PREVALENCE OF INTESTINAL PARASITES AMONG MEDICALLY
CERTIFIED FOOD-HANDLERS IN NAIROBI, KENYA

BY

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IN PARASITOLOGY IN THE SCHOOL OF PURE AND APPLIED SCIENCES OF
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DECLARATION

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

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SUPERVISORS’ APPROVAL

We confirm that the work reported in this thesis was carried out by the candidate under our supervision.

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DEDICATION

To my late mum, Jerusha; wife, Esther; daughters, Immaculate and Yvonne and son Kamau for their prayers, encouragement and patience throughout my study.
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ABBREVIATIONS AND ACRONYMS

AIDS .......... Acquired Immunodeficiency Syndrome
CAP .......... Chapter
CDC .......... Centres for Disease Control
DALYs ........ Disability Adjusted Life Years
HIV .......... Human Immunodeficiency Virus
HMIS .......... Health Management Information Systems
MoH .......... Ministry of Health
STC .......... Special Treatment Centre
TB .......... Tuberculosis
WHO .......... World Health Organization
Protozoa and helminthes are the commonest and widespread causes of infections and are a major public health problem with amoeba being the most frequent among food-handlers. Human beings acquire intestinal parasitic infections through several routes including faecal-oral and skin penetration and are facilitated by many factors including poor sanitation and overcrowding. Probable sources of infection include food outlets through food-handlers. The objective of the study was to determine the rate of intestinal parasitic infection in medically certified food-handlers within the validity period. The incubation period of these infections range from a few days for protozoa to three months for helminthes. Issued medical certificates have a validity period of six months and some food-handlers may get infected by intestinal parasites and can be sources of infection to people they serve or interact with during the six months validity period. Medically certified food-handlers were randomly selected from various categories of food outlets from Mathare, Central Business District of Nairobi and its environs. Three hundred and twelve food-handlers of which 131, 105 and 76 were studied from low, middle and high class food outlets respectively. Sampling was carried out on a weekly basis over a period of twelve months. During the study period 48, 17 and 5 food outlets from low, middle and high class were sampled. Stool samples collected from the food-handlers were processed and examined under a microscope for cysts, trophozoites, larvae and eggs of intestinal parasites using direct and concentration techniques. SPSS version 16 was used to analyze generated data. Fisher’s Exact test was used to test for significance between parasite associations. The parasites found in certified food-handlers were *Entamoeba histolytica*, *Ascaris lumbricoides* and *Giardia lamblia*. *E. histolytica* had slightly higher infection rates than *G. lamblia* and *A. lumbricoides* but no significant difference was observed (*F*=1.779, *P*= 0.248). Overall pattern of parasites per age stratum had significant statistical difference (*P*=0.039, $\chi^2= 4.20$). Low class food outlets had slightly higher parasite incidences but the different food class categories showed no significant differences (*F*=1.495, *P*= 0.297). Parasite infection had no significant statistical difference with education levels (*P*= 0.36, $\chi^2= 2.99$) but was statistically significant between the age strata (*P*=0.039, $\chi^2= 4.20$). The results of this study showed that certified food-handlers get intestinal parasitic infections during the certificate six month’s validity period. Therefore, it is recommended that the validity period of medical certificates be revised from six to three months; enforce high level of personal hygiene through regular food outlet inspections and health education. This will reduce the chances of infections from the food-handlers to their clients.
CHAPTER ONE

INTRODUCTION

1.1 Background information

Intestinal parasitic infections are globally endemic and constitute greatest single worldwide cause of illness and disease (Thiong'o et al., 2011; Bethony et al., 2006). They are great public health nuisance especially in the developing countries (Dianou et al., 2004; Gétaz et al., 2007). Studies indicate that intestinal parasitic infections cause malnutrition, morbidity, mortality and socioeconomic impact owing to treatment cost and hospitalization (Murray & Lopenz, 1996). Intestinal parasites, which have direct life cycle, are transmitted by faecal oral route to human through poor personal hygiene (Ukoli, 1990; Kaferstein & Abdussalam, 1999). Food-handlers with poor personal hygiene and have inadequate knowledge on food safety play a major role in the transmission of food borne diseases (Nyarango et al., 2008; Abera et al., 2010). Food-borne parasitic diseases are included in a new initiative of the World Health Organisation on estimating the burden of food-borne diseases (WHO, 2006, 2007). These diseases are widespread in developed and developing countries. An estimated 30% of the population in developed countries suffer from food borne diseases each year, whereas in developing countries up to 2 million deaths are estimated per year (WHO, 2007). Infections with intestinal helminthes are estimated to affect about two billion people worldwide, of whom 300 million suffer associated severe morbidity (Savioli et al., 2002; de Silva et al., 2003; Bethony et al., 2006). There are over 200 helminthes associated with human intestinal tract and include the trematodes, cestodes, nematodes and the acanthocephalans (Crompton, 1999). The cost of the infection by the intestinal worms is huge. The majority of the burden is due
morbidity rather than mortality. The 2001 Global Burden of Disease (GBD) study indicates that 58.1 million people suffered high intensity *A. lumbricoides* infection, 26.6 million with high intensity *T. trichiura* infection, and 59.9 million with high intensity hookworm infection. Only 3000 deaths were attributable to each species. Globally, *A. lumbricoides* is estimated to cause the loss of 1.817 million DALYs, *T. trichiura* 1.006 million DALYs, and hookworm 0.97 million DALYs. The majority of DALYs were lost in Southeast Asia (47%) and sub-Saharan Africa (23%) where poor people living in rural areas and urban slum lack adequate housing, safe water supplies and good waste disposal systems (Rosenfield et al., 1984). Parasitic diseases are thus contributing to inequalities that exist both within and between societies (Evans & Jamison, 1994).

Until recently, very few countries had conducted national surveys on intestinal worms, despite transmission by immigrants to previously unreported locations (Nüesch, 2005). National surveys have been conducted in a number of African countries, including Angola, Burkina Faso, Mali, Malawi, Mozambique, Niger, Sierra Leone and Uganda. In countries without comprehensive data, estimates of national prevalence are extrapolated from the available prevalence surveys that have been undertaken. In certain countries, however, very few surveys exist and these have typically been conducted in areas of known high prevalence, potentially over-estimating national prevalence (Brooker et al., 2000). However a global atlas of helminth infection was launched with the aim to describe the geographical distribution and prevalence of infection and to highlight areas for which prevalence data are absent (Brooker et al., 2000, 2009).
In Kenya outpatient morbidity estimate for four years from 1996-1999 indicated that intestinal worms ranked number 5 consecutively with 663,314 (4.2%), 600,660 (4.4%), 597,100 (4.4%) and 624,273 (4.5%) number of cases. In 1999, top ten causes of outpatient morbidity expressed in percentage intestinal worm infections gave 5%. On trends of top ten causes of outpatient morbidity by Province in Kenya from 1996-1999 intestinal worms was ranked at position 4 in Central and 5 in Coast, Eastern and North Eastern at position 3 and 6: Rift Valley and Western at position 5 and 6 and Nairobi and Nyanza- all others. In 1999, causes of hospitalization and deaths by major categories in Kenya infectious and parasitic diseases was ranked at position one with 118,641 (31.2 %) out of 379,839 total hospitalized and 58,543 (33.8%) and 60,098 (29%) as number of males and females respectively. Number of deaths reported in the same category was 7,418 (35.3%) males and 6,655 (35.9%) females.

During 2003- 2004 out-patient morbidity reports, intestinal worms accounted for 622,685 cases (4%) and 1,029,048 cases (5%) respectively countrywide. Nairobi Province had 8,115 (in 2003) and 13,861 cases in 2004. Unedited out-patient morbidity reports on intestinal worms in Kenya for the years 2005 and 2006 was 1,193,600 and 1,266,439 respectively, inclusive of Nairobi cases that were 17,858 for the year 2005 and 22,181 for the year 2006 (MoH, 2009). Food-handlers are transmission agents to some of these parasitic infections. They are however required to undergo medical examination and issued with medical certificates before they are allowed into food service outlets. Currently, government medical institutions in Kenya issue certificates of medical examination (Appendix VI) to food-handlers upon examination which are valid for six months. This is in line with Cap 254 “Food, Drug and Chemical Substances”, (Food
1.2 Problem statement
Currently there are no vaccines developed against any stage of intestinal parasites. The six months validity period of the issued medical certificates is long since most parasites have a life-cycle shorter than three months. Conventional methods currently in use for examination cannot detect the parasites during the incubation period in an infected person. Medical certificates are only issued to those who test negative to all the tests. Medically certified food-handlers have been found infected and may infect others and clients due to the nature of their work before the validity period has elapsed. Such food-handlers are carriers of parasites and could infect their uninfected colleagues and clients that they serve. Rampant insanitary practices cause food and water contamination due to factors including limited health education to some food-handlers and therefore lack personal hygiene which aid in the spread of these infections (Khurana et al., 2008). It is against this background that the present study was carried out to establish intestinal parasitic infections amongst food-handlers in different classes of food outlets.

1.3 Justification
Intestinal parasitic infections are major public health concern due to morbidity and economic burden. Sources of infection include person to person through ingestion of contaminated food and water. The parasites infect all classes of people including food-handlers with equal measure as long as the there is a conducive environment. One of the requirements that should be met by food-handlers before they are issued with medical certificates is that they should be free from intestinal infections. However, Cap 254 (Laws
of Kenya) is silent and does not expound on what happens during the period between 0-6 months of the validity of the medical certificate. Epidemics due to parasitic pathogens do occur with serious consequences some of which have been associated with contaminated food and food-handlers. Studies have indicated that risk factors associated with parasites are high and food-handlers are similarly exposed to such risks (US Treasury Dept. 1936). This demonstrates that such epidemics could occur in Kenya and can be effectively propagated by unhygienic food-handling procedures. In Kenya, studies on intestinal parasite infection during validity periods of issued medical certificates have not been carried out. The results of this study are therefore aimed at advising authorities to revisit or review the frequency of examination of medically certified food-handlers and renewal period of their medical certificates.

1.4 Research questions
a) What is the prevalence of intestinal parasitic infections in medically certified food-handlers in different food outlets in Nairobi?
b) What are the intestinal parasites infecting selected and medically certified food-handlers in Nairobi during six months’ validity period?
c) What is the pattern of parasitic infection among food-handlers in different food outlets in Nairobi?

1.5 Null hypotheses
a) Food-handlers with valid medical certificates are not infected with intestinal parasites.
b) Food-handlers with newer medical certificates are not infected with intestinal parasites.

1.6 Objectives

1.6.1 General objective

To determine the prevalence of intestinal parasites in food-handlers in different types of food outlets in Nairobi, Kenya.

1.6.2 Specific objectives

a) To determine the prevalence of parasitic infections in medically certified food-handlers in different types of food outlets in Nairobi.

b) To determine types of parasites infecting medically certified food-handlers in different types of food outlets in Nairobi during six months’ validity period.

c) To establish the pattern of parasitic infections among food-handlers in different types of food outlets in Nairobi.
CHAPTER TWO
LITERATURE REVIEW

2.1 Global distribution of intestinal parasitic infections

Intestinal parasitic infections are prevalent worldwide and in developing countries may even be more important than bacterial infections. Various pathogens have been identified as causing diarrhoeal disease and include protozoa and helminthes (Dhanurkar, 2005). The present distribution of intestinal parasitic diseases reflect the success of hygiene and control measures in the more developed parts of the world rather than any clear geographical or climatic restriction. A study model has been developed to measure the burden of risk of disability that diseases represent in age group and different regions of the world (Bundy et al., 2004). The measurement took into account both losses from premature death and the effects of losses caused by weakened health. The effects of the loss of healthy life were quantified for the diseases listed in the 109 categories included in the International Classification of Diseases (WHO, 1977) and for approximately 95% of possible causes of disability (World Bank, 1993). To evaluate losses from illness, the incidence of cases by age, sex and demographic region was estimated and the number of years of healthy life lost was obtained by multiplying the expected duration of the diseases by a severity weight that estimated the severity of the disability in comparison with loss of life caused by the illness (World Bank, 1993). The global burden of disease estimated in this manner was quantified into units called the disability adjusted life years (DALYS) (Fig. 2.1 A & B). The precise amount of morbidity and mortality caused by intestinal nematodes will never be known. This elusiveness is due to the non-specificity of clinical signs, difficulties in parasitological diagnosis and a paucity of reliable and
accurate epidemiological data, compounded by the fact that much of the burden is concentrated among countries with weak disease surveillance systems (Brooker, 2010).

In many communities, 15% of the infected population harbours more than 60% of the worms. As morbidity is usually related to intensity of infection, heavily infected individuals have greater risk of developing diseases symptoms, and they also cause the most contamination of the environment (Anderson, 1986). In addition, individuals with a heavy worm burden are more likely to re-acquire heavy infection after treatment and to suffer from polyparasitism (Anderson & Medley 1985; Bundy & Cooper, 1989). Worm infections are a significant burden in Africa and some other regions of the world (World Bank, 1993) (Fig. 2.2). People infected with hookworms suffer from iron deficiency anaemia (Roche & Layrisse, 1966). Studies from many countries including Kenya detected the relationship between declining blood haemoglobin concentration and increasing hookworm intensity (Hill & Anderson, 1942). It was estimated that at any given time, about thirty million women who were pregnant were infected with hookworms and those infections contributed greatly to about 200,000 deaths of women in developing countries ascribed to anaemia (Viteri, 1994).

Hookworm infection often causes iron deficiency which may reduce productivity in affected individuals. The infection and the resulting anaemia may impair mental development in children, negatively influencing their cognitive performance and in women may also complicate pregnancy (Stephenson, 1984; Prescott & Jacloes, 1984, WHO, 1987; Holland, 2000). In developing countries, widespread hookworm infection may make substantial contribution to the depressed economies of these countries.
Economic hardship in the rural areas of countries such as Kenya has stimulated migration to urban areas. As a result, 45% of the world's population now live in cities and diseases that were traditionally considered to be 'rural' have spread into cities such as Nairobi, usually affecting the most deprived populations.

**Fig. 2.1:** Percentage distribution of DALYs lost by overall cause in countries with established market economies, A and developing countries, B (World Bank, 1993)
Fig. 2.2: Distribution of DALYs lost by worm infections by region, 1990 (in hundreds of thousands of DALYs lost). Source: World Bank (1993).

2.2 Protozoa

Intestinal protozoa including amoeba, flagellates, coccidians and ciliates have the highest prevalence in the tropics and sub-tropics (WHO, 1995). Parasitic diseases that are intestinal related are mostly caused by *Entamoeba histolytica* and *Giardia lamblia* (WHO, 1993; Kucik *et al.*, 2004). These parasitic diseases are found in all the major regions of Africa (Buchy, 2003). It was estimated that 500 million people were infected with *E. histolytica* and that 40 - 50 million individuals had clinical symptoms of amoeba, which causes 40 - 100 thousand deaths per year worldwide (WHO, 1993). However, *E. histolytica* is often over reported since it is not often distinguished from the more common, non-pathogenic, *E. dispar* (Abd-Alla *et al.*, 2006). Since the introduction of molecular techniques, it is estimated that 500 million individuals with *Entamoeba* infection are colonized by *E. dispar* (Fotedar *et al.*, 2007).
In Kenya, morbidity caused by amoebiasis and giardiasis from unedited reporting for the year 2006 was 446 and 3 respectively while mortality of 35 was reported due to amoebiasis. Other species of amoebae commonly found in human intestinal tract include *Entamoeba hartmani*, *E. coli*, *E. dispar*, *Trichomonas hominis*, *Blastocystis hominis*, *Chilomastix mesnilli* and *Endolimax nana* (Thiong' et al., 2011). Data on specific protozoa in Nairobi are poorly reported.

### 2.2.1 Occurrence and morphology of *Entamoeba histolytica*

Several species of amoeba are common in humans in most parts of the world but only *E. histolytica* is an important pathogen, invading the small intestine and colon of humans, apes, monkeys, dogs, cats and rats (Cox, 1992). Sometimes the amoeba invade the mucosa and sub-mucosa and may be carried via the portal vein to the liver and other parts of the body causing damage to the wall of the bowel or the liver but, in most people, there is no tissue invasion. The mortality rate due to amebic liver abscess has fallen to 1-3% in the last century following the introduction of effective medical treatment. Nevertheless, amebic liver abscess is complicated by sudden intraperitoneal rupture in 2-7% of patients, leading to a higher mortality rate (Stanley, 2003). Its greatest impact is in Africa and Asia. In Africa, Egypt, Morocco and countries located between 10°N and 10°S are severely affected. Prevalence is high in Asia, particularly in Bangladesh, Myanmar, China, India, Iraq, the Republic of Korea and Vietnam. Amoebiasis is also a problem in Mexico and other Latin American countries. In Europe and the USA amoebic infection rate is between 2-5% and often asymptomatic (WHO, 1981).
The cysts of *E. histolytica* are round or slightly oval hyaline bodies 8-20 μm in diameter. They are surrounded by a retractile wall, 125-150 μm thick apparently composed of fibrillar material. The plasma membrane frequently has deep invaginations; polyribosomes and vacuoles containing dense fibrogranular material, which can be observed close to its cytoplasm face. Food vacuoles tend to disappear as the cyst matures. Staining with iron-haematoxylin makes the cytoplasm appear vacuolated with numerous glycogen deposits that decrease in size and number as the cyst matures. Chromatoid bodies, which are aggregated ribosomes can be identified inside the cytoplasm as rod shaped structures with blunt or rounded ends. Iodine stains allow the clear visualization of one to four small nuclei.

Trophozoites are the active forms of *E. histolytica* but are only found in diarrhoeic stools. They are highly dynamic and pleomorphic cells, however, their motility and shape are strongly affected by changes in temperature, pH, Osmolarity and redox potential. Actively motile amoebae are elongated, with protruding pseudopodia but spherical when at rest. The diameter of the cell varies between 10 and 60 μm, not only due to the pleomorphism of the parasite, but also to the feeding conditions. Amoebae obtained directly from intestinal or liver lesions are generally larger (20-40 μm) than those found in nondysenteric stools or in cultures (10-30 μm). The cell surface has numerous circular openings 0.2 – 0.4 μm in diameter that correspond to the openings of micropinocytic vesicles (Martinez-Palomo, 1982).
2.2.2 *Entamoeba histolytica* lifecycle

Trophozoites dwell in the colon, where they multiply and encyst typically producing four-nucleated cysts. These appear in formed stools of carriers as round or slightly oval hyaline bodies with a refractive wall. When a cyst is ingested, the cyst wall is dissolved in the upper gastrointestinal tract and the parasite excysts in the terminal ileum eventually giving rise to 4 uninucleated trophozoites. Cysts do not develop within tissues, but the invasive forms of the parasite, the trophozoites that can penetrate the intestinal mucosa and disseminate to other organs. They are also short-lived outside the body and do not survive passage through the upper gastrointestinal tract. In contrast, cysts may remain viable in a humid environment and remain infective for several days (Ravdin & Stauffer, 2005).

An epidemic of amoebic dysentery that occurred in the US in 1933 provided an insight into the incubation period of the disease. Contamination of the water supply to hotels was responsible for this outbreak, which resulted in 1400 cases with reliable history data of 391 cases. Severe infections tended to have a shorter incubation period, most of them being reported within 1-6 weeks after initial exposure (US Treasury Department, 1936).

2.2.3 Transmission of *E. histolytica*

Transmission of amoebiasis may be accomplished through a variety of mechanisms. Asymptomatic carriers pass large numbers of cysts in their stools and are important source of infection particularly if they are engaged in the preparation and handling of food. Transmission and infection occur through ingestion of as few as 10 viable cysts and can be acquired from water, food and person to person through faecal oral route (Ximénez *et al.*, 2010). The cysts then remain viable in faeces but are killed by desiccation and
temperatures above 68°C (Schmerin et al., 1978; Phillips et al., 1981; Garcia, 1992). This protozoon derives its name from its ability to lyse virtually every tissue in the human body and in the bodies of experimental animals. Lysis of target cells involves contact dependent as well as contact-independent mechanisms. Initial attachment of the trophozoite occurs via the amoebic adhesions. Once attached, it has been suggested that the parasite releases an active peptide, the amoeba-pore that is capable of inserting ion channels into liposomes that possess cytolytic and bactericidal activities (Leippe et al., 1995).

2.2.4 Clinical manifestations of *E. histolytica*

Depending on the affected organ, the clinical manifestations of amoebiasis are either intestinal or extra-intestinal (Tengku & Norhayati, 2011). There are four clinical forms of invasive intestinal amoebiasis all of which are generally acute dysentery or bloody diarrhoea, fulminating colitis, amoebic appendicitis and amoeboma of the colon. About 50 million people are infected by this parasite and several tens of thousands die each year as a consequence of fulminating colitis or amoebic liver abscess (Dhanukar, 2005; Samuel et al., 2001). Amoebic colitis affects both sexes equally. Amoebic liver abscess is 7-12 times more common in men than in women, with a predominance among men aged 18-50 years (Gunther et al., 2011). The reason for this sexual disparity is unknown, although hormonal effects may be implicated, as the prevalence of amoebic liver abscess is also increased among postmenopausal women. Alcohol may also been an important risk factor. The sexual distribution is equal in children (Stanley, 2003). Other factors including HIV seropositivity are risk factors for invasive extra intestinal amoebiasis. *E. histolytica* may interact and modulate the virulence of certain human viruses and is itself a
host for its own viruses. For example, AIDS accentuates the damage and pathogenicity of *E. histolytica* (Hung *et al.*, 2005). However, cells infected with HIV are often consumed by *E. histolytica*. Infective HIV remains viable within the amoeba, although fortunately there has been no proof of human re-infection from amoeba carrying this virus (Brown *et al.*, 1991).

A) Dysenteric and diarrhoeic syndromes account for 90% of invasive intestinal amoebiasis (Ryan & Ray, 2004). Their various clinical manifestations depend on where the lesion is located within the rectosigmoid or higher regions of the colon (Stenson *et al.*, 2001). In people with this form of the disease, rectosigmoidoscopy may reveal superficial ulcerations extending over limited areas of the terminal portion of the large intestine. Patients with dysentery have an average of 3-5 mucosanguineous evacuations per day, with moderate colic pain preceding discharge and they have rectal tenesmus (Sepulveda & Trevirio-Garcia 1986, Martinez-Palomo & Ruiz-Palacios, 1990).

B) Fulminating amoebic colitis is extremely severe, rapidly evolving clinical condition with necrotic ulcerous lesions extending over large areas, even the entire colon. Case fatality rates associated with amebic colitis range from 1.9%-9.1%. Amebic colitis evolves to fulminant necrotizing colitis or rupture in approximately 0.5% of cases; in such cases, the mortality rate jumps to greater than 40% (Aristizábal *et al.*, 1991). Evacuations, preceded by intense colic pain, are frequent (20 or more in 24 hours) and consist of faecal material mixed with blood or occasionally blood alone. Rectal tenesmus tends to be constant and acute. Systemic manifestations include abdominal discomfort, anorexia and nausea. High fever (39-40°C) is usually present, accompanied by a weak, rapid pulse and
low blood pressure. The patient suffers from dehydration and prostration and may even develop shock. Peritonitis is a common complication due to perforation of the intestinal wall (Sepulveda & Trevino-Garcia, 1986; Martinez – Palomo & Ruiz-Palacios, 1990).

C) The symptoms of amoebic appendicitis are similar to those of bacterial appendicitis; acute pain and rigidity in the lower right quadrant of the abdomen, fever, tachycardia and nausea. In more than two thirds of cases of amoeba appendicitis, patients have ulcerous lesions of the caecum. In these cases, diarrhoea, often with blood, is also present (Sepulveda & Trevino-Garcia, 1986; Martinez-Palomo & Ruiz-Palacios, 1990).

D) Amoebomas are pseudotumoral lesions whose formation is associated with necrosis inflammation and oedema of the mucosa and sub-mucosa of the colon. Amoebomas always co-exist with amoebic ulcerations (Stanley, 2003; Misra et al., 2006). These are generally single, but occasionally multiple, masses usually found in the vertical segments of the large intestine: the caecum, the rectosigmoid region of the colon, the ascending colon and the hepatic and splenic angles of the colon (Misra et al., 2006). The condition is usually acute, with dysentery or bloody diarrhoea, abdominal pain and a palpable mass in the corresponding area of the abdomen (Sepulveda & Trevino-Garcia, 1986; Martinez-Palomo & Ruiz-Palacios, 1990).

2.2.5 Occurrence and morphology of *Giardia lamblia*

Giardiasis is one of the most common intestinal parasitic infections in humans and is distributed worldwide in developed and developing countries (Lane & Lloyd, 2002; Thiongó et al., 2011). There are, however, differences in the number of infections not
only between countries, but within geographic regions. Infections seem to be more common in children than adults and other social, environmental, climatic and economic factors also play a role in disease prevalence. Susceptibility to infection with *Giardia* is influenced by sex, age, environmental conditions, socio-economic conditions, occupation, nutritional status, gastric acidity and overall host immune status (Lane & Lloyd, 2002; Ouattara *et al.*, 2010). The number of infective cysts shed in faeces is highly variable but has been estimated as high as 900 million a day for a human being (Yoder *et al.*, 2007). Transmission occurs through ingestion of as few as 10 viable cysts and can be acquired from food, water and person to person by the faecal-oral route (Yoder *et al.*, 2010). From available data, it appears the incubation time for giardiasis is 12-20 days (Schmerin *et al.*, 1978; Phillips *et al.*, 1981; Garcia, 1992).

The highest prevalence of *G. lamblia* occurs in the tropics and sub-tropics where sanitation is poor (Thiongó *et al.*, 2011). Travellers to tropical Africa, Mexico, Russia, South East Asia, Southern Asia and Western South America are at high risk of acquiring giardiasis (Wolfe 1992). Giardiasis, caused by *Giardia lamblia*, is a frequent cause of diarrhoea that can have a negative impact on growth and development of children (Yodallahie *et al.*, 2002; Simsek *et al.*, 2004). Giardiasis infects 200 million people worldwide and may produce symptoms in 500,000 individuals every year (Walsh & Warren, 1979; WHO, 1987, 1993; Mineno & Avery, 2003). In developing countries, *G. lamblia* is one of the first pathogens to infect infants and peak prevalence rates of 15-20% occur in children less 10 years old (Hill, 1993). In developed countries, outbreaks frequently occur in childcare settings (Thompson, 1994) and in adults who drink contaminated water. From 1977 to 1988, giardiasis was the leading cause of outbreaks of
water-borne diseases in the USA causing an estimated 1600 hospital admissions annually (Lengerich et al., 1994). Homosexual males constitute a special risk group.

2.2.6 Lifecycle of *G. lamblia*

The lifecycle of *G. lamblia* is similar to direct lifecycle of most intestinal protozoa (Fig. 2.3). Both the trophozoite and cyst are included in the lifecycle. Trophozoites divide by means of longitudinal binary fission producing 2 daughter trophozoites. The trophozoites are the intestinal dwelling stage and attach to the epithelium of the host villi by means of the ventral disc (Garcia et al., 1992). They may remain attached or may detach from the mucosal surface and since the epithelial surface sloughs off the villi every 72 hours, they apparently detach at that time. The most common location of the organism is the crypts within the duodenum.

Cyst formation takes place as the organism moves down through the colon. Based on results by Halliday et al. (1995), *Giardia* appears to take up conjugated bile salts by active and passive transport mechanisms like the mammalian ileum. As conjugated bile salts are known to promote encystation, these uptake mechanisms may play an important role in survival of the cyst stage and ultimate completion of the lifecycle of the parasite (Healy, 1990). Cholesterol starvation may also play a role in stimulating encystation (Lujan & Maria, 2003). As the trophozoites retract the flagella into the exonemes, the cytoplasm becomes condensed and as the cyst matures, the internal structures are doubled, so that when excystation occurs, the cytoplasm divides, thus producing 2 trophozoites. Excystation would normally occur in the duodenum or appropriate culture medium (Bingham & Meyer, 1979). If kept cool and moist, cysts can remain viable for several
months, but they cannot survive if moisture is lacking. The cysts may be either round or oval and contain 4 nuclei axonemes and median bodies (Garcia & Bruckner, 1997).

2.2.7 Clinical manifestations of *G. lamblia*

The onset of *Giardia lamblia* infection may be accompanied by nausea, anorexia malaise, low grade fever and chills. There may be a sudden onset of explosive, watery, foul-smelling diarrhoea with flatulence and abdominal distention. However, the type of diarrhoea and the lack of blood mucous and cellular exudate is consistent with giardiasis and as acute stage usually lasts only a few days, it may not be recognized as the cause of the symptoms observed, but may mimic acute viral enteritis, basically dysentery, bacterial or other food poisonings, acute intestinal amoebiasis or traveler’s diarrhoea (toxigenic *E. coli*). The acute phase is often followed by a sub-acute or chronic phase. Symptoms in these patients include recurrent, brief episodes of loose, foul stools; there may be increased distention and foul gases in the stomach and intestines (Ankarklev *et al*., 2010).
20

Contamination of water, food, or hands with infecrive cysts.

A Infective Stage
A Diagnostic Stage

Cyst

Trophozoites are also passed in stool but they do not survive in the environment.

Fig. 2.3: Lifecycle of *Giardia lamblia* (Adopted from CDC, 1999: [www.dpd.cdc.gov/dpdx/Giardiasis.htm](http://www.dpd.cdc.gov/dpdx/Giardiasis.htm))

2.3 Occurrence of helminthes

Nearly 200 species of helminthes have been found associated with human alimentary tract (Coombs & Crompton, 1991; Crompton, 1999), many of these probably being the result of accidental or spurious infections. In total there are 342 helminthe species reported from human hosts (Crompton, 1999). Helminthes which occur in humans comprise 4 major groups: 2 groups of flatworms, the trematodes and cestodes, the nematodes and the
acanthocephalans. The platyhelminthes or flatworms are bilaterally symmetrical, dosorventrally flattened worms with definite head end and lacking body cavity. These comprise 3 classes: the monogenea (mainly ectoparasites of fishes), the cestoidean (tapeworms, endoparasitic); and the trematode (endoparasitic flukes; mainly digeneans) and only the latter 2 classes infect humans. They are macroparasites and their bodies are covered by layers that are more complex and less vulnerable to immune attack (Anderson & May, 1985).

In Kenya, only 25% of the government health admission facilities reported on helminthiasis in the year 2006. The report indicates 5 and 4 morbidity cases occurred owing to ascariasis and ancylostomiasis respectively, but no mortalities. Due to low reporting and inconsistent recording, Nairobi Province had no specific data on helminthes as from 1996 to date. It was noted that, there are various problems that exist at different levels of the HMIS that explain in part, the incompleteness and lack of timeliness which characterize the Health Information Services data. At the source of data collection, there has been a severe shortage of reporting forms. In addition, in some areas there are inadequate appropriately trained personnel to compile data and factors that may be attributed to the varying response rates include inadequate reporting forms, insufficient funds for the follow-up and supervision purposes and inadequate staff for compiling the data particularly at the periphery (MoH, 2009).

2.3.1 Occurrence and distribution of Nematodes

Nematodes are probably the most abundant and widespread animal group, often occurring in huge numbers in environments ranging from the Polar Regions to hot springs. In
addition to free-living marine and freshwater forms, there are free-living forms in the soil and parasitic in both animals and plants. Of all known habitual parasites found in human alimentary tract, *Ancylostoma duodenale*, *Necator americanus* (soil transmitted Nematodes), *Trichuris trichiura* and *Ascaris lumbricoides* are the commonest infections (Crompton, 1990, 1999). The parasite *N. americanus* prevails in tropical and sub-tropical regions whereas *A. duodenale* tends to occur in the cooler and somewhat drier regions (Pawlowski *et al.*, 1991). The hookworm *N. americanus* occurs in the Americas, equatorial Africa, Southern Asia, South East Asia, Polynesia and Australia. It is more common in Africa and in northern and south-western Asia. The ranges of both species of hookworm often overlap (WHO, 1987).

Pinworms are the only species that are common in temperate countries and together with the soil transmitted species are widely distributed in countries with warmer climates, often occurring concurrently in individuals (Crompton, 1989, 1999). This means that 4 out of every 10 people in Africa, Asia and South America harbour worms. *A. lumbricoides* infections have been reported during the last few years from more than 150 of the 208 states and countries of the world. Hookworms, whipworms and *T. trichiura*, are as widely distributed as *Ascaris* (Bundy & Cooper, 1989, WHO, 1987). These parasites are soil transmitted helminthes of global importance. Strongyloidiosis has a patchy global distribution and is less prevalent than ascariasis.

In 1994, 1.47 billion people were infected by *A. lumbricoides*, 1.35 billion by *A. duodenale*, 735 million by *N. americanus* and 1.36 billion by *T. trichiura* while morbidity that was caused by these nematodes was 120 - 215 million cases by *A. lumbricoides*; 90-
130 by hookworms and 60-100 million by *T. trichiura* (Chan *et al.*, 1994, Crompton, 1999). Hookworm infection often causes iron deficiency anaemia, which may reduce productivity in affected individuals including food handlers (WHO, 1992). Unedited report for the year 2006, Nairobi Province reported 22,181 cases of intestinal worms but had no specific data on nematodes.

2.3.2 Morphology of Nematodes

Nematodes are symmetrically bilateral, unsegmented normally dioecious worms which are usually filariform; they have a body cavity with a high hydrostatic pressure, a straight digestive tract with an anteriorly terminal mouth and posteriorly sub-terminal anus, no circulatory system, a simple excretory system and body wall consisting of an outer layer of cuticle and an inner layer of longitudinal muscles.

2.3.3 Pathology of Nematodes

The pathogenicity of Nematodes in their final host varies considerably, usually being dependent upon the size of the infection. For intestinal nematodes, disability associated with higher worm burden thresholds persists as long as the individual is infected, whilst chronic disability which occurs in children with worm burdens above the lower threshold, is assumed to be life-long (Bundy *et al.*, 2004). Pathology due to *Ascaris* can result from pneumonia caused by the worm's migration through the lungs, blocking of the gastrointestinal track or the bile or pancreatic duct. *A. Lumbricoides* is physically indistinguishable from *Ascaris sum* (Crompton, 1999). Hookworms, which are heavily armed with teeth or other sclerotized mouth parts and browse upon the gut wall, can cause considerable damage. Those that migrate around the body both as adults and larvae can
cause serious problems, especially if they reach sensitive regions as the brain, liver or eyes. The growth and nutrition of children are adversely affected by *Ascaris* and *Trichuris* pertaining to appetite, food intake, digestion, absorption and growth (Cooper *et al.*, 1990, Stephenson *et al.*, 1980). *Ascaris* is mainly prevalent in young people (children aged 6-12 years). *Trichuris* infection peaks in early life and then generally remains steady, whereas hookworm prevalence peaks in adolescence. Heavy worm burdens of *Ascaris* and *Trichuris* are found in children of primary school age. Deaths due to *A. lumbricoides* and *T. trichuria*, are assumed to occur among 5–14 year olds while majority of hookworm-related deaths occur in sub-Saharan Africa (WHO, 1981, 1987, 2002).

### 2.3.4 Survival and transmission of *A. lumbricoides*

Adults of *A. lumbricoides* inhabit the small intestine in man. After males and females copulate, the latter lays eggs that have complex eggshells at the rate of 240,000 per day and are passed out in stools (Hoagland & Schad, 1978). The complex eggshells protect the larvae from mechanical and physical damage and the external mucopolysaccharide; that is, the cortical layer ensures that the eggs adhere to objects (Wharton, 1980). In areas where infection is endemic, infective eggs have been found adhering to money, furniture, door handles, fruits, and vegetables, fingers, cooking and eating utensils (Kagei, 1983) as well as contaminating soil in households, gardens and public parks (Wong & Bundy, 1990). Eggs hatch into larvae which moult once to attain second infective stage. Unfertilized and fertilized eggs may or may not possess a cortical layer. Typically, eggs in stool samples under microscopy observation in laboratory appear to be brown in colour, probably because they become stained by bile pigments while in the gastrointestinal tract.
The egg-shell of a fertilized egg consists of an inner lipid layer responsible for selective permeability (Perry & Clark, 1982), a chitin protein layer responsible for structural strength and an inner vitelline layer (Foor, 1967 & Wharton, 1980). The inner layer contains a remarkably resilient lipoprotein, known as ascaroide, which explains how the egg with enclosed infective larvae can survive formaldehyde, disinfectants and other destructive chemicals. The fertilized egg is frequently observed to have an even deposit of mucopolysaccharide on its outer surface. This deposit is obtained when the egg is passing through outer uterus of the female worm (Foor, 1967) and is responsible for its adhesive properties (Kagei, 1983).

Embryonated eggs of *A. lumbricoides* must usually be swallowed by the human host in order to initiate the outlet of an infection (Fig. 2.4). The larva escapes from the egg, passes through the intestinal wall and reaches the liver via the hepatic portal system (Rogers, 1960). The larval stages spend about 4 days in the liver and about 14 days in the lungs and then begin to re-enter the gut via the bronchi and trachea. At least 65 days must pass after infection before eggs are first detected in the stool (Takata, 1951). Some evidence suggests that the eggs can become airborne and be inhaled in dust, thus gaining access to the alimentary tract (Bidinger *et al.*, 1981). The eggs of *A. lumbricoides* are produced in vast numbers and although many will either perish during embryonation due to exposure to ultraviolet radiation or become inaccessible to humans through the activities of earthworm burrowing into the soil during rainfall, enough survive to ensure persistence of infection. A female lays 50-60,000,000 eggs in 9 – 12 months before it dies.
Transmission depends on swallowing this infective stage by uninfected human host in order to initiate the outlet of an infection from a contaminated environment (da Costa-Macedo & Rey, 1990; Nyarango et al., 2008). The morbidity is strongly related to the number of worms which an individual harbours (the intensity of infection) and that the presentation of morbidity is related to the chronology of infection, migration and development of parasites in the human host (Brooker and Bundy, 2008). Although there is no doubt that transmission nearly always depends on swallowing eggs from a contaminated environment, other routes of transmission may also exist (Crompton, 2001). Three case studies have shown that *A. lumbricoides* can occasionally pass the placenta (Chu *et al.*, 1972, Rathi *et al.*, 1981, da Costa – Macedo & Rey, 1990) and since larval *Ascaris* can be transplanted between hosts experimentally, there is even the slight chance that infections may be established inadvertently through organ transplant surgery. Any community lacking facilities for the safe disposal of human faeces will remain vulnerable to infection with *A. lumbricoides* and use of untreated night soil as a fertilizer for vegetable will probably increase the risk of infections (Mustafa *et al.*, 2001).
2.3.5 Transmission of *E. vermicularis*

Eggs are released through the female reproductive opening or when the worm dies and disintegrates on the surface of the skin, each female releasing an estimated 10,000 eggs.
Movement to the anal region causes local irritation and itching. When laid, the eggs already contain immature larvae and these complete developments very rapidly, maturing within 6 hours at body temperature (Cook, 1995). The eggs are sticky and readily adhere, but are easily dispersed since they are light. If the local environmental conditions are not too dry, they can survive for a considerable period of time.

The characteristics of the eggs, and the way in which they are released from females favour several routes of transmission. In children, the itching associated with the presence of female worms on the peri-anal skin prompts scratching as a result of which eggs adhere to the hands and fingers from where they are easily transferred to the mouth. Contamination of nightclothes or bedding can also result in eggs being transferred to the hands. The lightness of the eggs results in a wide dispersal in bedrooms and houses and accidental transmission to other family members can occur from these sources. In one school, counts of eggs present on the wall of the lavatory showed some 500 per square foot (Crompton, 1991). Eggs can become airborne when clothes, bedding or dust are disturbed. They may then contaminate food or hands of people in the vicinity, and it has also been suggested that airborne eggs can be inhaled and then swallowed.

2.3.6 Transmission of hookworms: *A. duodenale* and *N. americanus*

The third stage larvae of hookworms are responsible for transmission and the outlet of infections by skin penetration (Gibson, 1994; WHO, 1981) if present in the soil that has been contaminated by faecal matter, thereby infecting uninfected barefooted person (Fig. 2.5). They inhabit the small intestine of humans in their adult stage. After mating, a female hookworm lays eggs which are voided by human with hookworm infection.
Hoagland & Schad (1978) conclude that *A. duodenale* is an opportunistic species, partly because the third larval stages can penetrate skin or enter via the oral route. Recent evidence from China indicates that larval *A. duodenale* may pass from mother to infect the foetus in utero and therefore transmammary infection with *A. duodenale* cannot be ignored. This hookworm can also function as a food-borne infection. *N. americanus* is less of an opportunist than *A. duodenale* with less flexibility in infection process (Hoagland & Schad, 1978). However, soil transmitted helminthes have a common lifecycle (Appendix 4). Major complications associated with hookworms is anemia, but can be corrected through treatment (Brooker *et al.*, 2008).
2.3.7 Global Distribution of Cestodes.

The parasites, *Hymenolepis nana*, *Taenia saginata*, *Taenia solium* and *Diphylobothrium latum* are intestinal parasites that infect humans and are of regional or national relevance (WHO, 1987). Taeniasis, caused by *T. saginata* is cosmopolitan but most prevalent in African countries South of the Sahara, Eastern Mediterranean countries and in parts of the former Soviet Union. Lower prevalence are found in Europe, India and southern Asia, Japan, the Philippines and much of Latin America with lowest prevalence found in Australia, Canada and the USA (WHO, 1979). *Taenia solium* is mainly restricted to low
social economic areas of Central and Southern Africa, Mexico, Central and South America and Southern Asia with sporadic cases that are found in Southern Europe (WHO, 1979). Diphyllobothriosis, a parasitosis caused by flatworms of the genus *Diphyllobothrium*, is contracted by consuming raw or undercooked fish (Dupouy-Camet and Peduzzi, 2004). These parasites are found in Europe, North and South America and Asia. About 20 million people are infected worldwide (Chai *et al.*, 2005). Several species have been described that occur either in freshwater fish or sea fish; *D. latum* is the most common species infecting humans (Scholz *et al.*, 2009). This parasite is common in northern parts of North America and in the Great Lakes of USA (Gibson, 1994). Nairobi Province had no specific information on Cestodes despite reporting 17,858 and 22,181 cases of intestinal worms in unedited report for the years 2005 and 2006.

### 2.3.8 Prevalence of human intestinal Tapeworms

Prevalence of human intestinal tapeworms extent is not known, but it has been estimated that as many as 100 million people worldwide may be infected with either *T. solium* or *T. saginata* and about 20 million with the dwarf tapeworm, *H. nana*. None of the intestinal tapeworm infections is really life-threatening but the presence of the adult pig tapeworm *T. solium* increases the risk of acquiring cysticercosis by the uptake of ineffective eggs and an infection with the broad tapeworm *D. latum*, may give rise to pernicious anaemia.

Human adult tapeworms are flat worms with a length ranging from a few centimeters (*H. nana*) up to several metres (*Taenia* species) and are white or greyish to yellow. Tapeworms are found in the small intestines of humans in their adult stages. The adult
worm always consists of a head (scolex) and a neck region (the growth region) and if time and conditions allow, the neck continues in a series of proglottides (segments which together form a strobila or the body). The Cestodes or Tapeworms occur as intestinal parasites of all groups of vertebrates (Khalil et al., 1994). In some species, the scolex is dominated by bothria (tentacles), which are sometimes called "sucking grooves", and function like suction cups. Other species have hooks and suckers that aid in attachment. Cyclophyllid cestodes can be identified by the presence of four suckers on their scolex (Britannica, 2010). Tapeworms of sub-class Eucestoda which infect humans are all segmented as adults, with one or more copies of reproductive system in each segment (proglottid) along the body strobila. True tapeworms are exclusively hermaphrodites with complex reproductive systems. There is a common external opening for both male and female reproductive systems, known as genital pore, which is situated at the surface opening of the cup-shaped atrium (MacDougald, 2003). There are 2 orders involved: the Pseudophillidea, recognizable by the fact that the scolex lacks suckers and hooks, the attachment organ being a pair of bothria, and the Cyclophyllidea, whose scolex is armed with a ring of 4 suckers and sometimes one or more rings of optical hooks.

2.3.9 Global distribution of Trematodes

Trematodes are flatworms. Molluscs normally harbour their asexual generations and vertebrates harbour adult sexual stages. Fasciolopsis buski and Echinostoma ilocanum are intestinal flukes that infect humans and are distributed in Eastern Asia countries (WHO, 1996; Chai et al., 2009). In these countries, people eat fresh water plants like Eleocharis tuberosa, Trapa natans and fresh water snails which harbour the infective stages of these
trematodes. According to Health Management Information Systems based in Nairobi, Kenya had no report on trematodes.

Intestinal Trematodes which are food-borne infections and have their highest prevalence in South East Asia and in the Western Pacific; they are also present in areas of Africa, the Americas and Europe (Chai et al., 2009). The geographical areas involved are expanding as are the populations at risk as a result of improvements in transportation that favour population movements and trade. Of the approximately 70 species known to colonize the human intestine, only a few species are known to cause actual infection. Globally, it is likely that more than the estimated 40-50 million people are infected with intestinal trematodes, primarily infected via the food-borne route, of which approximately 15 million are in China (WHO, 1995; Lun et al., 2005; Chai et al., 2009).

2.3.10 Lifecycle and infection by Trematodes

Trematodes or flukes are flatworms which originally evolved as parasites of molluscs and virtually all species retain a molluscan element in their life history. There are 2 subgroups, the Digeneans and the Aspidogstrea, but only the former in humans and the latter in other mammals. They have asexual generations in molluscs and single sexual generation in vertebrates and are normally hermaphroditic, occasionally partly or entirely dioecious. Human infection occurs from ingestion of raw or uncooked fish, shellfish or aquatic plants that harbour the infective metacercariae (WHO, 1995; Toledo et al., 2006; Chai et al., 2009). Fasciolosis is caused by two species of liver fluke, *Fasciola hepatica* and *Fasciola gigantica*. *F. hepatica* has a cosmopolitan distribution due to its capacity to infect many different species and to the ability of the intermediate snail host to adapt to a
wide range of ecological niches (Mas-Coma et al., 2005). The lifecycle of trematodes usually involves three generations, two asexual generations in mollusc and one sexual generation in a vertebrate. Eggs leave the vertebrate with faeces and hatch to release a ciliated larva which penetrates a particular molluscan host. Within the mollusc, miracidium develops into mother sporocysts, then to daughter sporocysts and finally to rediae. Within this second pathogenetic generation, a larval stage (cercariae) develops. These larvae escape from the mollusc to the next host where they transform into metacercariae in invertebrates or upon vegetation. Humans get infected by feeding upon prey or vegetation harbouring metacercariae. Clinical signs of fasciolosis are caused by the migration of the young flukes through the liver causing abdominal pain, indigestion, weight loss, mild fever and malaise. Aberrant migrations of Fasciola can also occur (Le et al., 2007).

2.4 Diagnosis of intestinal parasites

2.4.1 Clinical diagnosis

Clinical features of helminthic infections include urticarial rashes, diarrhoea, abdominal pain, anaemia, dehydration and distended abdomen (Berger & Marr, 2006). Protozoal clinical symptoms include dysentery, fulminating colitis, amoebic appendicitis, rectal tenesmus, dehydration and nausea (Martinez-Palomo, 1982).

2.4.2 Laboratory diagnosis

This was done through qualitative direct stool examination and quantitative formal ether concentration methods. Smears were either iodine stained or examined without the stain. Identification of larvae, ciliates, helminthes eggs, and amoebic trophozoites and cysts was
done by use of a microscope (Cheesbrough, 2004). Identification of *G. lamblia* from non-pathogenic flagellates is that the former contains two nuclei while the latter have one nucleus. Identification of *E. histolytica* is based on the presence of trophozoites that contained ingested red blood cells or presence of cysts. Hookworms are identified by their motile larvae and/or eggs while *A. lumbricooides* is by the presence of their eggs which appear brown in colour since they are stained by bile pigments while in the gastrointestinal tract.

### 2.5 Control, prevention and treatment of parasitic diseases

Control and prevention of parasitic infections is possible through personal hygiene practices, health education, proper food preparation, improved water supply, improvement of environmental sanitation, poverty reduction and early detection and treatment of infected people (Martinez-Palomo, 1982). Broad spectrum drugs that are used to treat protozoal infections include Paramomycin, Diiodohydroxyquin, Metronidazole, Tinidazole and Omidazole (Ouattara *et al.*, 2010). Treatment for helminthic infections include Thiabendazole, Mebendazole, Pyrantel embonate, Albendazole and Praziquantel (Kurup & Hunjan, 2010).
3.1 Study site

The study was carried out in Nairobi, the capital City of Kenya. It lies at an attitude of 36° 50' East and latitude 1° 17' South. It is located 480 km inland Northwest of the Indian Ocean, 140 km south of equator and has an area of 684 sq km (Fig. 3.1). Except for the months of July and August, which are distinctly cool, the rest of the months are warm (Moss, 2000). The area generally receives average rainfall of over 1700 mm per annum distributed almost throughout the year although there are two rainy seasons, March to May (long rains) and October to November (short rains).
Map showing the spot location of Nairobi in Kenya

Key: A- Central Business District where high and middle class food outlets were sampled. B- Mathare area where low class food outlets were selected.

Fig 3.1: Study area where food outlets were selected and food-handlers recruited from the City of Nairobi (Moss, 2003- http://www.travelguide.com)

3.2 Study population

The Public Health Department in the City Council of Nairobi listed food-handlers working in food outlets by July 2006 to about 108,000. These people have been certified to work in food outlets and have been issued with medical certificates after being examined every six months. Food-handlers are people who are in direct contact with food, food ingredients or who come into contact with food contact surfaces.
3.2.1 Inclusion/exclusion criteria

Inclusion criteria included those who had valid medical certificates at the start of the study, who had consented and agreed to fill informed consent form (Appendix IV). Those who never consented, or were on antihelminthic/antiamoebic treatment and those who did not have valid medical certificates were excluded from the study.

3.3 Classification of food outlets

Food outlets were categorized into three classes; low, middle and high. They were represented by food kiosks, middle class eating-houses and hotels respectively. Kiosks are small shops where food and drinks are sold with an average of one to three food-handlers, while restaurants are medium eating place with an average of seven food handlers. Hotels are outlets where people are accommodated for a while and served with drinks and food. These usually have an average of 20-30 food-handlers. The high class food outlets sampled included Norfork, Sarova Panafric and Ambassadeur. Middle class sampled included Highland Restaurant in Afya Centre, Antonio’s along Kaunda street Nairobi and Malindi Dishes in Gaberone road. Low class eating places included Riverside, Rehema and Ghetto food kiosk among others in the expansive Mathare area. Lack of treated water, appropriate sanitary facilities and lack of personal hygiene can increase the risk of infection particularly protozoal related. The proximity to my working area and the University; and the diversity of social structure were the factors that made Nairobi City to be considered for this study.
3.4 Sampling method

Simple random sampling of food outlets was done. A simple numbering method was assigned to the outlets and a random number picked to represent the first sample and every third plant was selected starting from this. Systematic random sampling of food-handlers was used to get the required sample size and was based on the cooperation given by respective managements and their medically certified food-handlers. Where cooperation was poor, the next outlet in a given class was considered. Therefore, medically certified food-handlers who cooperated were considered for this study.

3.5 Sample size determination

The sample size (n) was determined using the formula as used by Fisher et al., (1998).

\[ N = \frac{Z^2 pqD}{d^2} \]

Where:

- \( Z \) = Critical value corresponding with the desired probability, \( P = 95\% \) (0.05)
- \( Z = 1.96 \)
- \( p \) = Characteristic probability (assume 0.5)
- \( q = 1 - p = 1 - 0.5 = 0.5 \)
- \( D \) = Design effect
- \( d \) = Level of significance (\( \alpha = 0.05 \))

\[ N = \frac{196^2 \times 0.5 \times 0.5}{0.05} = 385 \]

Statistics acquired from the City Council of Nairobi indicated that 50%, 30% and 20% of the food-handlers work in low, middle and high class food outlets. Therefore, probability
proportional to size sampling was done. Food-handlers from the three levels of food outlets; low, middle and high class at the ratios of 5:3:2 respectively were used to achieve required sample size. A sample size of 312 was arrived at: low, middle and high class food outlets were represented by 131, 105 and 76 food handlers respectively. Overall total was 183 males and 129 females (low- 81 males, 50 females; middle- 58 males, 47 females; high- 44 males, 32 females.

3.6 Sample collection and processing

Food outlets were visited according to the arranged schedule. The schedule was arranged such that specimens were collected in the morning only. Two low class eating places were sampled per week for 24 weeks to give a total of 131 food-handlers and one middle class food outlet per week for 17 weeks to give a total of 105 food-handlers. One high class food outlet was sampled every month for five months and 76 food handlers examined. The total number of food-handlers was 312.

On the first visit, the researcher introduced himself to the management and the food-handlers. On each sampling day, each participant was given one clean labelled specimen polypots to put fresh stools. Instructions were given on how to introduce samples into the polypots (Khurana et al., 2008). Pertinent data on name, age, sex, and specimen number of each participant were recorded on research report form (Appendix V). The fresh samples were packed into cool boxes and taken immediately to the laboratory and examined without delay using normal saline for amoebic trophozoites and live helminthes larvae. Specimens were later preserved in 10% formalin and smears of each stool sample done directly on the slide, stained with Lugol’s iodine solution and examined under the
microscope. The same samples were prepared using the concentration method, stained using Lugol's iodine and examined under the microscope. Two samples were collected from each food-handler in the morning at an interval of seven days (Guimarães & Sogayar, 1995) to capture light infections and irregular shedding/oviposition.

3.7 Laboratory investigations

All stool samples collected were subjected to both direct and formal-ether concentration techniques and tallied in the research report forms (Appendix V).

3.7.1 Direct microscopy technique

Direct technique of stool examination was carried out as outlined by Cheesbrough (2004). Two wet preparations were made as follows: a drop of fresh physiological saline was placed on one end of a microscopic glass slide and a drop of Lugol's iodine on the other. An amount of fresh stool specimen of about 0.25mg was picked with an applicator stick and mixed with formal saline and the same done with Lugol's iodine. The two were then covered with glass cover slips (22 x 22 mm) and examined under an ordinary electric light microscope under power 10 for ova and larvae, and power 40 for ciliates, trophozoites, cysts and oocysts. The microscopic examination commenced from the right hand corner of the cover slip horizontally to the left till the specimen covered was fully examined. The number of cysts and ova for each species found in the entire preparation was then reported as; scanty (1-3), few (4-10), moderate (11-20), many (21-40) and numerous (over 40) per preparation (Cheesbrough, 2004).
3.7.2 Formal ether concentration technique
Concentration technique was carried out as outlined by Cheesbrough (2004). Three grams of fresh stool sample were emulsified in 7 ml of 10% formal saline. The resultant suspension was sieved into a centrifuge tube using three layers of wet cotton gauze and 3 ml of diethyl ether added. The centrifuge tube was corked, shaken vigorously and then centrifuged at a centrifugal force of 906xg- 2516xg for 3 to 5 minutes. The plug of detritus at the interface between diethyl ether at the top and the formal saline was dislodged with an applicator stick and poured off. The deposit was resuspended by shaking the tube. A drop was put on a glass slide and covered using a glass cover slip (22 x 22 mm) and examined for larvae, ciliates, helminthe eggs, cysts and oocysts as in direct technique. Two preparations were examined for each sample.

3.8 Ethical clearance
Permission was granted by Kenyatta University Institutional Review Board, Ministry of Science, Research and Technology, respective management of the food outlets and Public Health Department, City Council of Nairobi (Appendix I, II & III).

3.9 Data analysis
SPSS version 16 was used to analyze the data obtained. Fisher’s exact test was used to test for any significant association between parasite prevalence, types and pattern in certified food-handlers working in low, middle and high class food outlets. Parasite relationships to age strata, education and gender were also determined.
CHAPTER FOUR
RESULTS

4.1 The types and number of food outlets that participated
Seventy food outlets were visited in Mathare, Nairobi Central Business District and its environs. In Mathare, 68.6% of food outlets represented low class while 24.3% and 7.1% of food outlets represented middle and high class respectively.

4.1.1 Types of parasites found infecting medically certified food handlers
A total of 81% of the expected food-handlers had their stool samples examined. This total represented 42%, 33.6% and 24.4% of the food-handlers in low, middle and high class respectively. Males were 58.7% and females 41.3% of the sampled population. Forty nine cases were found infected with intestinal parasites namely *E. histolytica*, 12.5%, *A. lumbricoides*, 1.9% and *G. lamblia*, 1.3% (Fig 4.1). Out of this, *E. histolytica* represented 74.4%, 23.1% and 2.6% cases in low, middle and high class respectively. There were also *A. lumbricoides* ova with 83.3%, 16.7% and 0% cases and *G. lamblia* cysts with 75%, 25%, and 0% cases in low, middle and high class respectively. Distribution of the parasites between gender and the three classes of the food outlets is as shown in table 4.1. Males found infected were 15.8% as compared to 15.5% of females. A total of 84.3% of the total population sampled was not infected. There was however no statistical significance between the infection and the classes of the food outlet sampled ($\chi^2 = 0.627$, $P = 0.735$).
Total intestinal parasites - 15.7%: *E. histolytica* - 12.5%, *A. lumbricoides*, 1.9%, *G. lamblia*, 1.3%

Fig. 4.1: Overall prevalence of intestinal parasites in the sampled food-handlers

Table 4.1: Distribution of *E. histolytica*, *A. lumbricoides* and *G. lamblia* in the different classes of food outlets and between genders

<table>
<thead>
<tr>
<th>Class</th>
<th>Gender</th>
<th><em>E. histolytica</em></th>
<th><em>A. lumbricoides</em></th>
<th><em>G. lamblia</em></th>
</tr>
</thead>
<tbody>
<tr>
<td>Low</td>
<td>Male</td>
<td>5.7</td>
<td>0.64</td>
<td>0.64</td>
</tr>
<tr>
<td></td>
<td>Female</td>
<td>3.53</td>
<td>0.96</td>
<td>0.32</td>
</tr>
<tr>
<td>Middle</td>
<td>Male</td>
<td>1.28</td>
<td>0.32</td>
<td>0.32</td>
</tr>
<tr>
<td></td>
<td>Female</td>
<td>1.6</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>High</td>
<td>Male</td>
<td>0.32</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>Female</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

N= 312 (*P*>0.05, Fisher's exact test)
4.1.2 Gender and age distribution of food-handlers and association with intestinal parasites

In the sampled population, there were 41% females, and 59% males in the three categories of food outlets indicating that more males were captured than females (Fig. 4.2). The gender distribution amongst the recruited from these food outlets was that males were more with 25.9% than females who had 16% in the low class, 18.6% and 15.1% in middle class, and 14.1% and 10.3% respectively in high class. The youngest age was 18 years and the oldest recorded was 70 years. Mean and Standard Deviation for gender was 1.41 and 0.493 respectively. The gender distribution was not statistically significant amongst the classes ($P = 0.49, \chi^2 = 0.96$) (Fig. 4.3) and the parasitic infection ($P = 0.449, \chi^2 = 0.575$).

![Gender Distribution of Male and Female Food-handlers in the Study Sample](image)

Fig. 4.2 Percentage of male and female food-handlers in the study sample
4.1.3 Level of education of food-handlers and association with intestinal parasites

Data on education levels indicated that 21.8%, 68.6% and 9.6% had primary, secondary and university education respectively. Education status in the three classes of food outlets are as shown in figure 4.4. Parasites detected in those who attended primary school were-

- *E. histolytica* 19.1%, *A. lumbricoides* 2.9% and *G. lamblia* 1.5%, while the ones detected in those who attended secondary education were 7%, 1.4% and 0.93%, and university, 36.6%, 3.3% and 3.3% for *E. histolytica*, *A. lumbricoides* and *G. lamblia* respectively.
However, education levels of the food handlers had no significant correlation with parasite infection rates ($P = 0.36, \chi^2 = 2.99$).

![Bar chart showing frequency of education levels of food handlers in three classes of food outlets.](chart)

**Fig. 4.4 Frequency of the levels of education of food-handlers in the three classes of food outlets**

**4.1.4 Observation period in relation to the last medical examination of food-handlers and the association with intestinal parasites**

Forty stool samples were collected from selected food-handlers on a weekly basis, examined and data collected and recorded. A second sample from the same individuals was also collected and analyzed after one week. More parasites were found infecting
those in age stratum 21-30 years of age. The ones with the least infections were those in
the over 40 years and above, but the number was comparatively low. Those that were in
their sixth month of the validity period were deliberately excluded from the study since
preliminary investigations revealed that those who did not consent had gone for
unscheduled medical check up and were unwilling to participate. Those who were in their
first month were 31.7%, 2nd month 19.9%, 3rd month 14.4%, 4th month 18.6% and 5th
month 15.4%. The study however, established that irrespective of the differences in
months, there was no statistical significant difference in the distribution of the parasites
\( \chi^2 = 5.38, P = 0.497, \) at 95%CI), and had no significant correlation with the classes of the
food outlets \( (P = 0.294, \chi^2 = 0.11). \)

4.2 Prevalence of intestinal parasitic infections in different food outlets

The intestinal parasites found in food-handlers were \( E. \ histolytica, \ A. \ lumbricoides \) and
\( G. \ lamblia. \) The positive cases were distributed in both males and females. It was noted
that the highest cases were those of \( E. \ histolytica \) in the low class food outlets with 9.29%
cases followed by the same in the middle class with 2.88% and high class with 0.32%
(Table 4.2). The types of parasites in the study population were varying in the occurrence.
The three parasites, \( E. \ histolytica, \ A. \ lumbricoides \) and \( G. \ lamblia \) were more prevalent in
those who had university education than primary and secondary. This was probably due to
the larger number of the respondents relative to the figures in the other levels. Among the
three levels of education, university level had the highest prevalence \( (E. \ histolytica-
36.7%, \ A. \ lumbricoides-3.3\% \) and \( G. \ lamblia-3.3\%). \) Secondary graduates were infected
with 7% \( E. \ histolytica, \ 1.4\% \ A. \ lumbricoides \) and 0.9% \( G. \ lamblia \) while primary had
19.1% positive cases for \( E. \ histolytica, \ 2.9\% \ A. \ lumbricoides \) and 1.5% \( G. \ lamblia. \) It was
evident that the occurrence of *E. histolytica* was higher than occurrence of *A. lumbricoides* and *G. lamblia* in the three levels. However, the difference was not statistically significant ($\chi^2 = 0.627, P = 0.735$) in all the outlets and levels.

Table 4.2: Prevalence of intestinal parasites in the various food outlets

<table>
<thead>
<tr>
<th>Class of food outlets</th>
<th>Low</th>
<th>Middle</th>
<th>High</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>E. histolytica</em></td>
<td>9.29*</td>
<td>2.88</td>
<td>0.32</td>
</tr>
<tr>
<td><em>A. lumbricoides</em></td>
<td>1.6</td>
<td>0.32</td>
<td>0</td>
</tr>
<tr>
<td><em>G. lamblia</em></td>
<td>0.96</td>
<td>0.32</td>
<td>0</td>
</tr>
</tbody>
</table>

*Figures are in percentages relative to the total samples examined*

4.2.1 Prevalence of parasites in different age strata

The prevalence was reported in clustered age strata of 18-20 years, 21-30, 31-40 and 41 years and above to both male and female groups, and pegged to the respective food outlets. It was noted that, the overall elevated parasite occurrence was mainly noted in those in the age bracket of 21 – 30 where males were found harbouring a higher percentage of *E. histolytica* than females. In the age stratum 18-20 years, *E. histolytica* cases were 16.7%; 7.2% males and 9.5% females, while 16.3% cases; 10.6% males and 5.7% females were detected in 21-30-year age stratum. In the 31-40 age stratum, only 7.9% cases were detected; 4.5% males and 3.4% females. In the 40 and above age stratum 4.9% were detected; 2.4% from each gender. Cases of *A. lumbricoides* were only detected
in the 21-30; 0.7% from each gender and in 31-40 age stratum; 2.3% from each gender.

One point four percent of *G. lamblia* were detected in males in the 21-30 age stratum while 2.3%; 1.1% males and 1.1% females in 31-40 age stratum. The respondents who were in the age stratum less than 20 years were 13.5% in total; low class- 6.4%, middle class- 4.5% and high class 2.6%. Those who were in age 21-30 years were 45.2%; 19.2% in low, 16.7% in middle and 9.3% in high classes. A total of 28.2% respondents were in the 31-40 age stratum; 12.2% in low, 6.7 in middle and 9.3% in high class, while those in the over 40 year-group were 13.1%; 4.2% in low, 5.8% in middle and 3.2% in high class.

There were more male respondents than females. The parasite infection rate was significantly correlated with the age strata (*P* = 0.039, *χ*² = 4.20) (Fig. 4.5).

![Fig 4.5: Prevalence of parasites in various age strata. *P*=0.039, *χ*² = 4.20, *E. h* = *Entamoeba histolytica*, *A. l* = *Ascaris lumbricoides*, *G. l* = *Giardia lamblia*. M= male, F= female](image-url)
4.3 Overall distribution pattern of parasitic infections in medically certified food-handlers

The overall distribution of the parasites in the different months in the three classes of food outlets is shown in figure 4.6. Data on parasite pattern showed no significant variation amongst food-handlers \( (F = 1.779, df = 2, P = 0.248) \) in the three different types of food outlets in Nairobi. In the low class, parasites were 59.2%, 10.2% and 6.1% for \( E. \) histolytica, \( A. \) lumbricoides and \( G. \) lamblia respectively. Middle class had 18.4%, 2.0% and 2.0% while high class had 2.0%, 0% and 0% for \( E. \) histolytica, \( A. \) lumbricoides and \( G. \) lamblia respectively. This study established that there was no significant difference in parasites distribution pattern \( (F = 1.779, P > 0.05) \) (Table 4.3) and between classes \( (P= 0.735, \chi^2 = 0.627) \).

Fig: 4.6: Overall parasite distribution pattern from 1st- 5th month in low, middle and high class food outlets
4.3.1 Types and prevalence of parasites encountered in the 5 months’ period in low, middle and high class food outlets

The findings from this study indicated that the parasite counts did not significantly vary with the span as from the last examination date ($r = 0.218, P > 0.05$). This shows that parasite infection in individual samples recorded after any given time within the five months period did not vary. However, the fifth month recorded the highest total parasite representing 34.7% while the least was recorded in the 1st month with 2.04 % of the total parasites encountered (Table 4.3)

Table 4.3: Types and prevalence in percentages of parasites encountered in the five months period in low, middle and high class food outlets (N=49)

<table>
<thead>
<tr>
<th>Class</th>
<th>Parasite</th>
<th>Duration* (months)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1st</td>
<td>2nd</td>
</tr>
<tr>
<td>Low</td>
<td>E. histolytica</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>A. lumbricoides</td>
<td>2.04</td>
</tr>
<tr>
<td></td>
<td>G. lamblia</td>
<td>0</td>
</tr>
<tr>
<td>Middle</td>
<td>E. histolytica</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>A. lumbricoides</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>G. lamblia</td>
<td>0</td>
</tr>
<tr>
<td>High</td>
<td>E. histolytica</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>A. lumbricoides</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>G. lamblia</td>
<td>0</td>
</tr>
</tbody>
</table>

*Duration = the number of months after the last medical examination

$P>0.05$, Fisher’s exact test.
CHAPTER FIVE
DISCUSSIONS

5.1 Intestinal parasitic infections identified in medically certified food-handlers

This study has shown that *E. histolytica* and *G. lamblia* are some of the intestinal protozoa which infect food-handlers in food outlets in Nairobi Kenya. These pathogenic intestinal parasites had a rate of 12.5% for *E. histolytica* and 1.28% for *G. lamblia*. An intestinal Nematode, *A. lumbricoides* was also found infecting the food-handlers with an overall of 1.92%. The finding from this study showed *E. histolytica*, *A. lumbricoides* and *G. lamblia* as the intestinal parasites found infecting some selected and medically certified food-handlers during validity period of their medical certificates in three classes of food outlets selected on the basis of the type of license issued. These classes were low, middle and high. The findings were of particular significance since infected food-handlers may be at risk of developing illness themselves, and may be a threat to the health of their clients (Esparar *et al.*, 2004).

Protozoans discussed in this study are transmitted via the faecal-oral route. *Giardia* has been reported to cause diarrhoea, and its presence is typically associated with upper gastrointestinal symptoms while *E. histolytica* is associated with invasive intestinal amoebiasis (Martinez-Palomo & Ruz-Palacios, 1990; Ouattara *et al.*, 2010; Thiongó *et al.*, 2011). *A. lumbricoides* and other intestinal helminthes are associated with intestinal disturbances, growth retardation in children and a host of other disorders. Although the overall parasite occurrence was 15.7%, this was not as high as the values of another similar study done in Northwest Ethiopia by Andargie *et al.*, (2008) which found 29.1% of
food-handlers infected. Also, Al-Lahham et al, (1990) conducted a similar study in Jordan where the frequency of infection by intestinal protozoa was 30.2%, far much higher than in the just concluded study where 13.8% represented the same. The reason could be due to dissimilar socio-economic status and environmental conditions associated with the two diverse populations.

Although education levels could influence the pattern of infection, not all of the participants had access to formal health education on food safety. Those who had access to primary school education were 21.8%, secondary education, 68.6% and only 9.6% had university education and were distributed among the food outlets. Many food-handlers did not seem aware of basic safety and health requirements to work with food and food products. A mere 14% conceded having undergone an intensive food safety course. There was, however, no statistical significance \((P > 0.05)\) between infection and education of the respondents. Studies by Babiker et al, (2009) found that the infectivity of intestinal helminthes in Sudanese food-handlers was 2.7%, and was similar to the findings of the present study which realized 1.92%.

5.2 The prevalence of intestinal parasitic infections in medically certified food-handlers

In this study, it has been shown that intestinal parasites are prevalent in food-handlers with valid medical certificates regardless of the class of the food outlet they work in.

In Kenya, medical certificates issued to food-handlers have a validity period of six months as per CAP 254; Food Drug and Chemical Substances Act. These certificates are normally issued to those food-handlers who test negative to intestinal parasites among other tests. It
is however, evident that within the six months' validity period of these issued certificates, the food-handlers can get infections and also be a source of infection to others. A study conducted in Malaysia showed that approximately 10-20% of food-borne disease outbreaks are due to contamination by food-handlers (Zain & Naing, 2002). Another similar study conducted by Abera et al, (2010) revealed that 25; 6.5% of the food-handlers working in kitchens in Bahir Dar Town, Northwest Ethiopia were suffering from diarrhoea. Active trophozoite forms of *E. histolytica*, *G. lamblia* and larva of *S. stercoralis* were found associated with diarrhoeic food-handlers. *Giardia lamblia* infected food-handlers can directly transmit *Giardia* to consumers if ingested via contaminated food and water since the parasite does not require environmental maturation. Moreover, Mintz et al, (1993) found that food-handlers infected with *G. lamblia* were vehicles for *Giardia* outbreak in commercial food outlets. Thus, food-handlers should be in good health and those suffering from diarrhoea must be excluded from work until they have been examined, treated and completely free of symptoms. It was noted that infection by *G. lamblia* (1.3%) was far much less than that of *E. histolytica* (12.5%) and *A. lumbricoides* (1.9%) in the just concluded study. This is in agreement with a study conducted by Abera et al, (2010) in Ethiopia although their figures were slightly higher (*G. lamblia* 7%, *E. histolytica*, 12.76 % and *A. lumbricoides*, 11.7%).

In the present study, male respondents were more; 59% than females; 41% in the three classes of food outlets. There was no statistical significance in infection due to gender or span. The prevalence was such that more parasites, particularly *E. histolytica*, were found in low class food outlets than in middle or high class. More males were found infected than females. There was, however, no significant statistical difference between parasite
occurrence and the degree of infection. The infection rate was elevated in the 21-30 age strata; 55.1% and least in the over 40 age; 4%. There was no significant statistical difference in the prevalence of intestinal parasitic infections amongst food-handlers in all the food outlets ($P > 0.05$). The techniques used, which were direct stool smear and formal-ether were both vital in that the former was convenient for motile amoebic trophozoites while the latter was convenient for helminthic ova and detection of light helminthic and amoebic infections. At least two stool specimens per individual were conveniently examined to capture light infections. The need for at least two specimens may be further justified by the observation that there are alternating periods of high and low excretion of protozoan cysts. Previous studies conducted have shown that the prevalence of infection varies and depends largely on the methods employed and the number of examinations made in individual cases (Danciger & Lopez, 1975, Esparar et al., 2004). The significant difference in the recovery rates between stool examinations done once against that, which is done twice, implies the probable need to examine more than one specimen on different days from food-handlers.

5.3 The pattern of intestinal parasitic infections among food-handlers

The pattern of the infections did not vary greatly although more parasites were detected towards the final months before the next examination was due. No association was established between the frequency of parasitic infection with age, sex, span and class of food outlet. This illustrates equal exposure to infection/s undoubtedly; continuous health supervision, regular medical examination and prompt treatment of infected food-handlers minimize the infection rates (Babiker et al., 2009).
A food-handler infected with helminthes that are soil-transmitted cannot be a source of infection to other people. This is due to the fact that stages that are produced within the host or pass to the exterior to complete development to the next stage have complex life cycles which may involve more than one host before infective stage is attained (Crompton, 1991). However, other helminthes including Enterobius vermiculuris and Taenia solium can be transmitted directly through contamination but were not encountered in the study. All food-handlers whose stool samples turned positive for parasitic protozoans were potential source of infection to uninfected persons. Transmissions and infections by G. lamblia and E. histolytica are similar, by ingestion of as few as 10 viable cysts that can be acquired from drinking water, food and person to person through faecal-oral route (Garcia et al., 1992; Thiongó et al., 2011). A person gets infected by ingestion of viable A. lumbricoides eggs in especially undercooked vegetables and salads. Parasite infection was shown to have no significant difference in variation in the three classes of food outlets although parasite pattern depended on the number of viable cysts/ova ingested.

Although it was expected that some seasonal variation in transmission and maintenance of intestinal helminthes may feature here, the present study failed to detect any significant variation between occurrences of infection in different months. This is in agreement with a similar study done in Khartoum by Babiker et al., (2009), but contradicts a study done in Nigeria by Nzeako, (1992) who reported seasonality of infection with intestinal helminthes.
In summary, the discrepancy in lack of personal hygiene may explain the current situation of these intestinal parasitic infections in the completed study. The prevalence of intestinal parasites may have been due to poor personal hygiene practices and environmental sanitation, lack of safe water, ignorance of health-promotion practices, and poor health services (Nyarango et al., 2008; Andargie et al., 2008; Thiongó et al., 2011).
CHAPTER SIX

CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusions

Based on the results of this study, the following conclusions have been made:

i. Intestinal parasites were found in medically certified food-handlers from the first to the fifth month of the validity period of the medical certificates and across all the classes of the food outlets.

ii. Intestinal parasites identified from the medically certified food handlers were *E. histolytica*, *A. lumbricoides* and *G. lamblia*.

iii. The parasite distribution pattern was found to be progressive from the first month to the fifth month of the validity period of the medical certificate, and more cases were found in the 21-30-year age stratum. Low class food outlets had more parasites than middle or high class.

6.2 Recommendations

Based on the above conclusions, the following are therefore recommended:

i. Encourage high level of personal hygiene through health education and improvement of environmental sanitation.

ii. Enforce hygienic practices in all classes of food outlets through scheduled inspections and food handling safety courses to ensure food safety during processing, preparation and storage in food service outlets.
iii. Validity period of the medical certificates should be revised from 6-3 months from the prescribed FORM ‘D’ of the first schedule; Regulation 15 (3)- Food Hygiene Regulations of CAP 254, Laws of Kenya.

iv. Further studies should be carried out especially on laboratory quality control in personnel competency, quality results, reagents and standard equipments.
REFERENCES


Kagei, N. (1983). Techniques for the measurement of environmental pollution by
infected stages of soil-transmitted helminthes. Collected papers on the control of soil transmitted helminthiases 24-46.


APPENDICES

Appendix I: Research authorization

MINISTRY OF SCIENCE & TECHNOLOGY

Telegrams: SCIENCE TEC", Nairobi

Fax No. Telephone: 318581
When replying please quote

MOST 13/001/36C 357/8

Paul Njuguna Kamau
Kenyatta University
P.O. Box 43844
NAIROBI

Dear Sir,

RE: RESEARCH AUTHORIZATION

Following your application for authority to carry out research on, 'Prevalence of Intestinal Parasites in Food Handlers with Medical Certificates in Nairobi, Kenya.'

I am pleased to inform you that you have been authorized to carry out research in Nairobi City for a period ending 30th October 2008.

You are advised to report to the Provincial Commissioner and Provincial Director of Education Nairobi before embarking on your research project.

On completion of your research, you are expected to submit two copies of your research report to this office.

Copy to:

The Provincial Commissioner
NAIROBI

The Provincial Director of Education
NAIROBI
Appendix II: Research clearance permit

CONDITIONS

1. You must report to the District Commissioner and the District Education Officer of the area before embarking on your research. Failure to do that may lead to the cancellation of your permit.

2. Government Officers will not be interviewed without prior appointment.

3. No questionnaire will be used unless it has been approved.

4. Excavation, filming and collection of biological specimens are subject to further permission from the relevant Government Ministries.

5. You are required to submit at least two (2) four (4) bound copies of your final report for Kenyans and non-Kenyans respectively.

6. The Government of Kenya reserves the right to modify the conditions of this permit including its cancellation without notice.

This is to certify that:

Prof./Dr./Mr./Mrs./Miss............PAUL NJUGUNA...........

of (Address)........ÊKENYATTA UNIVERSITY........

P.O. BOX 43844 NAIROBI

has been permitted to conduct research in..............

NAIROBI........Location.

NAIROBI........District.

NAIROBI........Province,

on the topic........PREVALENCE OF INTESTINAL

PARASITES IN FOOD HANDLERS

WITH MEDICAL CERTIFICATES IN

NAIROBI - KENYA

..................................................

for a period ending .........................., 2008.

Research Permit No............MOST13/001/37c 226

Date of issue............7/5/07

Fee received............SHS 500

PAGE 3

B.O. ADEWA

Applicant's Signature

Permanent Secretary

Ministry of Science and Technology
Appendix III: Research authorization, City Council of Nairobi

MEDICAL OFFICER OF HEALTH
TELEGRAMS: "MUNICIPALITY" NAIROBI
CITY HALL EXT. 2040...
P.O. BOX 30108
NAIROBI

KENYA

PUBLIC HEALTH DEPARTMENT

Ref: PHD/MOI/RI VOL.I(17)

14 March, 2006

Paul K. Njuguna,
Reg. No. 156/11497/04
Kenyatta University
NAIROBI.

SUBJECT: RESEARCH – PREVALENCE OF INTESTINAL PARASITES IN FOOD HANDLERS IN NAIROBI

Your letter dated 27th February, 2006 on the above subject matter refers.

I am pleased to inform you that your request for research has been accepted.

This is however subject to:

i. Payment of KSHS.1, 200/= research fee.

ii. Submission of a copy of your findings to the office of the undersigned after the completion of your research.

By a copy of this letter, the relevant section is requested to accord you the necessary assistance.

DR. L.I. MUNENE,
FOR: MEDICAL OFFICER OF HEALTH
C.C.:
Central Administration
EDC
Central District Administrator
MINISTRY OF PUBLIC HEALTH AND SANITATION
P.O Box 30016
Nairobi, Kenya

INFORMED CONSENT FORM

Informed consent form for certified food-handlers working in food outlets in the City of Nairobi and who we are inviting to participate in research on intestinal parasites infecting food-handlers.

Project title: PREVALENCE OF INTESTINAL PARASITES AMONG MEDICALLY CERTIFIED FOOD-HANDLERS IN NAIROBI, KENYA

Principal Investigator: Paul Njuguna Kamau

This Informed Consent Form has two parts:
- Information Sheet (to share information about the research with you)
- Certificate of Consent (for signatures if you agree to take part)

You will be given a copy of the full Informed Consent Form

PART I: Information Sheet

Introduction

I am Mr. Paul Kamau (Public Health Officer) working in this City for the Ministry of Public Health & Sanitation and attached/seconded to the Department on Environment, City Council of Nairobi, Kenya. We are doing research on intestinal parasites which infect food-handlers and other people who take food or drink in eating places causing diseases and are very common in this country. I am going to provide you with some information and invite you to be part of this research study. You have a right to decide whether or not you will participate in the research. Before you decide, you can talk to anyone you feel comfortable with about the research.

There may be some words that you do not understand. Please ask me to stop as we go through the information and I will take time to explain. If you have questions later, you can ask me, the study crew or other senior staff in this facility.
Purpose of this research

Intestinal parasitic infections are equally widespread to both children and adults and cause diseases which include stomachache, diarrhoea and headache. People who eat or drink in your outlets are in danger of contacting these parasites if you are infected since you can transmit to them. The Kenya Government also wants to eradicate diseases in line with the Vision 2030 for a healthy and productive nation. The reason we are doing this research is to find out if food-handlers like you get these parasites before the expiry of the medical certificates you have been provided so that a program can be put in place to stop any form of infection, and improve your health and that of your clients.

Type of Research Intervention

This research will involve collection of stool samples from the participating persons with valid medical certificates and will be done twice- one very early in the morning then the other one after seven days. You will be shown how to collect the samples.

Participant selection

We are inviting all men and women with valid medical certificates and those who have not taken any amoebic or helminthic drugs within the validity period of the certificate and who will agree to participate in this research.

Voluntary Participation

Your agreement to be a participant in this research is entirely voluntary. It is your choice whether to agree or not. If you choose not to agree all the services you receive at any health facility/user point will still continue and nothing will change.

Procedures

We are asking you to help us learn more about intestinal parasite infections in food-handling. We are inviting you to take part in this research project. If you accept, you will be asked a few personal questions and about residence, education and to provide us with your stool specimens. We will not ask you to share personal beliefs, practices or stories and you do not have to share any knowledge that you are not comfortable sharing. You may answer the questionnaire yourself, or it can be read to you and you can say out loud the answer you want me to write down. If you do not wish to answer any of the questions included in the survey, you may skip them and move on to the next question. The information recorded is confidential, your name is not being included on the data forms for analysis, only a number will identify you, and no one else except me and the researchers will have access to your information. Your name will only appear on the preliminary form for treatment purposes.

Duration

The research will only entail collection of two stool specimens per person. The results and treatment if any will be offered the next day after stool examination. Treatment will be administered in this facility once the results are released from this study to those who will be found infected with either worms or amoeba.
Risks

There is no imminent risk unless of course you feel uncomfortable when answering personal questions, which you can refuse to answer, or when collecting stool specimens.

Benefits

There will be no direct benefit to you, but your participation is likely to help us find out more about how to prevent and treat diseases caused by amoeba or worms in food-handlers. However, you will receive free treatment if found infected with amoeba or intestinal worms, and will be taught ways to prevent infection, better health care and quality hygiene practices.

Reimbursements

You will not be provided any incentive to take part in the research study. However, we will give you Ksh 200/= and reimburse your travel expenses when you come back to check on the results and treatment if you are not on duty that day.

Confidentiality

The research being done may draw attention and if you participate you may be asked questions by other people in this outlet. We will not be sharing information about you to anyone outside of the research team. The information that we collect from this research project will be kept private. Any information about you will have a number on it instead of names. Only the researchers will know what your number is and we will lock that information up with a lock and key. It will not be shared with or given to anyone.

Right to Refuse or Withdraw

This research is voluntary and you are under no obligation to participate. This will not affect in any way services rendered to you in any health institution.

Who to Contact

If you have any questions, you can ask them now or later. If you wish to ask questions later, you may contact any of the following:

1. Mr Paul Njuguna Kamau, Environment Department, City Council of Nairobi, P.O Box 30108-00100 Nairobi, Kenya. E-mail: pankam2005@yahoo.com
2. Prof. Peninah Oloo-Obudho, Moi University, P.O Box 1957-10101 Karatina, Kenya. E-mail: aloopenina@yahoo.com
3. Prof. Ephantus Kabiru, Kenyatta University, P.O Box 43844-00100 Nairobi, Kenya. E-mail: ewkabiru@yahoo.com

This proposal has been reviewed and granted by the Ministry of Education, Science and Technology; & the City Council of Nairobi, which are committees whose task are to make sure that research participants are protected from harm. If you wish to find about more about the Institutional Review Board, contact, The Chief Public Health Officer, Ministry of Public Health and Sanitation, P.O Box 30016, Nairobi, Kenya.

You can ask me any more questions about any part of the research study, if you wish to. Do you have any more questions please?
PART II: Certificate of Consent

I have read the foregoing information, or it has been read to me. I have had the opportunity to ask questions about it and any questions that I have asked have been answered to my satisfaction. I consent voluntarily to participate in this research.

Print Name of Participant ____________________________________________
Signature of Participant ____________________________________________
Date ____________________________________________________________
Day/month/year

Witness

I have witnessed the accurate reading of the consent form to the potential participant, and the individual has had the opportunity to ask questions. I confirm that the individual has given consent freely.

Print name of witness ___________________________ AND Thumb print of participant
Signature of witness ____________________________________________
Date ____________________________________________________________
Day/month/year

Statement by the researcher/person taking consent

I have accurately read out the information sheet to the potential participant, and to the best of my ability made sure that the participant understands that the following will be done:
1. A questionnaire will be filled
2. Stool samples will be collected
3. Sick food-handlers will be treated for free

I confirm that the participant was given an opportunity to ask questions about the study, and all the questions asked by the participant have been answered correctly and to the best of my ability. I confirm that the individual has not been coerced into giving consent, and the consent has been given freely and voluntarily.

A copy of this ICF has been provided to the participant.

Print Name of Researcher/person taking the consent ____________________________
Signature of Researcher/person taking the consent ____________________________
Date ____________________________________________________________
Day/month/year
Appendix V: Research report form

a) Name of food outlet .................................................................
b) Classification of food outlet: -
   Low class .................................................................
   Middle class ..............................................................
   High class .................................................................
c) Name of food-handler ..............................................................
d) Sex: male ( ) Female ( ) (Tick one) ................................................
e) Age of food-handler ..............................................................
f) Date of last medical examination ................................................
g) Food-handler's residential area ................................................
h) Highest level of education attained ................................................
i) Any proglottids in the stool (Yes/No) ........................................
j) Laboratory results:-

<table>
<thead>
<tr>
<th>Parasite stage and type</th>
<th>Scanty (1-3)</th>
<th>Few (4-10)</th>
<th>Moderate (11-20)</th>
<th>Many (21-40)</th>
<th>Very many (Over 40)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Larvae -</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ciliates -</td>
<td></td>
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<tr>
<td>Helminth-</td>
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<td>ova-</td>
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<tr>
<td>Cysts-</td>
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<tr>
<td>Trophozoites-</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No parasites seen-</td>
<td></td>
<td></td>
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<td></td>
<td></td>
</tr>
</tbody>
</table>

Remarks by Examiner .................................................................
Designation .................... Signature ....................... Date ..............
Appendix VI: Medical certificate sample

CITY COUNCIL OF NAIROBI
Public Health Department

THE FOOD, DRUGS AND CHEMICAL SUBSTANCES ACT
(CAP.254)

CERTIFICATE OF MEDICAL EXAMINATION - FORM D

I hereby certify that I have examined Mr./Mrs./Miss .................................................................

And that in my opinion HE/SHE is fit under the Food, Drugs and Chemical Substances
(Food Hygiene Regulations) to work at ..............................................................................................

Plot No. .................................................. Road .................................................................

Town ........................................................................................................................................

Examined at .......................................................... Lab Ref No./FHC/ ........................................

Date examined .................................................................

Receipt No. .................................................................................................................................

Official Seal ........................................................................................................................................

.......................................................... MEDICAL OFFICER OF HEALTH ........................................

.......................................................... Date ...................................................................................

NB: This certificate is valid for six months only. Expiry Date ................................................................

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