i) To determine occurrence of *Schistosoma mansoni* infections in school children aged 8-15 years in the non-endemic Kagio area of Kirinyaga County, Kenya.

(ii) To establish occurrence of *Schistosoma mansoni* infections in school children aged 8-15 years in the endemic Mwea irrigation scheme of Kirinyaga County, Kenya.

(iii) To establish the relationship between labour migration and the occurrence of *Schistosoma mansoni* infection among school children of ages 8-15 years in the non-endemic Kagio area.

1.7 Significance of the study

Schistosomiasis is of public health concern. It is endemic in some areas like Mwea and can easily be spread to non-endemic areas like Kagio due to water related activities by Kagio people in the endemic area. The current study was aimed at determining the occurrence levels of *Schistosoma mansoni* in non-endemic area and establishing the possible transmission risks of the disease. The results of the study will be useful to policy makers involved in control of the disease to target such population in order to effectively control the disease.

CHAPTER TWO: LITERATURE REVIEW

2.1 Epidemiology and transmission of schistosomiasis

Schistosomiasis is second to malaria as the most important tropical disease in terms of incidence of morbidity and mortality (WHO, 2012). It is of great public health and socio-economic importance in the developing world (Reich and Fenwick, 2001). It is a snail transmitted infection first discovered in 1890 by Theodor Bilharz in Egypt (Nelson, 1989). An estimated 249 million people residing mainly in rural communities in 78 countries of the world are infected while 700 million people are at risk of infection (Brunn *et al.,* 2008; WHO, 2014). It is estimated that at least 90% of those requiring treatment for schistosomiasis live in Africa (WHO, 2014). Factors contributing to schistosomiasis infection are related to human contact with parasite contaminated water in daily water related activities and often related to poverty, ignorance, hygiene practices and certain play habits of school-age children such as swimming and fishing in infested water make them especially vulnerable to infection (Mostafa *et al*., 1995; WHO 2014).

Schistosomiasis is a snail transmitted and dynamic infection spreading to new foci due to social dislocation or massive migrations caused by drought, war, famine, labour migration and effects of man-made ecological changes such as creation of dams and the cultivation of rice in paddy fields that produces expanses of water which are suitable breeding grounds for snails (Madsen *et al*., 1989; Dunne *et al*., 1995).

Transmission requires contamination of surface water by excreta from infected definitive hosts, specific fresh water snails as intermediate hosts which transmit the infective stage (cercariae)

and human contact with contaminated water through activities such as farming, fishing, and domestic chores (WHO, 2013).

Distribution of schistosomiasis is very focal and is determined by the geographical distribution of snail intermediate hosts which differ in their habitat preferences for slow-flowing or still waters. Availability of suitable snail host, the potential of infected humans to contaminate the local water and human activities like flood irrigation of crops in the contaminated water determines the endemicity of the particular species of *Schistosoma*. Schistosomisis has a wildwide distribution as shown in Table 2.1.

Table 2.1: Parasite species and geographical distribution of schistosomiasis

Form of schistosomiasis	***Schistosoma* species**	**Species Geographical distribution**
Intestinal schistosomiasis	*Schistosoma mansoni*	Africa, the Middle East, the Caribbean, Brazil, Venezuela, Suriname
	Schistosoma japonicum	China, Indonesia, the Philippines
	Schistosoma mekongi	Several districts of Cambodia and the Lao People's Democratic Republic
	Schistosoma guineensis and related *S. intercalatum*	Rain forest areas of central Africa
Urinary schistosomiasis	*Schistosoma haematobium*	Africa, the Middle East

Source: CDC, 2013

Schistosoma mansoni is found in Malasyia, Arabia, Egypt, and East Africa including Kenya. It is endemic in South America and the Carribean Islands (Hagan, 1987; CDC, 2013). However, the parasite has disappeared from some places due to ecological changes either man-made, natural or control programmes (WHO, 2002). Man-made water projects increase the number of snail habitats and the simultaneous concentration of people around them results in intensification of schistosomiasis (Sturrock, 1986). Major factors in focal geographical distribution of endemic schistosomiasis in Kenya includes sporadic distribution of intermediate hosts common in Mwea, Coastal region and Kisumu along the lake basin. Snail development is

favoured by tropical climate and availability of water which is either slow-moving or still (WHO, 2002).

Transmission involves snails which are intermediate hosts and in *Schistosoma mansoni* infection, fresh water pulmonate snails of the genus *Biomphalaria* (Brown, 1994) serve as the intermediate hosts. Three of the *Biomphalaria* species; *Biomphalaria pteifferi, Biomphalaria sudanica* and *Biomphalaria choamphala* occur in areas endemic with *Schistosoma mansoni* (Brown, 1994; CDC, 2013). In Africa and the surrounding regions, 12 *Biomphalaria* species exist and majority are known to be susceptible to the natural infection with *Schistosoma mansoni* (Brown, 1994). *Biomphalaria pteifferi* is the most wide-spread in Africa south of Sahara and the Lake Victoria region. Besides using humans as the definitive hosts, *Schistosoma mansoni* may also infect other mammalian hosts mainly non-human primates and rodents.

In Africa, baboons have been shown to be excellent definitive hosts of *Schistosoma mansoni*, acquiring infection by drinking water infested with cercariae (Fenwick, 1969; Nelson, 1990). Several rodent species and other mammals have been shown to be naturally infected with *Schistosoma mansoni* (Karoum *et al.*, 1985). All *Schistosoma* species demonstrate quite narrow intermediate host specificity (Table 2.2).

Table 2.2: *Schistosoma* species and their hosts

***Schistosoma* species**	**Definitive host**	**Snail vector**	**Geographical location**
Schistosoma haematobium	Humans and primates	*Bulinus*	Africa
Schistosoma mansoni	Humans, rodents and primates	*Biomphalaria*	Africa and America
Schistosoma japonicum	Humans, ruminants and carnivores	*Oncomelania*	South East Asia
Schistosoma intercalatum	Humans, rodents and cattle	*Bulinus* and *Physopsis*	West and Central Africa
Schistosoma mekongi	Humans, dog and cats	*Oncomelania*	South East Asia

Source: CDC, 2013

The reservoir hosts play an important role in the epidemiology of the disease since they are a source of infection even after the human population has been cured of the disease (Cheng, 2007). The reservoir hosts are naturally infected with the schistosomes and spread more eggs in the environment than human hosts and are considered to be the main sources of transmission of schistosomiasis after the human population has been treated (Shen, 1992).

Schistosomiasis is contracted through human contact with fresh water infested with schistosome larvae while performing domestic related, recreational, occupational, religious and

other activities (WHO, 2014). Parasites are transmitted between hosts by motile aquatic stages; miracidia and cercaria which actively seek intermediate and definitive hosts respectively. The miracidia penetrates the soft tissues of the snail, develop forming sporocysts which bursts releasing fork-tailed cercariae. The fork-tailed cercariae are rapid swimmers and they periodically swim to the surface of the water and then sink to the bottom for up to three days. They are attracted to skin secretions and when they come into contact with the prospective definitive host, they attach and actively penetrate the skin within minutes, losing their tails in the process (CDC, 2012).

Optimum transmission is subject to factors that affect any or all stages of transmission including egg hatching, miracidial survival and penetration, host seeking, sporocyst development, cercariae shedding and penetration into the definitive host. Physical, chemical and biological characteristics of water such as temperature, ions, dissolved gases and food availability affect suitability of the water for snail development (Madsen, 1995).

2.2 Life cycle, biology and infection of *Schistosoma mansoni*

Schistosomes are unisexual trematodes having an indirect digenetic life-cycle, involving sexual reproduction in vertebrate definitive host and asexual reproduction in snail intermediate hosts. The adults of *Schistosoma mansoni* live in the mesenteric veins draining sigmoido-rectal region. In human hosts and other susceptible vertebrate hosts, the male and females pair up and the female is held in the gynaecophoric canal of the male from where it extends its anterior end far into the smallest venules and deposits the eggs one at a time. Female worms produce numerous eggs between 200-3000 per day (CDC, 2013). Eggs then work their way through the

vessels and the mucosa of the large intestines and enter the lumen of the intestines. The embryonated eggs are then discharged with feaces into fresh water and under suitable conditions they hatch into free swimming miracidia which only live for several hours as they actively seek suitable intermediate hosts using chemotaxis and phototaxis. On encountering the right snail host, the miracidia penetrates into the soft tissue and make their way into the liver. In the snail host, the miracidia losses cilia, develops into a first stage sporocyst, mother sporocyst and through asexual multiplication it gives rise to second stage sporocyst in which free swimming fork- tailed cercariae develop. They break off from the sporocyst and escape from the snail into the water. Cercariae are released into the water beginning 4 weeks after the infection (CDC, 2013) (Figure 2.1).

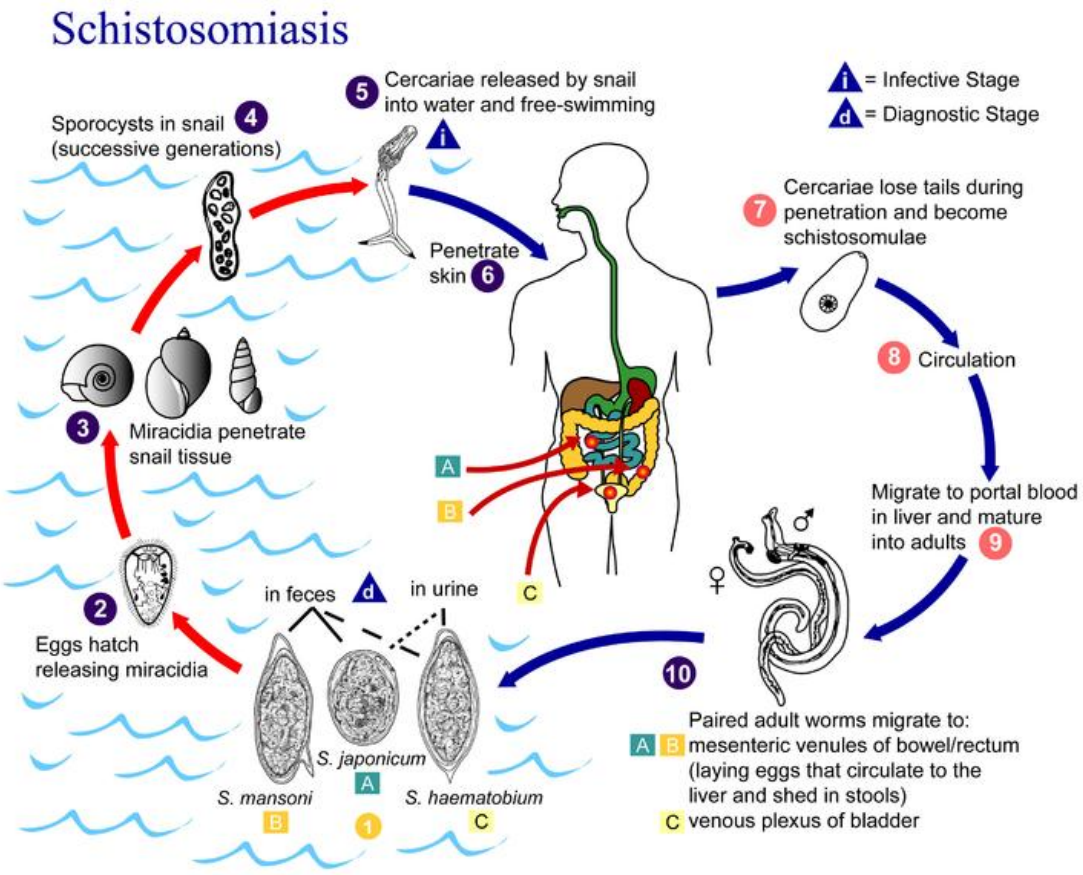

Fig 2.1 Lifecycle of *Schistosoma* spp. (Source: CDC, 2013)

The cercariae is infectious to humans and other susceptible vertebrate hosts and infections occurs when cercariae penetrates water-softened skin when individuals get into contact with contaminated water. Upon penetration, cercariae lose their tail and transform into a schistosomulae which enter the circulatory system and migrate through the vena cava, right heart, pulmonary circulation and through the left heart into the systemic circulation. Through the abdominal aorta they enter the mesenteric artery passing through the portal circulation into the liver where they develop for 3 weeks. In the hepatic portal vein, the schistosomulae grow, and on sexual maturation pairing takes place where the female is held in the gyaenocopholic

canal of the male (Figure 2.2). They migrate against the blood current into portal system venules of the intestines where they lay eggs which are voided out with feaces and the cycle is repeated (Rollinson, 1978; Arora *et al*., 2002). Egg production begins 4-8 weeks after infection and adult worms normally live for 2-5 years although some may survive much longer (CDC, 2013).

Fig 2.2 Paired adult Schistosome worm with the female held in the gyaenocopholic canal of the male (Source: CDC, 2013).

2.3 Pathology of schistosomiasis

Schistosomiasis causes morbidity and mortality (WHO, 2010). Most of the damage is caused by the eggs and not the adults and most of the pathology is caused by the host immune responses (delayed-type hypersensitivity and granulomatous reactions). The course of infection is divided into migratory, acute and chronic phases. The migratory phase occurs when the cercariae penetrate and migrate through the skin. It is normally asymptomatic but in sensitized patients, may cause transient dermatitis and occasionally pulmonary lesions and pneumonitis.

The acute phase (katayama fever) is coincident with the beginning of egg release and is characterized by allergic responses due to immune complex formation resulting in pyrexia, fatigue, aches, lymphadenopathy, gastrointestinal discomfort and eosinophilia.

The chronic phase occurs in response to the cumulative deposition of fluke eggs in the tissues and the host reactions that develop against them. In *Schistosoma mansoni* infections, many eggs lodge in the liver and elicit a cellular granulomatous reaction (polyps) which gives rise to symptoms of chronic infection (Dunne *et al.,* 1995; Odile *et al.,* 1995). Major pathological changes caused include cellular granulomatous reactions, intestinal polyposis, abdominal pain, diarrhea, cardiovascular problems including heart failure and periportal fibrosis (symmers fibrosis), progressive fibrosis causing obstruction of intestines and vessels, vascular lesions, portal hypertension which often leads to hepatomegaly, splenomegaly, ascites and gross enlargement of oesophageal and gastric vein (verices) and fatal bleeding from oesophagogastric varices when they burst (Lambertucci, 1987; Dunne *et al*., 1995). The severity of the pathology caused by eggs depends on the number of eggs trapped and the immune responses of the host which range from simple Katayama fever to chronic damage of the liver, kidney, intestines and sometimes brain and spinal cord. Reactions to eggs lead to granuloma formation and can lead to ulceration and thickening of bowel wall.

Damage of the central nervous system (CNS) sets in slowly though the impact is prolonged. Celebral granuloma have been associated with focal epileptic convulsions, while spinal cord granuloma may cause transverse myelitis. Chronic schistosomiasis in children causes stunted growth and reduced ability to learn and may affect peoples' ability to work and in some cases

may result in death. In sub-Sahara Africa, it is estimated that more than 200,000 deaths per year are due to schistosomiasis (WHO, 2014).

2.4 Diagnosis of schistosomiasis

2.4.1 Microscopy

Infections are conventionally diagnosed by the detection of fluke eggs in faecal or urine samples. Microscopic examination of stool or urine is the gold standard for diagnosis but requires the adult worms to be producing eggs (Gray, 2011). The extent to which eggs are shed varies therefore, for effective diagnosis, as many as three specimens of the first stool of the day, for three days are required in some patients. The eggs are sufficiently characteristic to facilitate specific diagnosis (Arora *et al.,* 2002). Diagnosis is done using a simple stool smear and staining technique using Lugol's iodine and the characteristic eggs identified and counted. Direct smears are done to identify the positive samples**.** *Schistosoma mansoni* eggs are large (114-180µ long by 40-70µ wide), brown or yellow in colour with a thin smooth shell, a rounded posterior end and the anterior end is somewhat pointed and curved (Figure 2.3). It also has a characteristic prominent lateral spine near the posterior end (Arora *et al.,* 2002).

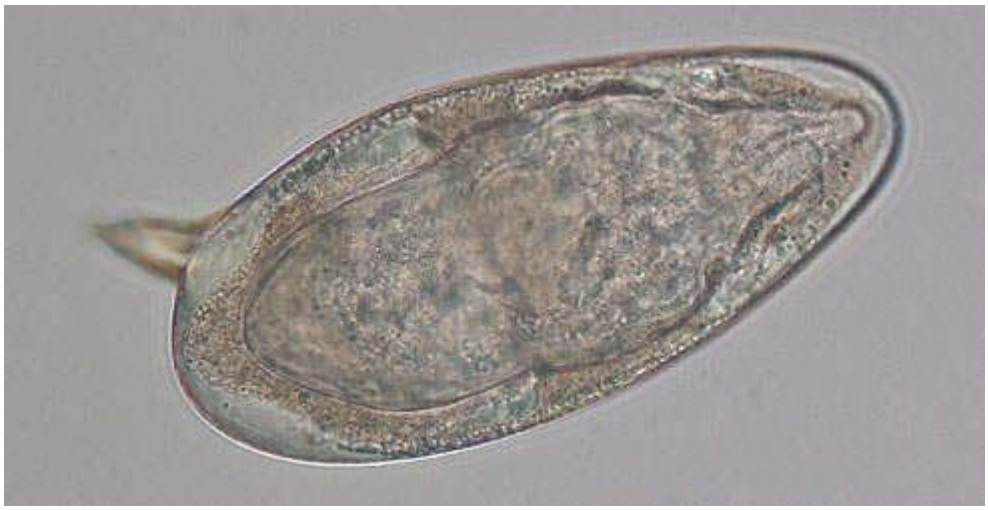

Fig. 2.3 *Schistosoma mansoni* egg. (Source: CDC, 2013)

2.4.2 Kato Kartz technique

This is a rapid, simple and inexpensive method recommended by WHO for diagnosis of intestinal schistosomiasis when the intensity of the infection is high and it is widely used in studies and requires 40-50 mg of feaces (Kartz, 1972). It has a specificity of 100% but the sensivity varies with prevalence and intensity of infection (Ross *et al*., 2002; Gryseels *et al*., 2006). *Schistosoma mansoni* diagnosis is done using Kato-kartz technique and the intensity of infection categorized as light, moderate or heavy based on the number of eggs per gram (EPG) of faeces (WHO, 2002). From population studies the mean egg burdens correlate with the severity of the disease (Ross *et al.,* 2002, Gryseels *et al*., 2006). Egg count is, however, variable hence the necessity to have repeated stool tests using the first stool of the day. In light infections, false negative results are common, often leading to underestimation of the intensity and prevalence of infection (De-Vlas *et al.,* 1992).

2.4.3 Immunological tests

Immunological tests have been developed to detect host antibodies against the infection but they have experienced cross-reactivity problems and cannot discriminate between previous and active infection and such tests are commonly used in surveys (Simpson *et al*., 1985). The tests are useful in patients who are not excreting eggs such as those with katayama syndrome. The tests are also useful in field studies for defining regions of low endemicity, where individual patients have low egg burdens and may also be beneficial in determining whether infection has re-emerged after an apparently successful control programme. Most techniques detect IgG, IgM and IgE against the soluble worm antigen or soluble egg antigens by enzyme-linked immunosorbent assay (ELISA), indirect haemagglutination or immunofluorescence (Ross *et al.,* 2002, Gryseels *et al.,* 2006). A cercarial antigen ELISA equivalent to the soluble egg antigen has been developed for serodiagnosis of schistosomiasis (Chand *et al*., 2010).

2.4.4 Molecular diagnosis

Diagnosis can also be done using molecular techniques such as Radio Immuno Assay (RIA) that involves measuring the concentration of parasite antigens or DNA in the host samples with some tests showing good correlation with the parasite burden. Specific and highly sensitive Polymerase Chain Reaction (PCR) has been developed for detection of schistosome DNA in feaces or sera and plasma (Chand *et al.*, 2010) and the approach has the potential to provide a test for diagnosing schistosomiasis in all phases of clinical disease, including the capacity to diagnose katayama syndrome and active disease and for the evaluation of treatment (Ross, 2002). For people living in non-endemic or low transmission areas, serological tests and immulogical tests may be useful in showing exposure to infection and the need for thorough

examination, treatment and follow-up (CDC, 2013). Detection of circulating adult worm and egg antigens is a promising technique that may supersede traditional diagnostic methods. There is recent development in immunoblot assay for the detection of adult worm antigens which reportedly has 95% sensitivity and 100% specificity. It is capable of detecting low levels of antigens from adult worms and eggs (Gray, 2011).

2.5 Control and treatment of schistosomiasis

Control of schistosomiasis is normally aimed at reducing infections and morbidity by interrupting the parasite life-cycle. This can be achieved through different methods directed on the hosts, parasites and the environment (CDC, 2012).

2.5.1 Chemotherapy

This is done through administration of praziquantel and oxamniquine to eliminate the parasite from the humans who are definitive hosts (Cioli, 2000). The two drugs are considered similar in efficacy and safety but praziquantel is used to a greater extent due to low cost. Praziquantel is effective against all adult *Schistosoma* species and according to WHO, it is considered safe in pregnancy, lactation and in children below the age of 24 months (Gryseels *et al*., 2006). It is administered at 40 mg/kg of body weight and it works by causing severe spasms and palalysis of the worm muscles, exposing worm antigens and allowing the body immune system to attack them. The drug achieves 60-90% cure rates with egg reduction of 90-95% in those not cured (WHO, 2012). However, the drug is uninfective against eggs and juvenile schistosomes; therefore, follow-up at 4-6 weeks is recommended with a repeat of treatment in 6-12 weeks

(CDC, 2013). It cannot be used for chemoprophylaxis because of its short half-life (1-1.5 hours) and it cannot kill schistosomula that are 3-21 days old.

Artemether is effective against all juvenile schistosomes during the first 21 days of infection in animals and humans (Utzinger *et al.*, 2007), and if given every two weeks, it should kill all the immature schistosomula. It has been used as a chemoprophylactic in schistosomiasis endemic areas for those at high risk of infection, such as flood relief workers and fishermen (Xiao, 2005).

In May, 2001 the World Health Organisation Assembly passed Resolution 54.19 which recommended regular treatment of high risk groups in endemic areas, particularly school age children, as the best means of reducing morbidity and mortality (WHO, 2002). The frequency of treatment is determined by the prevalence of the infection in school-age children. In high transmission areas, treatment may be repeated every year for a number of years with constant monitoring to determine the impact of the control measure (WHO, 2014). However, re-infections occur due to the cost of implementing the control measures. People of all ages can get re-infected following treatment, although older people re-acquire the infections at slower rates than younger ones (Kabatereine *et al*., 1999).

2.5.2 Snail intermediate hosts control

Intermediate hosts control is important in reducing reinfection. Intermediate hosts control using molluscicides is also done to control schistosomiasis (Katsivo, 1993). Snail control has been used with some success though a perfect molluscicide does not exist. A list of desirable

characteristics for molluscicides includes; toxicity to snails in low concentrations, absence of toxicity to mammals, lack of adverse effects when it enters the food chain and are stable in storage for at least 18 months (WHO, 2002). In addition, proven efficacy, specificity to snails and a variety of formulations and easy measurement of concentration in breeding sites is desirable (De souza, 1999). Niclosamide is currently the preferred molluscicide but the high cost of importing it limits the use. However, it has successfully been used in The People's Republic of China, Egypt, and Morocco (Yang *et al*., 2010). The berries of endod (*Phytolacca dodecandra*) have been shown to be natural molluscicide when they fall into the water (Hanelt *at al*., 2001) and its presence by the sides of the river in Ethiopia has been shown to be associated with reduction in local snail population (Sharma, 2009).

Biological control of snails using predators and competitor snails has had some success. Competitor snails such as *Marisa comuarietis* compete for food with the intermediate hosts and prey on snail eggs and has been used in Puerto Rico as a control agent. *Melanoides tuberculata* and *Thiara granifera* are also competitor snails. A cray fish *Procambarus clakii* feeds on *Biomphararia* species and can reduce snail population significantly. Snail eating fish have been cultured and released in the infected water with some success such as in The Peoples'Republic of China (Schimidt and Roberts, 2000; Zhou *et al*., 2010).

Environmental management has been practiced to control the snail vectors. This involves altering the rate of water flow by clearing the vegetation in the drainage canals, stream channelization, seepage control and canal lining. It makes the habitat unsuitable for snails which prefer stagnant shaded water. However, this method is not practical to apply since

altering the environment may make it suitable for other disease vectors such as *Simulium* that prefer fast moving water (CDC, 2013).

2.5.3 Public health education

Public health education is a very effective way of controlling many infectious diseases. Educating communities on: proper disposal of human waste, wearing shoes while in the fields and dangers of bathing, swimming, washing clothes or fetching water in canals and slow moving streams is effective in controlling schistosomiasis (WHO, 2014). In endemic countries such as Kenya and Nigeria it helps minimize infections in human hosts, significantly reduce contact with the infected water as well as prevent the contamination of the environment with human waste from infected persons. However, this has been hampered by high illiteracy levels of people living in infected environments such as irrigated land and lack of amenities such as toilets especially in the fields where they spend time working and also lack of piped water for domestic use (CDC, 2013; WHO, 2014). Behavioural change and public health education interventions should be tailored to children's understanding so that the goal of modifying the behavior to not urinating or defecating into open water, or being in contact with the open water while playing or washing to interrupt transmission can be met. Behavioural interventions should also target children's role models such as parents, older siblings and teachers so that they can exemplify adequate behavior change through their own life. Sensitizing children, parents and teachers on the importance and benefit of periodic deworming might increase the coverage of drug intake and this reduces worm burden in humans (Utzinger *et al.,* 2007).

2.5.4 Use of vaccination

Vaccines confer effective protection against infections. A vaccine could reduce worm fecundity and/ or prevent *Schistosoma* infection and re-infection not only in humans but also reservoir hosts such as water buffaloes that significantly contribute to transmission of *Schistosoma japonicum* (McManus *et al*., 2008). Over the past 20-30 years, multiple vaccine candidates based on recombinant-derived schistosome proteins (Loukas *et al.*, 2007), radiation-attenuated schistosome larval stages (Bickle, 2009; Lin *et al*., 2011) or DNA- derived proteins have been identified (McManus *et al*., 2008). Although protection against various schistosome species was achieved in wide range of host reservoir animals, there are currently only very few vaccine candidates such as recombinant Sm 14/FABP antigen and rSh28GST antigen which are studied in clinical trials (Tendler *et al*., 2008; Webster *et al.*, 2010). However, there is no vaccine for schistosomiasis though a number of parasite derived antigens confer partial protection against re-infection when used on mice. Research is currently underway to develop a vaccine for schistosomiasis (WHO, 2014).

CHAPTER THREE: MATERIALS AND METHODS

3.1 Study area

The area of study was Kagio, located in central Kenya, in Kirinyaga County south of Kerugoya town along Kerugoya –Sagana road. Administratively, the area is in Ndia Division of Kirinyaga West District. It is located at latitude -0.6166667ºS, longitude 37.255 and altitude of 1280 meters, and is situated 88 kilometers to the North-West of Nairobi (Appendix I).The largest group of residents are from the Kikuyu community.

The soil is predominantly red, well drained and of fine texture. Rainfall normally occurs in two seasons per year, with the main rainy season in the period April/June and short rainy season in August/September. The temperature is moderate throughout the year, ranging from 18-25^{0}C (www.meteo.go.ke). The main crops grown in the area are maize and rice. Horticultural crops grown include; French beans, tomatoes and carrots. Rice and horticultural crops trading are the main economic activities in the area. The population in Kagio town is high, approximately 46,179 persons (2009 Kenya population and housing census) due to the influence of the rice and horticulture growing as well as and rice trading.

The study area neighbours Mwea irrigation scheme and school children seek menial jobs in the irrigation scheme. There is poor sanitation in the irrigation scheme and this poses danger of transmission of *Schistosoma mansoni* to all persons visiting the irrigation scheme.

3.2 Study design

A cross-sectional study was carried out to determine the occurrence of schistosomiasis infections among primary school pupils in schistosome non-endemic area of Kagio and endemic Mwea irrigation scheme of Kirinyaga County. The data on the occurrence of schistosomiasis was obtained by collection of early morning stool and examining it for the presence of schistosome eggs. Data on the interaction of pupils from Kagio area with the Mwea irrigation scheme was collected using a questionnaire.

3.3 Description of variables

3.3.1 Dependent variables

The dependent variables are the outcome or response variables and in this study the dependent variable was the occurrence of schistosomiasis in school children aged 8-15 years from the selected schools.

3.3.2 Independent variables

Independent variables are the exposure or predictor variables and in this study the independent variables are labour migration from the non-endemic Kagio area to endemic Mwea irrigation scheme and the area of the irrigation scheme frequented by pupils from the non-endemic Kagio area recruited in the study.

3.4 Study population

The study population comprised primary school children aged 8-15 years from two schools around Kagio and one school in the Mwea irrigation scheme. The two schools that were

sampled in the non-endemic area of Kagio were Kagio primary school (1.5 km from the rice fields) with a total school population of 691 pupils of which 433 were aged 8-15 years and Kang'aru primary school (about 2.0 km from the rice field) with a total population of 381 pupils of whom 248 were aged 8-15 years. The two schools from the non-endemic area were close to kagio town approximately 0.5 km and 1.0 km respectively. One school in the irrigation scheme, Kandongu primary school was identified from the area frequented by the pupils who went to the rice fields to work or perform other activities. The school had an average population 150 pupils aged 8-15 years.

3.5 Inclusion criteria

Inclusion criteria were pupils in the selected schools of ages 8-15 years, who were willing to participate in the study and signed an ascent form after getting informed consent from their parents.

3.6 Exclusion criteria

Exclusion criteria were pupils from the sampled schools not willing to participate in the study and those below or above the specified age (8-15 years).

3.7 Sampling technique

Purposive sampling technique was used where two primary schools; Kagio and Kang'aru primary were sampled due to their close proximity to Mwea irrigation scheme. Kandongu primary school was sampled on the basis of the data obtained from the questionnaires filled by pupils from the two schools from Kagio area. Pupils aged 8-15 years from the selected schools were recruited for the study. The number of pupils sampled per school was based on the total

population aged 8-15 years where 175 pupils from Kagio, 88 pupils from Kang'aru and 59 pupils from Kandongu were sampled using a ratio of 6:3:2.

3.8 Sample size

Pupils were sampled randomly using random number generator. A total number of 322 respondents was calculated according to the formulae used by *Š*apoka *et al.* (2006). The calculation is as shown below;

$$n=\frac{Z^2P(1-P)}{e^2}$$

Where n= The desired sample size (population ≤10,000) (Kirinyaga-primary school enrolment report, 2012)

Z= The standard normal deviate usually set at 1.96 which corresponds to the 95% confidence level.

P= Population estimated to have a particular characteristic and in this study it is taken as 30% (Mutahi *et al*., 2005).

e = Is the level of precision taken as a percentage usually at 5%.

$$n = \frac{1.96^2 \times 0.3\ (1-0.3)}{0.05^2}$$

$$n = \frac{3.8416 \times 0.3 \times 0.7}{0.0025}$$

n =322**.**

The sample size was 322 school children**.** Based on the population of pupils aged 8-15 years from each school, (Kagio; 433, Kang'aru; 248 and Kandongu; 150) a ratio of 433:248:150 which simplified to 6:3:2, was used to get the number of pupils to be recruited in the study from each of the three schools respectively. Therefore, 175 pupils from Kagio primary school, 88 pupils from Kang'aru primary school and 59 pupils from Kandongu primary were sampled.

3.9 Data collection

Data collection was done though examination of stool specimen that were provided by a total of 322 pupils recruited in the study. The pupils from Kagio and Kang'aru primary schools were asked to fill questionnaires. After signing the ascent form, the contents of the questionnaire were explained by the researcher. Data on their interaction with the Mwea irrigation scheme such as their visit to the irrigation scheme, activities they performed in the irrigation scheme and how often they visited the irrigation scheme was collected (Appendix VI).

3.9.1 Sample collection

For stool collection, the recruited pupils were identified with a random numbers, each provided with a clean Elkay specimen cup labeled with the code for the sample and two applicator sticks. They were advised to place 30-50 grams (up to a particular marked level of the container) of morning faecal specimen directly into the cup. Only one sample per pupil was collected for the study. Samples were collected from each of the schools on separate days and ideally all samples from each school were collected in the same week. It took three months (September-November 2013) for all samples to be collected from the three schools. The collected samples were placed in a cool box and transported to Sagana Sub-District Hospital laboratory for

analysis that was carried out by the researcher assisted by qualified laboratory technologists. Examination was carried out within less than an hour after the collection of the specimen to avoid degradation. To preserve the samples 10% formalin was added and the contents mixed well.

3.9.2 Macroscopic examination

Macroscopically, the specimens were examined for presence of mucus and or blood and nature of the stool either formed or watery or loose and also the colour. The stool was examined for traces of mucus and blood that could be as a result of egg penetrating the wall of the intestine into the rumen. The presence of mucus and blood were used as an indicator of an infection and such samples were examined first to avoid distengration of the sample and the entire stool was examined macroscopically.

3.9.3 Direct microscopic examination of stool

Microscopic examination of the specimen was done by preparing a wet mount to demonstrate worm eggs (Arora *et al*., 2002). A drop of normal saline was placed at the center of the left half of a microscope slide and a drop of Lugol's iodine solution in the center of the right half of the slide. About 1-2 mg of stool sample was picked using an applicator stick and mixed with Lugol's iodine solution and an equal portion mixed with normal saline. The slide was covered using a cover slip that was dropped at angle to avoid trapping air bubbles. The slide was examined systematically under the light microscope at ×10 and also at ×40 magnification for the eggs and the total number of eggs counted in the entire field. Direct smears were done to identify the positive samples. Samples that were found to have the *S. mansoni* worm eggs were

recorded as positive. The results were tabulated showing the total number of positive samples per each of the schools sampled.

3.9.4 Intensity of infection

Kato Katz technique (Kartz, 1972) was used to determine the intensity of infection using the same samples (30-50g of stool sample provided by the recruited pupils). The specimen was placed on a nylon screen and using a plastic spatula, forced through the screen to separate the faecal material and the debris. The screened faecal material was transferred to a template laid centrally on a microscope slide. The template hole was completely filled with the screened fecal sample and leveled to the surface. Cellophane square film soaked in glycerol and 3% aqueous malachite green for at least 24 hours was placed over the faecal specimen to improve visibility of the schistosome eggs. The slide was inverted against another slide and the fecal specimen spread evenly under the cellophane. An additional drop of glycerol was added on the cellophane and on the edges to help clear any air bubbles that may have been trapped. The slides were placed under bright light in order to clear quickly. Eosine in saline was placed on the upper surface of the cellophane and left for 3-5 minutes then wiped off. This was to facilitate easy observation of *Schistosoma mansoni* eggs. After 24 hours, the entire slide was observed systematically for the eggs of *Schistosoma mansoni* at x40 magnification and the number of eggs counted was multiplied by 24 to obtain the number of eggs per gram. When full the template holds 41.7 mg of fecal material so when calculating eggs per gram, the number of eggs on the slide is multiplied by 24 which is calculated as follows; 1000 mg divided by 41.7 mg that gives 23.98 which is rounded to 24 (WHO, 2003).

Safety was ensured when processing samples by wearing gloves to avoid contact of the specimen with skin. After examination the specimen and the nylon screens used in Kato –Katz technique were placed in a 70% ethanol and buried in disposable specimen containers. The used microscope slides were discarded in a pot containing 1% hypochlorite solution and buried in disposable specimen containers. All used applicator sticks and remaining stool samples were soaked in a disinfectant solution and buried in disposable specimen containers.

3.9.5 Questionnaire administration

Questionnaires (appendix VI) were administered to each of the participating pupils from non-endemic area of Kagio to collect data on their activities in the irrigation scheme. The following variables were considered; whether their parents owned land in the irrigation scheme, frequency of their visits to the irrigation scheme, the particular part of the irrigation scheme they visited and other activities carried out by the pupils in the irrigation scheme such as bathing in canals, washing clothes and fetching water for domestic use. The contents of the questionnaires were explained to the pupils then they filled it with the assistance of the researcher and her assistants.

3.10 Data analysis

All data collected were entered into the computer using MsExcel software. It was then exported to SPSS for Windows version 10 for analysis. The data collected from stool examination including the number of positive samples from each school sampled and all the schools sampled were analyzed using t-test to check the difference in occurrence of schistosomiasis in the two schools located in non-endemic Kagio area and that lacated within the Mwea irrigation

scheme. The data collected using questionnaires were tabulated and analyzed using Chi-square statistics to test the relationship between labour migration from non-endemic Kagio area to endemic area, Mwea irrigation scheme and the occurrence of *Schistosoma mansoni* infections. Other variables considered were the activities carried out in the irrigation scheme by pupils from Kagio, location of the parents' land in the irrigation scheme and the area in the irrigation scheme the pupils frequently visited or worked in. Pearson correlation was used to determine the relationship between the area the pupils worked in and infection with *S. mansoni*. Chi-square test was the statistical tool of choice in order to test for homogeneity, randomness, association, independence and goodness of fit; this was done at $P < 0.05$ and 95% confidence level. Odds ratio was also calculated to test for the significance of other activities carried out by pupils in the irrigation scheme in contributing to infection. Data were presented using tables.

3.11 Ethical consideration

All ethical considerations were followed (WHO, 1975) in terms of protecting the rights and well being of the people studied. Ethical clearance was obtained from Sagana Hospital (Appendix VII). Also authority to conduct the study in the schools was sought from the County Education Officer, the head teachers of the schools and the Graduate school Kenyatta University. Participation for the pupils was voluntary and a written consent was obtained from their parents after reading and understanding the requirements for the study (Appendix IV). The participants were also informed that they could withdraw from the study at will and also filled an assent form (Appendix V). There were no risks of injury to the participants. The subjects were also examined for other helminthic infections and treated at Kerugoya General Hospital.

Data collected was treated confidentially and was used only for the purpose of this study. Pupils' identity was not revealed and they were identified using codes.

CHAPTER FOUR: RESULTS

4.1 Overall *Schistosoma mansoni* infection rates in non-endemic area of Kagio and Mwea

Pupils from the two primary schools sampled from Kagio non-endemic area had schistosome eggs in their stool samples. Kang'aru primary school had 9.1% (8 pupils out of 88) positive for *Schistosoma mansoni* eggs while Kagio primary school had 6.3% (11 pupils out of 175) positive for *S. mansoni* eggs.

The occurrence of *Schistosoma mansoni* infection in non-endemic area of Kagio was found to be 7.2% (19 out of 263). The study established that the occurrence of *Schistosoma mansoni* infection in the endemic area of Mwea was 22% (13 out of 59). The overall occurrence of *S. mansoni* infection was found to be 9.9% in all the three study schools.

Statistical analysis using t-test indicated there was no significant difference in *Schistosoma mansoni* prevalence between Mwea irrigation scheme and Kagio area (t=5.333, Cl=95%, df=1, P=0.118) (Table 4.1).

Table 4.1: *Schistosoma mansoni* infected pupils in schools sampled in Kagio area and Mwea irrigation scheme

Area	School name	Number sampled (n)	Infected cases	Percentage (%)
Kagio area	Kangaru school	88	8	9.1
	Kagio school	175	11	6.3
Total		**263**	**19**	**7.2***
Mwea irrigation scheme	Kandongu	**59**	**13**	**22***
	Total	**322**	**32**	**9.9***

*The overall infection rate of the pupils from non-endemic area of Kagio was 7.2% while that of Mwea irrigation scheme was 22% giving an overall prevalence of 9.9%.

A total of 19 pupils out of 263 from non-endemic area had *Schistosoma mansoni* eggs in their stool while from Mwea irrigation scheme, 13 out of 59 pupils had the *Schistosoma mansoni* eggs in their stool. The occurrence of *Schistosoma mansoni* infection in Mwea irrigation scheme was 22% and was significantly higher than in the schools in Kagio area. From the results (t=5.333, cl= 95%, df=1, P=0.118) there was no significant difference in the infection rates between the pupils from non-endemic area and those from the endemic area of Mwea irrigation scheme where the infection rate was 22 % in endemic area compared to 7.2 % non-endemic area.

4.2 Intensity of infection

Quantitative stool examination results revealed that 28 pupils out of 32 had light infection (10-100 eggs per gram) representing 87.5% of the infected pupils. Those who had moderate infestation (101-400 eggs per gram) were 4, representing 12.5% of the infected pupils. There were no pupils with heavy worm egg load. There was no significant difference between the light infection and the moderate infection (χ^2=0.0057, 95%Cl, P=0.9387); (Table 4.2).

Table 4.2: Intensity of *Schistosoma mansoni* egg infection (epg) in sampled pupils

Intensity (EPG) *	**No. of subject**	**Percentage (%)**
1-100	28	87.5
101-400	4	12.5
>400	0	0
Total	32	100

*Light (1-100epg), moderate (101-400epg) or heavy (>400epg). The mean number of eggs in subjects with light infection was 56 epg while those who had moderate infection had a mean of 104 epg.

4.3 Parent's land ownership in the irrigation scheme

The results revealed that 19.4% (51 out of 263) of the pupils sampled in Kagio area had their parents owning land within the irrigation scheme with 49% of the children helping their parents in the farms (Table 4.3).

Table 4.3: Parent's land ownership in the irrigation scheme

Response	Number of respondents	Overall Percentage
Parent owning land within the irrigation scheme	51	19.4
Parents not owning land within the irrigation scheme	192	73
Not sure	20	7.6
Total	263	100

4.4 Land location in the irrigation scheme where Kagio residents farmed

Much of irrigation land (82%) in Mwea frequented by pupils from non endemic area of Kagio was located around Kandongu primary school. Few pupils visited other areas of the irrigation scheme (Table 4.4).

Table 4.4: Land location and number of respondents from Kagio farming in Mwea in the irrigation scheme

Location	Number of respondents	Percentage (%)
Kirogo	1	2.0
Rwangondu	6	11.8
Kiine	2	3.9
Kandongu	42	82.3

Total	51	100.0

4.5 Labour export to the irrigation scheme

The results revealed that 213 pupils out of 263 representing 81% of the pupils from Kagio area worked in the irrigation scheme with the majority exporting their labour during the holidays. Majority (170) of the pupils mainly worked in Kandongu area representing 80% of child labour exported to the irrigation scheme.

The results showed that, 7.9% of the pupils exporting their labour to the irrigation scheme were found to have schistosome worm infestation compared to 4% of those that did not export labour but were infested with *Schistosoma mansoni* (Table 4.5). There was a significant positive correlation between pupils working in the irrigation scheme and infection with *S. mansoni* (r=0.99; cl=95%; P=0.01).

Table 4.5 Occurrence of *Schistosoma mansoni* infection in pupils from non-endemic area of Kagio working in the irrigation scheme

Test results	**No. of pupils who worked in the irrigation scheme**	**No. of pupils who did not work in the irrigation scheme**	**Total**
Positive	17 (7.9%)	2 (4%)	19
Negative	196 (92%)	48 (96%)	244
Total	213 (99.9%)	50 (100%)	263

4.6 Association between labour export and infection with *Schistosoma mansoni*

The overall schistosome prevalence rate in non-endemic Kagio area was 7.2%, therefore it was expected that 7.2% of the pupils from non-endemic area of Kagio would have schistosome eggs. A total of 213 pupils from Kagio area worked in the Mwea irrigation while 50 pupils did not work in the irrigation scheme. From the data, 15 pupils who worked in the Mwea irrigation scheme were expected to have schistosome eggs while 4 pupils who did not work in the Mwea irrigation scheme were expected to have schistosme eggs (Table 4.6). Statistical analysis indicated that there was a significant association between labour migration by children from Kagio area to Mwea irrigation scheme and the infection rate with *Schistosoma mansoni* of the school children of ages 8-15 years ($\chi^2 = 0.2604$; CI = 95%; df = 1; P = 0.01).

Table 4.6: Association between labour export and infection with *Schistosoma mansoni*

	No. of infected pupils who worked in the irrigation scheme	**No. of infected pupils who did not work in the irrigation scheme**
Observed	17	2
Expected	15	4

4.7 Other activities carried out by pupils who worked in the irrigation scheme

The results revealed that the pupils from Kagio area who exported their labour to the irrigation scheme also carried out other activities. A total of 213 pupils exported labour to the irrigation scheme and also performed other activities with 10 pupils swimming in the canals, 21 pupils

washing clothes, 23 pupils washing their legs and 159 pupils performing all the three activities combined (Table 4.6).

Table 4.7: Other activities carried out by pupils who worked in the irrigation scheme

Activity	**Frequency**	**No. infected**
Swimming	10	2
Washing clothes	21	1
Washing legs	23	4
Washing clothes, swimming and washing legs	159	12
Total	**213**	**19**

CHAPTER FIVE: DISCUSSION

5.1 Overall *Schisosoma mansoni* infection rate in non-endemic Kagio area

The study was carried out in Kagio area which is non-endemic and Mwea irrigation scheme which is endemic for *Schistosoma mansoni*. Rice farming is the major activity in Mwea irrigation scheme and it is also labour intensive and may require importation of labour from the neighbouring non-endemic regions. This may potentially introduce schistosomiasis in previously non-endemic areas. Pupils from non-endemic Kagio area export labour to Mwea rice irrigation scheme hence get exposed to infection with *S. mansoni.* The study revealed that pupils who visited the irrigation scheme were more infected than those who did not visit the irrigation scheme. The infection rate of 7.2% established for Kagio area was lower compared to previous study conducted previously in the same region (Kihara and Muhoho, 2007). However, the present results confirms earlier findings reported in Zimbabwe that showed at risk population lived at close proximity to the irrigation scheme and often visited the irrigation scheme (Mathys *et al.*, 2007).

From the questionnaire study it was found that the risk of infection of children from Kagio area who had contact with Mwea irrigation scheme was working in the rice paddies. Results from the present study revealed that 81% of the pupils worked in the paddy fields in different parts of the irrigation scheme and this may have increased their degree of exposure to the infective

stages of *Schistosoma mansoni*. These findings support recent reports done in Ethiopia that showed that frequent water contact lead to increased cases of infection (Alembrhan *et al*., 2013). Hygiene and playing in contaminated water make children vulnerable to infection (WHO, 2010). Poor sanitation and contact with contaminated water are factors that promote the

transmission of schistosomiasis (WHO, 2014). Other activities including washing the body, swimming and washing clothes done by 75% of the children in the farming fields may have exposed them more to the parasite since most of the water in the irrigation scheme is infested with the *S. mansoni* worm larva. Furthermore, results from study in Ethiopia showed that water and reasons for water contact such as swimming, working in irrigated agricultural field and bathing were significantly associated with *Schistosoma mansoni* infection (Fekadu *et al*., 1992).

5.2 *Schistosoma mansoni* infection in Mwea irrigation scheme

The infection rate of 22% in pupils from Kandongu primary school reported in the present study is lower than *Schistosoma mansoni* prevalence rate of 47% established previously by Kihara and Muhoho (2007). In addition, findings from the current study differed with a study done in Mbita District that showed a high infection rate of 76.8% (Sachiyo *et al*., 2014) and studies done in Kisumu town that showed high levels of *Schistosoma mansoni* among car washers working along shores of Lake Victoria (Karanja, 1997). However, the present findings are comparable with statistics established in Kisumu where the infection rate was 21% among school children in the endemic area located near Lake Victoria (Odiere *et al.*, 2011). The low infection rate in the current study as compared to previous findings may be attributed to regular chemotherapy using praziquantel that has been carried out annually in the present study area (MPHS, 2009). The relatively small population of pupils sampled may also have resulted in the

discrepancy in the results. Mwea rice irrigation scheme is endemic for schistosomiasis due to favourable conditions such as water bodies for snail development and poor sanitation in the rice paddies that favour the spread of the disease. Intestinal schistosomiasis is more prevalent and has high intensity in irrigated than non-irrigated areas (Mutahi and Thiong'o, 2005).

5.3 Relationship between labour migration and infection with *Schistosoma mansoni*

The identification of risk factors for the infection of *Schistosoma mansoni* contributes to a better understanding of the transmission process and for the identification of contol programmes in particular locality. The study evaluated the risk factors for infection comparing endemic and non-endemic areas. In the present study, prevalence of *Schistosoma mansoni* infection varied by school according to the proximity to the irrigated area with the school in the irrigated area presenting higher infection prevalence. It has already been established that distance from the water bodies affects infection rates whereby individuals closer to infected water bodies have a higher infection rate compared to those who live further from the infected water (Fekadu *et al*., 1992; Mathys *et al*., 2007).

Kagio area is an urban set-up with the largest population seeking menial jobs in Mwea irrigation scheme. The close proximity to the irrigation scheme makes it easy for school children to seek menial jobs in the irrigation scheme. Pupils from Kagio area worked in the irrigation scheme with some of those who worked in the irrigation scheme also performing other activities such as washing their body, washing clothes and swimming. Those that performed all the activities combined had a higher chance of infection compared to those that performed individual activities. Performing such activities exposed the pupils more to the

infested water. Pupils who exported their labour to the irrigation scheme showed higher infection rate with *Schistosoma mansoni* compared to those who did not export labour.

In the current study most of the pupils had light infection (1-100 epg). This is similar to previous reports that most individuals in endemic areas and its environs excrete low number of

eggs (Butterworth *et al*., 1991). However, it was in contrast to previous study done in Central Kenya by Mutahi and Thiong'o (2005) where 41% of the participating pupils had significant infection. Most the infected pupils worked in the irrigation scheme, results that support earlier studies done in Ethiopia that showed that children who assisted their family in irrigated farm works were more infected than those who did not participate in irrigated agricultural activity (Mathys *et al*., 2007). The general conclusion from the present findings is that there was an association between labour migration from the non-endemic area to the endemic area on transmission of *Schistosoma mansoni* infection.

CHAPTER SIX: CONCLUSIONS AND RECCOMENDATIONS

6.1 Conclusions

(i) The infection rate by *Schistosoma mansoni* in non-endemic Kagio area among the school children was 7.2% compared to 22% in Mwea irrigation scheme. Majority (85.5%) had light infection (1-100 epg).

(ii) The current study revealed that majority (80%) of the school children aged 8-15 years from Kagio area worked for pay in the Mwea irrigation scheme mainly in Kandongu area. Only 19.4% had their parents owning land in the irrigation scheme.

(iii) The results revealed that there was a strong relationship between labour migration and *Schistosoma mansoni* infection among school children from the non-endemic Kagio area to Mwea irrigation scheme. Therefore labour export was a significant transmission factor of *Schistosoma mansoni* in school children from Kagio area.

6.2 Recommendations

(i) Schistosomiasis control activities such as regular chemotherapy to reduce occurrence of the disease and regular suivaillance should be carried out by the Ministry of Health in both endemic and neighbouring non-endemic areas for effective control of the disease.

(ii) Public health education and awareness should be introduced by the Ministry of Health to persons in the non-endemic area who have an interaction with the endemic area on issues such as behavior change to reduce environment contamination with urine and feaces of infected persons, reduction of contact with contaminated water, wearing protective clothing and shoes while working in the rice paddies.

(iii) Majority of the pupils who had *S. mansoni* infection were found to export labour to Mwea rice irrigation scheme. There is a need to include issues related to schistosomiasis by the Ministry of Education in the school curriculum in both endemic areas and non-endemic areas neighbouring the endemic areas.

6.3 Further research

(i) Further research should be done to survey and identify snail intermediate hosts in the water bodies in Kagio and whether the snails are infected by the *S .mansoni* larvae.

(ii) Further research should be carried out to establish the occurrence levels of *S. mansoni* infection in the entire population in the non-endemic Kagio area. This will help understand the extent of the infection and therefore inform the Health Ministry and other stakeholders when formulating intervention measures to curb the disease in non-endemic areas adjacent to the endemic areas.

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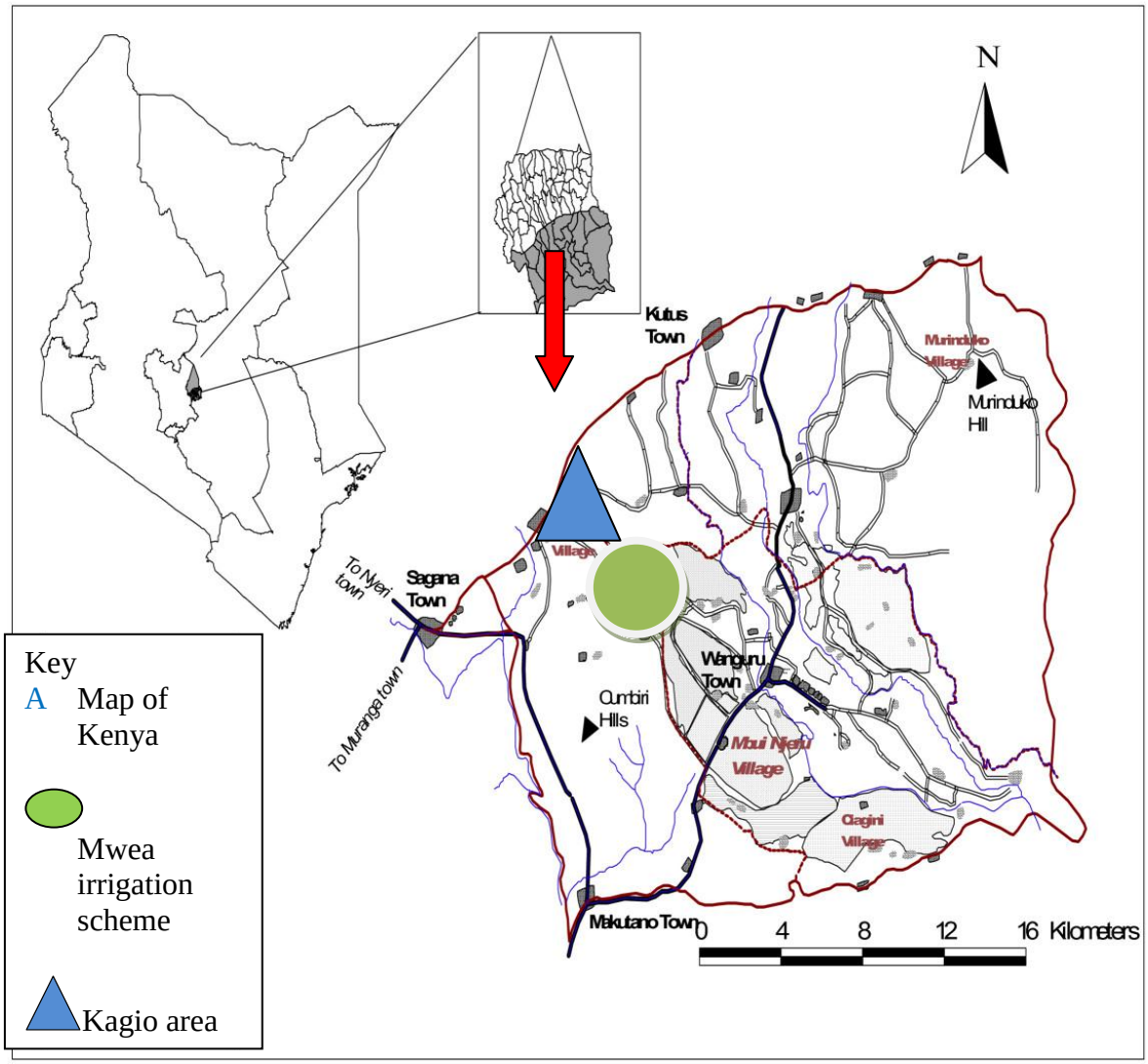
APPENDICES

APPENDIX I
MAP SHOWING MWEA IRRIGATION SCHEME AND KAGIO AREA



APPENDIX II

QUESTIONNAIRE GUIDE; TRANSMITTAL LETTER

DEAR RESPONDENT,

Am Ruth W.Munene Reg.No.I56/CE/22426/2010, a Master of Science student in the Department of Zoological sciences at Kenyatta university carrying out a research project on the influence of labour migration on the occurrence of *Schistosoma mansoni* infection in three primary schools of Kagio area and Mwea irrigation scheme of Kirinyaga County. I hereby invite you to participate in this research by filling in the attached questionnaire. Your views will be kept confidential and will be used solely for the purpose of this research.

Thank you in advance.

Signature……Date……

APPENDIX III

CONSENT EXPLANATION

Study title: Occurrence of schistosomiasis and its transmission risks in school children in non-endemic Kagio area in Kirinyaga County.

Researcher: Munene Ruth Wanjiku.

Reg. NO. I56/CE/22426/2010

Sschool of Pure and Applied Sciences,

Departmentof Zoological Studies

P.O.BOX 43844, Nairobi, Kenya.

Supervisors: 1. DR.LucyKamau

Department of Zoological sciences, Kenyatta University

2. DR. Muhoho Ng'ethe

Department of Pathology, Kenyatta University

PARTICIPATION INFORMATION

You child is being asked to participate in a medical research being conducted by the student named above. Your child's participation is voluntary. You can withdraw him/her without any penalty and can ask questions for better understanding and for the betterment of the study.

AIM OF THE PROJECT

Schistosomiasis is a parasitic disease that is prevalent in the tropical and sub-tropical countries. It is transmitted by a snail and water is very crucial in its transmission. It affects people of all ages and infection occurs when the larvae enters through the skin when one gets into contact with infested water.

RISKS, HAZARDS AND DISCOMFORTS

There will be no risks, hazards or discomforts on your child since they will collect the specimen themselves. To avoid any contamination of hands when collecting the specimen, the child will be provided with sterile applicator sticks.

BENEFITS

If you allow your child to participate in the study he or she will also be tested for other helminthes and treated at Kerugoya District Hospital.

PROCEDURE

If you allow your child to participate in the study he or she will be asked to provide an early morning stool sample. Each child will be provided with a sterile specimen cup and two applicator sticks. The sample will be labeled using the child's random code and will only be used for the purpose of the study. The participating pupils will be requested to fill a questionnaire

APPENDIX IV

PARENT CONSENT

INFORMED CONSENT AGREEMENT FOR SUBJECTS UNDER 18 YEARS.

I, Mr/Mrs/Miss/Ms……, being an adult and being the lawful parent/guardian of

Child's

name…….Age…….school……..,

Do hereby give permission to Dr/Mr/Mrs/Miss/Ms……

to include him/her in the intended study as detailed in the protocol that has been explained to me in……the language that I understand and understood by me. I have also understood the implications and benefits of the test. I accept the test to be carried out on my child.

Parent/guardian

signature…….....

Date……

Name of the person obtaining the consent……

APPENDIX V

ASSENT FOR CHILD

You are being asked to give a stool sample that will be examined for schistosome eggs and other helminthes. You do not have to do this if you do not want to but there is no danger in doing it.

Do you agree to give your stool specimen to be examined for schistosome eggs and other helminthes? Yes ( ) No ( )

Child's name …….

Child's signature…….

Signature of the person obtaining the consent…….

Witness name ……

Signature……

Date……

APPENDIX VI

QUESTIONNAIRE

Please fill the questionnaire as accurately as possible. The information in the questionnaire will be treated confidentially and will be used only for the purpose of this study.

Subject code/number……

School……

Age……

Instructions: tick where applicable

1. Do your parents have farm in the rice irrigation area/scheme?

   Yes ( ) No ( ) I do not know ( )

2. If yes, where is the farm located?

   (a) Kirogo

   (b) Rwangondu

   (c) Kiine

   (d) Kandongu

   (e) Others (specify)……

3. Do you help your parents in farming in the rice growing area?

   Yes ( ) No ( )

4. Do you go to work in the rice paddies for any payment?

   Yes ( ) No ( )

5. If yes, when do you go to work in the rice paddies?

   School holidays ( ) Weekends ( )

6. How often do you go to farm?

(a) Every weekend.

(b) Most weekends.

(c) Only holidays.

(d) Seldom.

7. Which area of the irrigation scheme do you frequently go to farm?

(a) Kirogo.

(b) Rwangondu.

(c) Kandongu

(d) Any other (specify)……

8. What other activity do you do when you go to the irrigation scheme?

(a) Swimming.

(b) Washing legs.

(c) Washing clothes.

(d) All the above activities.

(e) Any other (specify).

9. Do you have friends or relatives living in the irrigation scheme?

Yes ( ) No ( ) I do not know ( )

10. If yes, do you visit them?

Yes ( ) No ( )

11. If yes, how often do you visit them?

(a) Daily

(b) Weekly

(c) Monthly

(d) Seldom

APPENDIX VII
PROPOSAL APPROVAL LETTER FROM GRADUATE SCHOOL, KENYATTA UNIVERSITY

KENYATTA UNIVERSITY
GRADUATE SCHOOL

E-mail: dean-graduate@ku.ac.ke
Website: www.ku.ac.ke

P.O. Box 43844, 00100
NAIROBI, KENYA
Tel. 810901 Ext. 57530

Internal Memo

FROM: Dean, Graduate School

DATE: 18th September, 2013

TO: Ms. Munene Ruth Wanjiku
C/o Zoological Sciences Dept.
Kenyatta University

REF: 156/CE/22426/10

SUBJECT: APPROVAL OF RESEARCH PROPOSAL

This is to inform you that Graduate School Board at its meeting of 11th September, 2013 approved your Research Proposal for the M.Sc. Degree, Subject to editing the title to read "Occurrence of Schistosomiasis and its Transmission Risks in School Children in Non-Endemic Kagio Area in Kirinyaga County".

Thank you,

REUBEN MURILIKI
FOR: DEAN, GRADUATE SCHOOL

c.c. Chairman, Zoological Sciences Dept.

Supervisors:

1. Dr. Lucy Kamau
   C/o Zoological Sciences Dept.
   KENYATTA UNIVERSITY

2. Dr. Muhoho Ng'ethe
   C/o Pathology Dept.
   KENYATTA UNIVERSITY

RB/cao

Committed to Creativity, Excellence & Self-Reliance

APPENDIX VIII
ETHICAL CLEARANCE BY SAGANA SUB-COUNTY HOSPITAL

SAGANA SUB-COUNTY HOSPITAL,

P.O.BOX 117,

SAGANA.

TO,

RUTH W. MUNENE,

P.O.BOX 33,

EMBU.

RE: ETHICAL CLEARANCE

This is to inform you that our Review Committee went through your research proposal and considered you to conduct the research at our hospital laboratory.

Our technical team at the hospital laboratory department will assist you to ensure that all the recommended standards and procedures are followed in the course of your research process.

We wish you well in your research work.

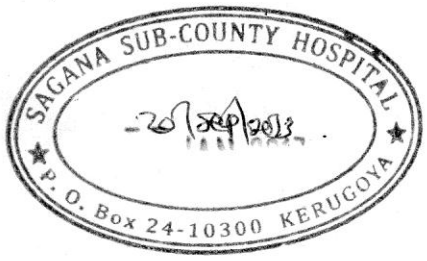
Thankyou.

Yours faithfully,

Dr. Nancy Kuria

Medical Supperitendent,

Sagana sub-county Hospital.



CC

Laboratory in-charge Sagana sub-county hospital.

APPENDIX IX
LETTER OF PERMISSION TO COLLECT DATA FROM SAGANA HOSPITAL

THE MEDICAL SUPPERITENDANT,

SAGANA SUB-COUNTY HOSPITAL,

P.O.BOX 117, SAGANA.

TO,

RUTH MUNENE,

KENYATTA UNIVERSITY,

I56/CE.22426/2010.

DEAR SIR/MADAM,

RE: PERMISSION TO COLLECT RESEARCH DATA

This is to certify that the above named Master of Science student has been permitted to do stool sample analysis and data collection at Sagana sub-county hospital.

As part of her Master of Science programme she will carry out stool sample analysis and collect the data regarding the same. Please accord her the necessary assistance to carry out the data collection.

Thankyou.

Yours faithfully,

SAGANA SUB-COUNTY HOSPITAL
P. O. Box 24-10300 KERUGOYA
25/ Dec /2013

Dr. Nancy Kuria.

Medical Supperitendant,

Sagana sub-county hospital.

CC

Zoological sciences department, Kenyatta University.

Lab in-charge Sagana sub-county hospital