GEOPHAGY AND OTHER SELECTED FACTORS AS POTENTIAL RISKS FOR ENHANCING TRANSMISSION OF INTESTINAL HELMINTHS IN PREGNANT AND LACTATING WOMEN IN WESTERN KENYA

PhD Thesis

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**A thesis submitted in fulfilment for the degree of Doctor of Philosophy (PhD) in the Department of Medical Microbiology
Faculty of Medicine**

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DECLARATION

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

Signed ....

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This thesis has been submitted for examination with our approval as university supervisors.

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DEDICATION

This thesis is dedicated to my wife Florence, my children and the entire family for their endurance and understanding during my long absence from home while undertaking this research.

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Acknowledgements

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Abstract

A cohort study was conducted to assess geophagy and other potential risks for enhancing the transmission of intestinal helminths in pregnant and lactating women in thirty-nine selected villages in Usigu Division, Bondo District, Nyanza Province in Western Kenya. Eight hundred and twenty-seven pregnant women whose gestational age was 14-24 weeks were recruited between October 1998 and February 2001 and followed up for six months after delivery. Recruitment was carried out with the assistance of a qualified medical nurse in four recruitment centres within the study area. The gestational age was determined by calculation of previous menstrual history and confirmed by ultrasonography at the health centre.

Recruited women, who were solely from an area that had been surveyed demographically, were interviewed for obstetric and socioeconomic history such as age, parity, level of education, marital status, income and sanitation. They were further interviewed about their earth eating habits. Before any tests were performed, the women had to sign a consent form accepting to participate in the study. All the procedures pertaining to antenatal clinics were performed according to Ministry of Health guidelines. A clinical examination to assess the health status of the individual was carried out. Vital medical information including body temperature and other conditions related to any previous and current pregnancy was obtained. Laboratory investigations carried out at baseline included blood for haemoglobin concentration (Hb), serum ferritin estimation and malaria parasitaemia. Stool samples were taken, processed and examined for intestinal helminths and a sub-sample used for silica estimation. Earth samples were collected for estimation of silica and iron concentration. All infected women were then treated at baseline for helminths and malaria.

After recruitment, the women were followed at mid-term (30-32 weeks), delivery, three months postpartum and six months postpartum for more interviews, examinations and sample collection for reinfection. The mean age of the recruited women was 24.8 years; their gestational age was 17.5 weeks and their median parity two. Twenty-two percent of the participants were primigravidae. Out of the recruited women, 378 (45.7%) were geophagous at the time of recruitment. There were no differences in geophagic behaviour among the women in different obstetric and socioeconomic groups. The prevalence of those eating earth dropped from 45.7% at baseline to 29.6% at six months postpartum. The amount of earth eaten

daily also decreased during the follow-up period. The types of earth most frequently eaten were soft stones and that from anthills.

Intestinal helminths were common among the pregnant women, with the prevalence of hookworm (49.8%) being higher among geohelminths, followed by *Trichuris trichiura* (24.1%) and *Ascaris lumbricoides* (16.3%). However, *Schistosoma mansoni* (63.0%) was the highest. There was no relationship between most of the helminths and geophagy at baseline except the intensity of *A. lumbricoides*, which was higher in the geophagous women than in the non-geophagous ones and more so among those who preferred anthill mounds and other types, but not soft stone (P=0.01). The prevalence and intensity of *S. mansoni* was higher in primigravidae than in multigravidae (P=0.02 and P=0.008 respectively). Primigravid women also had higher prevalence and intensity of malaria than multigravid (P<0.001). Infection with *Ascaris* was high in women who had no latrines. Education had some effect on hookworm infection; the prevalence of hookworm among those with primary education was 53% and among those with secondary education only 36%. Likewise, intensity was higher in those with primary education than among those with secondary education (135 epg compared to 90 epg). Distance from the lake had an effect on *S. mansoni* infection with those closer to the lake having higher prevalence and intensity than those living further inland (P< 0.001). In contrast, hookworm was higher in those living further inland than in those living closer to the lake (P=0.08).

Among the 827 women whose blood was taken for haemoglobin (Hb) and serum ferritin (SF) concentration, 48.1% had moderate anaemia (Hb ≥7.0 g/dl <11.0 g/dl). The mean haemoglobin was 10.9 g/dl. The geometric mean serum ferritin (SF) was 19 µg/L and prevalence of women with depleted iron stores (SF <12 µg/L) was 32.8%. Geophagous women had lower Hb (10.6 g/dl), compared to non-geophagous women (11.1 g/dl) (P<0.001). The prevalence of anaemia was much higher in the geophagous group than in the non-geophagous group (P=0.001). Serum ferritin was lower in the geophagous women compared to the non-geophagous ones (P<0.001). Hb was lower in primigravidae than in multigravidae (P<0.001), while SF was higher in primigravidae than in multigravidae (P<0.001).

Hookworm infection had a positive correlation with Hb and SF; *Trichuris* had a positive correlation with Hb but not SF, while malaria had a negative correlation with Hb and a positive correlation with SF. Iron depletion was more common in women with hookworm infections than among those who were malaria positive. When stratified by gravidity, no infection related differences in Hb or SF concentration were seen among the primigravidae, whereas these were marked among the multigravidae.

Seven hundred women examined at mid-term had either been treated successfully or had initially been free from *Ascaris*, 670 were *Trichuris* negative and 479 hookworm negative. These were therefore followed for reinfection and examined at delivery, three months postpartum and six months postpartum. At the last follow-up, when 405 women were successfully followed up for hookworm, the re-infection rate was 16% or 32% of the baseline prevalence; for *Ascaris* 593 women were followed and the reinfection rate was 9.4% or 57.7% of the baseline rate; for *Trichuris* 561 women had been followed up and 5.9% or 24.5% of the baseline prevalence, were found positive. Geophagous women had a higher rate of reinfection with *Ascaris* compared to non-geophagous women (13.3 vs. 5.8%, $P<0.001$). Anthill earth was associated with reinfection with *Ascaris* ($P<0.001$). Intensity, but not prevalence, of *Trichuris* was higher in geophagous women than in non-geophagous women ($P=0.005$). Reinfection of hookworm was higher in those women without latrines ($P=0.007$), and also in women with lower educational status (0 ving far from the lakeshore ($P=0.04$).

The study has shown that there is an increase in geophagy during pregnancy in the study population. It was confirmed that geophagy enhances helminth infection during pregnancy and lactation. It was highly associated with high intensity of *Ascaris lumbricoides* at recruitment. It was also shown that there was a rapid build up of intensity of *Ascaris* in geophagous women at reinfection than in non-geophagous ones. Earth from anthills was mostly associated with high reinfection of *Ascaris*.

It was concluded that women with low educational level stood a higher chance of being infected with hookworm than those with secondary education. Those without latrines were also at a higher risk of infection with hookworm than those with latrines. Geophagy had a negative association with both haemoglobin and serum ferritin. Finally the results show that women with depleted iron stores and anaemia are likely to be geophagous.

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Chapter 1

Introduction and literature review

1.1 Introduction

1.1.1 Intestinal helminths

Global estimates suggest that soil transmitted helminthiases and schistosomiasis are among the most common infections in the world. According to recent estimates by the World Health Organization *Ascaris lumbricoides* infects approximately 1.45 billion persons, hookworms (*Necator americanus* and *Ancylostoma duodenale*) 1.3 billion persons and *Trichuris trichiura* over 1 million people (WHO 2002). The annual mortality is estimated to be 60,000 due to *A. lumbricoides*, and 65,000 and 10,000 due to hookworms and *T. trichiura* respectively (WHO 2002). The three major schistosomes infect 200-270 million persons (Crompton & Nesheim 1982; Doumenge & Mott 1984; WHO 1998). This includes 78 million *Schistosoma haematobium*, 57 million *S. mansoni* and 69 million *S. japonicum*. Worldwide up to 0.6 billion people are at risk of infection with schistosomiasis (WHO 1998). Current estimates of total number of individuals with morbidity due to *Schistosoma mansoni* in sub-Saharan Africa is 393 million at risk and 54 million infected (WHO 1999). In 1998 WHO estimated that schistosomiasis and soil transmitted helminths were responsible for more that 40% of the disease burden due to tropical diseases (excluding malaria) (WHO 1999). Not only are the absolute numbers of persons infected with these helminths staggering but the prevalence of these infections is also high compared to other infectious diseases in the third world (Pawlowski 1984). Intestinal helminths are particularly widespread within the tropics and subtropical regions of the world. Africa alone was thought to have 132 million cases of hookworm by 1991 (Pawlowski *et al.*1991), the cases are now estimated to be over 200 millions (WHO 2002). In terms of morbidity and mortality, intestinal worms are among the top 17 infections (Walsh & Warren 1979). The contribution to mortality and morbidity are probably underestimated because of under-reporting (Cooper *et al.* 1986). More recent global studies estimate the overall prevalence and distribution of soil transmitted helminths at 2 billion people. 350 million of the infected associated with *A. lumbricoides* morbidity. 220 million with *T. trichiura* and 150 million with hookworm morbidity (WHO 2002).

The impact of these parasites on public health has been grossly underestimated. partly because although they cause considerable morbidity. mortality is very low (WHO 1990). The latest figures show the annual mortality for *A. lumbricoides* as 60.000 people. for *T. trichiura* as 10.000 people and for hookworms as 65.000 people (WHO 1998c. WHO 2002). Intestinal helminth infections are known to cause protein energy malnutrition and iron deficiency anaemia. which globally affects 2.15 billion people of whom 51%. are pregnant women and in sub-Saharan Africa alone over 206 million people suffer from anaemia with 52% of them pregnant women (WHO 2002).

Studies have been carried out on the epidemiology of helminths in Kenya especially in Nyanza and Coast Provinces (Hall *et al.* 1982; Latham *et al.* 1982; Magnussen *et al.* 1997; Brooker *et al.* 1999). In western Kenya. studies have shown that hookworm is the commonest helminth. followed by *T. trichiura* and *A. lumbricoides* (Kinoti *et al.* 1971; Pamba 1980; Chunge *et al.* 1985; Geissler *et al.* 1997; Brooker *et al.* 2000; Olsen *et al.* 2000). A study carried out in schoolchildren in Bondo District in 1995 showed a similar trend. The overall prevalence for hookworm. *T. trichiura* and *A. lumbricoides* infections in this area was >36%. 21% and 16.5% respectively. Apart from the three helminths. *Schistosoma mansoni* was also found to be highly endemic with a prevalence ranging between 16.8-60% (Thiong'o *et al.* 2001).

1.1.2 *Ascaris lumbricoides*

Ascaris lumbricoides infection constitutes a public health problem and is probably of major nutritional significance from the point of view of prevalence. The morbidity and mortality caused by *Ascaris* have most likely been grossly underestimated (Stephenson 1987). *Ascaris* represents a global worm burden of about 8.7 billion adult worms or 1.45 billion persons multiplied by the expected mean of six worms per host (WHO 1981; WHO 2002).

Human *Ascaris* is cosmopolitan in distribution and occurs in temperate as well as tropical and subtropical environments although it is more common in areas which have a combination of predisposing factors including poverty, high population density, agricultural activity, poor sanitary practices and favourable climatic conditions.

Most of *A. lumbricoides* infections are thought to occur in Asia (73%) while about 12% occur in Africa and the rest in Latin America (8%) (Peters 1978). In Africa *A. lumbricoides* infects an estimated 25% of Kenyans, 21% of rural people in rural Nigeria and has been reported from almost all African countries (Stephenson *et al.* 1980; Nwosu 1981; Crompton and Stephenson 1985). The prevalence of *Ascaris* in Kenya has apparently not decreased in over 60 years (Chunge *et al.* 1985).

Direct global mortality due to *Ascaris* is difficult to assess. In 1964 WHO experts calculated the mortality rate to be 0.006%, while Walsh and Warren put it at 0.003% (WHO 1964b; Walsh & Warren 1979b). More recent studies put mortality at 60,000 people per year (WHO 1998c). Case fatality due to intestinal obstruction reported in the last decade varied from 2.4% to 23%. In the USA the incidence of intestinal obstruction due to *Ascaris* in children 1-5 years was 0.2% among 1000 infected children (Blumenthal & Schultz 1975). Most of this mortality is due to complications such as intestinal obstruction, billiary ascariasis, intestinal perforations and hepatic abscess (Pinus 1985).

Morbidity due to *Ascaris* has probably been grossly underreported due to lack of data. In Bangladesh the overall prevalence of *Ascaris* is about 80% and is a leading cause of hospitalization for both in and out-patients, while in a children's hospital in Rangoon, Burma, 3% of all admissions were due to *Ascaris*, while in Kenya it was 2.6%. In Mexico, children admitted due to intestinal obstruction ranked fifth in all hospitalization and in Cape Town, South Africa, *Ascaris* was the most common cause of abdominal emergencies (WHO 1987).

1.1.3 *Trichuris trichiura*

Trichuris trichiura or whip worm has a cosmopolitan distribution. although it is more common in the warm. moist, tropical and subtropical countries. where prevalence can be as high as 90% (Peters 1978). About 63% of the infected persons are found in tropical and subtropical regions of Asia, about 11% in Africa and about 14% in the Americas (Pawlowski 1984).

Man becomes infected directly by ingesting the embryonated eggs from contaminated hands. water. food or soil. The number of clinically significant cases is often greatest in the 5-15 year age group (Wolfe 1978; Pawlowski 1978; Bundy *et al.* 1985). Studies carried out in St. Lucia. have shown that prevalence of *Trichuris* increases rapidly with age from early childhood. attains a plateau of 80 to 100% and remains high and relatively constant throughout adulthood (Bundy *et al.* 1987). In contrast the average worm burden in St Lucia declines significantly in adults (Bundy *et al.* 1987). This decline suggests that there is an age related reduction in either the rate of establishment of infection or the rate of exposure to infection (perhaps due to acquired immunity) or changes in behaviour with age (Bundy *et al.* 1987). *T. trichiura* is frequently found in multiple infections with *A. lumbricoides* and hookworm (Kamath 1973; Jung & Beaver 1951; Stephenson *et al.* 1980, 1987). Like the other intestinal helminths. the burden of *Trichuris* has an even distribution with the majority of the population harbouring few worms. while a few people are heavily infected (Croll & Ghadrian 1981; Bundy *et al.* 1985). It has been observed that young children on average harbour the heaviest worm burdens (Bundy 1986).

Many light infections appear to be asymptomatic. but a more serious syndrome called massive infantile trichuriasis can develop (Kouri & Valdez Diaz 1952). This is characterized by severe symptoms. including anaemia. Infection with *Trichuris* is divided into three categories of light. moderate and heavy infection. The severity of the disease is not only dependent on the intensity of infection itself and the location in the gastrointestinal tract but also on the state of the host including age. general health. iron reserves and experience with past infections (Pawlowski 1984). In light to moderate infections most patients will have epigastric and lower abdominal pains, diarrhoea, but rarely bloody, vomiting, headaches and weight loss, while in heavy infections the above symptoms will be severe and will include severe anaemia, rectal prolapse, clubbing of fingers and moderate eosinophilia (Wolfe 1978, Markell *et al.* 1986, Beaver *et al.* 1984). All these will lead to reduced food intake, increased nutrient loss and increased blood and iron loss. Infections with *T. trichiura* usually show a very strong, positive correlation between intensity of infection, whether by worm count or eggs per gram of faeces (Jung & Beaver 1951; Gilman *et al.* 1983). Severe morbidity is associated with burdens exceeding 500 worms, but infections with a few hundred worms may initiate disease (Bundy 1986). Diarrhoea is caused by inflammation and irritation of the colon and the presence of lesions which then interfere with colonic water re-absorption (Mathan & Baker 1970). Rectal prolapse is thought to be a consequence of straining while at defecation in the presence of massive numbers of worms and/or the possible irritation of nerve endings with increased peristalsis (Ramirez-Weiser 1971).

1.1.4 Hookworm infection

The prevalence of hookworm varies from 60 to 90% in rural unsanitary conditions in the moist tropics to 10 to 20% in dry unsanitary conditions such as in Iran and Pakistan (Miller 1979; Kochar *et al.* 1976; Schad & Banwell 1984). *Ancylostoma duodenale* and *Necator americanus* are the two most common human hookworm species. *A. duodenale* is predominant in Europe, the Mediterranean region, the Middle East, Iran, Pakistan and some parts of Northern India. Mixed infections of both species are found in Brazil and some parts of Africa, India and South East Asia. *N. Americanus* is the only hookworm in North and Central America; it is predominant in most parts of South America, Central Africa, Southern India, Indonesia and the Pacific (Beaver *et al.* 1984; Schad & Banwell 1984; Markell *et al.* 1986).

Hookworm infection is associated with faecal contamination of soil where sanitary facilities are inadequate. Prevalence and intensity generally increase with age, but levels of infection often decline in adulthood. Studies in

Africa and India indicate that worms appear to be acquired steadily through early life but the levels remain stable at a slightly higher rate for men than women from the second to fifth decade. This trend is not applicable where both men and women work on the fields. as in sub- Saharan Africa the prevalence will be similar (Bruce-Tagoe *et al.* 1977). After the fifth decade the infection tends to increase again (Schad *et al.* 1975). This change has been attributed to acquired immunity. though there is no conclusive evidence that man develops a functional immunity to hookworm infection (Miller 1979).

The first stage of infection. cutaneous invasion of the larvae. results in transient dermatitis and associated symptoms referred to as 'ground itch'. *N. americanus* larvae may migrate in the skin and this produces creeping eruption or cutaneous larva migrans. Larvae migrating through the lungs may cause coughing and wheezing and if the infections are heavy occasionally bronchitis and pneumonitis may develop (Beaver *et al.* 1984). The clinical state of hookworm infection is now called 'hookworm disease'. a term that refers to a microcytic. hypochromic anaemia caused by intestinal infection and is not related to any cutaneous or pulmonary manifestations (Beaver *et al.* 1984). In the older literature. the term ancylostomiasis was used. and in some parts of the word local terms are used for hookworm disease. for example the Kiswahili word *safura* in East Africa. The clinical severity of the disease is closely related to the worm burden and also to the condition of the host. including the iron reserves of the host. pregnancy. iron needs. the adequacy of iron intake and its bioavailability and the general state of the patient's health. Despite this, good nutrition does not necessarily protect the individual (Roche & Layrisse. 1966)

The most important nutritional impact of the infection on the host is anaemia due to the presence of adult worms in the small intestine and the blood and iron loss that the worms cause (Rochem & Layrisse 1966; Banwell & Schad 1978; Variyam & Banwell 1982). It has been estimated that a single *N. americanus* is responsible for a mean blood loss of 0.031 ± 0.015 per day while *A. duodenale* will cause a mean blood loss of 0.08 ± 0.02 ml per day (WHO 1994). These measurements of feeding activity explain the pathogenicity of hookworms and account for their contribution to the development of iron deficiency anaemia. In acute adult worm infections, nausea, vomiting, diarrhoea and cramping abdominal pain can occur (Cline *et al.* 1984). In chronic hookworm infection, the common symptoms are those of iron deficiency anaemia, which include breathlessness, palpitations, headache and depression. In addition to low haemoglobin levels, iron deficiency anaemia is accompanied by low mean corpuscular volume, elevated free erythrocyte protoporphyrin, low transferrin saturation, and low serum ferritin levels. The peripheral blood film is microcytic and hypochromic. Bone marrow staining for iron indicates the absence of iron stores.

1.2 Modes of transmission of intestinal helminths

Ascaris lumbricoides is the largest of the common nematode parasites of man. It has a relatively simple life cycle involving the egg, four larval stages, immature worms and adult worms of both sexes. The adult worm normally lives in the small intestines (Makidono 1956). The female adult worms measure 20-35 cm in length and about 3-6 mm in diameter, while the males are smaller and are from 15-31 cm in length and 2-4 mm in diameter (Beaver *et al.* 1984). Female worms produce eggs which are excreted in the faeces of the host. One female *A. lumbricoides* can discharge an average of 200,000 to 240,000 eggs per day (range 134,000 to 360,000) (Cram 1926). However, only two eggs per female worm are needed to infect man and develop into adult male and fertile female worms in order for the population of *A. lumbricoides* in a given area to remain reasonably stable (Croll *et al.* 1982).

Humans contract *Ascaris* by ingestion of embryonated eggs. This commonly occurs through faecal contamination of food, water or other beverages, eating utensils or fingers. Cooked food is usually safe but can become contaminated after cooking. Consumption of earth by young children, pregnant women and others may lead to infection.

The eggs are passed in the faeces in an un-embryonated state. In the presence of sufficient oxygen, moisture and warmth, the larva develops inside each egg in about two to three weeks and becomes infective one week later.

The eggs are broad and ovoid and measure 45-70 x 35-50 µm; they have a tough protein coat and prior to embryonation, are extremely resistant to drying or to destruction by various chemicals. They have been known to remain in soil for up to six years in Germany and 14 years in Russia (Mueller 1953). However, the majority of them are thought to be destroyed after passage in faeces to the environment. Eggs deposited on heavy clay soils in moist, shady locations with temperatures of 22 to 32°C typical of tropical climates, develop rapidly into infective-stage larvae (Beaver *et al.* 1984). Development is much slower at low temperatures and can take up to 45 to 55 days at 16-18°C (Pawlowski & Arfaa 1984b). Development of embryos generally ceases bellow 15°C and above 38°C. Heating and prolonged exposure to ultraviolet rays from sunlight kills *Ascaris* eggs (Fitzgerald & Ashley 1977).

Embryonated eggs ingested by the host hatch in the small intestine and liberate larvae which burrow through the mucosa and are carried to the liver via the enterohepatic circulation. The larvae develop further and roam about in the liver for about five days, then proceed via the blood vessels to the right side of the heart and on to the lungs. They continue growth in the lungs for 9-15 days, finally break out into the alveoli, pass up the trachea, and are either swallowed or sometimes spat out by the host. The swallowed larvae are apparently not affected by the gastric juice in the stomach and most survive to reach the small intestine where they grow to become juvenile and then to mature adult worms of both sexes which mate to produce fertilized eggs (Fig. 1.1). Many of the larvae are lost in inappropriate tissues and do not survive to become adult worms. The entire cycle from ingestion of eggs to production of fertilized eggs by mature *A. lumbricoides* take two to three months (Vogel & Minning 1942).

The life cycle of *Trichuris* is simple. Most stages of development occur in the caecum of the host, from the first cuticular moult of the larva to adulthood and sexual reproduction. Female adults shed an average of 3,000 to 20,000 eggs per day. The eggs are not embryonated when passed out in stool and therefore are not infective. In the soil and within ambient temperatures and humidity, embryonic development starts with the first stage larva. This stage takes between 15 to 30 days and development may abort or be inhibited by direct sunlight or desiccation. The larva hatches when the eggs are ingested by the host. The larva then penetrates the intestinal villi where it remains temporarily and later migrates to the caecum where it burrows and undergoes cuticular moults to maturity (Fig. 1.2). This development to adult will take 30 to 90 days and after copulation the female will start to lay eggs. The eggs are barrel shaped and have transparent polar plugs. They measure 50 to 54 µm in length and 22 µm in width. In heavy infection worms can be found in the posterior level of the ileum or on the walls of the appendix. The length of the adult worm varies from 3-5 cm and females are larger than males. Worms feed on enterocyte syncytium, but may also ingest erythrocytes, leucocytes and mucosal tissue fluids. (Pawlowski 1984).

Adult hookworms usually live in the upper small intestines where the female deposits eggs into the intestinal lumen. *A. duodenale* has been estimated to produce an average of 20,000 eggs per female worm per day and *N. americanus* about half that number. Egg production can be related to several factors including age of infection, the nutritional status of the host and the number of worms present. The immunological response to the infection by the host may also affect egg production (Schad & Banwell 1984).

Eggs leave the host through the faeces and if the conditions are favourable will hatch within 24 hours. Ideal conditions are aerated, moist soil and a temperature range of 23 to 33° C (Gilman 1982). Eggs of the two hookworm species are similar and difficult to distinguish. They are thin shelled, transparent; ovoid with round ends and measure 64-70 x 36-42 µm. The eggs hatch into first stage or rhabditiform larvae which feed on bacteria and faecal matter. For approximately seven days, the larvae grow, moult once, and then moult again after following further growth. This third stage or filariform larva is infective, non-feeding and capable of active vertical movement in the soil and on vegetation. These larvae are very sensitive to desiccation, and the mortality rate is 90% in their first three weeks of life (Gilman 1982). Larvae are attracted to a potential host by warmth and carbon dioxide. Both species of hookworm can penetrate the skin effectively and usually between the toes or hands. Following penetration. the larvae migrate within the circulatory system to the lungs where they enter the alveoli and migrate then to the pharynx where they are swallowed or pass through the oesophagus into the stomach. Maturation is completed in the small intestine, and the eggs of *A. duodenale* first appear in faeces between 43 and 105 days; those of *N. americanus* appear between 40 and 60 days after initial infection (Fig. 1.3) (Schad & Banwell 1984).

Both *A. duodenale* and *N. americanus* have been recorded to be fairly long-lived (Miller 1979), for *A. duodenale*, a maximum of six to seven years and a maximum of worm egg output of 12 to 18 months (Kendrick 1934). The absolute longevity of *N. americanus* was reported to be five to six years. The adult worms are somewhat stout, cylindrical nematodes, off-white or rusty red in colour. *A. duodenale* is the larger of the two, measuring 5-10 mm for the males and 10-18 mm for the females. *N. americanus* females measure about 10 mm and males 5-9 mm in length. Both parasites have a well developed sub-globular buccal capsule. In *A. duodenale* this bears two pairs of teeth and in *N. americanus* a pair of semicircular cutting plates. Eggs of the two worms are difficult to distinguish unless precise measurements of a series of eggs are made.

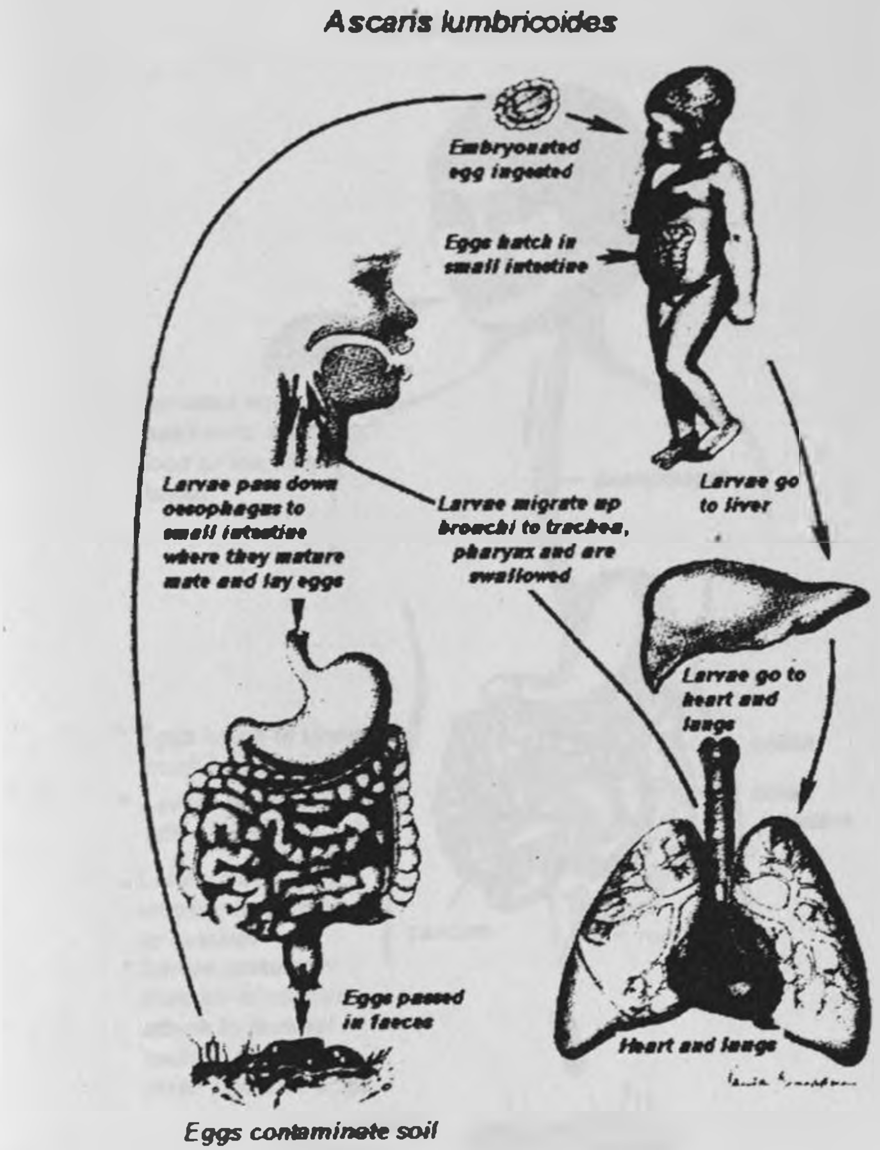

Figure 1.1 Life cycle of *Ascaris lumbricoides* (from Stephenson 1987).

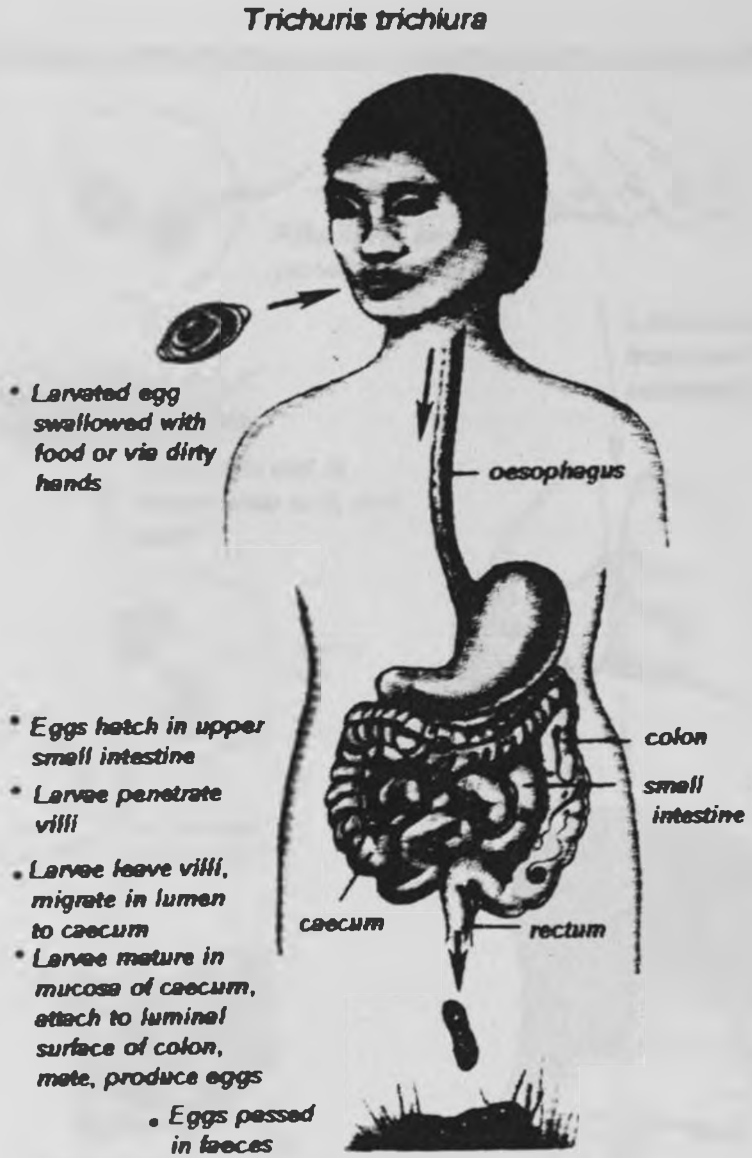

Figure 1.2 Life cycle of *Trichuris trichiura* (from Stephenson 1987).

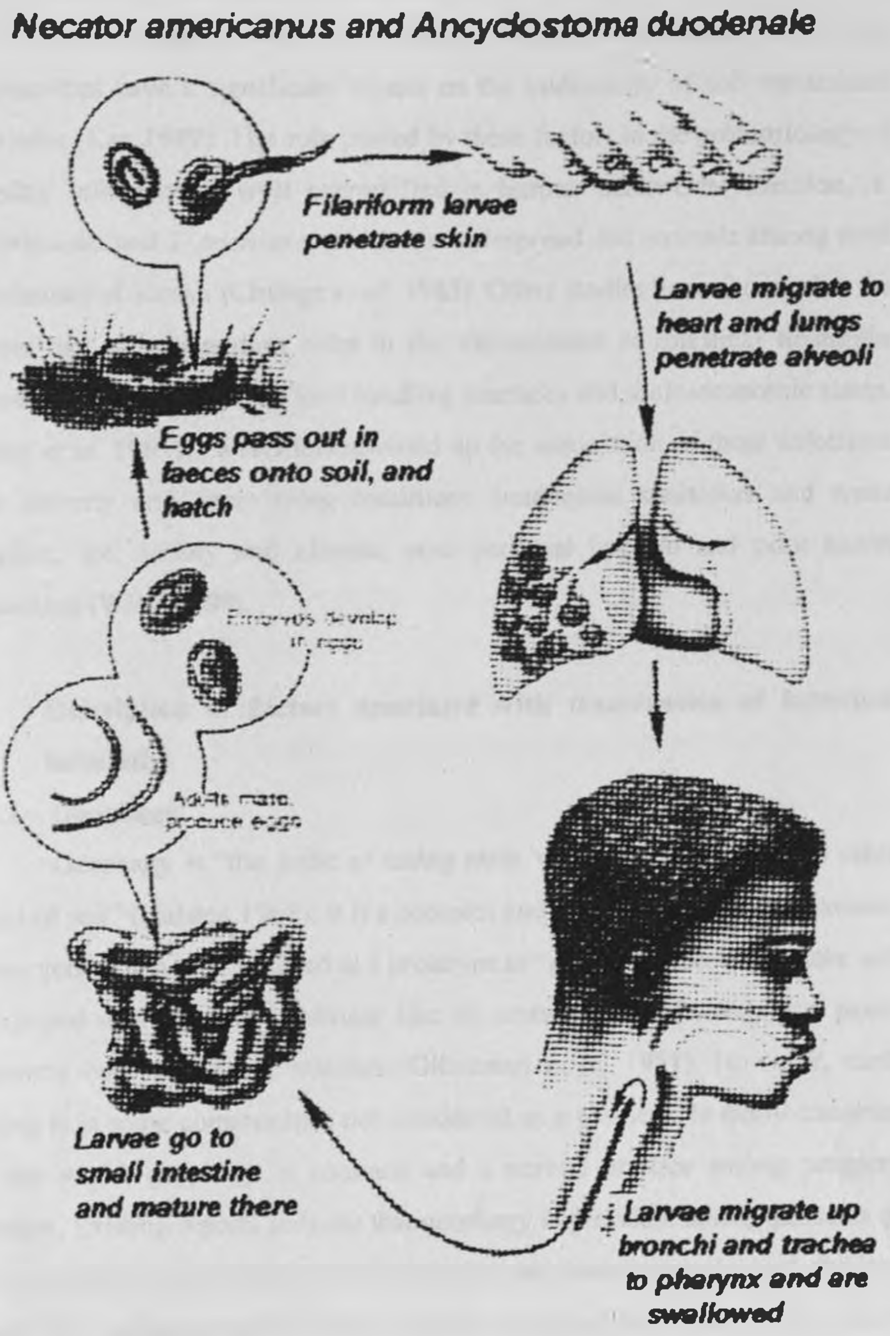

Figure 1.3 Life cycle of hookworm (from Stephenson 1987).

1.2.1 Factors influencing transmission of intestinal helminths

The environment and socio-economic factors of inhabitants of the rural communities have a significant impact on the endemicity of soil transmitted helminths (Kan 1989). The role played by these factors in the epidemiology of parasitic infections is well exemplified in human hookworm infection, *A. lumbricoides* and *T. trichiura*. which are widespread and endemic among rural populations of Kenya (Chunge *et al.* 1985). Other studies have shown that the factors that play important roles in the transmission of intestinal helminths include geophagy, hygiene. food handling practices and socio-economic status. (Esrey *et al.* 1991). WHO has summed up the association of these infections with poverty and poor living conditions. inadequate sanitation and water supplies. soil quality and climate. poor personal hygiene and poor health awareness (WHO 1998).

1.3 Description of factors associated with transmission of intestinal helminths

1.3.1 Geophagy

Geophagy is "the habit of eating earth which includes clay and other types of soil" (Halsted 1968); it is a complex and perplexing human behaviour. It has generally been classified as a prototype of 'pica'. *i.e.* non-food intake and is lumped with aberrant behaviour like the consumption of starch, ice, paint. cigarette butts and burnt matches (Gilickman *et al.* 1981). However. earth eating is in some communities not considered as a problem. In many countries of the world, geophagy is common and a normal practice among pregnant women. Existing reports indicate that geophagy is prevalent among peasants of the rural areas in the third world countries and among women and children from low socio-economic status in urban societies anywhere in the world (Anell & Lagercrantz 1958) (Fig. 1.4). However. systematic studies of this subject. including epidemiological data. are scarce. Geophagy in young children (Calabrese & Stanek 1990) and in women during pregnancy (Horner *et al.* 1991) has been studied in populations in industrialized countries. Studies on this behaviour in developing countries are. however. still few (Wong *et al.*

1993. Geissler *et al.* 1997). Further research is therefore necessary in order to determine its distribution in different populations. and to assess its possible impact on health. Geophagy is widespread in the animal kingdom. particularly herbivores (Johns 1990), and is common among primates including chimpanzees (Hladik *et al.* 1974). While nutritionists tend to look at geophagy as maladaptive or as a symptom of metabolic dysfunction. animal behaviourists look for adaptive functions of such phenomenon.

1.3.1.1 Nutritional explanation of geophagy

The most prevalent explanation for human geophagy is that it is a response to alleviate a nutrient deficiency (Danford 1982: Danford *et al.*1982a). However. in clinical studies where geophagy has been associated with mineral deficiencies of pathological degree. it has been implicated both as a cause and as a result of particular deficiencies (Halstead 1968). Following the pica model of pathological craving. deficiencies of specific nutrients have been the focus of the search for the cause of geophagy (Danford 1982).

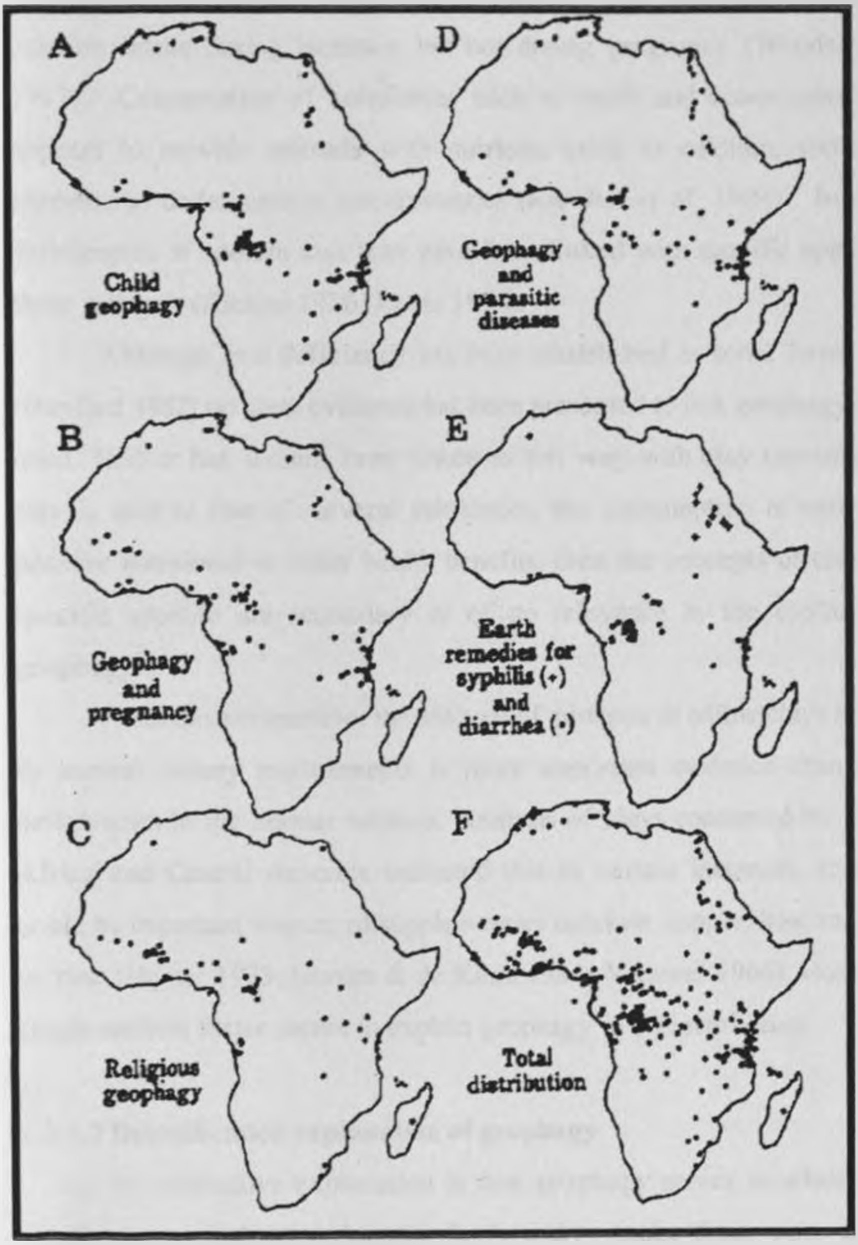

Figure 1.4 Map of geophagy in Africa showing various case studies and recorded observations (actual distributions are much more extensive) (from Lagercrantz. 1958).

Experimentally induced deficiencies in iron and potassium are known to evoke changes in dietary behaviour in animals. Rats selectively increase their calcium intake during lactation but not during pregnancy (Woodside *et al.* 1987). Consumption of substances such as earth and bones (osteophagia) appears to provide animals with nutrients such as calcium, sodium, and phosphorus under certain circumstances (Kreulen *et al.* 1984). In humans, deficiencies of sodium and iron have been linked with specific appetites for these minerals (Richter 1936; Rozin 1976).

Although iron deficiency has been established in some forms of pica (Danford 1982) no clear evidence has been presented to link geophagy and iron need. Neither has sodium been linked in this way with clay consumption. If clay is seen as one of several substances, the consumption of which offers positive nutritional or other health benefits, then the concepts of craving and specific appetite are secondary or of no relevance in the explanation of geophagy.

From this perspective, the analysis of minerals in edible clays in relation to normal dietary requirements is more important evidence than data on deficiencies in the human subjects. Analysis of clays consumed by people in Africa and Central America indicated that in certain instances, these clays could be important sources of supplementary calcium, copper, iron, magnesium or zinc (Hunter 1973; Hunter & de Klein 1984; Vermeer 1966). However, no single nutrient factor seems to explain geophagy in a general sense.

1.3.1.2 Detoxification explanation of geophagy

An alternative explanation is that geophagy serves to adsorb dietary xenobiotics and thereby detoxify foods and/or make them more palatable. Ecologists have suggested adsorption of alkaloids, tannins, oxalates, and other natural plant constituents as an explanation for many cases where animals in natural settings are observed to eat clay (Kreulen 1985). Under laboratory conditions, rats eat clay in response to gastrointestinal malaise induced by poisoning and other stresses (Mitchell *et al.* 1977).

The potential role of clay ingestion in neutralizing plant toxins in primate diets has important implications for the understanding of the evolution of human dietary behaviour, suggesting that plant foods may have been more important to our ancestors than was previously thought (Johns 1986, 1990). However, the hypothesis that primate geophagy serves a detoxification function in relation to plant secondary compounds (Oats *et al.* 1977; Davies *et al.* 1988) is difficult to test experimentally.

1.3.1.3 Relevance of geophagy to human health

There is clinical evidence of an association between geophagy, anaemia and iron deficiency (Moore & Sears 1994). Recent field studies in western Kenya and Kilifi have documented this among school children and pregnant women (Geissler *et al.* 1998a, 1998b). Existing cross-sectional data do not, however, explain the causality linking the two phenomena. There is inconclusive evidence whether geophagy leads to anaemia, whether it is induced by anaemia or iron deficiency, or whether it is a source of extra-dietary iron (Johns & Duquette 1991). Anaemia is one of the most prevalent complications in pregnancy in most developing countries, and is likely to affect pregnancy outcome (Horner *et al.* 1991; DeMeyer & Adiels-Tegman 1985). Given these medical implications, it is important to understand people's motivations for eating earth and their knowledge of its links to illness.

Geophagy might be a source of infection, in particular with helminths (Halstead 1968; Wong *et al.* 1990, 1991; Glickman *et al.* 1999). Medical research has shown that earth eating is linked to health problems. It contributes to geohelminths among children (Wong *et al.* 1991; Geissler *et al.* 1998a). *Ascaris* and whip worm infection are associated with poor nutritional status and impaired growth; they also affect certain cognitive functions among children (Nokes *et al.* 1992; Simeon *et al.* 1994). A recent study among schoolchildren in the proposed study area of Usigu has shown the same (Geissler *et al.* 1998a). However, no investigations into geophagy during pregnancy and its possible role as a source of infection exist, although it is known from ethnographic records that women in many parts of the world do eat earth during pregnancy.

1.3.2 Environmental hygiene and food handling practice

Water and sanitation has been the subject of considerable recent attention as a result of the declaration by the United Nations General Assembly that the 1980s would be a decade of international drinking water supply and sanitation. A major objective was to improve the health of populations through improved water supply and sanitation. Most of the research on water and sanitation projects has focused on incidences of diarrhoeal diseases, malnutrition, mortality in young children and the health impact associated with infections with *A. lumbricoides*. *T. trichiura*. hookworms (*Ancylostoma duodenale* and *Necator americanus*) and *Schistosoma mansoni* (Esrey *et al* 1991).

A measure of good sanitation is the availability and use of pit latrines. In most areas of Nyanza Province, pit latrine availability and use has been low due to socio-cultural beliefs and the type of soil. The soil texture in some areas is rocky while in others it is a black cotton soil that does not allow latrine construction. This combined with other socioeconomic factors led to exposure to helminthic infection. In their literature review, Esrey *et al*. (1991) have shown that where water and sanitation were improved, prevalence of these infections decreased by up to 29% for *Ascaris*, 77% for schistosomiasis and 4% for hookworm. These reviews have also shown that lack of water for domestic hygiene and proper human excreta disposal increases the risk for *Ascaris*, *Trichuris* and *S. mansoni* infections, while improper disposal of human excreta is a major risk factor for hookworm infection. The presence of water supply was associated with large reductions in the prevalence of schistosomiasis in Zimbabwe where the prevalence of *S. mansoni* among schoolchildren in areas with piped water was 0.8% as compared to 4.8% in areas without piped water (Mason *et al* 1986). It appears that environmental, socioeconomic and immunological factors conjointly play important roles in the dynamic process of infection and reinfection after treatment, however immune responses may limit the number of parasites in the host at any one time and acquired immunity does not seem to be related to re-infection in a direct way (Pawlowski 1982). The environmental and socioeconomic factors are more commonly implicated in the reinfection through contamination and spread of the parasites (Henry 1988).

There is very little information on the study of food handling practices as a risk factor on its own. A few studies have combined it with personal and environmental hygiene. Some studies have associated hygiene behaviour with the education of the mother, social and physical environment, individual factors and finally health education (Valerie *et al.* 1995).

1.3.3 Socioeconomic factors

Living standards, possession of some valuable objects, education levels and psycho-social factors and income of the family play an important role and have direct relationship with the health behaviour of the individual. Valerie *et al.* (1995) in their study in Burkina Faso concluded that socioeconomic factors played a role as predictors of a woman's hygiene behaviour; these included the husbands' occupation, education, some family ownership of certain valuable objects and cultural factors. Henry (1988), in a study for reinfection with *Ascaris* in St. Lucia, has shown that crowding was significantly associated to reinfection. He also noted that income per person was significantly associated to reinfection with *A. lumbricoides*.

1.4 Intestinal helminths and pregnancy

Intestinal parasites cause special problems during pregnancy. Environmental, nutritional and immunological factors influence the clinical manifestations (D'Alauro *et al.* 1985). Pregnant women are predisposed to helminthic infection due to behavioural changes during this period. The changes include dietary intake. Studies on intestinal helminths have been carried out in all categories of people with more emphasis on schoolchildren since they are the most vulnerable and active group that acquire infection early in their life. In a report, Bundy *et al.* (1995) estimated that 7.5 million pregnant women in sub-Saharan Africa alone were at risk of hookworm infection and about one million of these were estimated to be at risk of clinically demonstrable morbidity with a worm burden of >40 worms. Studies on the impact of helminthic infection on pregnant women have mostly been based on anaemia and iron status (Nurdiati *et al.* 2001) A cohort study carried out in Indonesia found that pregnant women infected with intestinal worms have high prevalence of low iron stores in the third trimester and a negative association was found between hookworm and serum ferritin in the first trimester (Nurdiati *et al.* 2001). Further studies on the aetiology of severe anaemia in pregnancy have indicated that up to a third of cases reported are due to hookworm infection (Fleming 1989). A study on risk factors for anaemia in pregnancy in a hospital in Tanzania found that about 10% of all cases with severe anaemia were caused by malaria and hookworm infection (Mlay *et al.* 1994). In a study in western Kenya, it was reported that hookworm infection was a predictor of low haemoglobin in pregnant women (Olsen *et al.* 1998).

The incidence of intra-uterine growth retardation (IUGR) increased with the number of intestinal parasites detected as shown in a study where *Ascaris* was predominant. This high rate was attributed to helminth infection or malnutrition due to high intensities of intestinal helminths (Villar *et al.* 1989). *Ascaris* infections have been associated with severe complications such as intestinal obstruction, cholangitis or hepatitis caused by migration of adult worms and cases of billiary *Ascaris* during pregnancy have been reported in the United States (Asrat *et al.* 1995). Other cases with gall bladder ascariasis have been confirmed by ultrasound in pregnant women in Ecuador (Gomez *et al.* 1993). In Kashmir, India, where 2% of 665 cases of hepatobilliary ascariasis were diagnosed by sonography, it was shown that pregnancy facilitated the migration of adult worms from the papilla of Vateri to the invasion into the gall bladder (Khuroo *et al.* 1992).

1.5 Control of intestinal helminths

Many countries in Africa have projects aimed at improving sanitation, water supplies and health education which if successful, help in the control of intestinal helminthiasis (Stephenson 1989). In recent years major efforts have been made to reduce the morbidity that accompanies the helminthic infections with the benefit of considerable experience of the Rockefeller sanitary commission to eradicate worms (Ettling 1990). The primary objective here is to use antihelminthic drugs available to lower worm burdens so that significant reduction in morbidity is gained and sustained (Albonico *et al.* 1998). There is a general agreement in the public that de-worming children is justified. It has been confirmed by many that mass chemotherapy at community level is the most effective method of control of intestinal helminths (Stephenson *et al.* 1983; Arfaa 1986). This has been ranked highly as a worthy cause for the poorer and more deprived communities where greater impact of intervention is required (World Bank 1993). Apart from more successful projects of helminth eradication in Japan (Yokogawa 1985), other countries have formed National Control Programmes aimed at both reducing morbidity due to schistosomiasis and helminthic infections (WHO 1993). Since the burden of intestinal helminthic infection is linked to the type of lives people lead, improvement in housing, appropriate sanitation at household and community level, provision of clean water, greater access to health care and education, personal hygiene and personal earning are recommended (UNICEF 1998).

1.5.1 Chemotherapy

Chemotherapeutic control of intestinal helminths is based on the principle that killing of the adult worm will reduce environmental contamination, and hence the rate of transmission, as well as directly treating the disease in individuals. Therefore with appropriate drugs, chemotherapy has an immediate impact on both transmission and morbidity (Bundy *et al.* 1989). The most commonly used drugs on the market are in the group of benzimidazole carbamates: Albendazole and Mebendazole given as a single 400 mg and 500 mg dose respectively (WHO 1995, 1996, 1998b). These drugs are considered most effective and multiple dosages will virtually ensure complete expulsion of the worm from the host (Royal Society of Medicine 1984; Bundy *et al.* 1985). With benzimidazole drugs, single dose chemotherapy is convenient for control programmes and for treating asymptomatic infection; for symptomatic cases however, therapy for three days will ensure complete expulsion of the worms (Bundy *et al.* 1989).

Benzimidazole drugs are considered safe, but their use in pregnancy was previously contraindicated because of uncertainty over possible embryo toxicity and teratogenicity (Bundy *et al.* 1989; Rossignol 1990). These drugs have now been recommended for use in pregnancy by the World Health Organization but only during the second trimester (WHO 1986, 1996, 1998). Mebendazole has been used successfully in pregnancy although it was recommended to be used from the second trimester where studies have shown that it was not associated with congenital defects of new born babies (de Silva *et al.* 1999). Mebendazole is also recommended for deworming by the Ministry of Health in Kenya from the second trimester (personal communication Medical Officer of Health Bondo district). Although the other benzimidazole drugs are also recommended for treatment from the second trimester, the MOH cautioned that only a single dose should be administered in order to minimize any chances of severe symptoms during pregnancy. Although benzimidazole drugs have varying degrees of side effects including nausea, vomiting, abdominal pain and discomfort and sometimes diarrhoea, these are rare. They cause little inconvenience when given as single doses (Rossignol 1990).

- To determine the prevalence and intensity of helminthic infection among pregnant and non-pregnant women at baseline and follow ups.

1.8 Hypothesis

Null hypothesis: Geophagy and other factors such as socio-economic. environmental and food handling techniques and nutritional deficiencies especially of iron do not act as risk factors in the transmission of intestinal helminths in pregnancy and lactating mothers.

Alternate hypothesis: Geophagy and other factors such as socioeconomic. environmental and food handling techniques and nutritional deficiencies especially of iron act as risk factors in the transmission of intestinal helminths in pregnancy and lactating mothers.

Chapter 2

Materials and methods

2.1 Study area

2.1.1 Bondo District

The study was carried out in Usigu Division of Bondo District in Nyanza Province in Kenya (Fig. 2.1). Bondo District lies between the Equator to the north and 0° 25′ latitude south and between longitude E34° 0′ and E34° 33′. It borders on the shores of Lake Victoria to the south. Busia District to the west and Kisumu District to the east. To the north the District borders Siaya District with Yala River and swamp forming the boundary. The District covers an area of 2330 km^2 of which 194 km^2 is covered by Lake Victoria. Kenya is administratively divided into eight provinces of which Nyanza is one. The province is in turn divided into twelve districts. among which is Bondo. The district is situated at the northern tip of Lake Victoria which is the largest freshwater lake in Africa. The lake is also commonly referred to as Winam by the local people. The district is divided into divisions which are further divided into locations and sub-locations. Among the divisions of the district is Usigu. which is divided into five locations. The present study was carried out in 39 villages located in four sub-locations of Usenge. Got-Agulu. Usigu and Got-Ramogi in West Yimbo and Central Yimbo Locations (Fig. 2.2).

Figure 2.1 Map of Kenya showing the study district (shaded area).

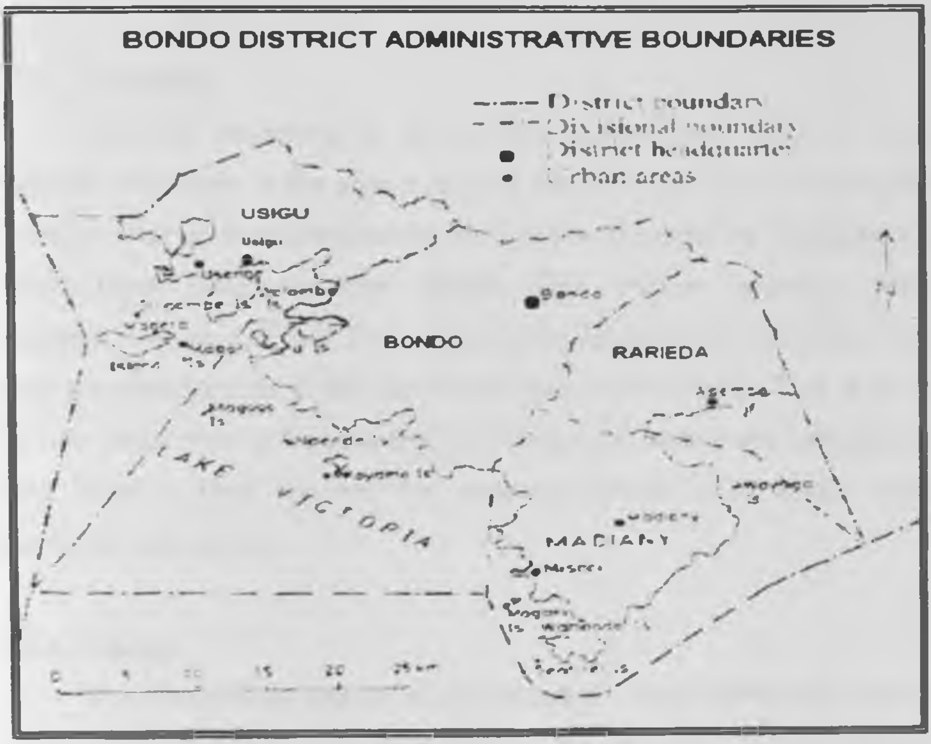

Figure 2.2 Map of Bondo District showing divisional boundaries.

2.2 Physical features

2.2.1 Topography

This area stretches from the lake shore which is ringed by reeds with some few rocky areas to the banks of Yala River. There are beaches which are mostly used as landing points for passenger and fishing boats. The altitude is between 1100-1300 metres above sea level. The highest points are Usenge and Ramogi hills, locally referred to as 'Got'. There are several rocky hills scattered over the area. The soil type in the area is of volcanic origin, belonging to the Nyanzian system and is dominantly red to strong-brown friable clay with laterite horizon. The red clay is overlaid by a greyish brown layer (A horizon).

The iron content increases with depth. Close to the lake. stretches of "black cotton soil" grumosolic black clay. are found (Survey of Kenya 1970: 26-27).

2.2.2 Vegetation

The main vegetation is the *Lantana camara* bush which is found virtually everywhere in the area. Along the lake shore are reeds in expansive swamps. while on the mainland indigenous shrubs and trees like *Euphorbia* are found. Other indigenous trees include *Ober* (*Albizia coriaria*). *Othoo* (*Balanites aegyptiaca*) and *Siala* (*Markhamia lutea*) while eucalyptus trees were introduced by the forest department around Got-Ramogi. Yala River is the only major river in the area and flows though the swamp and Lake Sare to Lake Victoria. There are very few temporary streams which collect water during the rainy season.

2.2.3 Climate

The area is very close to the equator and the largest freshwater body in Africa. and therefore has two main seasons. The climate is distinctly divided into the rainy and dry seasons. The rainy season is from March to May (long rains) which are relief and September to November (short rains) which are convectional. The rains are sometimes erratic and will vary from season to season. as well as having local small-scale variation. There is usually less rainfall closer to the lake shore. In between the seasons. December to February, there may be little rainfall or none. The average annual rainfall is about 1200 mm. The temperatures are constantly high during the day between 26 and 32 degrees centigrade (Fig.2.3). The minimum temperature at night is 17ºC. The relative humidity ranges between 60 and 80% in the rainy season and is lower during the dry season. 40-50% (Ministry of Agriculture Bondo Annual Report 2001).

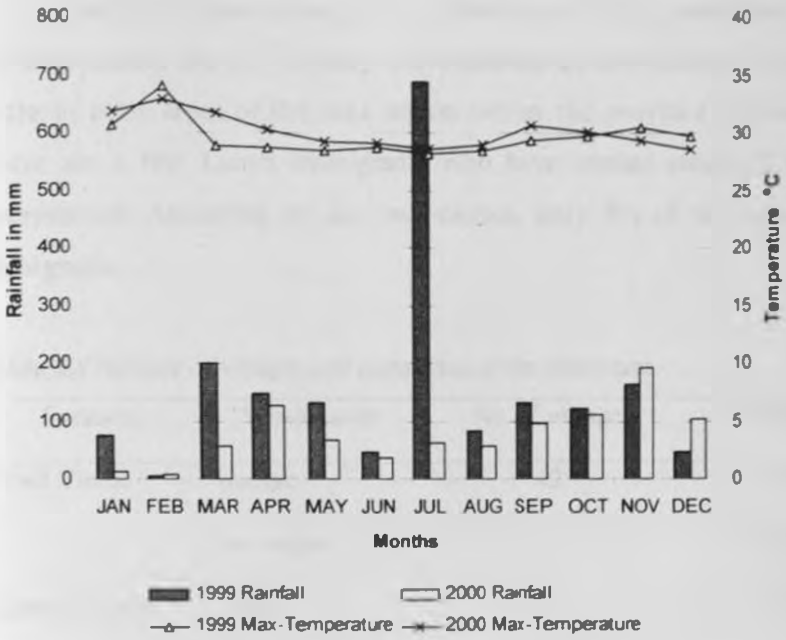

Figure 2.3 Rainfall and temperature for Bondo District 1999 and 2000.

2.3 The people

In the 1989 national census the population for the district was estimated at 640.000 and was projected to reach 866.000 by 1999. The density was 220 persons per km^2. There were 159.000 women of child bearing age (15-49). The population growth has been 3.5% in the past decade (CBS). According to the same census. the total population for Usigu Division was 32.000 inhabitants: however the 1999 census put the population at 45.864 (District Health Strategic plan for Bondo 2001). The overall density for Usigu was 160 persons per km^2 but it was more populated at the beaches. The population in the two locations of West and Central Yimbo was 25.459 (GOK 1999) (Table 2.1). The population ratio of male to female is approximately 1:1.1 due to labour migration. The total number of women is 13.330 out of which 6.900 (52%) are of child bearing age (14-49).

The main ethnic group is Luo which is a Nilotic group that arrived in the area around the 16th century and dispersed at Got-Ramogi and moved to settle in other areas of the lake region within the province (Ochieng 1974). There are a few Luhya immigrants who have settled among the Luo and intermarried. According to our own census, only 8% of the population are immigrants.

Table 2.1 Number of villages and population of the study area.

Location	Sub-location	No. of villages	Population
West Yimbo	Usenge	12	13,611
	Got-Agulu	5	4,589
Central Yimbo	Usigu	12	4,117
	Got-Ramogi	10	3,142
Total		39	25,459

2.3.1 Occupation

Most of the people in the study area are peasant farmers. They cultivate maize, millet, sweet potatoes, sorghum and cassava for local consumption. They keep zebu cattle, mainly for milk, and sheep, goats and chickens. There is no cash crop grown in this area. The main occupations are fishing and small-scale businesses. The fishing industry relies mainly on Nile-perch (*Lates niloticus*, local name mbuta) and sardines (*Limnothrissa* spp., local name omena), which are the main source of income. *Tilapia* and *Oreochromis* spp. (ngege) are caught mainly for the local market. Most of the male adults are employed in the fishing industry or work as civil servants; others are working in cities and are relied upon by the family members as a source of income. Migration is common among the families as the wives either join the husbands in the towns or the husbands come home over weekends or at the end of the month.

2.4 Health statistics

There are two government health facilities at Usigu and Got-Agulu. The health centre at Got-Agulu is equipped with 10 beds and is the only delivery point in the area. The staffing is not adequate at the health centre where they have three enrolled community health nurses (ECHN) and one support staff. Usigu dispensary is manned by one ECHN and one support staff. All the facilities offer maternal and child care clinics. There are a few private clinics within the trading centres and the beaches but they only offer outpatient care. Referral of patients is made to Bondo District Hospital which is 30 km away. The most complicated cases are referred to the Provincial Hospital in Kisumu which is 95 km from the study area. Referral is usually a problem as none of the facilities in the area has transport and some people do not have money for car hire. The women and children in this area are deficient in most micronutrients including iron. due mainly to the typical cereal-based diet with few animal products (Mwaniki *et al* 2002). The infant mortality rate is 110 per 1000 (Bondo District strategic health plan 2001). Home deliveries account for up to 60-70% and are carried out by traditional birth attendants (TBA). Women of child bearing age (15-49) account for 52% of the total female population. Leading causes of morbidity in the district are malaria. diseases of the respiratory system. diarrhoeal diseases. skin conditions including ulcers. intestinal worms. urinary tract infections. eye infections and anaemia. ranked from the highest to the lowest. respectively (District Health Management Report 2000). Immunization coverage is 45% (Bondo District health strategic plan 2001-2006).

2.5 Water and sanitation

According to national statistics. the coverage of safe water supply in the larger Siaya District was 4% and in Usigu Division 8%. There are three water supply pumping stations but their use is limited to schools and health facilities and a small fraction of the population. According to our own survey. the main source of water was the lake (93.7%) and River Yala for the people living further inland. Some of the villages depend on dams for water during the dry season. Only 4.4% of the study population have access to tap water. Electricity is available to people living in the main commercial centre. Usenge and markets which are along the main tarmac road. Apart from Usenge the other trading centres are Usigu, which is the Divisional Headquarters, and Uhanya, which is a major landing beach. The latrine coverage is 63.2%, and those in use were 52.8 % (KEDAHR survey 1999-2000). Most of the pit latrines are ordinary with very few vent improved ones (1.0%).

2.6 Education

A government survey states that 50.4 % of the adult population of western Kenya can read and write (GOK 1988). According to the Kenyan Danish Health Research project surveys (unpublished), most of the adult males and females in the study area had at least attained primary education levels (93%). Out of the population, 32.6% of the males had secondary education while only 15.1% of the women had secondary education. This drop in secondary education could be attributed to lack of funds for all groups, to pregnancy and early marriages in girls and to fishing activities in older boys. Another factor could be that some parents give their boys an education preference over education for girls. The area is served by four secondary schools, two mixed and one each for boys and girls only. There are 16 primary schools in the two locations and one village polytechnical school.

2.7 Transport

The area is well served by all-weather roads. There is a main tarmac road traversing the study area from east to west which serves most of the villages and is the road leading to the District and Provincial headquarters at Bondo and Kisumu respectively. There are all-weather feeder roads which lead to various villages inland and to beaches. Boats are also used for transport between villages through fish landing beaches. This network makes movement of fish from landing beaches to the market easy.

2.8 Methodology

The study was carried out as part of a longitudinal birth cohort study conducted by the KEDAHR project. This was an intervention and reinfection study carried out among pregnant and lactating women who were either geophagous or not. The women were recruited at 14-24 weeks gestational age. They were examined for worm infection, treated and followed up for reinfection up to six months post partum.

2.8.1 Community mobilization

The community was informed of the study through public meetings which are organized by the local administration in all the sub-locations in the study area: these are commonly referred to as "Chief's Baraza". Seminars were held for community leaders, who included administrators, heads of government departments, beach leaders, all church representatives and village health committee members who represented community health workers and traditional birth attendants (TBA). During the meetings and seminars, the objectives of the study were explained in detail, putting stress on the interventions and specimen collections which needed to be understood clearly prior to the start of the field work. The mobilization was intended to create awareness about the need for antenatal care for the expectant mothers and their unborn babies.

2.8.2 Training

Training was organized for several categories of personnel who assisted in the field operations. The existing TBAs were mobilized through the District Medical Officer's office and trained for two weeks at a local church. The training included safe home delivery in case of emergencies, referral of mothers to the health facility, monitoring of the gestational age of the pregnant woman and assistance with the recruitment of the clients for the study. Forty-five TBAs were trained and represented all the villages. At the end of the training they were given a certificate and TBA delivery kit.

The second lot to be trained were 12 field assistants who were recruited from villages within the study area. They were at least form 4 school leavers and well conversant with the area. The field assistants were trained in communication skills. demography. the filling in of questionnaires and first aid principles. Since this was a group used to identify clients and assist in the follow-up of cases. they were given bicycles to use in the field.

Health personnel were also trained to assist the principal investigator in specimen collection and follow-up of clients. The nurses were given on-the-job training on the use of an ultrasound machine which enabled the confirmation of gestational age. All the people mentioned assisted the investigator in carrying out the field and laboratory work in this study.

2.8.3 Sample size

The estimation of the sample size for the study population was based on the census and the percentage of geophagy in the study area. The number recruited also depended on the demographic data and the migration pattern. Based on this information a random sample of 10% of the community was screened for geohelminths to determine the prevalence and intensity. A minimum sample size of 550 pregnant women was required to show a difference of 50% between the re-infection rates of geophagous and non-geophagous women with the power of 80% and 5% level of significance. A total of 827 women were therefore recruited. This represented 10% of the expected pregnant women in the area. This figure also allowed up to 20% loss due to follow up. migration. desertion and serious illness. The recruitment took 15 months to complete.

2.8.5 Recruitment

The initial stage involved recruitment of study clients from the area. This was done after the demographic survey and sensitization meetings with the administration and the local community leaders. The field assistants and TBAs assisted in the identification of pregnant women from the villages. The potential clients were then screened at the five recruitment centres which had been set up within the study area. These centres were chosen so that screening and other follow-up activities were closer to the community. The main recruitment centre was at Got-Agulu Health Centre. At the recruitment centres. the traditional birth attendants accompanied their clients for enrolment by the field worker and eventual screening and recruitment. The inclusion criteria for recruitment were:

- Permanent residency within the study area and likely to stay for more than 2 years.
- Pregnancy at gestational age of 14-24 weeks.
- A haemoglobin level of 7.0 g/dl and above.
- No chronic disease
- Those giving informed consent to participate.

The exclusion criteria were:

- Non-consenting women
- Non-residents:
- Those with haemoglobin less than 7.0 g/dl.
- Those who were pregnant with gestational age above 24 weeks.

At the health centre. there were several stages before registration. The client. after being confirmed as being from the study area through the demography booklet. was given a small card with her name and personal number. The next stage involved determining her gestational age. This was done through ultrasound examination by a nurse trained in ultrasonography. The client was referred to the laboratory where a blood sample was collected for haemoglobin estimation using a portable. battery-operated β-haemoglobin photometer detection system (HemoCue^R AB. Ängelholm. Sweden) which gave results in g/dl of blood. Only those with a haemoglobin level above 7.0 g/dl were considered for recruitment. Those who did not meet the criteria were advised to continue with normal antenatal clinics. Those whose gestational age was below 14 weeks were re-booked to come at a later date for recruitment.

After confirmation of eligibility, the client was registered and personal data including obstetric history recorded. History of earth eating (geophagy) was recorded on geophagy questionnaire. Informed consent was taken after a verbal explanation of the purpose of the study.

2.8.6 Field organization and questionnaires

The client was given an appointment card in a folder on which the date for the next visit was indicated. All appointments for various tests were indicated on this card. Whenever the client came to the clinic, a sticker was placed in the appropriate space as a marker for her attendance. A file was opened at the institution in which all information about the client was kept. The ultrasound room was used by the nurse for ultrasonography and medical examinations. In the laboratory, record books were kept for all tests carried out on the client. All material leaving the field, including questionnaires and specimens for analysis and register books, were recorded, with whoever collected them signing for them.

2.8.7 Assessment of geophagy among pregnant women

Qualitative assessment of geophagy was carried out by field assistants in local language (dholuo) to identify women who ate soil. This was done through interviews as indicated in the questionnaire. The questions about soil eating were repeated during follow-up visits at midterm and after delivery. The quantity of soil consumed was assessed by weighing the amount of soil indicated by the client.

2.8.8 Sample collection

2.8.8.1 Soil

The second stage of the study involved sample collection which was carried out by individuals trained and supervised by the principal investigator. Earth samples were collected from all clients who reported eating earth. An appointment was given to the client at the earliest convenience for the field assistant to accompany her to the place where she collected the earth. About

100 g of earth were collected from the spot and placed in black plastic bags. The earth was labelled with the name of client, date, place of collection, type of earth and village. For those who ate the soft stone locally known as "odowa", usually purchased from the local market or kiosk, the client had to offer leftovers or be accompanied by the field assistants when purchasing some of it. The collected soil was then taken to the laboratory for further analysis. This earth was collected by the client in the same way she collected hers for consumption (Geissler *et al.* 1998a).

2.8.8.2 Determination of earth contamination with geohelminth eggs

The sugar floatation method was used to isolate helminth eggs from the earth samples. Earth samples from the field were dried on brown Manila paper in the laboratory at room temperature. The soil was crushed between the papers into fine powder and sieved through 100 µM mesh. The sieved soil was weighed in two portions of 2 g each and put in two test tubes. The earth was mixed with a bleaching solution then mixed on a shaker for one hour. Five ml of saturated sugar solution was added to the mixture and shaken on a vortex for 30 minutes. The tubes were further centrifuged for 15 minutes at 1500 rpm. The eggs were picked by placing a cover slip on top of the contents in the test tube and examining them under the microscope at power x10 and x40. The last procedure was repeated in order to pick out all eggs available in the sample (Schultz & Kroeger, 1992). The remaining soil samples were stored in sealed plastic papers to keep them free of moisture.

2.8.8.3 Determination of silica in earth

The sieved earth samples were taken to Nairobi where the investigator processed them for silica content. Using an analytical weighing balance, 0.05 g of earth was weighed and placed in a nickel crucible containing 10 ml of 15% evaporated sodium hydroxide. The crucibles were placed in a muffle furnace 10 at a time. The contents were heated to 800°C maintaining the temperature at 800°C for five minutes. The contents were removed and allowed to cool before adding water to about three quarters full and allowed to stand overnight. The contents were then diluted to 1 litre with a 1% acid solution.

The sample was prepared for reading by pipetting 10 ml of the sample. then adding a reducing agent and a colour development solution and measured at wavelength 815 nm by a spectrophotometer (Perkin-Lambda 15 uv/vis Spectrophotometer). The blank and the standard reference solution were treated in the same way as the test. A standard reference curve was drawn from the optical density readings obtained by diluting a standard silica stock solution in seven dilutions of 1.0. 1.5. 2.5. 5.0. 7.5. 10.0 and 13 ml in 100 ml of water. and the absorbance obtained.

2.8.8.4 Determination of iron concentration in earth

Earth was collected and prepared as stated earlier. The already pulverized earth was stored in sealable plastic bags. Five grams of the earth sample were weighed into an acid washed plastic container. Iron was extracted from the earth by adding 5 ml of 0.1 NHCL to the sample and mixing thoroughly by use of a mechanical mixer for one hour. The mixture was filtered through Whitman filter paper into an acid washed plastic container.

The concentration of iron in the earth was determined by aspiration of the filtrate to an atomic absorption spectrophotometer (Flame emission spectrophotometer AA-68. Shimadzu model) at 248.3 nm.

2.8.8.5 Stool collection and examination

During recruitment. plastic poly pots were given to the registered women with instructions to put in a portion of stool not less than 15 grams. This was done by demonstrating with a small lump of earth the least amount to be collected. Stool was brought in and handed over to the laboratory technicians who labelled it accordingly. The label included name of client. serial number. date and category of stool according to when it was collected.

Duplicate 50 mg modified Kato smears of stool specimens were prepared (WHO 1991) from two samples collected on consecutive days.

The slides were read microscopically at objective x10 by two qualified technicians immediately for hookworm and then kept overnight in order to clear for other helminth eggs. The eggs were counted and the results recorded as eggs per gram of stool (epg) after obtaining the mean of the two slides. To obtain the number of eggs in one gram, the average of the two was multiplied by 20. The rest of the sample was kept in the freezer at -30°C for the estimation of silica. A 10% sample from the examined slides was processed for quality control.

Stool sample for silica was dried in an oven at 100°C to dry weight. This was to ensure that no moisture remained in the sample. The samples were analysed for silica in Nairobi at the Centre for Public Health Research (CPHR) using the same procedure as for earth. The only difference was that the weight of the stool sample used for analysis was 0.1 g instead of 0.05 g for soil, because soil has a higher concentration of silica than stool.

2.8.8.6 Blood sample

Blood samples were collected at the main Health Centre. This was carried out by the investigator assisted by qualified laboratory staff. The initial sample was a finger prick sample meant for screening, in order to determine the level of haemoglobin before recruiting the client into the study. The method used was convenient because as little as one drop of blood was enough to fill the cuvette for the haemocue machine that was used. Haemoglobin (Hb) concentration, was measured using a coulter counter machine.

The remaining blood samples were centrifuged and the plasma separated and placed in cryo-tubes covered with aluminium foil and kept in liquid nitrogen for further analysis of serum ferritin which was carried out in Copenhagen, Denmark by the ELISA technique.

2.9 Data analysis

Data were entered using SPSS^{TM} software in duplicate, validated and cleaned. Categorical variables were analysed using frequency distributions and differences in proportions were assessed using the chi-square tests. The significance limit was chosen at $P<0.05$. Group means were calculated for continuous variables. Skewed data of densities of intestinal helminth eggs were logarithmically transformed and geometric means determined. Significant differences were determined by *t*-test for paired data while analysis of variance (ANOVA) was used to compare those of more than two groups. Logistic regression was used to analyse determinants of geophagy and occurrence of parasite infections at given time points.

2.10 Ethical considerations

Prior to initiating the field work in the study area. authority was sought from the Kenyan Government for permission to undertake the research. An application was made to the National Ethical Committee for approval of the proposal and consent to carry out the study. The proposal was approved by the Kenyatta National Hospital Scientific and Ethical Committee on the Kenyan side. and the Danish National Ethical Committee on behalf of the collaborating institutions in Denmark. This was followed by the introduction of the study to the local leaders in the study area through seminars organized by the provincial administration in Nyanza Province. All the leaders from the Provincial Commissioner. the District Commissioner and down to the division and location chiefs were briefed in detail. The local administrators in turn organized barazas. where community members were briefed. During recruitment of clients. a written consent form was read to the clients who were encouraged to sign after understanding the purpose of the study and to accept to join voluntarily. All medical procedures were carried out according to Ministry of Health policies. The District Medical Officer of Health participated as one of the local consultants in case of emergencies. Cases needing medical attention were referred to the clinician. Finally. the confidentiality of the respondents was taken fully into consideration.

Chapter 3

Geophagy among pregnant and lactating women in Bondo District, Western Kenya

3.1 Introduction

Geophagy is the habit of deliberate eating of earth. which is common in many societies in the world (Gelfand 1945: Anell & Lagercrantz 1958: Halsted 1968: Abrahams & Parsons 1996). It has sometimes been classified as an "aberrant" behaviour or a "perversion of appetite" (Wong *et al.* 1988). Research on geophagy has relied on the western common sense assumption that eating 'dirt' is detrimental to health. Because of this view. geophagy has not been fully studied and in recent reviews. questions have been raised as to its potential importance for health (Lacey 1990: Reid 1992). Existing studies on geophagy have been based on hospital and institution cases and mentally disturbed individuals (Wong and Simeon 1993). Only in recent years. have studies on geophagy been carried out among schoolchildren in western Kenya (Geissler *et al.* 1997) in KwaZulu-Natal South Africa (Saathoff *et al.* 2002) and in Guinea (Glickman *et al.* 1999) and in pregnant women in Kilifi District. Kenya (Geissler *et al.* 1998) and Namibia (Rainville 1998). The aim of the present study is to investigate the epidemiology and health impact of geophagy in a longitudinal study conducted in pregnant and lactating women in western Kenya. This chapter reports the first results of the study.

3.2 Study area and population

The study was conducted in two Locations in Usigu Division. Bondo District. Nyanza Province. Kenya. Detailed description of the area and population is as explained in chapter two.

3.3 Study design

The study was part of a longitudinal intervention cohort study undertaken by the Kenyan-Danish Health Research Project (KEDAHR) between 1998 and 2001. Expectant women were recruited between week 14 and 24 of gestational age. Questionnaire interviews were administered at baseline and followed up at mid-term. delivery. three and six months postpartum.

3.4 Methods

At recruitment. pregnancy was confirmed by the nurse through screening and the gestational age was determined by ultrasonography. Questionnaires were administered after the woman had signed the consent form. The interviews from a pre-tested questionnaire were carried out at Got-Agulu health centre where ultrasonography facilities were available. All recruited women were interviewed by a trained female field assistant in a secluded area and their obstetric history taken by a nurse. The individual interviews were conducted by a female field assistant after an introductory group discussion about earth eating behaviour with the women. as outlined by Geissler and co-workers (1997). After the interview. the women were asked to estimate their daily earth intake by picking an appropriate amount from an earth sample. which was then weighed. Information collected from pregnant women through structured interviews at recruitment and subsequent follow-up examinations included history of earth eating. socioeconomic factors and personal hygiene. This information was later linked to parasitological data on intestinal helminth infections. Where information about sanitation was not clear. the women were followed to their homes for further confirmation. During the home visits the presence of latrines and their usage was noted.

3.4.1 Silica estimation in stool

A total of 204 stool samples were picked randomly from the 821 samples collected from the recruited pregnant women in order to assess their silica content. The stool was dried at 100^{0}C in a hot air oven to remove moisture. The dry stool was crushed and stored in plastic containers. Using an analytical weighing balance, 0.1 g of stool was weighed and placed in a nickel crucible containing 10 ml of 15% evaporated sodium hydroxide. The crucibles were placed in a muffle furnace, heated to 800^{0}C maintaining the temperature for 5 minutes and then allowed to cool before adding water to about three quarters full and allowing to stand overnight. The contents were then diluted to 1 litre using 1% hydrochloric acid solution.

The sample was prepared for reading by pipetting 10 ml of the sample, adding a reducing agent and a colour development solution, and measured at wavelength 815 nm by a spectrophotometer (Perkin-Lambda 15 uv/vis Spectrophotometer). The blank and the standard reference solution were treated in the same way as the test. A standard reference curve was drawn from the optical density readings obtained by diluting silica stock solution in seven dilutions of 1.0, 1.5, 2.5, 5.0, 7.5, 10.0 and 13 ml in 100 ml of water and the absorbance obtained (Fig. 3.1)

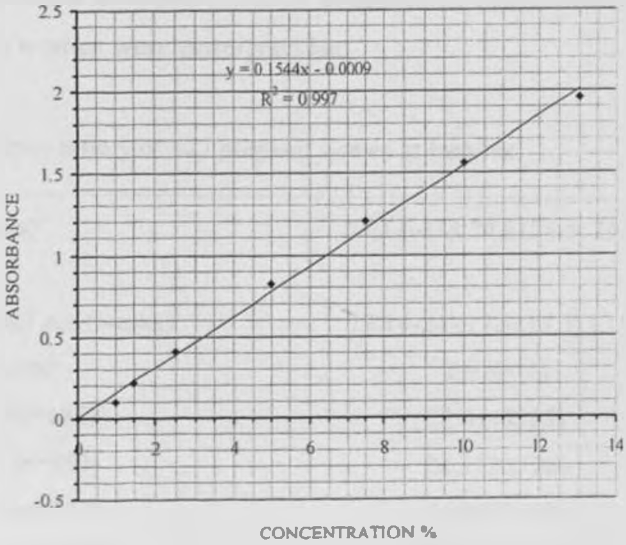

Figure 3.1 Standard reference curve for silica concentration in percentage (at 815 nm), optical density (◆) and linear OD.

3.4.2 Data analysis

Data were entered and analysed using SPSS^{TM} software. The χ^2 test was used to test for differences in proportions, the McNemar test was used to test for difference in paired observations and Wilcoxon signed ranks test was used to test for difference in means of paired observations. A significance level of 0.05 was used for all tests.

3.5 Results

A total of 1,418 eligible pregnant women reported for interviews at recruitment centres between September 1998 and February 2001. Out of these, 511 were excluded for not having permanent residence in the study area; 35 women had passed the required gestational age; 15 did not consent; 13 came for the second pregnancy after delivery and had therefore been registered earlier and 9 absconded after recruitment. This left 827 women who met all the criteria and were included in the study.

As seen in Table 3.1, the age range of those recruited was 14-47 years with a median of 23. The gestational age range was 14-24 weeks with a median of 17. Parity ranged between 0 and 11 pregnancies with a median of 2; 22% (n=182) of the women were primigravidae.

Table 3.1 Obstetric history of 827 pregnant women at baseline.

Age (years)[1]	23(14-47); 24.8 (24.3; 25.2)
Gestational age (weeks)[1]	17(14-24);17.5 (17.3; 17.7)
Parity[2]	2.0 (0-11)
0 (n=182)[3]	22.0 (19; 25)
1 (n=193)	23.3 (20; 26)
2 (n=132)	16.0 (13; 19)
3~ (n=320)	38.7 (35.3; 42.1)

[1] Median (range) Mean (95% confidence interval);
[2] Median (range);
[3] Percentage (95% confidence interval)

3.5.1 Socioeconomic characteristics

As shown in Table 3.2. only 2.0% of the women were in paid employment as civil servants. including teachers. compared to 54.2% without specific occupation: the rest were involved in small-scale business. Only 7.8% were in the highest bracket of income of Ksh. 5000: 16.1% had attended school beyond primary level. while 6.3 % did not have any formal education. The recruited women identified themselves mainly as Luo (91.1%) and most of them were married (82.8%). The number of dependent children living with the women ranged from 0-11. the median being three. As for the quality of housing in the area. most of the houses were typical mud-walled huts thatched with grass. 60.7%. and only 24.3 % (n= 201) had cement plastered walls: 63.8% of the homes had pit latrines (n=523).

Table 3.2 Socioeconomic characteristics of 827 pregnant women at baseline[1].

Occupation	
Small business (n=259)	31.3 (28.0; 34.0)
House wife (n=418)	50.5 (47.0: 54.0)
Farmer (n=94)	11.4 (9.2; 13.6)
Tailor (n=9)	1.1 (0.8: 1.4)
Civil service (n=16)	2.0 (1.0: 3.0)
No occupation (n=31)	3.7 (2.4: 5.0)
Income per month (KSh)	
<1000 (n=273)	33.0 (29.7; 36.3)
1000-2999 (n=174)	21.0 (18.2: 23.8)
3000-4999 (n=233)	28.2 (25.0; 31.3)
≥5000 (n=147)	7.8 (15.1; 20.5)
Education level	
None (n=52)	6.3 (4.6; 8.0)
Primary (n=642)	77.6 (74.7: 80.5)
Secondary (n=133)	16.1 (13.6; 18.6)
Marital status	
Single (n=102)	12.3 (10.0: 14.6)
Married (n=685)	82.8 (80.2; 85.4)
Widows (n= 40)	4.8 (3.3; 6.3)
Tribe	
Luo (n=753)	91.1 (89.0; 93.0)
Luhya (n= 65)	7.9 (6.0; 9.8)
Others (n=9)	1.0 (0.3; 1.7)
Presence of latrine	
No (n=304)	36.8 (33.3: 40.3)
Yes (n=523)	63.8 (60.3: 67.3)
Roof of house made of	
Grass thatch (n=502)	60.7 (57.3: 64.1)
Iron sheets (n=325)	39.3 (35.9: 42.7)
Wall of house made of	
Mud (n=626)	75.7 (72.7: 78.7)
Cement (n=201)	24.3 (21.3; 27.3)
Number of dependants	
0 (n=126)	15.2 (12.7; 17.7)
1 (n=43)	5.2 (3.6: 6.8)
2 (n=130)	15.7 (13.2: 18.2)
3 (n=143)	17.3 (14.7; 19.9)
4 (n=385)	46.6 (43.1; 50.1)

[1] Percentage (95% confidence interval).

3.5.2 History of earth eating

Among the 378 women who ate earth at recruitment, 65.3% had eaten earth before pregnancy, while only 6.5% of the non-geophagous had eaten earth before this pregnancy (*P*<0.0001) (Table 3.3). Most of the geophagous as compared to non-geophagous women had eaten earth as young girls (*P*<0.0001).

Table 3.3 History of earth eating among 827 pregnant women at baseline.

	Geophagous (n = 378)	Non-geophagous (n = 449)	*P*- value
Ate earth before pregnancy[1]			<0.0001
No (n=551)	34.7 (29.8; 39.6)	93.5 (91.2; 95.8)	
Yes (n=276)	65.3 (60.4; 70.2)	6.5 (4.2; 8.8)	
Ate earth as young girls[1]			<0.0001
No (n=660)	61.4 (56.4; 66.4)	95.3 (93.3; 97.3)	
Yes (n=167)	38.6 (33.6; 43.6)	4.7 (2.7; 6.7)	

[1] Percentages (95% confidence interval).

3.5.3 Background characteristics of pregnant women by geophagy

As shown in Table 3.4, the prevalence of geophagy was higher in those below compared to those above 30 years of age (47.4 vs. 40.6%) and in those with only primary compared to secondary education (47.0 vs. 39.1%), although the differences were not statistically significant (*P*=0.08 and *P*=0.09, respectively). Likewise, there was an indication that those with dependants were more geophagous than those without dependants (46.9 vs. 38.9, *P*=0.09). Civil servants had a much lower prevalence of earth eating than the non-salaried women. Otherwise, the prevalence of geophagy did not differ in the categories of parity, gestational age, income, marital status, ethnic group or other socioeconomic characteristics.

Table 3.4 Background characteristics among 827 pregnant women by geophagy at baseline.

	Geophagous[1] (n=378)	Non-geophagous[1] n=449)	*P*- value
Age (years)			0.08
Below 30 (n=620)	47.4 (43.4; 51.4)	52.6 (48.6; 56.6)	
30 and above (n=207)	40.6 (33.8; 47.4)	59.4 (52.6; 66.2)	
Parity			0.65
0 (n=182)	45.1 (37.7; 52.5)	54.9 (47.5; 62.3)	
1 (n=193)	45.6 (38.5; 52.7)	54.4 (47.3; 61.5)	
2 (n=132)	52.3 (43.6; 61.0)	47.7 (38.7; 56.4)	
3 (n=95)	42.1 (32.2; 52.1)	57.9 (47.9; 67.9)	
4 (n=87)	46.0 (35.4; 56.6)	54.0 (43.4; 64.6)	
5+(n=138)	42.8 (34.4; 51.2)	57.2 (48.8; 65.6)	
Gestational age (weeks)			0.98
14 (n=189)	45.5 (38.3; 52.7)	54.5 (47.3; 61.7)	
15-16 (n=217)	46.1 (39.4; 52.8)	53.9 (47.2; 60.6)	
17-20 (n=259)	46.3 (40.1; 52.5)	53.7 (47.5; 59.9)	
21-24 (n=162)	44.4 (36.6; 52.2)	55.6 (47.8; 63.4)	
Number of dependants			0.09
0 (n=126)	38.9 (30.8; 47.5)	61.1 (52.5; 69.7)	
1+ (n=701)	46.9 (42.9; 50.9)	53.1 (49.1; 57.1)	
Main occupation			0.25
Business (n=259)	49.0 (43.0; 55.0)	51.0 (45.0; 57.0)	
Housewife (n=418)	45.5 (40.7; 50.3)	54.5 (49.7; 59.3)	
Farmer (n=94)	44.7 (34.7; 54.7)	55.3 (45.3; 65.3)	
Tailor (n=9)	33.3 (2.0; 64.6)	66.7 (35.4; 98.0)	
Civil servant (n=16)	18.8 (3.0; 34.6)	81.3 (61.8; 100.3)	
No occupation (n=31)	41.9 (24.2; 59.6)	58.1 (40.4; 75.8)	
Income per month: (KSh)			0.38
< 1000 (n=273)	43.2 (37.2; 49.2)	56.8 (50.8; 62.8)	
1000-2999 (n=174)	46.0 (38.5; 53.5)	54.0 (43.5; 61.5)	
3000-4999 (n=233)	50.2 (43.6; 56.8)	49.8 (43.2; 56.4)	
=> 5000 (n=147)	42.9 (34.9; 50.9)	57.1 (49.1; 65.1)	
Educational level			0.09
Primary (n=694)	47.0 (43.3; 50.8)	53.0 (49.2; 53.8)	
Secondary (n=133)	39.1 (30.7; 47.5)	60.9 (52.5; 69.3)	
Marital status			0.16
Single (n=102)	39.2 (29.6; 48.8)	60.8 (51.2; 70.4)	
Married (n=725)	46.6 (43.0; 50.2)	53.4 (49.8; 57.0)	
Tribe			0.82
Luo (n=753)	45.7 (42.1; 49.3)	54.3 (50.7; 57.9)	
Luhya (n=65)	44.6 (32.3; 56.9)	55.4 (43.1; 67.7)	
Others (n=9)	55.6 (22.6; 88.6)	44.4 (11.4; 77.4)	
Presence of latrine			0.19

Yes (n=523)	48.7 (43.0; 54.4)	51.3 (46.6; 57.0)	
No (n=304)	44.0 (39.7; 48.3)	56.0 (51.7; 60.3)	
Roof of house made of			0.93
Grass (n=502)	45.8 (41.4; 50.2)	54.2 (49.8; 58.6)	
Iron-sheet (n=325)	45.5 (40.0; 51.0)	54.5 (49.0; 60.0)	
Wall of house made of			0.13
Mud (n=626)	44.2 (40.2; 48.2)	55.8 (51.8; 59.8)	
Cement (n=201)	50.2 (43.2; 57.2)	49.8 (42.8; 56.8)	

[1] Percentages (95% confidence interval).

3.5.4 Earth eating habits before and after pregnancy

The percentage of women eating earth decreased after delivery as shown in Table 3.5. At baseline 45.7% ate earth and at mid-term 46%. This percentage dropped significantly, to 34.5% at three months postpartum and to 29.6% at six months postpartum (*P*<0.001). Likewise, the mean earth intake per day declined significantly between mid-term and three months postpartum to six months postpartum (*P*=0.001) (Fig.3.2). There were significant differences in the frequency of daily earth intake between the third and sixth month postpartum and baseline, but no difference was noted between mid-term and baseline. The number of women eating earth more than once daily seemed to decrease after delivery. At baseline, 54.2% (n=205) of the women ate soft stone purchased at the local market where it is found cheaply. Those who ate earth from ant hills, which is readily available in the fields and sometimes in homesteads, were 42.8% (n=158). The remaining 4.0% (n=15) ate earth from the wall of huts (11), gullies along paths (2) and from streams, as well as black cotton soil (1). At mid-term, the percentage of those who ate the different types of earth was similar to the baseline, with a slight increase in those who ate from ant hills.

3.5.5 Interview results and assessment of silica as a soil-tracer

Two hundred and four stool samples were analysed. Out of these, 126 were non-geophagous and 78 geophagous (38.2%) according to the interviews. The mean silica content was 2.05% of the dry weight of stool (median 1.0%, range 0.08-12.2%). Geophagous women had a higher mean than the non-geophagous (3.1 vs. 1.4%, *P*<0.001). The mean silica content was higher in those who ate soft stone as compared to anthill, 3.4 vs. 3.0%. Using 1% as the cut-off point to distinguish geophagous from non-geophagous women, 42.6% of the women had high silica content in stool and 57.4% low silica content. Faecal silica and reported geophagy were strongly associated (*P*<0.001). As a test of geophagy - taking the interview results as the golden standard - its sensitivity would be 53.0% and its specificity 72.6%.

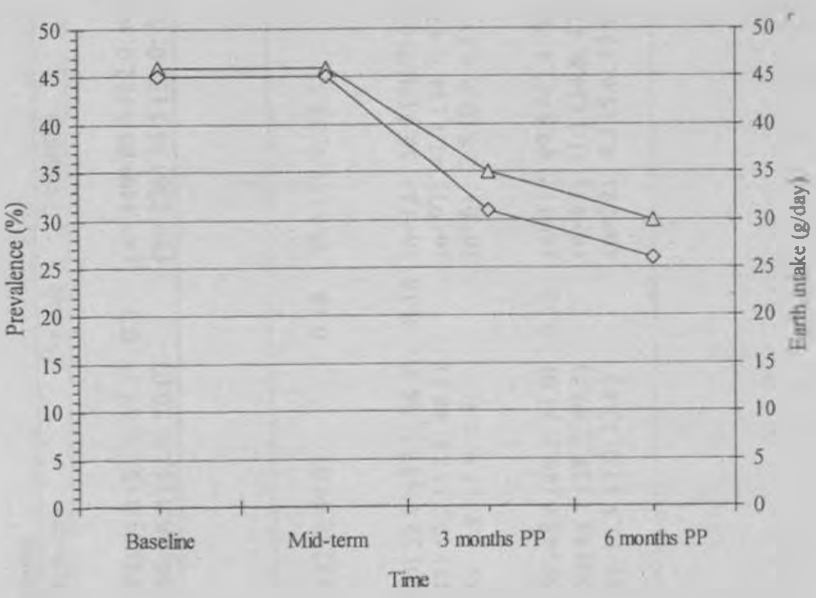

Figure 3.2 Prevalence of geophagy (Δ) and amount of earth intake (◊) among women from baseline to six months postpartum.

Table 3.5 Earth eating habits during and after pregnancy.

	Pregnancy			Postpartum			
	Baseline	Mid-term	*P*-value	3 months	*P*- value	6 months	*P*- value
Earth eating[1]							
No	(n= 449) 54.3 (50.8; 57.8)	(n= 399) 54.0 (51.3; 57.7)	0.5	(n= 449) 65.5 (62.0; 69.0)	< 0.001	(n= 457) 70.4(66.8; 74.0)	< 0.001[3]
Yes	(n= 378) 45.7 (42.2; 49.2)	(n= 340) 46.0 (42.0; 50.0)		(n= 236) 34.5 (31.0; 38.0)		(n= 192) 29.6(26.0; 33.0)	

Among earth eating women only							
Earth intake (g/day)[2]	44.5 (41.5; 47.4)	45.4 (42.0; 49.0)	0.18	30.6 (27.3; 33.3)	< 0.001	25.5 (23.0; 28.0)	< 0.001[4]
Type[1]							
Soft stone	(n= 205) 54.2 (50.7; 57.7)	(n=180) 52.9 (47.5; 58.3)	0.16	(n=133) 56.4 (50.0; 62.8)	< 0.001	(n=100) 52.1 (44.9; 59.3)	< 0.001[4]
Anthill	(n=158) 42.8 (39.4; 46.2)	(n=147) 43.2 (37.8; 48.6)		(n=97) 41.1 (34.7; 47.5)		(n= 87) 45.3 (38.1; 52.5)	
Others	(n=15) 4.0 (2.0; 6.0)	(n=13) 3.9 (1.9; 5.9)		(n=6) 2.5 (0.5; 4.5)		(n=5) 2.6 (0.3; 4.9)	
Frequency[1]							
1/day	(n=180) 47.6 (44.2; 51.1)	(n=155) 45.6 (40.2; 51.0)	0.55	(n=117) 49.6 (43.1; 56.1)	< 0.001	(n=98) 51.0 (43.8; 58.2)	< 0.001[4]
2/day	(n=164) 43.4 (40.0; 46.8)	(n=150) 44.1 (38.7; 49.5)		(n=97) 41.1 (34.6; 47.4)		(n=76) 39.6 (32.6; 46.6)	
3+/day	(n= 34) 9.0 (6.1; 11.9)	(n=35) 10.3 (7.0; 13.6)		(n=22) 9.3 (5.6; 13.0)		(n=17) 9.4 (0.6; 18.2)	

[1] Percentages (95% confidence interval)
[2] Mean (95% confidence interval)
[3] McNemar Test
[4] Wilcoxon signed rank test

3.6 Discussion

Geophagy is very common among pregnant women in the study area. This is corroborated by earlier reports from East Africa (Anell and Lagercrantz 1958; Geissler *et al.* 1998; Prince *et al.* 1999), where it appears to be widely accepted as a common pregnancy practice (Gelfand 1945; Hunter *et al.* 1984; Horner *et al.* 1991). In a study conducted on the Kenyan coast at the ANC, 56% of the pregnant women gave a history of eating earth (Geissler *et al.* 1998b) and a study among the Ewe of Ghana found that 63% of pregnant women were geophagous (Vermeer 1971). In the study presented here, the prevalence of geophagy in pregnant women was slightly lower than the 54 % we reported from a pre-study conducted in health institutions in the same area and differed significantly from the 71% prevalence for the women interviewed in their homes in that study (Prince *et al.* 1999). Although the sample size was smaller (57) in the pre-study compared to the current study (827), it is possible that there was a response bias in the study presented here due to the interview location, as women in rural western Kenya tend to be inhibited by the hospital setting.

The mean daily earth intake as estimated in this study was similar to the amount reported for pregnant women from the coast of Kenya (41.5 g by Geissler *et al.* 1998b) and for women in West Africa (40-80 g by Hunter 1984b and 30 g by Vermeer 1971). Schoolchildren in our study area in Yimbo, western Kenya were reported to consume an average of 28 g of earth in a day with a range of 8-108 g (Geissler *et al.* 1997); 45% of the women ate earth 1-2 times daily, similar to women at the Kenyan coast (Geissler *et al.* 1998b). The earth types were similar to those eaten by schoolchildren in the study area (Geissler *et al.* 1997) and have been described earlier in East Africa (Anell & Lagercrantz 1958; Kilbride & Kilbride 1993). The pregnant women in western Kenya ate earth from termite mounds more frequently than coastal women, probably because of the scarcity of ant hills in the coastal study area (Geissler *et al.* 1998b). More of the pregnant women ate soft stone as compared to schoolchildren in the same area, who ate more commonly of the termite hill earth (Geissler *et al.* 1997), probably because soft stone was purchased from markets at a price of 2 KSh. for a 100g lump (0.015 £) and the children could not afford it. while ant hills are common in the area. The women said the soft stone was smooth in the mouth and free from sand. Some thought it was more hygienic than collecting earth from ant hills in the fields and that it was easier to carry around because it was solid. unlike ant hill earth which is sometimes loose. However those who ate from termitaria said it had a very good taste. The women may have over-reported the consumption of soft stone because it was seen as cleaner and smarter than picking ant hills from the fields. The increase of ant hill earth as compared to soft stone between the first and second survey might then be explained by the growing familiarity and openness of the women. A relatively small percentage of the women ate earth from mud walls in the study area as compared to coastal women (Geissler *et al.* 1998). possibly because they are smeared with cow dung to protect them from cracking and because women in western Kenya really value their walls after decorating them and would not allow any interference.

Earth eating habits of older and younger women corroborate earlier findings from the study area that there were no age-related differences in earth eating among the child bearing age group (Prince *et al.* 1999) and from the USA where no difference in pica in respect to age could be shown (Smulian *et al.* 1995). In this community it is assumed that earth eating is normal in women. hence the prevalence remains the same in girls and changes in boys as they mature (Geissler *et al.* 1997). This has also been reported in Zambian schoolchildren where more girls than boys ate earth (Nchito *et al.* 2004). Older boys or men are rarely seen eating earth in this community because they felt it was shameful (Geissler *et al.* 2000). Most of the women said they started eating earth in the second trimester. This was confirmed by the quantitative analysis since most of the women ate earth at recruitment (14-24 weeks gestational age) and the number only declined after delivery. This agrees with other studies in Africa that associated geophagy with pregnancy. but without any quantitative evidence (Anell & Lagercrantz 1958). There was no significant difference between geophagous and non-geophagous women by parity. This differs with results from the coast. among which those with higher parity were more likely to eat earth (Geissler *et al.* 1998). Our earlier report in the area did not show differences in parity (Prince *et al.* 1999). The women who had gone to school beyond primary level were less often geophagous than those below primary level of education. Although this was only borderline significant. it could be attributable to the knowledge and understanding of the risks involved in earth eating. However. it could also be a consequence of greater inhibition in reporting earth-eating among the more educated women.

The agreement between interviews and stool silica assessments as a means of determining geophagous and non-geophagous women was satisfactory. Taking the silica estimation results as the golden standard. the sensitivity was 59% and specificity 67.5%. We preferred interviews to soil tracer methods (Wong *et al.* 1988: Stanek & Calabrese 1995) because our primary objective was to identify geophagous women and this approach was more appropriate for field conditions than the more expensive and time consuming soil tracer methods. The distribution of earth in the stool could be uneven and taking only one stool sample would not give satisfactory results. Moreover the method would trace those with high dietary intake of silica and not necessarily the geophagous women. It has been shown that silica is passed out in the first 24 hour's stool and that the percentage of stool silica is heavily dependent on the time after ingestion. This makes it difficult to quantify the individual intake based on stool.

The silica content in the women's stool was higher than that of schoolchildren from the area (Geissler *et al.* 1997). This can probably be attributed to the fact that the amounts of earth ingested by the women were also higher than those of children.

3.7 Conclusion

Geophagy is very common among pregnant and lactating women in our study area in western Kenya. This could lead to a higher risk of infection with soil transmitted intestinal helminths. which would be likely to have a negative impact on the health of the pregnant woman and the child. On the other hand. earth eating might also fulfil positive functions. such as nutritional supplementation or the countering of pregnancy-related nausea. It is therefore important to carry out more studies on the subject. on its role as a risk factor in the transmission of intestinal helminths and its impact on nutrition. particularly iron.

Owing to the diversity of earth eating habits and its deeply rooted nature in communities like the one we studied. research should be planned to cater for both socio-cultural and biomedical understanding of the potential effects of geophagy on individuals and communities.

Chapter 4

Risk factors for intestinal helminth infections among pregnant women

4.1 Introduction

Intestinal helminths are widespread within the tropical and subtropical regions of the world. Recent WHO estimates have shown that total numbers of people infected with helminths could be over 3 billion: of these. 1.45 billion people are infected with *Ascaris lumbricoides*. 1.05 billion with *Trichuris trichiura*. and 1.3 billion with hookworm. Morbidity of 350 million. 220 and 150 million for *Ascaris*. *Trichuris* and hookworm respectively (WHO 2002). The annual mortality for *A. lumbricoides* is estimated to be 60.000. hookworm 65.000 and for *T. trichiura* 10.000 (WHO 2002). Up to 0.6 billion people are at risk of infection with schistosomiasis while over 200 million are infected worldwide (WHO 1998). Current estimates of the total number of individuals at risk of infection with *Schistosoma mansoni* in sub-Saharan Africa is 393 million while 54 million are infected (WHO 1999). In 1998 WHO estimated that schistosomiasis and soil transmitted helminths were responsible for more that 40% of the disease burden due to tropical diseases (excluding malaria) (WHO 1999). In Kenya the prevalence of these infections ranges between 10 and 80% in different regions from the coast to the shores of Lake Victoria (Hall *et al*. 1982; Latham *et al*. 1982; Chunge *et al*. 1985: Magnussen *et al*. 1997; Geissler *et al* 1997; Brooker *et al* 2000: Olsen *et al* 2000: Thiong'o *et al*. 2001).

Studies on the relationship between geophagy and the transmission of intestinal helminths have been carried out among schoolchildren in western Kenya (Geissler *et al*. 1998.) and KwaZulu Natal. South Africa (Saathoff *et al*. 2002). finding a positive association between geophagy and infections. particularly with *Ascaris lumbricoides*. Various other studies have found environmental. occupational and socioeconomic factors to be positively associated with intestinal helminth infections (Henry *et al.* 1988; Valerie *et al.* 1995; Olsen *et al.* 2000).

In this study, we aimed at determining the prevalence and intensity of intestinal helminths in pregnant and lactating women and related them to potential risk factors such as geophagy, environmental and socioeconomic conditions.

4.2 Materials and methods

4.2.1 Study area and population

The study area and population are as described previously in chapters 2 and 3.

4.2.2 Parasitological examination

Stool samples were collected on two consecutive days and duplicate 50 mg Kato Katz cellophane thick smears made (WHO 1991). The slides were examined within an hour for hookworm eggs, and on the following day for other intestinal helminth eggs. Intensity was expressed as the mean eggs per gram of faeces (epg) of the two samples. Individuals infected with geohelminths were treated with Mebendazole (Vermox) 500 mg given as a single dose as per instruction from the District Medical Officer. This was done after the second sample was examined. Those infected with *Schistosoma mansoni* were not treated during pregnancy or lactation following the Kenyan Ministry of Health drug policy on the use of praziquantel. The women were later treated during lactation under the supervision of the Health Centre staff who instructed them not to breastfeed for at least 24 hours after taking the drug. Those who could not adhere to this instruction were treated after the babies were weaned.

4.2.3 Obstetric history

Information on obstetric history was gathered in a questionnaire administered by a nurse at recruitment. The data included the age of the woman, her gestational age and her history of previous deliveries. Apart from this. they were screened for any chronic disease and high risk cases noted or excluded from the study depending on the severity of the risk.

4.2.4 Earth eating

The women's histories of earth eating habits were collected through interviews which were carried out by a trained female field worker. To have the confidence of the client. the interviews were carried out at secluded areas within the Health Centre. The information included the type of earth eaten. the amount eaten and the frequency of eating in a day. Other information included previous history of earth eating in or outside pregnancy.

4.2.5 Environmental sanitation

During the interviews. the women gave information on environmental sanitation. Data was collected on the presence of pit latrines. types of latrines and whether they were clean if used. Distance from water sources was noted and availability of clean drinking water also determined. Other factors included food handling and storage within the household. Homesteads were visited to verify some of the information given during interviews and to observe contamination of homesteads with faeces. usage of latrine and general sanitary conditions of the homestead.

4.2.6 Socio-economic information

Information on the level of education of the women and their spouses was obtained and recorded. Other socio-economic factors that were of interest included the number of dependants in the household (family size). marital status. occupation. type of house. household property and total income per month.

4.2.7 Data analysis

Data were entered in duplicate and analysed using SPSS^{TM} version 10 software. Descriptive statistics were used to get the prevalence of infections. Intestinal helminth egg counts were $\log_{10}$ transformed to get normal distribution before analysis for intensities. Chi-square tests were used to test for differences in proportions while *T*-test was used to assess differences in intensities of infection in categories with two groups and One-way Analysis of Variance (ANOVA) in those with three or more groups. A significance level of P=0.05 was used in all tests.

4.3 Results

Out of the 827 women aged between 14 and 47 years. 821 managed to produce stool for examination at recruitment. The prevalence and intensity of infection with intestinal helminths is shown in Table 4.1. *Schistosoma mansoni* was the commonest (63.0%) followed by hookworm (49.8%). *T. trichiura* (24.1%) and *A. lumbricoides* (16.3%). The mean intensity was highest in *A. lumbricoides* (815 epg) followed by hookworm (129 epg). *S. mansoni* (98 epg) and *T. trichiura* (52 epg).

Table 4.1 Prevalence and intensity of intestinal helminths in 821 pregnant women at baseline.

Hookworm		
Prevalence (%)[1]	(n=409)	49.8 (46; 53)
Intensity (epg)[2]		129 (111; 149)
A. lumbricoides		
Prevalence (%)[1]	(n=134)	16.3 (14; 19)
Intensity (epg)[2]		815 (535; 1145)
T. trichiura		
Prevalence (%)[1]	(n=198)	24.1 (21; 27)
Intensity (epg)[2]		52 (43; 64)
S. mansoni		
Prevalence (%)[1]	(n=517)	63.0 (60; 66)
Intensity (epg)[2]		98 (86; 110)

[1] Percentage (95% confidence interval); [2] Geometric mean (95% confidence interval) eggs per gram faeces in infected only;

4.3.1 Prevalence and intensity of intestinal helminths by geophagy

The intensity of *A. lumbricoides* eggs in geophagous women was higher than in non-geophagous ones but not significantly different (1000 vs. 637 epg (*P*=0.24)). The rest of the geohelminths did not show any notable difference in intensities between geophagous and non-geophagous women as seen in Table 4.2. The prevalence of *S. mansoni* was lower in geophagous women compared to non-geophagous women (59.5 vs. 65.9%) although the difference was not significant (*P*=0.06).

Table 4.2 Prevalence and intensity of intestinal helminths by geophagy in 821 pregnant women at baseline.

Infection	Geophagous (n=375)	Non-geophagous (n=446)	*P* -value
Hookworm			
Prevalence (%)[1]	48.5 (43.4: 53.6)	50.9 (46.6: 55.2)	0.50
Intensity (epg)[2]	138 (110:173)	123 (101: 148)	0.44
A. lumbricoides			
Prevalence (%)[1]	16.0 (12.2: 9.8)	16.6 (13.1: 20.1)	0.80
Intensity (epg)[2]	1000 (562:1830)	637 (383; 1059)	0.24
T. trichiura			
Prevalence (%)[1]	22.9 (18.6: 27.2)	25.1 (21.1: 29.1)	0.47
Intensity (epg)[2]	50 (38: 67)	54 (42: 71)	0.68
S. mansoni			
Prevalence (%)[1]	59.5 (54.5: 64.5)	65.9 (61.4: 70.4)	0.06
Intensity (epg)[2]	96 (79:115)	99 (83: 117)	0.81

[1] Percentage (95% confidence interval)

[2] Geometric mean (95% confidence interval): eggs per gram faeces in infected only.

4.3.2 Prevalence and intensity of intestinal helminths by type of earth eaten

Table 4.3 shows the overall prevalence and intensity of intestinal helminths. Overall there was no significant difference in prevalence and intensity of the types of intestinal helminths examined between the types of earth eaten. except for the intensity of *Ascaris* (*P*=0.01). This was as a result of a higher intensity in those who ate earth from ant hills (and other types) as opposed to soft stone.

Table 4.4 shows the prevalence and intensities of intestinal helminths by frequency of eating earth. However there were no significant differences noted in the prevalence and intensity of the helminths according to the frequency of eating soil in all.

Table 4.3 Prevalence and intensity of intestinal helminths by type of earth eaten in 821 pregnant women at baseline.

Infection	Soft stone (n=204)	Ant hill (n=156)	Other Types (n=15)	None (n=446)	*P* -value
Hookworm					
Prevalence (%)[1]	45 (38; 51)	54 (46; 62)	47 (18; 75)	51 (46; 56)	0.32
Intensity (epg)[2]	124 (89; 173)	155 (112; 220)	121 (43; 340)	123 (101; 149)	0.65
A lumbricoides					
Prevalence (%)[1]	14 (9; 18)	19 (13; 25)	13 (-06; 33)	17 (13; 20)	0.55
Intensity (epg)[2]	421 174;1013)	1905 (916; 4013)	13182 (321; 539510)	631 (384;1059)	0.01
T. trichiura					
Prevalence (%)[1]	22 (16; 27)	25 (18; 32)	21.5 (-03; 43)	25 (21; 29)	0.76
Intensity (epg)[2]	44 (29; 66)	59 (38; 93)	39 (6; 265)	55 (42; 70)	0.74
S. mansoni					
Prevalence (%)[1]	58 (52; 65)	60 (52; 67)	74 (48; 99)	66 (62; 70)	0.17
Intensity (epg)[2]	85 (66; 110)	107(78;135)	143 (77; 265)	99 (83; 117)	0.53

[1] Percentage (95% confidence interval); [2] Geometric mean (95% confidence interval) eggs per gram faeces in infected only.

Table 4.4 Prevalence and intensity of intestinal helminths by frequency of earth eating in a day in 821 pregnant women at baseline.

Infection	Once (n=178)	Twice (n=163)	Three times + (n=34)	None (n=446)	*P* -value
Hookworm					
Prevalence (%)[1]	48 (40; 55)	52 (44; 60)	38 (21; 55)	51 (46; 55)	0.46
Intensity (epg)[2]	140 (96; 204)	122 (90; 166)	180 (72; 453)	125 (103; 152)	0.78
A. lumbricoides					
Prevalence (%)[1]	16 (10; 21)	16 (10; 22)	15 (0.2; 27)	17 (13; 20)	0.97
Intensity (epg)[2]	897 (352; 2287)	1201 (506; 2851)	521 (33; 8299)	658 (398; 1091)	0.64
T. trichiura					
Prevalence (%)[1]	24 (18; 31)	23 (16; 24)	21 (06;35)	25 (21; 29)	0.90
Intensity (epg)[2]	55 (36; 84)	47 (30; 75)	43 (16; 112)	53 (42; 70)	0.92
S. mansoni					
Prevalence (%)[1]	57 (50; 65)	60 (52; 67)	71 (54; 87)	66 (62; 70)	0.12
Intensity (epg)[2]	97 (75; 127)	89 (66; 121)	112 (57; 218)	99 (83; 118)	0.89

[1] Percentage (95% confidence interval); [2] Geometric mean (95% confidence interval) eggs per gram faeces in infected only.

4.3.3 Prevalence and intensity of intestinal helminths by parity

Table 4.5 shows that primigravidae were more likely to have higher *A lumbricoides* intensity than those who had given birth before (1170 vs. 704 epg: *P*=0.28). The prevalence and intensity of *S. mansoni* were higher in para 0 compared to para 1+ respectively (*P*=0.02: *P*=0.008) respectively. There was no difference observed in the other parasites.

Tables 4.6a and 4.6b are similar to Table 4.5 but stratified by geophagy. The purpose was to find out if the effects seen in Table 4.5 were different in geophagous and non-geophagous. It was noted that the prevalence in *S. mansoni* 64.6 vs. 58% in para 0 and para 1+ respectively were not significantly different between geophagous and non-geophagous. A similar trend was seen in intensity of *S. mansoni* where a significant difference was noted in geophagous group.

Table 4.5 Prevalence and intensity of intestinal helminths by parity in 821 pregnant women at baseline.

Infection	Para 0 (n=182)	Para 1+ (n=639)	*P* -value
Hookworm			
Prevalence (%)[1]	46.2 (39: 54)	51 (47: 55)	0.26
Intensity (epg)[2]	126 (89: 179)	130 (110; 153)	0.87
A. lumbricoides			
Prevalence (%)[1]	15.4 (10: 21)	16.6 (14; 20)	0.70
Intensity (epg)[2]	1170 (635; 2158)	704 (447;1109)	0.28
T. trichiura			
Prevalence (%)[1]	25.8 (19: 32)	23.6 (20: 27)	0.54
Intensity (epg)[2]	49 (31; 76)	54 (43: 67)	0.66
S. mansoni			
Prevalence (%)[1]	70.3 (64; 78)	61 (57: 64)	0.02
Intensity (epg)[2]	131 (101; 171)	89 (77: 102)	0.008

[1] Percentage (95% confidence interval)

[2] Geometric mean (95% confidence interval): eggs per gram faeces in infected only

Table 4.6a Prevalence of intestinal helminths in 821 pregnant women by geophagy and parity at baseline.

Infection	Para 0 (n=182)	Para 1+ (n=639)	*P* -value
Hookworm[1]			
Geophagous (n=375)	41.5 (30.4; 52.3)	50.5 (44.7: 56.3)	0.15
Non-geophagous (n=446)	50.0 (40.0; 60.0)	51.2 (45.9: 56.5)	0.84
A. lumbricoides[1]			
Geophagous (n=375)	15.9 (7.9; 23.9)	16.0 (11.7: 20.3)	0.97
Non-geophagous (n=446)	15.0 (8.0; 23.0)	17.0 (13.0: 21.0)	0.62
T. trichiura[1]			
Geophagous (n=375)	23.2 (13.9; 32.5)	22.9 (18.0; 27.8)	0.95
Non-geophagous (n=446)	28.0 (19.1; 37.9)	24.3 (19.9: 28.8)	0.45
S. mansoni[1]			
Geophagous (n=375)	64.6 (54.1; 75.1)	58.0 (52.2: 63.8)	0.28
Non-geophagous (n=446)	75.0 (62.4; 83.6)	63.3 (58.3: 63.8)	0.03

[1] Percentage (95% confidence interval).

Table 4.6b. Intensity of intestinal helminths in 821 pregnant women by geophagy and parity at baseline.

Infection	Para 0 (n=182)	Para 1+ (n=639)	*P* -value
Hookworm (epg)[1]			
Geophagous (n=375)	128 (72: 230)	140 (109: 180)	0.78
Non-geophagous(n=446)	124 (79; 197)	122 (99; 151)	0.94
A. lumbricoides (epg)[1]			
Geophagous (n=375)	2027 (1011:4064)	830 (405; 1703)	0.21
Non-geophagous (n=446)	727 (272; 1939)	616 (339: 1122)	0.79
T. trichiura (epg)[1]			
Geophagous (n=375)	41 (19; 91)	53 (39: 72)	0.48
Non-geophagous (n=446)	54 (31; 94)	54 (40: 73)	0.99
S. mansoni (epg)[1]			
Geophagous (n=375)	117 (77; 177)	90 (73: 112)	0.25
Non-geophagous (n=446)	143 (100: 204)	87 (72: 108)	0.01

[1] Geometric mean (95% confidence interval) eggs per gram faeces in infected only.

4.3.4 Prevalence and intensity of intestinal helminths by socioeconomic factors

As seen in Table 4.7. the prevalence and intensity of intestinal helminth infections were not affected by the presence or absence of a pit latrine in the household. likewise neither the use of the latrine nor its cleanliness had any effect on the prevalence and intensity of the helminths. Clean homes provision of clean water and proper storage of food did not show any significant difference in the prevalence and intensity of helminths. As reported in chapter three. the less educated women were more geophagous than the ones with higher education: this is reflected by the higher prevalence of helminths among the less educated ones as compared to those with higher education especially prevalence of hookworm (*P*=0.001) as seen in Table 4.8. Although most women with no employment and with bigger family sizes were more geophagous. there were no significant differences in prevalence and intensity of helminths noted in the same groups. There were no significant differences in prevalence and intensity of helminths between older women (30+ years) and younger ones (bellow 30 years).

4.3.5 Prevalence and intensity of intestinal helminths in 821 pregnant women by distance from the lake

Six hundred and sixty-four women were recruited from villages bordering the lake shore and 157 from villages 5-10 km from the lake. As seen in Table 4.9. the prevalence and intensity of hookworm were higher in women from villages further from the lake than in those closer to the lake 57% vs. 49%. *P*=0.08 and 14.8 vs. 9.5. *P*=0.08: the prevalence and intensity of *S. mansoni* were higher in villages closer to the lake (67% vs. 46% *P*<0.001 and mean (log x+1) 20.9 vs. 7.1 *P*<0.001). There was no difference in the prevalence and intensity of the other parasites which were evenly distributed within the study area.

Table 4.7 Prevalence and intensity of intestinal helminths by presence of latrine in 821 pregnant women at baseline

Infection	Latrine present (n=520)	No latrine (n=301)	*P* -value
Hookworm			
Prevalence (%)[1]	50 (45; 54)	50 (44; 56)	0.99
Intensity (epg)[2]	121 (111; 145)	164 (112; 186)	0.26
A. lumbricoides			
Prevalence (%)[1]	15 (12;18)	19 (14; 23)	0.12
Intensity (epg)[2]	966 (590;1581)	588 (322; 1076)	0.20
T. trichiura			
Prevalence (%)[1]	22 (19; 26)	27 (22; 32)	0.11
Intensity (epg)[2]	51 (39; 65)	55 (41; 74)	0.65
S. mansoni			
Prevalence (%)[1]	61 (56; 65)	67 (62; 72)	0.62
Intensity (epg)[2]	92 (77;109)	200 (89; 128)	0.24

[1] Percentage (95% confidence interval)

[2] Geometric mean (95% confidence interval) eggs per gram of faeces in infected only.

Table 4.8 Prevalence and intensity of intestinal helminths by level of education in 821 pregnant women at baseline.

Infection	Primary and below (n= 689)	Secondary and above (n=132)	*P* -value
Hookworm			
Prevalence (%)[1]	53 (49; 56)	36 (27; 44)	0.001
Intensity (epg)[2]	135 (116; 159)	90 (56; 145)	0.08
A. lumbricoides			
Prevalence (%)[1]	17 (14; 20)	14 (8; 20)	0.36
Intensity (epg)[2]	818 (537; 1245)	590 (236:1472)	0.56
T. trichiura			
Prevalence (%)[1]	25 (22; 28)	19 (12; 26)	0.13
Intensity (epg)[2]	57 (45; 66)	39 (20; 76)	0.27
S. mansoni			
Prevalence (%)[1]	64 (60; 68)	58 (50; 66)	0.16
Intensity (epg)[2]	102 (89; 117)	73 (53; 101)	0.07

[1] Percentage (95% confidence interval)

[2] Geometric mean (95% confidence interval) eggs per gram faeces in infected only.

Table 4.9 Prevalence and intensity of intestinal helminths by distance from lake shore in 821 pregnant women at baseline.

Infection	<5 km (n=664)	>5km (n=157)	*P* -value
Hookworm			
Prevalence (%)[1]	49.0 (45; 53)	57.0 (49; 65)	0.08
Intensity (epg)[2]	9.5 (7.5; 11.9)	14.8 (9.0; 23.0)	0.08
A. lumbricoides			
Prevalence (%)[1]	17.0 (14; 19)	15.0 (10; 21)	0.70
Intensity (epg)[2]	2.1 (1.5; 2.8)	1.6 (1.0;2.6)	0.41
T. trichiura			
Prevalence (%)[1]	25.0 (22; 29)	19.0 (13; 26)	0.10
Intensity (epg)[2]	1.8 (1.4; 2.3)	1.1 (1.0; 1.7)	0.08
S. mansoni			
Prevalence (%)[1]	67.0 (63; 70)	46.0 (39; 54)	<0.001
Intensity (epg)[2]	20.8 (17.2; 25.0)	7.1 (4.5; 11.0)	<0.001

[1] Percentage (95% confidence interval)
[2] Geometric mean $\log_{10}(x+1)$ (95% confidence interval).

4.4 Discussion

Geohelminths and *S. mansoni* are prevalent in the study area. due to favourable climatic conditions including high temperatures. high humidity. adequate rainfall during the year. good soil types conducive for parasite existence and socio-cultural behaviour of the local people favouring parasite transmission.

While geohelminths were directly associated with geophagical behavior. *S. mansoni* was associated with other risk factors. This. therefore. necessitated its inclusion in the analysis of the results in this chapter.

The prevalence of *A. lumbricoides* and *T. trichiura* in this area were similar to other studies in the same region (Geissler *et al.* 1997: Olsen *et al.* 2000: Thiong'o *et al.* 2000). However. the prevalence of *Trichuris* was lower compared to that found in schoolchildren by Geissler in 1997. The prevalence of hookworm was high and compared well with other studies carried out in the country (Pamba *et al.* 1980: Chunge *et al.* 1985: Magnussen *et al.* 1997: Geissler *et al.* 1998: Brooker *et al.* 2000: Olsen *et al.* 2000).

While the prevalence of *Ascaris* and *Trichuris* was evenly spread in the study area, hookworm was higher further inland and lower closer to the lake shore. This corroborates results from Thiong'o *et al.* in the same area and Olsen *et al.* in Kisumu District. Inversely the prevalence of *S. mansoni* was higher closer to the lake and lower inland which also agrees with results from schoolchildren in the same area (Thiong'o *et al.* 2001) and in schoolchildren in Busia District (Brooker *et al.* 2000). The latter situation is due to the transmission of schistosomiasis that is taking place on the lake shore; hence the people who live closer are likely to be more highly infected than people who live further inland. It has not been understood why hookworm prevalence and intensity should be lower at the lake shore and yet the soils and sanitary conditions are more or less the same in all the study area and even worse closer to the beaches due to the higher population density. A possible factor could probably be that it is drier close to the lake due to microclimate patterns.

4.4.1 Association between geophagy and geohelminths infection

As seen in the previous chapters, geophagy is common among pregnant women in the study area, a fact that confirms earlier reports from the area and elsewhere in the country (Hunter *et al.* 1984; Willey 1998; Geissler *et al.* 1998; Prince *et al.* 1999). The most commonly eaten earths were soft stone and ant hills which were eaten raw. Since these soils are picked from the fields or purchased from the open markets, chances of contamination with helminth eggs are quite high, in particular for ant hills. Geophagy has been linked to helminth infections in many studies (Hunter 1973; Wong *et al.* 1988; Horner *et al.* 1991; Geissler *et al.* 1997). There was some association in the intensity of *Ascaris* with geophagy and more so with the ant hill soil. This corroborates findings of Geissler *et al.* (1997) in the same study area among schoolchildren and findings from a study in schoolchildren in KwaZulu Natal in South Africa (Saathoff *et al.* 2000). This is also biologically and epidemiologically true since it is only *Ascaris* eggs with the sticky proteinous coating on the surface that could stay longer in the soil and also be transmitted through ingestion. The lack of association between *T. trichiura* which has a similar transmission mode to *Ascaris* can be explained by the smooth surface on the egg shell that will not adhere to soil. Also, the high sensitivity to desiccation of these eggs as compared to those of *A. lumbricoides* will reduce the chances of contamination leading to low intensity.

4.4.2 Association between socioeconomic factors and geohelminth infection

There was no association between latrines and geohelminths infections in this area. This contradicts findings by Olsen *et al.* (2001) in the neighbouring district of Kisumu. This could however be attributed to the study design where the current study was selective whereas Olsen's work included all age groups. Whereas most geohelminths are household infections, the public places play an important role in hookworm infection (Feacham *et al.* 1983). There was an association between the education of the mother and hookworm infection. This confirms studies in Nigeria where education had an effect on geohelminths infections (Adekunle *et al.* 1986). The education of women could have improved their income, since most of them were working as civil servants. However, most of the women in the area earned less than 3000 Kenya shillings monthly and income did not have any effect on the infections of helminths in the area.

4.5 Conclusion

Intestinal helminths are prevalent in the study area. All factors that may enhance transmission have been demonstrated in this study and may lead to high transmission potential of the helminths. These factors are essential in the control programmes and should be considered especially in maternal and child welfare clinics.

Chapter 5

Effects of geophagy and intestinal helminth infections on haemoglobin and iron status in pregnancy

5.1 Introduction

Earth eating (geophagy) is common among people on all continents and particularly in Africa (Anell & Lagercrantz 1958: Johns & Duquette 1991: Abrahams & Parsons 1996). In many societies it is accepted as a normal practice (Gelfand 1945: Hunter 1973: Reid 1992). and it is often regarded as a common "craving" for non-food substances during pregnancy (Lacey 1990). Cross-sectional studies assessing geophagy among pregnant women and its possible impact have been carried out in Kenya (Geissler *et al.* 1998). The relationship between geophagy. iron status and anaemia is still debated (Lackey 1979: Parry-Jones & Parry-Jones 1992) and it is not clearly understood whether earth eating results in low iron status (Cavdar *et al.* 1980: Horner *et al.* 1991: Prasad *et al.* 1993) or is a result of infections associated with earth eating (Rainville 1998: Geissler *et al.* 1998: Olsen *et al.* 2001; Saathoff *et al.* 2002) or whether low iron status induces the craving for soil (Reimann & Koptugel 1980: Moore & Sears 1994).

Intestinal helminth infections affect millions of women of reproductive age in developing countries (Gillespie & Johnston 1998). Intestinal helminths may cause anaemia through reduced food intake. mal-absorption and endogenous nutrient loss. The main anaemia-causing helminths are the hookworms *Ancylostoma duodenale* and *Necator americanus*. *Trichuris trichiura* and the schistosomes. Hookworms cause chronic blood loss by attaching themselves to the mucosa of the small intestines and ingesting tissue and erythrocytes (Banwell & Schad 1978). In 1990. it was estimated that worldwide 44 million women were both pregnant and infected with hookworm (Bundy *et al.* 1995). Infection during pregnancy could compromise maternal and infant health and nutrition status (Villar *et al.* 1989. Weigel *et al.* 1996: Santiso 1997).

Iron deficiency is the most common cause of nutritional anaemia. It is estimated that 600 to 700 million people suffer from anaemia due to iron deficiency and in developing countries it may affect a half the population. mainly children and women of reproductive age (Herceberg & Galan 1992). Anaemia in pregnancy causes considerable incapacity in women due to tiredness. breathlessness and decreased ability to work (WHO 1979). It is also associated with adverse perinatal outcomes such as low birth weight and premature births (Brabin 1991). The aetiology of anaemia in pregnancy is multi-factorial. with prevalence and causes varying considerably in different areas of the world. Causes that have been reported in sub-Saharan Africa are malaria and hookworm infections (Fleming 1989). Studies on predictors of iron status and anaemia among pregnant women have been carried out on the Kenyan coast (Shulman *et al.* 1996: Geissler *et al.* 1998). in school children (Brooker *et al.* 1998 and Geissler *et al.* 1998b) and in a community in western Kenya (Olsen *et al.* 1998). All these studies associated hookworm with low iron status.

Iron deficiency is a slowly evolving condition which passes through progressive stages of pre-latent. latent and manifest iron deficiency that lead to anaemia (Cook 1982: Finch & Cook 1984). The first stage involves the loss of sequestered iron reserves without a decrease in iron supply to the developing red blood cell (storage iron depletion). The second stage (iron deficient erythropoesis) results from diminished erythroid iron to the bone marrow. but occurs in the absence of a significant effect on circulating haemoglobin levels. Continued blood loss and lack of iron replacement then lead to the development of the final stage. manifest iron deficiency anaemia. These three stages of iron deficiency anaemia can be measured in the laboratory by the measurement of serum ferritin. free erythrocyte protoporphyrin and haemoglobin respectively (Cook 1982: Finch & Cook 1984). However. in this study only haemoglobin and serum ferritin were estimated. We know that Hb can show the delayed stage of iron deficiency while reduction in serum ferritin will reveal the inadequacy existing between iron supply and iron requirements which is the most sensitive indicator for assessing iron status in a population (Hercberg & Galan 1992). There is, however, no single iron parameter that can monitor the entire spectrum of iron deficiency. In some instances we may report false iron stores in possibly iron-depleted subjects, for example in inflammatory processes and other pathological conditions. In such cases there is need to control for acute phase response [α_1- antichymotrypsin (ACT)] which regrettably is a weakness we are aware of in this chapter.

The present study aimed at determining the role of geophagy and helminth infections in haemoglobin and iron status among pregnant women in western Kenya.

5.2 Materials and methods

5.2.1 Study area and population

The study area and population are as described previously in chapters 2 and 3.

5.2.3 Haematological examination

Out of the 827 recruited women, venous blood samples were collected from 824. Haemoglobin (Hb) concentration was estimated using a battery operated β-haemoglobin photometer detection system (HemoCue^{TM} AB, Ängelholm, Sweden). Serum samples from 644 women were analysed for ferritin using an enzyme-linked immunosorbent assay (ELISA) Novapath^{TM} ferritin kit (Bio-Rad, Anheim, California, USA). Women with haemoglobin below 11.0 g/dl (Stoltzfus & Dreyfus 1998) were considered anaemic and those with serum ferritin levels below 12 μg/L were considered to have depleted iron stores.

5.2.4 Parasitological examination

Eight hundred and twenty one stool samples were collected and prepared according to the KATO-KATZ technique (WHO 1991) to assess the number of eggs per gram (epg) of stool for all intestinal helminths. Hookworm eggs were counted immediately and the preparation was left overnight at room temperature for the clearance of the slide. and enumeration of *Ascaris lumbricoides. Trichuris trichiura* and *Schistosoma mansoni* eggs was carried out the following day. In this part of the study. malaria as one of the major causes of anaemia in pregnancy was considered. Thick and thin blood slides from the study subjects were therefore prepared and stained by Giemsa stain for malaria parasite examination. Parasites were counted against 200 white blood cells and the counts calculated per micro-litre of blood from the WBC count. To determine a negative blood smear. 100 high power oil immersion fields were examined. Parasites were reported as number of trophozoites per micro-litre of blood.

5.2.5 Iron estimation in earth samples

Earth samples were collected by the geophagous women from where they usually collected them daily. assisted by field assistants. A random sample of 30 earth samples was selected from the three main types. soft stone. ant hill and earthen wall of houses. The samples were pulverized and stored in sealable plastic bags. Five g of the soil sample was weighed into an acid washed plastic container. Iron was extracted from the soil by adding 5 ml of 0.1N HCL to the sample and mixing thoroughly by use of a mechanical mixer for 1 hour. The mixture was filtered through Whitman filter paper into an acid washed plastic container. The concentration of iron in the earth samples was determined by aspiration of the filtrate to an atomic absorption spectrophotometer (Flame emission spectrophotometer AA-68. Shimadzu model) at 248.3 nm.

5.2.6 Data analysis

Data were analysed using SPSS software. χ^2 tests were used to test for differences in proportions. while Student's *t*-test and Mann-Whitney *U*-test were used to assess differences in intensities of infections. A significance level of 0.5 was used for all tests. To identify predictors and confounders. multiple linear regression was used with haemoglobin and serum ferritin as dependent variables. Due to the skewed distribution. serum ferritin values were log-transformed to obtain normally distributed data.

5.3 Results

Among the 827 women recruited. data were available on haemoglobin (Hb) concentration for 824 (99.6 %). serum ferritin (SF) concentration for 644 (78.0%) and intestinal helminth based on duplicate smears for 821 (99.3%). The mean Hb in the study population was 10.9 g/dl and the prevalence of anaemia (Hb <11.0 g/dl) was 48.1%. The geometric mean of SF was 19.0 µg/L and the prevalence of women with depleted iron stores (SF<12 µg/L) was 32.8%. The prevalence and geometric mean intensity of hookworm was 49.8% and 129 epg respectively: *S. mansoni*. 63% and 98 epg: *A. lumbricoides*. 16.3% and 815 epg: *T. trichiura*. 24.1% and 52 epg. The prevalence and intensity of malaria (*P. falciparum*) was 45.8% and 590 parasites per µL of blood. The prevalence of geophagy was 45.7% and the mean earth intake 44.5 g/day.

5.3.1 Geophagy

As shown in Table 5.1. geophagous women had a significantly lower Hb as compared to non-geophagous women (10.6 vs. 11.1 g/dl. *P*<0.001). The prevalence of anaemia was significantly higher among the geophagous women than the non-geophagous women (55.8 vs. 41.5%. *P*<0.001). The geometric mean SF was lower among the geophagous women (15.7 µg/L) as compared to the non-geophagous women (22.5 µg/L). and the proportion of women with depleted iron stores was significantly higher among geophagous women compared to non-geophagous women (48.8 vs. 22.5%. *P*<0.001).

5.3.2 Parity

The mean Hb was lower among primigravidae than among multigravidae (10.4 vs. 11.0 g/dl. *P*<0.001). The prevalence of anaemia was higher among the primigravidae than among multigravidae (69.4 vs. 42.1%. *P*<0.001) (Table 5.2). Conversely the prevalence of women with depleted iron stores was higher in the multigravidae than in the primigravidae and mean SF higher in the primigravidae than in multigravidae (33.0 vs. 16.5 µg/L. $P<0.001$).

Table 5.1 Haemoglobin and serum ferritin concentration and proportion with low values among geophagous and non-geophagous women.

	Geophagous (n=378)	Non-geophagous (n= 446)	*P*-value
Haemoglobin (g/dl)[1]	10.6 (10.5; 10.8)	11.1 (11.0; 11.2)	<0.001
<11.0 (%)[2]	55.8 (56.8: 60.8)	41.5 (36.9: 46.1)	<0.001
Serum ferritin (µg/L)[3]	15.7 (12.2; 18.2)	22.5 (20.0; 25.0)	<0.001
<12 (%)[2]	48.8 (42.9; 54.7)	22.5 (18.1; 26.9)	<0.001

[1] Mean (95% confidence interval): [2] (95% confidence interval): [3] Geometric mean (95% confidence interval). geophagous. n=288: non-geophagous. n=356.

Table 5.2 Haemoglobin and serum ferritin concentration and proportion with low values among primigravid and multigravid women.

	Primigravidae (n=180)	Multigravidae (n=644)	*P*-value
Haemoglobin (g/dl)[1]	10.4 (10.1; 10.6)	11.0 (10.9; 11.1)	<0.001
<11.0 (%)[2]	69.4 (62.5; 76.3)	42.1 (38.1: 46.1)	<0.001
Serum ferritin (µg/L)[3]	33.0 (30.2; 35.8)	16.5 (14.2; 18.8)	<0.001
<12 (%)[2]	17.3 (10.9: 23.7)	36.6 (32.3: 40.9)	<0.001

[1] Mean (95% confidence interval): [2] (95% confidence interval): [3] Geometric mean (95% confidence interval). primigravidae. n=139: multigravidae n=505

5.3.3 Infection

Table 5.3. shows the mean Hb and serum ferritin concentration and the proportions below normal values among women infected with intestinal helminths and malaria. From this table. the mean Hb concentration in the hookworm negative women was significantly lower than in the hookworm positive ones (11.0 vs. 10.7 g/dl. $P<0.001$). Conversely the proportion of women with anaemia was significantly higher in hookworm negative than in those who were hookworm positive (52.8% vs. 42.4%. $P<0.001$). There was no significant difference noted in the concentration of serum ferritin between hookworm positive and hookworm negative women. but there was a significant difference in proportion of depleted iron stores

between the hookworm positive women and hookworm negative ones (28.9 vs. 36.4%. *P*=0.04). There was a significant difference in the Hb between women who were *T. trichiura* positive and those who were negative (11.1 vs. 10.8 g/dl. *P*<0.001). but no difference was observed in the prevalence of anaemia in the same groups. Likewise there was no difference in the prevalence of depleted iron stores and SF between the same groups. There were no differences in the Hb and proportions with anaemia between women who were infected with *A. lumbricoides* and *S. mansoni* and those who were not infected. Similarly there were no differences in SF and proportions of depleted iron stores between those infected with the two parasites and the negative ones.

The women who were infected with malaria had lower Hb and a higher SF concentration than those who did not have malaria infection. The malaria positive women also had a higher proportion of anaemia than those who were malaria negative. The proportion of depleted iron stores was lower among malaria positive women compared to malaria negative women.

Table 5.3a. is a modification of table 5.3 with intensities of positive cases stratified in light. moderate and heavy infection. From the same table it is again seen that as in table 5.3. Hb concentration in heavy hookworm infection was still higher than in the negative group although not significantly so. In infections with malaria. the mean Hb was significantly higher in negative than in the positive (11.1 vs. 10.6 g/dl. *P*<0.001). On the contrary the proportion of anaemia cases was significantly lower in the negative than in the positive (40.8 vs.58.1 %. *P*<0.001). On serum ferritin the mean SF was significantly lower in the negative than in the positive (14.6 vs. 25.4 μg/L. *P*<0.001). while the proportion of those with depleted serum ferritin (<12 μg/L) was higher in the negative than in the positive (45.0 vs. 19.2%. *P*<0.001).

Table 5.3 Haemoglobin and serum ferritin concentration and proportion with low values among women infected with intestinal helminths and malaria.

Infection	N	Negative	N	Positive	*P*-value
Hookworm					
Hb (g/dl)[1]	411	10.7 (10.5: 10.8)	408	11.0 (10.9: 11.2)	<0.001
<11.0 (%)[2]		53.8 (48.9: 58.7)		42.4 (37.5: 47.3)	<0.001
SF (μg/L)[3]	313	18.2 (15.6: 20.8)	325	20.1 (17.7: 22.5)	0.17
<12 (%)[2]		36.4 (31.0: 41.8)		28.9 (23.9: 33.9)	0.04
S. mansoni					
Hb (g/dl)[1]	303	10.8 (10.6: 10.9)	516	10.9 (10.8: 11.0)	0.15
<11.0 (%)[2]		48.2 (42.5: 53.9)		48.1 (43.7: 52.4)	0.97
SF (μg/L)[3]	226	19.5 (17.1: 21.9)	412	19.0 (16.4: 21.6)	0.74
<12 (%)[2]		33.2 (26.9: 39.5)		32.3 (27.7: 36.9)	0.82
A. lumbricoides					
Hb (g/dl)[1]	685	10.8 (10.7: 10.9)	134	11.0 (10.8: 11.2)	0.14
<11.0 (%)[2]		49.1 (45.3: 52.9)		43.3 (34.8: 51.8)	0.22
SF (μg/L)[3]	537	19.2 (16.7: 21.7)	101	19.3 (16.8: 21.8)	0.95
<12 (%)[2]		33.1 (29.1: 37.1)		29.7 (20.7: 38.7)	0.50
T. trichiura					
Hb (g/dl)[1]	621	10.8 (10.6: 10.9)	198	11.1 (10.9: 11.3)	<0.001
<11.0 (%)[2]		49.9 (45.9: 53.9)		42.4 (35.4: 49.4)	0.06
SF (μg/L)[3]	484	18.7 (16.2: 21.2)	154	20.7 (18.2: 23.2)	0.24
<12 (%)[2]		34.1 (29.8: 38.4)		27.9 (20.7: 35.1)	0.15
Malaria					
Hb (g/dl)[1]	434	11.1 (10.9: 11.2)	365	10.6 (10.4: 10.7)	<0.001
<11.0 (%)[2]		40.8 (36.1: 45.5)		58.1 (53.0: 63.2)	<0.001
SF (μg/L)[3]	333	14.6 (12.4: 16.8)	297	25.4 (22.8: 28.0)	<0.001
<12 (%)[2]		45.0 (39.6: 50.4)		19.2 (14.0: 24.4)	<0.001

[1] Mean (95% confidence interval): [2] (95% confidence interval): [3] Geometric mean (95% confidence interval)

Table 5.3a. Haemoglobin and serum ferritin concentration and proportion with low values in different intensities among infected with Intestinal helminths and malaria

Infection		Negative	Positive			*P*-value
			Light	Moderate	Heavy	
Hookworm(n)		411	293	74	41	
	Hb (g/dl)^1	10.7 (10.5, 10.8)	11.0 (10.8, 11.1)	11.3 (11.0, 11.5)	10.9 (10.4, 11.4)	0.004
	<11.0 (%)^2	53.8 (48.9, 58.7)	43.7 (37.9, 49.4)	37.8 (26.5, 49.1)	41.0 (25.7, 57.2)	0.01
Hookworm (n)		313	235	57	33	
	SF (µg/l)^1	18.2 (16.4, 20.3)	20.0 (17.7; 22.3)	22.2 (17.5, 28.3)	18.3 (14.0, 24.0)	0.40
	<12 (%)^2	36.4 (31.0, 41.8)	31.9 (25.3, 37.5)	7.5 (7.6, 28.6)	27.3 (11.2, 43.3)	0.04
A lumbricoides(n)		685	107	12	15	
	Hb (g/dl)^1	10.8 (10.7; 10.9)	11.0 (10.7, 11.3)	10.8 (9.9; 11.8)	11.3 (10.7, 11.8)	0.40
	<11.0 (%)^2	49.1 (45.3, 52.9)	43.0 (33.4; 52.6)	58.3 (25.6; 91.0)	33.3 (9.0; 57.6)	0.36
A lumbricoides(n)		537	81	7	13	
	SF (µg/l)^3	19.2 (17.7; 20.7)	19.3 (15.6, 24.0)	22.2 (11.1, 42.2)	17.7 (10.7, 29.1)	0.96
	<12 (%)^2	33.1 (29.1, 37.1)	28.4 (18.4, 38.4)	28.6 (-16.5, 73.7)	38.5 (11.5, 65.0)	0.09
T trichiura(n)		621	166	20	12	
	Hb (g/dl)^1	10.8 (10.6, 10.9)	11.1 (10.9, 11.3)	11.7 (11.0, 12.4)	10.9 (9.9, 12.0)	0.001
	<11.0 (%)^2	49.9 (45.9, 53.9)	43.4 (6.3, 51.8)	30 (9.5, 50.5)	50 (15.8, 83)	0.17
T trichiura(n)		484	130	15	9	
	SF (µg/l)^3	18.7 (17.2, 20.3)	21.3 (18.0, 25.2)	21.2 (16.5, 27.3)	13 (6.2, 27.8)	0.29
	<12 (%)^2	34.1 (29.8, 38.4)	29.2 (22.0; 38.3)	6.7 (-6.2, 19.6)	44.4 (11.4, 77.4)	0.097
S mansoni(n)		303	251	172	93	
	Hb (g/dl)^1	10.8 (10.6; 10.9)	11.0 (10.8, 11.1)	10.9 (10.7, 11.1)	10.9 (10.6, 11.1)	0.45
	<11.0 (%)^2	48.2 (42.5, 53.9)	48.2 (41.9, 54.5)	47.7 (40.1, 55.3)	48.4 (38.0, 58.8)	0.99
S. mansoni(n)		226	196	143	73	
	SF (µg/l)^1	19.5 (17.3, 21.9)	18.0 (15.9, 20.7)	20.0 (17.2, 23.3)	19.2 (15.4, 24.8)	0.78
	<12 (%)^3	33.2 (26.9, 39.5)	34.2 (27.4, 43.0)	30.1 (22.5, 37.7)	31.5 (20.7, 42.3)	0.87
Malaria(n)		439	212	106	42	
	Hb (g/dl)^1	11.1 (10.9; 11.2)	10.8 (10.7, 11.0)	10.3 (10.1, 10.9)	9.9 (9.6, 10.3)	<0.001
	<11.0 (%)^2	40.8 (36.1; 45.4)	49.5 (42.7, 56.3)	66.0 (57.0, 75.5)	83.3 (71.5, 95.0)	<0.001
Malaria(n)		338	170	84	38	
	SF (µg/l)^1	14.6 (13.4, 15.8)	21.5 (18.7, 24.6)	29.5 (23.7, 36.6)	40.5 (29.6, 55.3)	<0.001
	<12 (%)^2	45.0 (39.7, 50.5)	23.5 (17.3, 30.3)	14.3 (6.7, 22.7)	7.9 (-0.8, 16.6)	<0.001

[1] Mean (95% confidence interval). [2] (95% confidence interval); [3] Geometric mean (95% confidence interval)

When stratified by parity (Table 5.4). Hb was still lower among hookworm negative compared to hookworm positive women. but only in multigravidae. The proportion with anaemia was more pronounced in the hookworm negative than in the hookworm positive women among the multigravidae. There was no difference in the proportion with anaemia and concentration of Hb among the primigravidae. There was a significant difference in the proportion of women with depleted iron stores between hookworm positive and negative among the multigravidae (32.4 vs. 41.8%, *P*<0.03). There were no differences either in the proportions of women with depleted iron stores among the primigravidae or in the concentrations of SF between hookworm positive and negative women. Haemoglobin was lower but not significantly different among women who had no Trichuris infection in both primigravidae and multigravidae (Table 5.5).

Table 5.4 Haemoglobin and serum ferritin concentration and proportion with low values. by parity among women infected with hookworm.

	Hookworm negative	Hookworm positive	*P*-value
Primigravidae	97	83	
Hb (g/dl)[1]	10.4 (10.1; 10.7)	10.3 (10.0: 10.7)	0.83
<11.0 (%)[2]	70.1 (60.8: 79.4)	68.7 (58.6: 78.8)	0.84
SF (µg/L)[3]	30.8 (27.8: 33.8)	35.8 (33.3; 37.8)	0.38
<12 (%)[2]	19.7 (10.6; 28.8)	14.3 (5.6: 23.0)	0.40
Multigravidae	314	325	
Hb (g/dl)[1]	10.8 (10.6; 10.9)	11.2 (11.0; 11.3)	<0.001
<11.0 (%)[2]	48.7 (43.1; 54.3)	35.7 (30.4: 41.0)	0.001
SF (µg/L)[3]	15.4 (13.1; 17.7)	17.5 (15.2: 19.8)	0.08
<12 (%)[2]	41.8 (35.4: 48.2)	32.4 (26.7: 38.1)	0.03

[1] Mean (95% confidence interval): [2](95% confidence interval): [3]Geometric mean (95% confidence interval). primigravidae (hookworm negative. n=76: hookworm positive. n=63): multigravidae (hookworm negative. n=237: hookworm positive. n=262)

Table 5.5 Haemoglobin and serum ferritin concentration and proportion with low values, by parity among women infected with *T trichiura*

	T. trichiura negative	*T. trichiura* positive	*P*-value
Primigravidae	133	47	
Hb (g/dl)[1]	10.2 (10.0; 10.5)	10.8 (10.3; 11.3)	0.02
<11.0 (%)[2]	71.4 (63.6; 79.2)	63.8 (49.8; 77.8)	0.33
SF (µg/L)[3]	32.4 (29.7; 35.1)	34.7 (31.8; 37.6)	0.73
<12 (%)[2]	17.6 (10.2; 23.1)	16.2 (5.7; 26.7)	0.84
Multigravidae	488	151	
Hb(g/dl)[1]	10.9 (10.8; 11.0)	11.3 (11.1; 11.5)	0.002
<11.0(%)[2]	44.1 (35.6; 49.6)	35.8 (28.0; 43.6)	0.07
SF (µg/L)[3]	16.1 (13.8; 18.4)	17.5 (14.2; 19.8)	0.34
<12 (%)[2]	38.5 (33.5; 43.5)	31.6 (24.0; 40.2)	0.18

[1] Mean (95% confidence interval); [2] (95% confidence interval); [3] Geometric mean (95% confidence interval). primigravidae (*T trichiura* negative, n=102; *T trichiura*, positive n=37); multigravidae (*T. trichiura* negative, n=382; *T.trichiura* positive, n=117)

As seen in Table 5.6, there were significant differences in Hb and SF between malaria positive and malaria negative women among both primigravidae and multigravidae. There was also a significant difference in the proportion with anaemia between malaria positive and malaria negative among both primigravidae and multigravidae ($P<0.001$ and $P=0.03$) respectively; the proportion was much higher among malaria positive than malaria negative women. Significant differences were also noted in the proportion of women with depleted iron stores between malaria positive and malaria negative in both groups of gravidity. Among both geophagous and non-geophagous women, there was a significant difference in Hb between hookworm positive and hookworm negative cases. There was also a significant difference in the proportion of anaemia between the same groups. No difference was noted in SF and in the proportion with depleted iron stores between the groups (Table 5.7).

Table 5.6 Haemoglobin and serum ferritin concentration and proportion with low values, by parity among women infected with malaria.

	Malaria negative	Malaria positive	*P*-value
Primigravidae	56	117	
Hb (g/dl)[1]	11.0 (10.6; 11.5)	10.0 (9.8; 10.2)	<0.001
<11.0 (%)[2]	50.0 (36.6; 63.4)	80.3 (73.0; 87.6)	<0.001
SF (µg/L)[3]	17.1 (13.8; 20.4)	42.5 (39.9; 45.1)	<0.001
<12 (%)[2]	41.5 (26.2; 56.8)	7.7 (2.1; 13.3)	<0.001
Multigravidae	378	248	
Hb (g/dl)[1]	11.1 (10.9; 11.2)	10.8 (10.7; 11.0)	0.042
<11.0 (%)[2]	39.4 (34.4; 44.4)	47.6 (41.3; 53.9)	0.026
SF (µg/L)[3]	14.3 (12.1; 16.5)	20.0 (16.6; 22.4)	<0.001
<12 (%)[2]	45.5 (39.7; 51.3)	24.6 (18.6; 30.6)	<0.001

[1] Mean (95% confidence interval); [2] (95% confidence interval); [3] Geometric mean (95% confidence interval). primigravidae (malaria negative, n=41; malaria positive, n=91); multigravidae (malaria negative, n=286; malaria positive, n=203)

Table 5.7 Haemoglobin and serum ferritin concentration and proportion with low values, by geophagy among women infected with hookworm.

	Hookworm Negative	Hookworm positive	*P*-value
Non-geophagous	218	226	
Hb (g/dl)[1]	10.9 (10.7; 11.1)	11.2 (11.0; 11.0)	0.034
<11.0 (%)[2]	48.6 (41.8; 55.4)	35.0 (38.7; 41.3)	0.003
SF (µg/L)[3]	20.9 (18.1; 24.2)	24.0 (21.1; 27.3)	0.16
<12 (%)[2]	28.4 (21.4; 35.4)	17.4 (11.8; 23.0)	0.014
Geophagous	193	182	
Hb (g/dl)[1]	10.4 (10.2; 10.6)	10.8 (10.6; 11.0)	0.017
<11.0 (%)[2]	59.6 (52.6; 66.6)	51.6 (44.2; 59.0)	0.12
SF (µg/L)[3]	15.5 (13.2; 18.0)	16.0 (13.9; 18.4)	0.75
<12 (%)[2]	45.8 (37.6; 54.0)	44.0 (35.6; 48.4)	0.75

[1] Mean (95% confidence interval); [2] (95% confidence interval); [3] Geometric mean (95% confidence interval). non-geophagous (hookworm negative, n=169; hookworm positive, n=184); geophagous (hookworm negative, n= 144, hookworm positive, n=141)

There was higher SF among the *S. mansoni* negative women than the positive ones in the non-geophagous group (*P*=0.03). There were no differences noted in the proportions with anaemia and depleted iron stores between the positive and negative women in all groups, neither was there any difference in the Hb between all the groups and SF between those in the geophagous group (Table 5.8). No difference was observed in any of the groups in the *A. lumbricoides* positive and negative women (Table 5.9).

Table 5.8 Haemoglobin and serum ferritin concentration and proportion with low values, by geophagy among women infected with *S. mansoni*.

	S. mansoni negative	*S. mansoni* positive	*P*-value
Non-geophagous	151	293	
Hb (g/dl)[1]	11.0 (10.8; 11.2)	11.1 (10.9; 11.2)	0.45
<11.0 (%)[2]	43.0 (35.0; 51.0)	41.0 (35.3; 46.7)	0.67
SF (µg/L)[3]	26.2 (22.4; 30.8)	20.8 (18.8; 23.5)	0.03
<12 (%)[2]	16.5 (9.6; 23.4)	25.6 (20.0; 31.2)	0.06
Geophagous	152	223	
Hb (g/dl)[1]	10.5 (10.3; 10.8)	10.7 (10.5; 10.8)	0.40
<11.0 (%)[2]	53.3 (45.3; 61.3)	57.4 (50.8; 64.0)	0.43
SF (µg/L)[3]	14.3 (12.3; 16.6)	16.7 (14.5; 19.3)	0.15
<12 (%)[2]	50.5 (41.0; 60.0)	41.1 (33.7; 48.5)	0.13

[1] Mean (95% confidence interval); [2] (95% confidence interval); [3] Geometric mean (95% confidence interval), non-geophagous (*S. mansoni* negative, n= 115; *S. mansoni* positive, n=238); geophagous (*S. mansoni* negative, n=111; *S. mansoni* positive, n=174)

Significant differences were noted in the Hb between *Trichuris* positive and negative women in both groups of geophagy. There were no differences in the proportions with anaemia and depleted iron stores between the two infection status (positive and negative) of *Trichuris*, neither were there differences in SF in the same categories among both geophagous and non-geophagous women (Table 5.10).

Table 5.9 Haemoglobin and serum ferritin concentration and proportion with low values, by geophagy among women infected with *A. lumbricoides*.

	A. lumbricoides negative	*A. lumbricoides* positive	*P*-value
Non-geophagous	370	74	
Hb (g/dl)[1]	11.0 (10.9; 11.1)	11.3 (10.9; 11.5)	0.24
<11.0 (%)[2]	42.4 (35.8; 49.0)	37.8 (26.5; 49.1)	0.46
SF (μg/L)[3]	23.1 (20.8; 25.6)	19.4 (15.0; 24.9)	0.20
<12 (%)[2]	22.7 (17.9; 27.5)	22.6 (11.1; 34.1)	0.99
Geophagous	315	60	
Hb (g/dl)[1]	10.6(10.4; 10.8)	10.8 (10.4; 11.1)	0.38
<11.0 (%)[2]	56.8 (51.2; 62.4)	60.0 (53.6; 56.4)	0.33
SF (μg/L)[3]	15.1 (13.4; 16.9)	19.1 (14.5; 25.3)	0.09
<12 (%)[2]	46.4 (39.9; 52.9)	37.5 (23.5; 51.5)	0.26

[1] Mean (95% confidence interval): [2](95% confidence interval): [3]Geometric mean (95% confidence interval). non-geophagous (*A. lumbricoides* negative. n=300: *A. lumbricoides* positive. n=53): geophagous (*A. lumbricoides* negative. n= 237: *A. lumbricoides* positive. n=48)

Table 5.10 Haemoglobin and serum ferritin concentration and proportion with low values, by geophagy among women infected with *T. trichiura*.

	T. trichiura negative	*T. trichiura* positive	*P*-value
Non-geophagous	332	112	
Hb (g/dl)[1]	11.0 (10.9; 11.1)	11.3 (11.0; 11.6)	0.02
<11.0 (%)[2]	42.8 (37.4; 48.2)	38.4 (29.4; 47.4)	0.42
SF (μg/L)[3]	22.3 (19.8; 25.0)	23.2 (19.5; 27.4)	0.73
<12 (%)[2]	24.7 (19.4; 30.0)	16.7 (8.6; 24.5)	0.12
Geophagous	238	86	
Hb (g/dl)[1]	10.5 (10.3; 10.7)	10.9 (10.7; 11.2)	0.008
<11.0 (%)[2]	58.1 (52.3; 63.9)	47.7 (36.9; 58.2)	0.08
SF (μg/L)[3]	15.2 (13.6; 17.0)	17.7 (13.5; 23.0)	0.24
<12 (%)[2]	45.2 (38.5; 51.9)	43.8 (31.4; 50.5)	0.83

[1] Mean (95% confidence interval): [2](95% confidence interval): [3]Geometric mean (95% confidence interval). non-geophagous (*T. trichiura* negative. n=263: *T. trichiura* positive. n=90): geophagous (*T. trichiura* negative. n= 221: *T. trichiura* positive. n=64)

As shown in Table 5.11. the Hb was lower in malaria positive women among both geophagous and non-geophagous groups. The proportion with anaemia was therefore higher among the malaria positive women within the stratified geophagy groups. The mean SF concentration was higher in the malaria positive cases than the malaria negative ones in both groups of geophagy. The proportion with depleted iron stores was higher in malaria negative cases as compared to malaria positive cases within the two geophagy groups.

In Table 5.12. most of the differences in Hb and SF and proportions with anaemia and depleted iron stores could be due to parity rather than geophagy. As observed in the same table. there were significant differences noted in all comparisons while in the group stratified by parity. only the ones in multigravidae showed significant differences. while the geophagous and the non-geophagous in the primigravidae did not show any difference at all.

Table 5.11 Haemoglobin and serum ferritin concentration and proportion with low values. by geophagy among women infected with malaria.

	Malaria negative	Malaria positive	*P*-value
Non-geophagous	221	212	
Hb (g/dl)[1]	11.4 (11.2; 11.5)	10.8 (10.6; 10.9)	<0.001
<11.0 (%)[2]	31.7 (15.4; 38.0)	53.3 (46.5: 60.1)	<0.001
SF (μg/L)[3]	17.6 (15.6; 19.7)	28.9 (25.0: 33.5)	<0.001
<12 (%)[2]	32.8 (25.8: 39.8)	11.7 (6.7; 16.7)	<0.001
Geophagous	213	153	
Hb (g/dl)[1]	10.8 (10.6: 11.0)	10.3 (10.1: 10.5)	0.002
<11.0 (%)[2]	50.2 (43.4: 57.0)	64.7 (57.0: 72.4)	0.006
SF (μg/L)[3]	11.9 (10.5: 13.4)	20.8 (17.6: 24.6)	<0.001
<12 (%)[2]	58.8 (50.8: 66.8)	30.1 (21.9: 38.3)	<0.001

[1] Mean (95% confidence interval): [2](95% confidence interval):
[3] Geometric mean (95% confidence interval). non-geophagous (malaria negative. n=174. malaria positive. n=171): geophagous (malaria negative. n= 153: malaria positive. n=123)

Table 5.12 Haemoglobin and serum ferritin concentration and proportion with low values. by geophagy among pregnant women.

	Primigravidae	Multigravidae	*P*-value
Non-geophagous	98	348	
Hb (g/dl)[1]	10.4 (10.2; 10.6)	11.3 (11.1: 11.4)	<0.001
<11.0 (%)[2]	71.4 (62.4; 80.4)	33.0 (28.0; 38.0)	<0.001
SF (μg/L)[3]	38.0 (30.5: 47.5)	19.5 (17.7: 21.6)	<0.001
<12 (%)[2]	12.0 (4.5; 19.5)	25.3 (20.1; 30.5)	<0.014
Geophagous	82	296	
Hb (g/dl)[1]	10.3 (10.0: 10.6)	10.7 (10.5; 10.8)	0.034
<11.0 (%)[2]	67.1 (56.6: 87.5)	52.7 (46.9; 58.5)	0.020
SF (μg/L)[3]	28.0 (21.4: 36.5)	13.3 (12.0: 14.7)	<0.001
<12 (%)[2]	23.4 (12.8: 34.0)	50.9 (44.2; 58.6)	<0.001

[1] Mean (95% confidence interval): [2](95% confidence interval): [3]Geometric mean (95% confidence interval). non-geophagous (primigravidae. n=75. multigravidae. n=281): geophagous (primigravidae. n= 64: multigravidae. n=224)

Multiple regression analysis was conducted. using haemoglobin as the dependent variable and geophagy. hookworm intensity and *T. trichiura* intensity as independent variables. Malaria intensity was controlled for separately by primigravidae and multigravidae. In the final multiple regression model presented in Table 5.13. geophagy (and malaria) were negative. and hookworm and *Trichuris* were positive predictors of Hb. Since the relationship between hookworm and *Trichuris* egg output and Hb was not linear. the relationship was assessed using dummy variables. In contrast. the relationship between malaria parasitaemia and Hb was linear and malaria was therefore assessed as a continuous variable. after $\log_{10}(x+1)$ transformation. However the effect of malaria parasitaemia was assessed separately for primi- and multigravid women. due to the finding of a significant interaction ($P<0.001$). The regression coefficient of ant hill soil of -0.58 reflects a 0.58 (0.33: 0.82) g/dl lower mean Hb in women eating ant hill earth compared to non-geophagous women. Similarly moderate hookworm was associated with a 0.47 g/dl Hb compared to hookworm negative women. Since malaria parasitaemia was log-transformed. the regression coefficients reflect a 0.35 and 0.13 g/dl decrease in Hb per log increase in malaria parasitaemia in primigravidae and multigravidae respectively.

5.3.4 Estimation of iron

Ten samples of each of the 3 types of earth were analysed for iron. The mean intake for soft stone was 45.1 g per day, anthill 43.7 g, and earth from wall of house 38 g per day. The mean concentration of iron in the different types of earth and their 95% confidence intervals are given in Table 5.14. Soft stone had low iron content (27 mg/kg) while wall of house had the highest iron content (189 mg/kg). This could be attributed to contamination of the earth during its preparation from topsoil near human dwellings. The mean amount of iron in the daily intake of soft stone in geophagous women was 1.2 mg which was 4% of the recommended dietary intake (RDI) in pregnant women (Herbert 1987), ant hill 5.7 mg (19% of RDI) and wall of house 7.2 mg (24% RDI).

Table 5.13 Final multiple regression model with haemoglobin concentration as the dependent variable.[1, 2]

	B	95% CI	*P*- value
Geophagy[3]			
Soft stone	-0.50	-0.73; -0.28	<0.001
Anthill	-0.58	-0.82; -0.33	<0.001
Other earth	-0.38	-1.05; 0.30	0.27
Hookworm intensity[4]			
Light	0.20	0.001; 0.40	0.05
Moderate	0.47	0.14: 0.80	0.005
Heavy	0.33	-0.106: 0.76	0.14
T. trichiura intensity[5]			
Light	0.36	0.13: 0.59	0.003
Moderate	0.86	0.27; 1.47	0.005
Heavy	-0.01	-0.77; 0.75	0.98

[1] Adjusted R^2=0.13. n=793: [2] Malaria separately by primigravidae and multigravidae were controlled for in the model: [3] Using earth eating as reference category: [4] Using hookworm uninfected as reference category: [5] Using *Trichuris* uninfected as reference category:

Table 5.14. Iron content of different types of earth and the estimated daily intake.

	Mean earth intake per day[1]	Mean iron concentration[2]	Estimated daily iron intake[3]	% RDI[4]
Soft soil	45.1 (40.9: 9.3)	27 (21; 33)	1.2	4
Ant hill	43.7 (39.3; 48.1)	131 (68: 196)	5.7	19
Wall of house	38 (22.3; 53.7)	189 (65; 312)	7.2	24

[1] Mean g/day (95% confidence interval): [2] Mg/kg (95% confidence interval): [3] Mg/day: [4] Recommended dietary intake for women during pregnancy: iron 30mg/d (Herbert 1987)

5.4 Discussion

5.4.1 Geophagy

As seen in Chapter 3. geophagy is prevalent among pregnant women in the study area. This confirms earlier findings in the region (Kilbride 1993: Geissler *et al.* 1998). The common types of earth eaten here are ant hill and soft stone. This corroborates reports in schoolchildren by Geissler *et al.* (1997) in the same area. In our study we found geophagy to be negatively associated with iron status among pregnant women in the area. Our findings agree with those of a study among pregnant women from the Kenyan coast where geophagy was significantly associated with low haemoglobin levels and low serum ferritin concentrations (Geissler *et al.* 1998). Our results are also in agreement with those of a study among schoolchildren in western Kenya where iron depletion was associated with geophagy (Geissler *et al.* 1998b) and among Zambian schoolchildren (Mbiko *et al.* in press) (for similar findings see Berkel *et al.* 1970: Crosby 1976: Arcasoy *et al.* 1978: Arbiter & Black 1991). Minnich (1968) associated geophagy with reduced absorption of iron in the gut. However. apart from the adverse effects of mal-absorption of iron as quoted above. there is evidence that the earth eaten contains iron that may alleviate the deficiency if absorbed. Previous investigations in the study area have shown that both young people and women tend to prefer ant hill earth which has been shown to have high concentrations of iron. Earth from the walls also had a very high content of iron, and this earth type has been confirmed to be a favourite of women at the coast (Geissler *et al.* 1998). Our findings cannot explain why the women who ate earth had depleted iron stores. Possibly they started eating earth in order to meet the iron requirements during pregnancy or they were already deficient in iron and hence required replenishment. These arguments are supported by the fact that earth eating habits changed in some women after delivery, which might be attributable to their reduced requirement of iron after delivery and the improved nutrition. This view of anaemia inducing soil eating is supported by various studies which have confirmed that vulnerable groups of individuals who are most likely to be anaemic are the ones most prone to geophagy (Crosby 1976; Moore & Sears 1994; Geissler *et al.* 1997, 1998; Saathoff *et al.* 2002).

5.4.2 Helminth infections

In pregnant women severe iron deficiency anaemia increases morbidity and mortality and implies an increased risk to the foetus (Crompton & Stephenson 1990). Several factors contribute to iron deficiency, such as insufficient dietary intake, reduced absorption and severe infections. Heme iron is better absorbed than the non-heme iron. Iron is absorbed mainly in the duodenum and proximal jejunum (Schafer & Bunn 1929), and its absorption can be impaired by medicinal antacids such as tricilicate or by clay. Helminth infections may influence iron status by reducing nutrient intake (Latham *et al.* 1990; Stephenson *et al.* 1993) and may also influence iron status by interfering directly or indirectly with metabolism and transport (Stephenson 1987). Hookworm infection has been closely implicated in the aetiology of anaemia in the tropics (Woodruff, 1982; Foy & Nelson, 1993). While it is evident that blood is lost due to hookworm infection, the development of this anaemia is influenced by other factors of which iron status of the host is the most important (Stephenson & Crompton 1990). The widespread prevalence of iron deficiency is due in part to the inefficient absorption of dietary iron due to damage caused to the intestinal mucosa (Prasad *et al.* 1983). Hookworms are known to attach themselves to the mucosa of the ileum where they suck blood but can move to the other parts of the intestines depending on their density.

5.4.3 Hookworm

Although hookworm has been documented as a major cause of iron deficiency, studies have shown that it is not possible to determine the hookworm intensity levels which cause anaemia (Shield *et al.* 1981: Latham *et al.* 1983: Hercberg *et al.* 1986: Srinivasan *et al.* 1987: Foo 1990: Pritchard *et al.* 1991; Lwambo *et al.* 1992: Bakta. *et al.* 1993: Allemann *et al.* 1994: Stoltzfus *et al.* 1997). There have been attempts to estimate the number of adult hookworms in relation to anaemia. It has been estimated that 1000 eggs per gram would be indicative of 32 adult worms of *Necator americanus*, the most common hookworm in the tropics (Pawlowski *et al.* 1991). In our study the mean intensity of hookworm was 129 epg which translates to only 4 adult worms. This shows that most of our study participants had a light intensity infection according to the WHO standards.

It was surprising to note that hookworm and *T. trichiura* were associated with high rather than low Hb. These findings are difficult to explain, but the possible explanation could be confounding. This may have occurred even if lack of hookworm infection was associated with some factors that reduce Hb, of which some include dietary intake, absorption, body iron distribution and physiological losses. The dietary intake requires food that is rich in iron and that can be absorbed. Heme iron found in animal food is better absorbed than non-heme found in vegetable food (Crompton & Whitehead 1993). Hercberg and Galan (1992) have explained that anaemia might have causes other than iron deficiency (possibly nutrition aetiologies) which might lead to false positives, thereby overestimating the true prevalence of iron deficiency. Other factors that would be linked to decreased Hb and serum ferritin are inflammatory syndromes even to a mild degree (Fleming 1982: Hercberg *et al.* 1987). In absorption, there are luminal factors which enhance absorption of iron from the duodenum and jejunum and include animal protein (cystein) and ascorbic acid (Crompton & Whitehead 1993). Further to this, the authors state that absorption increases as the body iron decreases and up to 42% of iron is reabsorbed during hookworm infection. During the feeding stage, hookworm causes blood leakage into the lumen which is reabsorbed. When the absorption exceeds the loss due to the few worms, then the result would be an increase in Hb. It should be noted that in Kenya women are given iron supplements during pregnancy. Although a study in Kenya has indicated that even at low intensities hookworm was associated with iron deficiency anaemia (Olsen *et al.* 1998), the present study which included only women who had an Hb above 7.0 g/dl, contradicted this theory. There have been reports that the threshold number for any demonstrable morbidity with hookworm is 40 worms, but because of the nonlinear relationship between burden and risk of morbidity, the threshold can vary from 50 to 200 worms without changing the population at risk of disease (WHO 1994).

5.4.4 *Ascaris lumbricoides*

We found no relationship between intensities of *A.lumbricoides* and Hb or iron status. This agrees with other studies (Blumenthal & Shultz, 1976; Islek *et al.* 1993; Geissler *et al.* 1998b; Olsen *et al.* 1998). In Zanzibari school children, *Ascaris* infection was associated with lower Hb but not with lower serum ferritin. Iron is absorbed through the intestinal mucosa in the duodenum and jejunum, and possibly iron absorption is impaired by the presence of *Ascaris* in this part of the intestine (Islek *et al.* 1993).

5.4.5 *Trichuris trichiura*

We did not find any relationship between *T. trichiura* and iron status. *T. trichiura* is known to burrow its anterior portion of the body in the luminal epithelium of the colon and heavy intensities have been associated to erythrocyte loss from the gut (Bundy & Cooper 1989). Some studies have confirmed an association between low Hb in cases of heavy *T. trichiura* infections (Ramdath *et al.* 1995; Cooper *et al.* 1997) while others have found no relationship despite heavy intensities (Stoltzfus *et al.* 1997b). However, the intensities of *Trichuris* in our study population were very low and had no relationship with iron status which agreed with the findings of Olsen *et al.* (1998) in the neighbouring district.

5.4.6 *Schistosoma mansoni*

There was no relationship between Hb and intensity of *S. mansoni* infection, which corroborated earlier findings (Latham *et al.* 1982; Geissler *et al.*, 1998b; Olsen *et al.* 1998) and contradicts other studies where heavy intensities of *S. mansoni* had negative association with Hb (Sturrock *et al.* 1996). We did not find any association between iron status and *S. mansoni* intensities. This result contradicted findings by Olsen *et al.* (1998), who found reduced SF in adults in their study.

The results of our study could probably be due to the fact that the participants were selected among healthy individuals with a haemoglobin concentration >7.0 g/dl. Women are usually given iron tablets during pregnancy with a view to alleviating their excess needs during this time. We therefore missed the severely anaemic ones. However, since even those with mild anaemia resorted to geophagy, and it has been established already that over 52% of pregnant women in Africa are anaemic (WHO 1994), further research including all categories of anaemia in the vulnerable groups should be carried out to determine whether geophagy is the cause of anaemia or whether anaemia leads to geophagy.

Chapter 6

Reinfection with intestinal helminths among pregnant and lactating women

6.1 Introduction

The role of geophagy or earth eating as a source of geohelminth infection has been suspected for a long time (Anell & Lagercrantz 1958; Halstead 1968). but only more recently has the practice been shown to be associated with infection with *Ascaris lumbricoides* and. though less conclusively. *Trichuris trichiura* among institutionalized orphans (Wong *et al*. 1991) and among schoolchildren in western Kenya (Geissler *et al*. 1998). KwaZulu-Natal in South Africa (Saathoff *et al*. 2002) and Zambia (Nchito *et al*. 2004). Various other studies have found environmental. occupational and socioeconomic factors to be positively associated with intestinal helminth infections (Henry *et al*. 1988: Valerie *et al*. 1995: Olsen *et al*. 2000). Most studies have been cross-sectional. whereas only a few have utilized the reinfection design to evaluate the role of these factors as determinants of helminth infection after treatment (Albonico *et al*. 1995 Magnussen *et al*. 1997: Geissler *et al*. 1998: Olsen *et al*. 2000). mostly in children. The present chapter looks at the role of geophagy and other factors on helminth reinfection among pregnant and lactating women in western Kenya recruited at 14-24 weeks gestation and followed up to six months postpartum.

6.2 Materials and methods

6.2.1 Study area and population

The study area and population are as described previously in chapter 2 and 3.

6.2.2 Recruitment and follow-up

Expecting women were recruited at 14-24 weeks gestation after meeting the criteria of the study as outlined in Chapter 2. The women were interviewed about their earth eating habits and socioeconomic status. Baseline parasitological examinations were carried out at this time. All information collected at recruitment was repeated at subsequent follow-up visits. The recruited women were followed at 28-32 weeks gestation. delivery. three months postpartum and six months postpartum. During these visits. stool samples were collected and examined for intestinal worms.

6.2.3 Parasitological examinations

Stool samples were collected on two consecutive days and duplicate 50 mg Kato Katz cellophane thick smears were made (WHO 1991). The slides were examined within an hour for hookworm eggs and on the following day for other intestinal helminth eggs. Intensity was expressed as mean eggs per gram of faeces (epg) of the two samples. After the second examination. all the women were treated with a single dose of Mebendazole 500 mg (Vermox) by a clinician. This was the drug and dosage recommended by the local Medical Officer. They were then booked for the first follow-up visit.

6.2.4 Data analysis

Data were entered in duplicate and analyzed using SPSS™ version 10 software. Descriptive statistics were used to assess the prevalence of infections. Intestinal helminth egg counts were $\log_{10}$ transformed to obtain normal distribution before analysis for intensities. McNemar test was used for the prevalence and Chi-square tests were used to test for differences in proportions. while a paired *t*-test was used to assess differences in intensities of infection in categories with two groups and One-way Analysis of Variance (ANOVA) in those with three or more groups. A significance level of P=0.05 was used in all tests.

6.3 Results

6.3.1 Reinfection

During the first follow-up examination at midterm. 700 women were declared negative or were not infected by *Ascaris lumbricoides*. 670 were not infected with *Trichuris trichiura* and 479 were not infected with hookworm. They were therefore included in the analysis for reinfection. As shown in Table 6.1. at delivery 11.2% of the women had been reinfected with hookworm. 4.6% with *A. lumbricoides* and 3.8% with *T. trichiura*. During the second follow-up at three months postpartum. the reinfection rates were 14.8% for hookworm. 6.6% for *A. lumbricoides* and 5.2% for *T. trichiura*. however there was no significant difference between the reinfections at delivery and three months post partum. At six months postpartum. hookworm reinfection had increased to 16.0% but showed no significant difference between the rate at delivery (16.0 vs. 11.2%. *P*=0.09). The rate of reinfection with *A. lumbricoides* was significantly different from reinfection at delivery (9.4 vs. 4.6%. *P*=0.001). There was no significant difference between reinfection of *T. trichiura* at 6 months postpartum and at delivery (5.9 vs. 3.8%. *P*=0.12). There were no significant differences between intensities of *A. lumbricoides* at different follow-up times; however there was a borderline difference between intensities at delivery and six months post-partum (239 vs. 828 epg . *P*=0.06). Likewise there were no significant differences between intensities of *T. trichiura* or hookworm infection during the follow-up.

Table 6.1. Reinfection rates and intensities of intestinal helminths among women at delivery. three months and six months postpartum follow-up.[1]

	At delivery	3 months postpartum	*P*-value[4]	6 months postpartum	*P*-value[4]
Hookworm					
Prevalence[2]	11.2 (8.2:14.2)	14.8 (11.0; 18.0)	0.118	16 (12.0; 20.0)	0.087
Intensity[3]	147 (59: 365)	57 (26: 103)	0.034	74 (44: 125)	0.325
A. lumbricoides					
Prevalence[2]	4.6 (2.6; 6.6)	6.6 (4.6: 8.6)	0.080	9.4 (6.4: 12.4)	0.001
Intensity[3]	239 (57: 1014)	1429 (192: 10641)	0.081	828 (100: 6883)	0.063
T. trichiura					
Prevalence[2]	3.8 (2.0; 6.0)	5.2 (3.2. 8.2)	0.230	5.9 (4.0; 8.0)	0.117
Intensity[3]	61 (24; 160)	62 (40: 96)	0.920	53 (30: 96)	0.780

[1] Only women who were found uninfected or successfully treated at 32 weeks gestation

[2] Percentage (95% confidence interval). n at delivery. 3 and 6 months postpartum. are: hookworm 411. 411 and 405: A *lumbricoides* 590. 606 and 593: *T trichiura* 573. 575. and 561:

[3] Geometric mean (95% confidence interval) eggs per gram for infected women only:

[4] McNemar χ^2 test applied for the prevalence and the paired *t*-test for the intensities (all compared to values at delivery)

6.3.2 Hookworm

The proportion of women found free of hookworm infection at mid-term after treatment was smaller compared to other worms. Thus. only 479 women were included in the follow-up for hookworm reinfection. which was attributed to the fact that hookworm had the highest prevalence at baseline and many cases might not have been cured by the single dose of mebendazole. As seen in Table 6.2. there were no significant differences noted in the reinfection rates of hookworm between the two groups at all levels of follow-up: however there was a significant difference in the intensity of hookworm egg count between geophagous and non-geophagous women at delivery (153 vs. 77 epg. P=0.03). There were no significant differences in hookworm reinfection rate or intensity between the two geophagy groups at the three months and six months follow-up. The highest relative risk for reinfection with hookworm was at six months. (1.14 (95% confidence interval [95% CI] 0.87: 1.39) comparing geophagous and non-geophagous women. However these differences were not statistically significant. When geophagous women were stratified by type of earth (those eating anthill earth and those eating other earth) and compared to non-geophagous women. reinfection among the anthill group remained higher: however there was no statistical difference in the rates of reinfection of hookworm between geophagous and non-geophagous women at all levels of follow-up (Table 6.3). There was still a significant difference in intensity of hookworm between women eating different types of earth and non-geophagous at delivery (*P*=0.02). No significant difference was noted in the intensities of hookworm between the groups at three months and six months postpartum.

Table 6.2. Reinfection rates and intensities of hookworm among geophagous and non-geophagous women at delivery. three months and six months postpartum follow-up.[1]

	Geophagous (n=225)		Non-geophagous (n=254)		*P*-value
	N		N		
At delivery					
Prevalence[2]	187	9.6 (5.3; 13.9)	224	12.5 (8.1; 16.9)	0.35
Intensity[3]		153 (83: 282)		77 (55; 106)	0.03
3 months pp[4]					
Prevalence[2]	190	16.8 (11.6; 22.0)	221	13.1 (8.6: 17.6)	0.29
Intensity[3]		57 (42: 78)		54 (37: 77)	0.76
6 months pp[4]					
Prevalence[2]	184	17.4 (11.8: 23.0)	221	14.9 (10.1; 19.7)	0.50
Intensity[3]		67 (52: 86)		60 (44: 83)	0.62

[1] Only women who were found uninfected or successfully treated at 32 weeks gestation:

[2] Percentage (95% confidence interval)

[3] Geometric mean (95% confidence interval) eggs per gram for infected women only

[4] Postpartum

Table 6.3. Reinfection rates and intensities of hookworm among women eating anthill earth, compared to those eating other earth or no earth at all at delivery, three months and six months postpartum follow-up.[1]

	Geophagous		Non-geophagous (n=254)	*P*-value
	Anthill (n=95)	Other earth (n=130)		
At delivery				
Prevalence[2]	15.4 (7.4: 23.4)	5.5 (1.1; 9.9)	12.5 (8.1; 16.9)	0.07
Intensity[3]	208 (90: 478)	83 (35; 198)	77 (55: 106)	0.02
3 months pp[4]				
Prevalence[2]	21.5 (12.3: 30.7)	13.5 (7.0: 20.0)	13.1 (8.6: 17.6)	0.17
Intensity[3]	55 (34: 89)	61 (39; 93)	54 (37: 77)	0.9
6 months pp[4]				
Prevalence[2]	19.5 (10.5; 28.5)	15.9 (8.9: 22.9)	14.9 (10.1; 19.7)	0.64
Intensity[3]	61 (42: 88)	72 (49: 105)	60 (44: 83)	0.75

[1] Only women who were found uninfected or successfully treated at 32 weeks gestation:
[2] Percentage (95% confidence interval). at delivery (anthill. n=78: other earth. n=109: non-geophagous. n=224): 3 months postpartum (anthill. n=79: other earth. n=111: non-geophagous. n=221): 6 months postpartum (anthill. n=77: other earth. n=107: non-geophagous. n=221):
[3] Geometric mean (95% confidence interval) eggs per gram for infected women only
[4] Postpartum

6.3.3 *Ascaris lumbricoides*

As seen in Table 6.4. geophagous women had higher reinfection rates of *A. lumbricoides* during follow-up than non-geophagous ones. Reinfection with *A. lumbricoides* in geophagous women increased from 6.0% at delivery to 10.5% at three months postpartum compared to 3.3 to 3.4% in non-geophagous women during the same period (*P*<0.001). The increase in prevalence was even wider between the two groups at six months postpartum (13.8% vs. 5.8%. *P*<0.001) (Fig. 6.1). There were no significant differences noted in intensities of *A lumbricoides* between geophagous and non-geophagous women during the follow-up. The relative risk for reinfection with *A. lumbricoides* at delivery comparing geophagous and non-geophagous was 1.48 (0.84: 2.61) compared to 2.16 (1.28: 3.66) at three months postpartum and 2.44 (1.50: 3.98) at six months postpartum. However. only the last two were statistically significant (*P*<0.001).

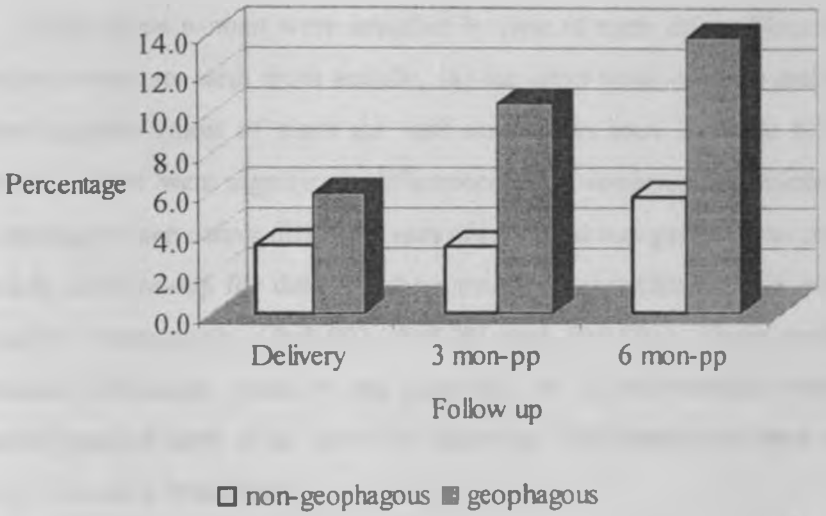

Figure 6.1. Prevalence of *Ascaris lumbricoides* infection by geophagy among lactating women examined for re-infection from pregnancy (second trimester) to 6 months postpartum

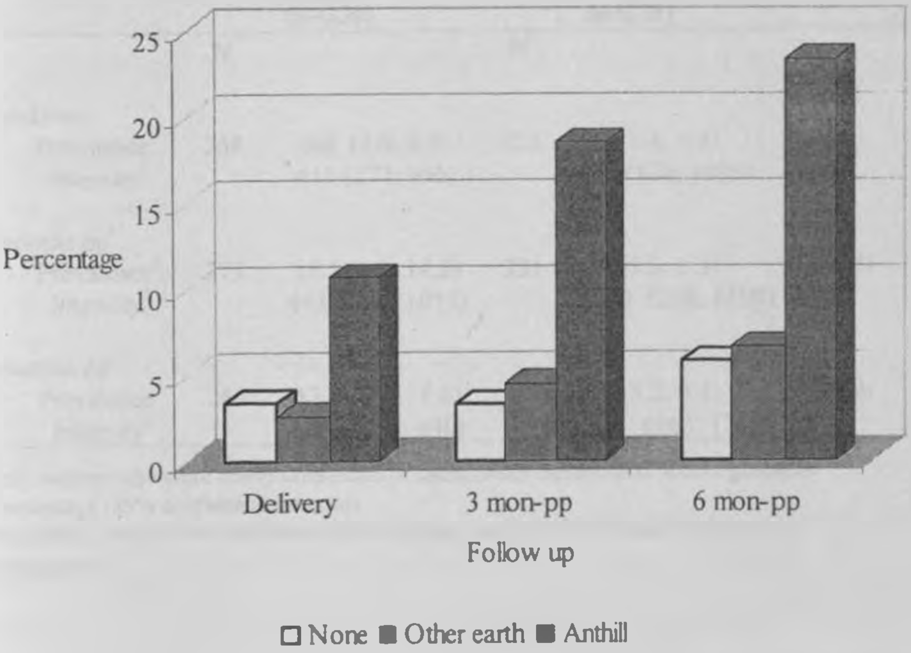

Figure 6. 2. Prevalence of *Ascaris lumbricoides* infection by type of earth eaten among lactating women examined for re-infection from pregnancy (second trimester) to 6 months postpartum.

Geophagous women were stratified by type of earth eaten. There were 139 women who ate earth from anthills. 183 ate other types of earth and were grouped together (most of them ate 'soft stone'). As seen in Table 6.5 and Figure 6.2, there were significant differences in *A. lumbricoides* reinfection rates among women eating different types of earth and non-geophagous ones at all levels of follow-up for delivery, three months postpartum and six months postpartum, respectively ($P=0.002$, $P<0.001$ and $P<0.001$). There were no significant differences noted in the intensities of *A. lumbricoides* between different types of earth at all levels of follow-up. The reinfection rates in all groups followed a linear trend.

Table 6.4. Reinfection rates and intensities of *Ascaris lumbricoides* among geophagous and non-geophagous women at delivery, three months and six months postpartum follow-up[1]

		Geophagous (n=320)		Non-geophagous (n=378)	*P*-value
	N		N		
At delivery					
Prevalence[2]	268	6.0 (4.0; 9.0)	323	3.4 (1.4; 5.4)	0.14
Intensity[3]		411 (177; 956)		360 (126; 1024)	0.83
3 months pp[4]					
Prevalence[2]	275	10.5 (6.8; 14.2)	331	3.3 (1.3; 5.3)	<0.001
Intensity[3]		443 (193; 1017)		1080 (264; 4410)	0.25
6 months pp[4]					
Prevalence[2]	268	13.8 (9.6; 18.0)	325	5.8 (3.2; 8.4)	<0.001
Intensity[3]		404 (190; 640)		536 (164; 1745)	0.46

[1] Only women who were found uninfected or successfully treated at 32 weeks gestation

[2] Percentage (95% confidence interval)

[3] Geometric mean (95% confidence interval) eggs per gram for infected women only

[4] Postpartum

Table 6.5. Reinfection rates and intensities of *Ascaris lumbricoides* among women eating anthill earth. compared to those eating other earth or no earth at all at delivery. three months and six months postpartum follow-up.[1]

	Geophagous		Non-geophagous (n=378)	*P*-value
	Ant hill (n=138)	Other earth (n=182)		
At delivery				
Prevalence[2]	10.7 (4.9; 15.6)	2.6 (0.1; 5.1)	3.4 (1.4: 5.4)	0.002
Intensity[3]	451 (151; 1350)	311 (46-2118)	360 (126; 1024)	0.90
3 months pp[4]				
Prevalence[2]	18.5 (11.4: 25.6)	4.5 (1.2; 7.8)	3.3 (1.3; 5.3)	<0.001
Intensity[3]	404 (150: 1090)	589 (80: 4346)	1080 (264: 4410)	0.48
6 months pp[4]				
Prevalence[2]	23.3 (15.5; 31.1)	6.6 (2.6; 10.6)	5.8 (3.2: 8.4)	<0.001
Intensity[3]	349 (160: 761)	349 (127; 959)	536 (164: 1745)	0.76

[1] Only women who were found uninfected or successfully treated at 32 weeks gestation

[2] Percentage (95% confidence interval). at delivery (anthill. n=112: other earth. n=156: non-geophagous. n=323): 3 months postpartum (anthill. n=119: other earth. n=156: non-geophagous. n=331): 6 months postpartum (anthill. n=116: other earth. n=152: non-geophagous. n=325)

[3] Geometric mean (95% confidence interval) eggs per gram for infected women only

[4] Postpartum

6.3.4 *Trichuris trichiura*

As seen in Table 6.6. there was no significant difference in the reinfection rates of *T. trichiura* between geophagous and non-geophagous women at all levels of follow-up: however. there was a small increase in the rates among the geophagous group. There was a significant difference in intensity of *T. trichiura* between geophagous and non-geophagous at six months follow-up (83 vs. 37 GM epg. *P*=0.005). There were no significant differences noted in intensities between the two groups of geophagy at delivery and three months postpartum. The relative risk for infection with *T. trichiura* was highest at three months postpartum but was not significantly different between the two groups.

When stratified by the earth eaten. there was still no significant difference in the reinfection rates of *T. trichiura* between geophagous and non-geophagous (table not shown). There was however a significant difference in the intensity of *T. trichiura* between the women eating different types of earth and non-geophagous ones at six months follow-up (*P*=0.02). There were no significant differences in intensities between the different groups of women at delivery and three months postpartum.

Table 6.6. Reinfection rates and intensities of *Trichuris trichiura* among geophagous and non-geophagous women at delivery, three months and six months postpartum follow-up.[1]

		Geophagous (n=315)		Non-geophagous (n=355)	*P*-value
	N		N		
At delivery					
Prevalence[2]	267	3.4 (1.2; 5.6)	306	4.2 (1.9; 6.5)	0.58
Intensity[3]		54 (19; 152)		58 (27; 126)	0.90
3 months pp[4]					
Prevalence[2]	269	6.3 (3.3; 9.3)	306	4.2 (1.9; 6.5)	0.26
Intensity[3]		73 (41; 130)		42 (29; 62)	0.12
6 months pp[4]					
Prevalence[2]	262	6.4 (3.9; 9.9)	299	5.3 (2.5; 7.5)	0.35
Intensity[3]		83 (57; 118)		37 (25; 55)	0.005

[1] Only women who were found uninfected or successfully treated at 32 weeks gestation;

[2] Percentage (95% confidence interval)

[3] Geometric mean (95% confidence interval) eggs per gram for infected women only

[4] Postpartum

6.3.5 Socioeconomic factors

Three socioeconomic factors were considered important in the transmission of geohelminths: latrine coverage, the woman's educational standard and the number of dependent children in the household. Distance from the lake was also included due to variations in the transmission patterns of hookworm in the area as noted in Chapter 4. There was a significant difference in reinfection with hookworm between women who had latrines and those who did not have latrines at delivery (7.9% vs. 16.5%, *P*=0.007), but not at three months and six months postpartum. Similarly there was no difference in the intensities of hookworm between these groups at delivery, three months and six months post partum (Table 6.7).

There were no statistically significant differences in the prevalence of *Ascaris* between women with latrines and those without latrines. However, the trend showed that those without latrines were likely to be reinfected earlier than those with latrines (6.5% vs. 3.5. P=0.09). Similarly, there were no significant differences in reinfection rates and intensities of *Trichuris* infection.

Table 6.7. Reinfection rates and intensities of hookworm among women with and without a pit latrine at delivery, three months and six months postpartum follow-up.[1]

	With latrine		Without latrine		P-value
	N		N		
At delivery					
Prevalence[2]	253	7.9 (4.5; 11.3)	158	16.5 (10.6; 22.4)	0.007
Intensity[3]		93 (57; 149)		107 (68; 167)	0.65
3 months pp[4]					
Prevalence[2]	253	14.6 (10.2; 19.0)	158	15.2 (9.5; 20.9)	0.87
Intensity[3]		50 (37; 69)		65 (46; 90)	0.29
6 months pp[4]					
Prevalence[2]	252	15.9 (11.3; 20.5)	153	16.3 (10.3; 22.6)	0.91
Intensity[3]		65 (50; 86)		60 (45; 81)	0.66

[1] Only women who were found uninfected or successfully treated at 32 weeks gestation
[2] Percentage (95% confidence interval)
[3] Geometric mean (95% confidence interval) eggs per gram for infected women only
[4] Postpartum

Table 6.8 shows the educational level of the women which was negatively associated with reinfection with hookworm at delivery (12.7 vs. 4.9%, P=0.05). However there was no significant differences noted in the reinfection and intensities with hookworm between women in the two educational categories. Similarly there were no significant differences in reinfection rates or intensities of both *Ascaris* and *Trichuris* between the two educational levels.

Table 6.8. Reinfection rates and intensities of hookworm among women with different levels of education at delivery, three months and six months postpartum follow-up.[1]

		Primary		Secondary	P-value
	N		N		
At delivery					
Prevalence[2]	330	12.7(9.1; 16.3)	81	4.9 (0.1; 9.7)	0.046
Intensity[3]		95 (68; 132)		179 (36; 855)	0.27
3 months pp[4]					
Prevalence[2]	327	16.5 (12.4; 20.6)	84	8.3 (2.3; 14.3)	0.06
Intensity[3]		55 (43; 71)		59 (38; 92)	0.85
6 months pp[4]					
Prevalence[2]	325	17.5 (13.3; 21.7)	80	10.0 (4.0; 16.0)	0.10
Intensity[3]		62 (49; 77)		78 (50; 121)	0.42

[1] Only women who were found uninfected or successfully treated at 32 weeks gestation

[2] Percentage (95% confidence interval)

[3] Geometric mean (95% confidence interval) eggs per gram for infected women only

[4] Postpartum

Having dependent children or not did not have any effect on the reinfection rates or intensities of hookworm and *Ascaris*. There was, however, a significant difference in the reinfection rates with *Trichuris*, but not in the intensities, between women with and without dependent children at delivery (2.4 vs. 6.5%, P=0.015) Most of the villages in the study area were close to the lakeshore with a few situated further inland. As seen in Table 6.9, women who lived far from the lakeshore were twice as likely to get re-infected with hookworm than those residing closer to the lake (18.3 vs. 9.7%, P=0.04) at delivery and (24.7 vs. 12.7%, P=0.009) at three months postpartum. There was no effect of distance from the lake on the intensities of hookworm reinfection. Distance from the lake had no effect on the reinfection rates or intensities with *Ascaris* and *Trichuris*.

Table 6.9. Reinfection rates and intensities of hookworm among women in relation to the distance from the lakeshore at delivery, three months and six months postpartum follow-up.[1]

	N	> 5km	N	< 5km	*P*-value
At delivery					
Prevalence[2]	71	18.3 (9.3; 27.3)	340	9.7 (6.5; 12.9)	0.036
Intensity[3]		117 (48; 286)		94 (69; 128)	0.535
3 months pp[4]					
Prevalence[2]	73	24.7 (14.7; 34.7)	338	12.7 (9.1; 16.3)	0.009
Intensity[3]		41 (28; 61)		63 (47; 83)	0.095
6 months pp[4]					
Prevalence[2]	76	22.4 (12.8; 32.0)	329	14.6 (10.7; 18.5)	0.096
Intensity[3]		80 (51; 125)		48 (47; 72)	0.148

[1] Only women who were found uninfected or successfully treated at 32 weeks gestation
[2] Percentage (95% confidence interval)
[3] Geometric mean (95% confidence interval) eggs per gram for infected women only
[4] Postpartum

6.4 Discussion

Geohelminths are prevalent in the study area as reported by other studies by Geissler *et al.* (1998) and Thiong'o *et al.* (2001). The area has all the factors that favour transmission and maintenance of intestinal helminths in the community: humid conditions, optimal temperatures for larval development and egg viability, and a generally low socioeconomic status. The association of geophagy with geohelminth infections has been studied (Hunter 1973; Wong *et al.* 1988; Horner *et al.* 1991; Geissler *et al.* 1997) and studies have been carried out on reinfection by helminths comparing drug efficacies or risk factors (Albonico *et al.* 1995; Magnussen *et al* 1997; Geissler *et al.* 1998b; Olsen *et al.* 2000; Saathoff *et al* 2000). However, most of these studies have been carried out on schoolchildren with a few cross-sectional studies on pregnant women (Geissler *et al.* 1998).

The prevalence of *A. lumbricoides* at baseline was 16.3% with an intensity of 815 epg. The prevalence was 9.4% at the close of the follow-up period, which was 58% of the baseline rate. This reinfection rate is low compared to that reported from schoolchildren in the same area and in the neighbouring district (Geissler *et al.* 1998b: Olsen *et al.* 2000). The increase was linear and had a high association with geophagy. This positive association of *A. lumbricoides* and geophagy has been documented in other studies (Wong *et al.* 1991: Geissler *et al.* 1998: Glickman *et al.* 1999: Saathoff *et al.* 2000). This association is likely to be causal as *Ascaris* infection is mainly through the ingestion of material contaminated with the eggs. In this study it was found that the type of earth most associated with *Ascaris* infection stemmed from anthills. which corroborates findings of other studies (Geissler *et al.* 1998: Saathoff *et al.* 2000). Anthills were common in the study area and often situated near to or in the homes. These mounds are sometimes used by children as screens behind which to defecate. thereby contaminating the surrounding area with *Ascaris* eggs. The contamination can be spread by wild and domestic animals. for example cattle. poultry and monkeys which are common near homesteads. It was observed that anthill earth is sometimes eaten at the source. a habit which is a direct ingestion of the eggs from the field. The different infection rates of the women and schoolchildren could be explained by the fact that the children are very active: pregnant women at this stage are a bit tired and confined to the home at the time of delivery and also women tend to be more hygiene conscious than the children.

Ascaris intensities at six months were comparable to the ones at baseline. This agrees with studies that reported higher intensities in children during reinfection than pre-treatment (Henry 1988: Albonico *et al.* 1995: Olsen *et al.* 2000). but not among adults (Olsen *et al.* 2000). Among schoolchildren in the same region intensities did not attain the baseline means after 11 months (Geissler *et al.* 1998b). Likewise. studies in Kwale District did not reach the baseline mean intensities even at 18 months after treatment (Magnussen *et al.* 1997). The mean intensities of infections among the women were not as high as has been reported in children and unlike the prevalence they were not significantly different between geophagous and non-geophagous groups. as previously found by Geissler *et al.* (1998b).

The finding that reinfection with *Ascaris* was not associated with access to a latrine disagreed with findings by Henry (1988) who found a significant difference in reinfection between those with latrines and those without and showed that *Ascaris* infections could be reduced by 30% by introduction of latrines (Henry 1988). Similarly, studies in Nigeria and Lubumbashi (Tshikuka *et al.* 1995; Asaolu *et al.* 2002) found that people using the bush for defecation had a higher prevalence of *Ascaris* than those with pit latrines and Sorensen and his co workers in Sri Lanka (1994) established that *Ascaris* was more prevalent in families with poor sanitary conditions (latrine coverage <20%) than in those with good sanitary conditions (latrine coverage >70%).

There was a 5.9% prevalence of *Trichuris* at 6 months postpartum which converted to 24% of the baseline prevalence. This was low compared to the reinfection rates found by Olsen *et al.* (2000) and Magnussen *et al.* (1997) in other areas of Kenya, and by Geissler *et al.* (1998b) in schoolchildren in the same area. There was a significant difference in the intensities between geophagous and non-geophagous women, which corroborated findings by Geissler *et al.* (1989b). This is biologically plausible since the mode of infection is through ingestion of contaminated material. However, there being no difference in reinfection between geophagous and non-geophagous women could be explained by the fewer eggs of *Trichuris* in the environment. *Trichuris* eggs do not last longer in the soil because they are highly sensitive to desiccation, and unlike *Ascaris* eggs, they lack the thick sticky albumin coating that would have allowed them to adhere on earth particles. They are easily swept away owing to the smooth surface coating (Geissler *et al.* 1998b).

It was found that hookworm cure rate was poor after treating with Mebendazole as a 500 mg single dose. This has also been documented by studies among schoolchildren in Tanzania and Kenya and in all age groups in Mali (Albonico *et al.* 1995; Muchiri *et al.* 2001 and De Clercq *et al.* 1997). The rate of reinfection with hookworm was 16% at 6 months postpartum which was about 32% of the baseline prevalence. This agreed with results within the same range obtained in other studies in Kenya (Magnussen *et al.* 1997; Olsen *et al.* 2000). Likewise, the intensities were low during reinfection and compared well with the findings of the same studies in Kenya. There was a significant difference in intensities of hookworm between geophagous and non-geophagous at delivery which could have been coincidental or egg aggregation in the stool sample. However it was shown that the high rate of reinfection and intensity was confined to women who ate anthill earth. One would speculate that since children in the area have a tendency to defecate around anthills. these women might have acquired the infection in the course of their search for earth from these mounds. The moist earth around the mounds would be conducive for the survival of hookworm filariform larvae. Women who did not have latrines in their homesteads had higher reinfection rates than those with latrines. This corroborates findings by Sorensen and his co-workers in Sri Lanka (1994). Olsen *et al.* in western Kenya (2001) Arfaa *et al.* in Iran (1977) and Holland *et al.* in Panama (1988). While comparing educational levels. the women with lower standards of education had higher reinfection rates than those with secondary or higher education. This agrees with findings by Olsen *et al.* (2001) in the same region and Holland *et al.* (1988) in Panama. This might be linked to the fact that the more educated women in the area tend to wear shoes. which could in turn protect them from getting infection. Distance from the lake has been associated with higher prevalence of hookworm in the area (Olsen *et al.*, 2001: Thiong'o *et al.* 2001). Women living further inland had higher reinfection rates than those living closer to the lakeshore. The mode of transmission of hookworm is through larval penetration. It is therefore surprising that although sanitary conditions are similar throughout the study area and sometimes worse closer to the lake due to the type of rock or earth. hookworm is more prevalent inland than closer to the lake shore. There has been a negative correlation between hookworm and *S. mansoni* prevalence along the lakeshore (Thiong'o *et al.* 2001). The explanation would be based on further studies modelled on GIS findings by Saathoff *et al.* (2002) where they demonstrated association between hookworm and normalized difference vegetation index (NDVI)

6.5 Conclusion

This study has shown that geophagy is associated with *Ascaris lumbricoides* reinfection among pregnant women. It was found that geophagy intensities built up more rapidly among geophagous women than non-geophagous within six months. This calls for repeated treatment of the women after delivery. In view of the fact that transmission is localized and probably within the household domains. treatment should be extended to other members of the family and more so to the high risk groups such as the younger age groups of pre-school and school age children (WHO 1996: Montresor *et al.* 2002). Since there is interaction among community members. It is known that high reinfection would be an indication of relatively high infection in the community (Brooker *et al.* 2000). The study has also shown that a single dose of mebendazole 500 mg was not an effective tool in hookworm treatment. albendazole should be used in future control measures and more effectively twice a year. Since geophagy is an important factor in the transmission of *Ascaris*. further studies should be undertaken to find ways of stopping the behaviour by safer supplements or a combination of health education coupled with prompt treatment of the high risk groups.

Chapter 7

General Discussion, Conclusion and Recommendations

7.1 Geophagy

This study has confirmed that geophagy or earth eating is common among pregnant women in the area studied. This universal cross-cultural phenomenon has been reported from different parts of the world such as Latin America (Hunter & de Kleine 1984). USA (Hunter 1973). Iran. India. China. Indonesia and Oceania (Anell & Lagercrantz 1958: Hunter 1979). but also in the Mediterranean. and up to the recent past. in western Europe (Halstead 1968). It appears to be particularly widespread in Africa (Lagercrantz 1958. Fig.1.4 Map of Africa). In his review of earth eating in Africa. Hunter (1979) noted that earth was eaten in connection with local religious practices and as medicine. In many African societies craving for earth during pregnancy is considered a normal and satisfying need for the mother and the foetus. The study area residents were no exception in accepting this habit which is perceived as a normal practice in women of all age categories (Geissler *et al.* 1997; Geissler 1998).

7.1.1 Types of earth eaten

The preferred earth eaten by pregnant women in the study came from ant hills. This type of earth is thought to be rich in minerals (Hunter 1979). Other sources included walls of houses made from mud from deeper soil. and presumably richer in minerals. and soft stone which is mined from particular hills in the Province. The latter has been commercialized and is thought by women to be cleaner. During his studies of geophagy in west Africa. Vermeer (1966) found that the Tiv people in Nigeria mined soil and sold it as an anti-diarrhoeic and mineral supplement. while in Ghana the Ewe mined clay and sold it through markets (Vermeer 1971). However. unlike in Kenya. their clay was processed and sun baked in special shapes before being sold. Edible clays in Ghana were analysed for mineral content: the highest concentration of iron was 39 mg/kg while the lowest concentration was 4 mg/kg. These concentrations were almost comparable to those found in the soft stone (27 mg/kg) but much lower compared to those in the ant hill mounds in our study area. The craving for earth and its link to pregnancy where iron need is increased due to physiological changes necessitated the need for determining the association with parasitic infections and iron deficiency anaemia among the women.

7.1.2 Geophagy and iron nutrition

Earth eating has been thought to be related to nutrition. Studies to this effect have been undertaken in different countries (Halstead 1968: Hunter 1973; Arcasoy *et al.* 1978) but inconclusive as far as the causal relationship is concerned. Iron deficiency anaemia is a severe stage of iron deficiency in which haemoglobin falls below cut–off points (7.0g/dl) as set by WHO expert committee on health matters. This occurs when iron stores are exhausted and the supply of iron to the tissues is compromised. Biochemical indicators of iron deficiency include serum ferritin. transferrin saturation. transferrin receptor and erythrocyte protoporphyrin. Iron deficiency generally develops slowly and is not clinically apparent until anaemia is severe. even though functional consequences already exist. The physiological requirement for iron in pregnancy is difficult to meet with most diets. There is need to increase iron intake to compensate for the development of the foetus. In the present study geophagous women took extra iron through earth eating of about 20% of the recommended dietary intake: however. it is not known how much of it was absorbed. Decreased iron absorption has been reported in several case studies linking it to geophagy and iron deficiency. In Turkey these tests included children with prolonged geophagy which showed a high rate of mal-absorption of iron associated to geophagy (Arcasoy *et al.* 1978: Sayar *et al.* 1975: Ayan *et al.* 1983).

Geophagy was found to be negatively correlated with haemoglobin and serum ferritin among the pregnant women. It was also found that the concentration of iron was higher in the few earth samples picked from mud walls (189 mg/kg) of huts and the ant hill earth (131 mg/kg) than the soft stones (27mg/kg) (see Chapter 3) – implying that there was more iron in earth from the surface than the soft stones mined deeper in the hills.

7.1.3 Geophagy and helminth infections

The present study endeavoured to throw light on the role that geophagy might have on the lethal but marginalized parasitic infections in the risk population. In the study area a few individuals harboured heavy helminth infections while most of them had very light infections or were negative. This phenomenon of over dispersion and aggregation of parasites in few individuals has been studied (Croll & Ghadrian. 1981; Keymer 1982: Upatham *et al.* 1992: Show & Dobson 1995). It should be noted that Croll and his co-worker did not find any correlation between pre-treatment and post-treatment worm burdens 12 months later. Over dispersion could have led to low mean egg counts of intestinal worms in our study area.

Reinfection with *A. lumbricoides was* 58% of the pre-treatment prevalence seven months earlier. which suggests that at 12 months post-treatment. the rate would be higher. This shows that worm acquisition was rapid and dependent on relationship between worm burden and rates at pre-treatment and the behaviour of the host after chemotherapeutic treatment. It also shows that reinfection with *Ascaris* is attained faster in high risk groups and decays also faster as reported in children and adults (Elkins *et al.* 1986).

Predisposition to *T. trichiura* was low in this study area. Bundy *et al.* (1987) working in St. Lucia noted that this parasite was highly aggregated in the community with a few people harbouring heavy burdens and also concluded that reinfection was higher than in other parasites. which was not the case in this study. This would be attributed to the fact that we had a particular selected age group while we know that predisposition to *T. trichiura* is age related.

Reinfection rates with hookworm were low in the present study. This could have been due to the state of the cohort participants who were rendered less active due to pregnancy. However, predisposition to hookworm is normally low while the hookworm burden may be higher. A study in India revealed that a group treated in 1988 did not have a difference in reinfection in 1996: the prevalence was lower in 1996 than 1990 and 1988 (Quinnell *et al.* 2001). This could be true in the sense that hookworm infection reaches its peak in adults and may plateau or decrease a bit with age.

In the present study it was concluded that geophagy was prevalent in women and especially in pregnant ones (45.7%) and that the commonly eaten earths were from soft stone (54.2%) and ant hill mounds (42.8%), which were abundant in the area. The prevalence of hookworm in the study area was 49.8%, *A. lumbricoides* 16.3%, *T. trichiura* 24.1% and *S. mansoni* 63.0%.

It was also shown that earth eating, and especially ant hill earth, was related to high intensity of *A. lumbricoides* whose transmission is through ingestion.

The most interesting observation was the association of predisposition to *A. lumbricoides* with geophagy where intensities rapidly built up. The prevalence in the geophagous women was higher during reinfection at six months: geophagous (13.8 %) and non-geophagous (5.8%). Among the earth eaters, ant hill earth was highly responsible for *Ascaris* reinfection (23.3%), with other types of earth only 6.6%.

7.2 Other risk factors for helminth infection

Education had an effect on prevalence of hookworm: there was low prevalence among highly educated women (36%) as compared to those with low education (53%).

Women without latrines in the homes were likely to become re-infected with hookworm during pregnancy. The intensities of hookworm were also higher in this category of women than in those women with latrines.

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There was a high reinfection rate of *A. lumbricoides* among women with primary education during pregnancy than among those with secondary education. There was also a higher prevalence of hookworm among women residing farther from the lake than those closer to the lake. Conversely there was a higher prevalence of *S. mansoni* among women residing closer to the lake than those residing farther inland.

7.3 Recommendations

7.3.1 General lessons for helminth control

Control measures should target high risk groups for different types of helminths: *A. lumbricoides* and *T. trichiura* in the younger age groups and among women in the reproductive age groups who become victims during pregnancy. All adults should be targeted for hookworm and *S. mansoni* infections.

Treatment should be carried out two times a year in order to cater for the rapid reinfection rates in the risk age groups. Furthermore, the appropriate drug should be used, in this case Albendazole, since Mebendazole was not as effective against hookworm in the study area.

Control measures in the area should be based on prevalence rather than intensity of infections. This would reduce the rate of reinfection in the whole area rather than a few targeted individuals.

7.3.2 Geophagy and pregnancy

Pregnant women should be examined for helminths and their Hb estimated during their first visit to the maternal child health clinics in order to detect any abnormalities early and to take the necessary steps to correct.

Health education should be introduced in all clinics to emphasize the dangers of earth eating and other potential risk factors that may predispose the women to helminth infection. Furthermore, women should be encouraged to visit the clinic in early pregnancy (first trimester) instead of coming late in the second trimester as noted in the study area.

Since geophagy is a culturally rooted practice that cannot easily be discouraged. and as we do not sufficiently understand the potential benefits of this practice. relevant health education should find ways of promoting improvements of the practice. For example. women could be encouraged to bake the earth before eating it. which already some are doing. and which is common in areas in Nigeria (Vermeer 1966). in order to reduce the potential of helminth transmission.

Rather than dismissing earth eating as an obsolete (and thus embarrassing) practice. it should be treated as a normal habit. which. however. should not be overdone. The potential risks should be explained to pregnant women. as well as how these can be avoided without abandoning the practice altogether.

7.4 Study Limitations

During the study a few limitations were noted. The common constraint was migration of the study subjects out of the study area. This made follow up cumbersome. Other limitations included:

- The inability to treat *S. mansoni* cases during pregnancy due to guidelines from the Ministry of Health.
- The use of a single dose of mebendazole for the treatment of geohelminths. which did not clear hookworm infection.
- The technique for egg recovery from the earth was cumbersome and did not produce expected results.

7.5 Further studies

As the potential benefits of earth eating have yet to be elucidated. further studies should be undertaken in order to assess whether the nutritional benefits outweigh the bad effects of geophagy.

Further studies are needed to determine whether it is geophagy that has an effect on iron absorption or whether iron deficiency leads to geophagy and hence causes damage to the mucosa in the intestines.

A more detailed study on geophagy and iron deficiency anaemia should be undertaken. which should include as many tests as possible for determining the true state of anaemia while taking care of confounders.

The origins and quality of 'soft stone' should be studied in different settings. This earth type is increasingly commonly eaten. especially among urban women. and unless its quality is monitored. health risks such as exposure to environmental poisonous materials may be encountered.

Since geophagy is common. the practice ought to be studied in urban settings. Here. the risk of exposure to environmental pollutants is much greater than in the rural areas. Urban schoolchildren in Zambia have been shown to be avid earth eaters (Nchito *et al* 2004). and there is no reason to assume that adult women in the city would easily abandon this habit. Exposure to pollutants such as heavy metals could have grave consequences for the health of the mother and the child. especially during pregnancy. and the child's long-term development. Thus. studies on the prevalence of geophagy in urban areas in Kenyan as well as the sources and quality of earth eaten should be done.

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