EVALUATION OF COCCIDIAL PREVALENCE AND INTENSITY IN POULTRY BIRDS IN MINNA METROPOLIS, NIGER STATE, NIGERIA

A survey to determine the prevalence and intensity of coccidiosis among poultry birds within Minna metropolis Niger State, Nigeria, was carried out between July and October 2013. A total of 450 fresh faecal droppings and 80 carcasses were collected from 12 different poultry farms within Minna, and analyzed using parasitological techniques. From the 450 faecal samples collected, a total of 166(36.3%) were infected with oocyst of Coccidia. An overall geometric mean intensity of 8.12 oocyst/3g of faeces was recorded. Prevalence of infection was higher in the adult birds (37.2%) than in the younger birds (36.5%). There was no significant difference ($P<0.05$) in the intensity of infection between the age groups. The *Eimeria* species isolated from the post mortem examination included: *Eimeria tenella, E. maxima, E. acevulina, E. necatrix* and *E. mitis.* An overall prevalence of 66.3% infection was recorded from the post mortem examination. *E. tenella* prevalence was higher than the other *Eimeria* species. The overall geometric mean intensity of 8.33oocyst/1g of faeces was recorded. However, the intensity of oocysts was not significant ($P<0.05$). An overall prevalence (55.6%) of single infection and a prevalence (44.4%) of mixed infections of Eimeria species isolated from the post-mortem examination was also recorded. The present study showed that coccidiosis is an important disease of poultry in the study area and suggest that appropriate control and treatment strategies be designed to reduce the effect of the disease.

CHAPTER ONE

1.0 INTRODUCTION

1.1 Background

Poultry refers to domestic birds such as Chickens, Turkeys, Ducks, Guinea fowl, Peasants, Geese, Quills kept for meat or egg production. Poultry are kept in backyards or in commercial production systems in most areas of the world. Compared to a number of other livestock species, fewer social and religious taboos are related to the production, marketing, and consumption of poultry products. For these reasons, poultry products have become one of the most important protein sources for people throughout the world (Anders and Jordan, 1998).

Diseases results when normal body functions are impaired, and the degree of impairment determines the severity of the disease. It may result from the consequences of harmful actions of infectious and parasitic agents, or it may be caused by injury or physical stress with which the bird cannot cope. A disease resulting from parasitism depends on the number, type, and virulence of the parasite, the route of entry to the body, the defense status and capabilities of the host. (Saif, Barnes, Glisson, Mcdougald, and Swayne, 2003).

Coccidiosis is an important poultry disease caused by a protozoan parasite belonging to the Apicomplexa phylum, the family of the *Eimeridae* and genus of *Eimeria*. There are 7 species of coccidia of pathological importance in chicken: *Eimeria acervulina, Eimeria brunetti, Eimeria maxima, Eimeria necatrix, Eimeria tenella, Eimeria praecox* and *Eimeria mitis* (Ovington, Alleva, and Kerr, 1995). In all parts of the world where confinement rearing is practiced, coccidiosis represents

a major disease problem that requests attention of poultry producers, feed manufactures and poultry disease experts. The infectious form of the parasite is the oocyst, which sporulates two days following excretion through the feaces in natural environment and can be ingested by a susceptible host organism (Williams, 1999).

Coccidiosis is a gastrointestinal avian disease characterized by enteritis and diarrhea, which can become bloody with certain *Eimeria* species. The sporozoites replication in the epithelial cells of the intestinal tract causes tissue damage with resulting interruption of feeding, digestive processes or nutrient absorption, dehydration and blood loss (McDougald, 2003). The macroscopic lesions in the digestive tract are some predisposing factors to many gastrointestinal bacterial poultry diseases such as Clostridiosis, Salmonellosis and Colibacillosis (Bostvironnois and Zadjian, 2011). Certain immunosuppressive viral diseases such as Infectious bursal disease, Marek disease and Chick aneamia infectious viral disease (Lanckriert *et al*., 2010) also exacerbate coccidiosis. The economic importance of the disease is due to its high rate of morbidity and mortality in young and adult birds, reduced feed conversion efficiency and egg production especially in sub-clinical cases.

Epidemiological studies in Nigeria have established the economic importance of coccidiosis as a major parasitic disease of poultry (Majaro, 1980, 1983 and 2001; Adene and Oluleye, 2004; Abdu, 2007). The disease occurs throughout the year in Northern Nigeria but with higher prevalence rate from May to September have been reported (Etuk, Okoli, and Uko, 2004; Abdu, 2007).

Coccidiosis has been shown to be common to intensively managed commercial poultry farms especially where management or hygienic standards are compromised (Adene and Oluleye, 2004). The increasing interest in commercial poultry production in Nigeria evidenced by the proliferation of poultry farms, suggests increased risk of outbreak of coccidiosis. The current study came up with the prevalence and intensity of coccidiosis and the preponderance of *Eimeria* species affecting the intensively managed commercial exotic chickens in Minna.

1.2 Justification

The poultry industry occupies an important position in the provision of animal protein (meat and egg) to human and generally plays a vital role in the national economy as a revenue provider. Coccidiosis is a health problem resulting in significant economic losses in the world. The impacts of disease on animal agriculture include, for example, lost in revenues, costs of vaccination, prevention, eradication, decontamination and restocking. Knowledge of the prevalence of coccidiosis is essential in understanding the epidemiology of the disease and on which basis the designing of appropriate control measures that would increase poultry productivity to meet the local seasonal demand for chicken and chicken products during religious and cultural festivals.

1.3 Aim

The aim of this study to investigate the prevalence and intensity of coccidiosis infection in poultry birds within Minna.

1.4 Objectives

I. To determine the prevalence of coccidial infection among poultry in Minna metropolis.

II. To determine the species of coccidia infecting poultry in Minna metropolis.

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Background history

Poultry is one of the most intensively reared of the domesticated species and one of the most developed and profitable animal production enterprises (Obiora, 1992). Its importance in national economies of developing countries, in the improvement of the nutritional status and income of many small scale farmers particularly those with small land holdings as well as landless, whose contribution has been recognized by various scholars and rural development agencies for the last two decades. (FAO, 1987; Creevey, 1991; Kitalyi, 1998).

Poultry production has been constantly increasing over the past decades, and a recent survey made by FAO shows that the whole poultry population in the world reaches about 14 billion, among these 75% are in the developing countries (FAO, 2000). In most African countries, backyard poultry account for more than 60% of the total national poultry flocks accorded an asset value of more than 5.75 billion US$ (Sonaiya , 1990). It is estimated that these provide 12kg of poultry needs per inhabitants per year whereas cattle provides 5.3kg (Forsido, 1990).

According to (Calnek, et al., 1997) chicken production is constrained by many extrinsic factors among which malnutrition, poor management and the absence of biosecurity are outstanding. Losses have also been attributed to limited housing and veterinary care services. Furthermore, poor genetic potential due to lack of selection and predation are also potential threats to productivity. Parasitism like coccidiosis ranks high among the factors that threaten village chicken production (Adene and Dipeolu, 1997). Moreover, it has been reported that parasitic infection or their

concurrent infections result in immune-suppression, especially in response to vaccines against some poultry diseases. Studies in other countries had shown that the prevalence of parasitic infestations in village chicken flocks is close to 100%, and in most cases individual birds' harbor more than one parasite type (Permin, Bojesen, Nansen, Bisgaard, Frandsen, and Pearman, 2007).

2.2 Morphological features of chickens

2.2.1 The digestive system of chickens

The small intestine of poultry is relatively simple and short but highly efficient Never the less, it is easily divided into 3 parts; duodenum, proximal small intestine (jejunum) and distal small intestine (ileum). The proximal and distal small intestines do not show distinguishing histological differences that define the jejunum and ileum of other vertebrates. The ileocaecal junction is found at the base of the distal small intestine and top of the large intestine. This junction is the site where twin caecal pouches join the linear portion of the intestine and is the location of the largest element of the gut immune tissue in the caecal tonsils. The caeca are thin-walled pouches that contain the anaerobic microflora responsible for fermentation in the bird. Finally, the large intestine, short and simple in poultry, joins the small intestine with the cloaca, the common receptacle for urinary, fecal, and reproductive products.

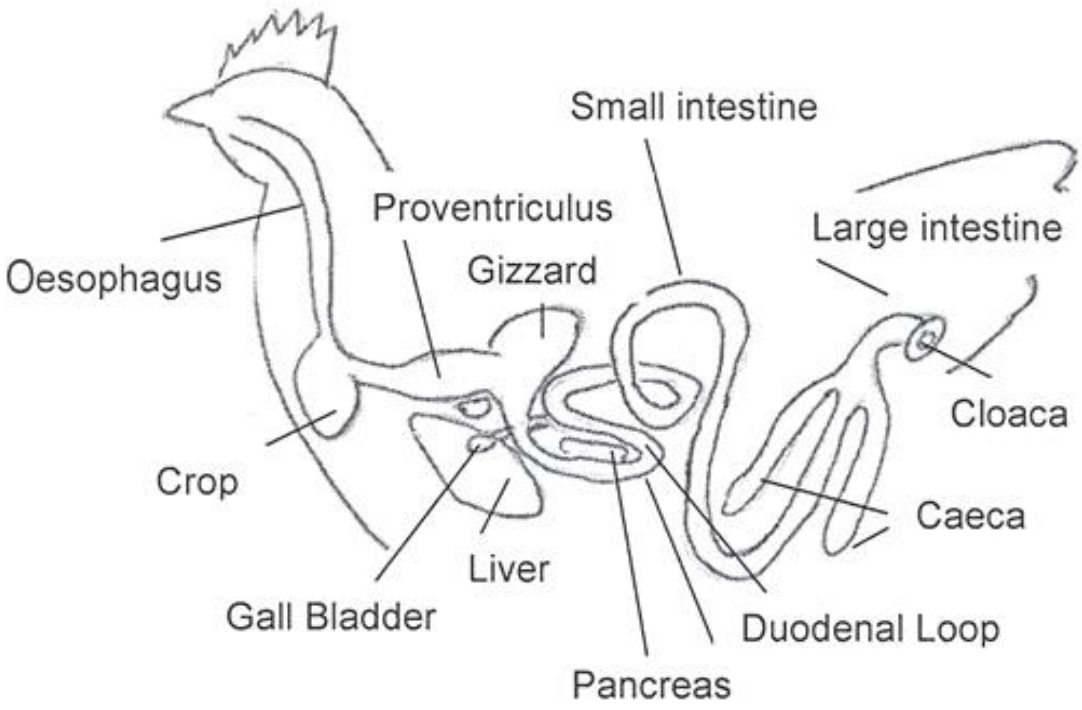

Plate 2.1 Digestive system of chicken (Dibner and Richards, 2004).

2.2.2 Infection by coccidiosis in chicken

Coccidiosis is one of the most common and costly diseases in poultry. The death rate can be quite high, both in chicks and in adults. It is characterized by droopiness, paleness of the comb, diarrhea and occasional appearance of blood in droppings. Coccidiosis in chickens, is caused by several different species of coccidia (Genus *Eimeria*), which are single celled protozoans that live in the gut wall of poultry. They are host-specific: Eg. Turkeys and other species are not infected by chicken coccidia and vice-versa. The different species of coccidia live in different parts of the gut and can be divided into those causing intestinal coccidiosis (the majority) or caecal coccidiosis (one species). For this to effectively occur, coccidia undergo a life cycle in the host (Murray and Hanson, 1998).

2.3 General life cycle of coccidia

Stages of coccidia in chickens appear both within the host as well as outside. The developmental stages in the chicken give rise to a microscopic egg (called an oocyst) which is passed out in the droppings. Under proper conditions of temperature and moisture the oocyst develops within one to two days to form a sporulated oocyst which is capable of infecting other chicken. At this stage the ooycst contains eight bodies (called sporozoites), each of which is capable of entering a cell in the chicken's intestine after the oocyst is eaten. When sporozoites enter the cells, they divide many times producing either a few or many offspring (merozoites). The numbers produced depend on the species of coccidia involved. Each merozoite in turn may enter another intestinal cell. This cycle may be repeated several times. Because of this cyclic multiplication, large numbers of intestinal cells

are destroyed. Eventually, the cycle stops and sex cells (male and female) are produced. The male fertilizes the female to produce an oocyst which ruptures from the intestinal cell and passes in the droppings(Woltgan, Uphoff, Hofmann, Kerlen, Seack, and Hornig, 1999). Thousands of oocysts may be passed in the droppings of an infected chicken. Therefore, poultry raised in crowded or unsanitary conditions are at great risk of becoming infected by *Eimeria* species.

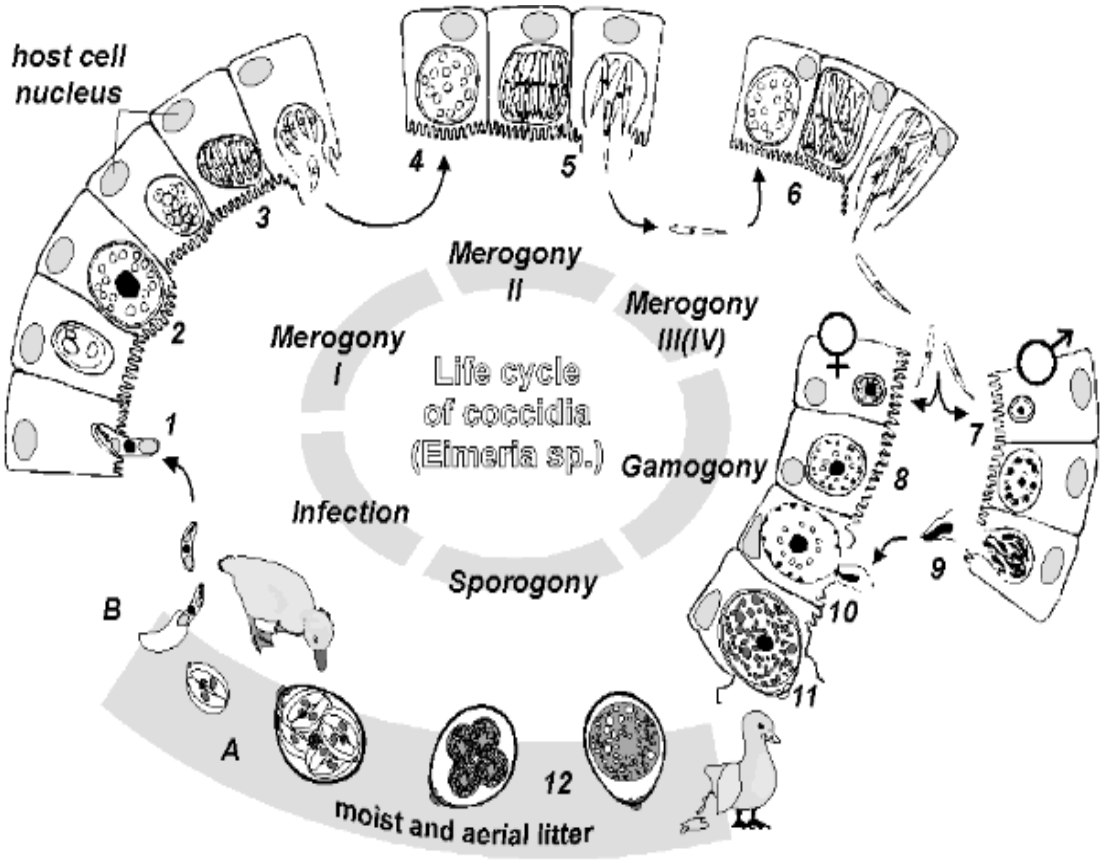

Plate 2.2: Life cycle of *Eimeria sp.* (Anne, 2006).

2.3.1 Coccidian parasites

Different *Eimeria* species occupy different ecological sites of the alimentary canal (Long, 1980).The small intestine is the most popular site for most coccidia in chickens. This is because the organ is very long, providing a large number of enterocytes and allowing enormous multiplication of the parasites usually without causing too much damage (Long,1980). These strains include; *E. tenella, E. necatrix, E. brunetti, E. maxima, E. acervulina, E. mitis, E. hagani, E. praecox. Eimeria tenella* affects young chicks most commonly from the age of 6-12 weeks. The affected chick suffers from acute coccidiosis a disease in which bloody diarrhea is conspicuous and death losses are often high. The oocysts are small, broadly ovoid, with a little difference between the two ends. They measure 19.5 to 16.5 to 22 microns with an average of 19 by 22.6 microns . Gross lesions of *Eimeria tenella* are confined to the caeca and consist of the presence of haemorrhages on the outside or inside of the wall of the caeca, free-blood or a chocolate-colored fluid content inside the caeca with a thickening of its wall or the presence of a large core of cellular debris and blood. *Eimeria tenella* can kill birds so dead birds in a flock with increased mortality should always be examined for the presence of lesions compatible with *E. tenella* infection or caecal coccidiosis (Murray and Hanson, 2007). *Eimeria necatrix* species is the most pathogenic for chicken. The damage caused by this species is most likely to occur in birds and immediate death losses are not so frequently found. The disease takes a longer course and there for called chronic coccidiosis. The oocysts are broadly oval. Some are somewhat egg shaped, with one end more pointed than the other, measuring11.3 to 18.3 by 13.2 to 22.7 microns, averaging to14.2 by 16.7 microns. *Eimeria necatrix* develops in the small

intestine (early stages) and later in the cecum (sexual stages) like E. tenella, it develops within deeper tissues of the small intestine and is a major pathogen of poultry. However, *E. necatrix* causes a more chronic disease than*E. tenella* and does not produce as many oocysts. Therefore, a longer time is usually required for high levels of environmental contamination. Birds heavily infected with *E. necatrix* may die before any marked change is noticed in weight or before blood is found in the feces (Murray and Hanson, 2007). *Eimeria brunette* has somewhat egg shaped oocysts with a thick lining on the inner surface of the oocysts. They measure 20.7 to 30.3 microns in length and 18.1 to 24.2 microns in width. *Eimeria maxima* receive its name from the size of its oocysts, which are larger than any other *Eimeria* species. They have a yellow colour and the shells often have a rough surface. *E. maxima* produce few marked changes in the small intestine until the fifth day after infection. After which, in severe infections, numerous small hemorrhages occur along with a marked production of thick mucus. The intestine loses tine and becomes flaccid and dilated. The inner surface is inflamed and the intestinal content consists of a pinkish mucoid secretion. Lesions of *E. maxima* comprise multiple petechial (pin-point size) haemorrhages often seen from the outside of the mid-gut area, in addition, segmental ballooning or enlargement of the mid-gut with presence of orange-tainted mucous may be noted. However, unless the lesions are typical they are harder to identify than those caused by *E. acervulina* and *E. tenella* and therefore it is highly desirable to confirm its presence by identifying the presence of coccidial oocysts (eggs) in a scraping from the mid gut under a microscope (Murray and Hanson, 2007).

Eimeria acervulina has egg shaped oocysts whose diameters are not known. *E. acervulina* is less pathogenic than *E. tenella* or *E. necatrix*. *E. acervulina* is responsible for sub-acute or chronic intestinal coccidiosis in broilers, older birds andchickens at the point of lay. The clinical signs consist of weight loss and a watery, whitish diarrhea. At postmortem, greyish-white, pin-point foci or transversely elongated areas are visible from the outer (or serous) surface of the upper intestine. The foci consist of dense areas of oocysts and gamete (male and female sex cells) production. Gross lesions caused by *E. acervulina* are usually the most prevalent and are usually confined to the upper small intestine (duodenum), although they may extend to the mid-gut (jejunum). The lesions have a unique appearance, consisting of white patches or transverse white lines inside the gut that may already be observed from the outside. *E. accervulina* and *E. maxima* develop in epithelial cells, primarily in the upper part of the small intestine (Murray and Hanson, 2007).

Eimeria mitis species is occasionally pathogenic. It develops in the upper part of the small intestines and to a lesser extent in the lower part and sometimes even in the ceaca. The oocysts are nearly spherical in shape and their diameters are not known. *Eimeria praecox* species is practically nonpathogenic with oocysts which are oval in shape .

2.4 Transmission

Chickens become infected with *Eimeria* spp. by ingesting infective oocysts (eggs) from litter, soil and contaminated feed and water. The infected birds excrete oocysts into their faeces and are a source of infection for other birds. As *Eimeria* spp. Can survive for long periods in infected birds and the environment (Khan, Irshad, Anjum, Jahangir, and Nasir, 2006). The oocysts in faeces become infective through the process of sporulation in about two days (Jeurissen, Janse, Vermeulen, and Vervelde, 1996). Birds in the same flock may ingest the oocysts through litter pecking or the contamination of feed or water. Although no natural intermediate hosts exist for the *Eimeria* spp. many different animals, insects, contaminated equipment, mice, wild birds, and dust can spread oocysts mechanically. Oocysts generally are considered resistant to environmental extremes and to disinfectants, although survival time varies with conditions oocysts may survive, for many weeks in soil, but survival in poultry litter are limited to a few days because of the heat and ammonia released by composting and the action of molds and bacteria. Viable oocysts have been reported from the dust inside and outside broiler houses, as well as from insects in poultry litter. The darkling beetle, common in broiler litter, is a mechanical carrier of oocysts. Transmission from one farm to another is facilitated by movement of personnel and equipment between farms and by the migration of wild birds, which may mechanically spread the oocysts. New farms may remain free of coccidia for most of the first grow out of chickens until the introduction of coccidia to a completely susceptible flock. Such outbreaks, often more severe than those experienced on older farms, are often called the new house syndrome. Oocysts may survive for many weeks under optimal conditions but will be quickly

killed by exposure to extreme temperatures or drying. Exposure to 55°C or freezing kills oocysts very quickly. Even 37°C kills oocysts when continued for 2-3 days. Sporozoites and sporocysts can be frozen in liquid nitrogen with appropriate cryopreservation technique, but oocysts cannot be adequately infiltrated with cryoprotectants to effect survival. Threat of coccidiosis is less during hot dry weather and greater in cooler damp weather (Saif *et al*,. 2003). Recovered chickens shed oocysts representing a problem in multi-age operations (Sheriff *et al*,. 2008). Factors contributing to outbreaks of clinical coccidiosis include; litter moisture content exceeding 30% due to ingress of rain or leaking waterier, immunosuppression (Marek's, IBD and Mycotoxins diseases), suboptimal inclusion of anticoccidials or incomplete distribution (poor mixing) in feed and environmental and managemental stress such as overstocking, inoperative feeding systems, inadequate ventilation (Simon, 2005).

2.5 Epidemiology and economic impacts

Coccidiosis remains one of the major disease problems of poultry in spite of advances made in prevention and control through chemotherapy, management and nutrition. *Eimeria tenella* and *E. necatrix* are the most pathogenic species (Getachew, Getachew and Dorchies, 2008). The disease causes high mortality, morbidity and adverse effects on the growth of infected birds (Anjum, 1990). The incidence of cocidiosis in commercial poultry has increased due to higher stocking densities and intensive husbandry practices (Ruzica, 2005). Coccidiosis occurs worldwide and is a major cause of mortality and suboptimal growth and feed

conversion efficiency in immature flocks unless appropriate preventive measures are implemented. The cost of anticoccidial feed additives and treatment is estimated to exceed 400 million US$ annually in all poultry producing areas of the world (Simon, 2005).

Hence, particular vaccines may be designed for rearing standard broilers for up to about 6 weeks or for breeding stock (Williams,1998). Coccidia have been found wherever15 poultry are raised. The spread of this parasitic disease is enhanced by poor bio-safety and management practices. While the *Eimeria* spp. that are known to infect chickens, typically a poultry facility will contain only 1-3 species at a time. In the United States (US) the species *E. acervulina, E. maxima* and *E. tenella* are found most often. However, reports of increased incidence of *E. mitis* and *E. praecox* have been surfacing. There is evidence that protection against the major species of coccidia will allow for the emergence of minor species in a poultry operation. Thus, vaccines using oocyst based or subunit must provide protection against other species that are pathogenic for chickens. In Pakistan whereas in layers and breeders, *E. tenella* showed the highest prevalence, 38.88 and 65% respectively (Khan *et al*,. 2006).

2.6 Signs and symptoms

Infected birds exhibit depression, loss of weight condition, paleness, ruffled feathers, drooping wings, pale and dry flanks and occasional slight whitish soiling around the vent, diarrhea and bloody droppings. Often, a large percentage of the chickens are sick die suddenly before the above symptoms become obvious. The general performance of birds may be affected without the disease causing obvious signs. Post-mortem findings vary depending on the species of *Eimeria* responsible for the infection. In caecal coccidiosis, which is caused by *Eimeria tenella*, the blind gut (caeca) becomes swollen, filled with blood and cheesy plugs. In intestinal coccidiosis, the damage will vary depending on the *Eimeria* species. Other signs include white streaks or spots in the upper part of the intestine, a ballooned and blood-filled intestine, reddish spots, inflammation and dead tissue in the lower part of the small intestines. Chickens that are infected with high levels of coccidia display symptoms such as droopiness and emaciation and may never achieve weight gain equal to their uninfected counterparts**.**

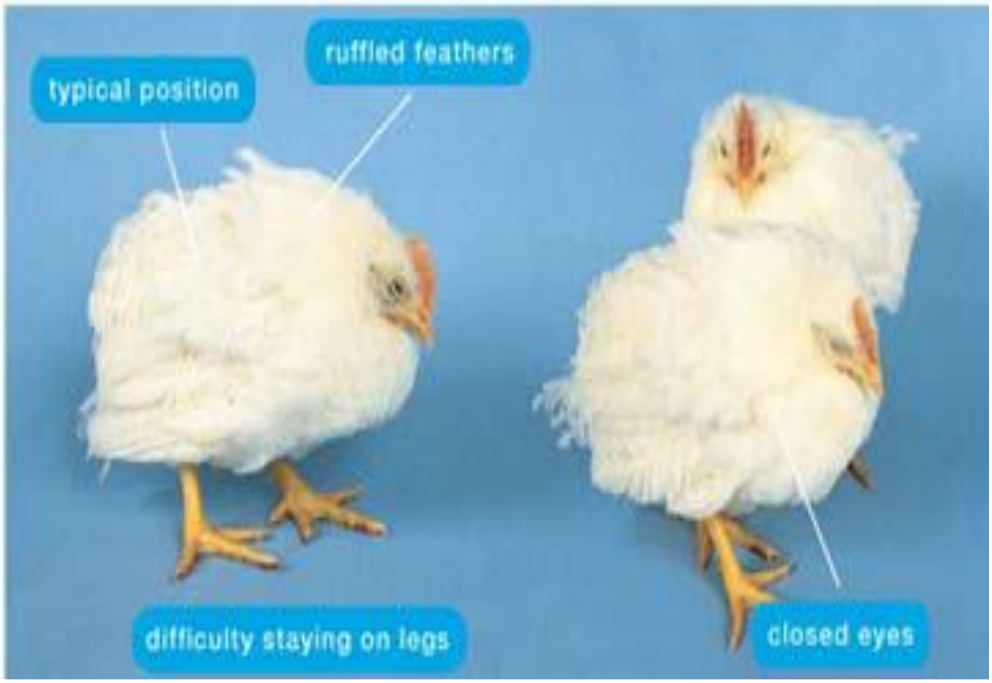

Plate 2.3 Clinical signs of coccidiosis in chicken. (Graat, Ploeger, Henken, Vriesreilingh, Noordhuizen, and Beek,1996).

2.7 Diagnosis

2.7.1 Clinical and postmortem inspection

Eimeria tenella is the best known of poultry coccidia, because of the easily recognizable Lesions and often-spectacular losses it causes in commercial broilers or layer pullets. This species inhabits the caeca, causing a severe disease characterized by bleeding, high morbidity and mortality, lost weight gain, emaciation, loss of skin pigmentation, and other signs. Diagnosis is dependent upon finding caecal lesions with prominent blood and often-firm bloody cores and accompanying clusters of large schizonts and oocysts (Saif *et al*,. 2003).

2.7.2 Faecal examination

Identification of *Eimeria* spp. oocysts in faeces is an easy and cheap way to diagnose many *Eimeria* spp. infections and to get an impression of the infection level, direct smear method and both qualitative and quantitative techniques can be done to faecal sample (Anders and Jorgen 1998).

2.7.3 Direct smear method

Identification of coccidia oocysts is possible by using a direct smear method, where a thin smear of emulsified faeces is examined under a microscope. Direct microscopic examination of intestinal mucosa can only be used in animals, which have been culled or found dead. It can be used to find the intracellular and extracellular stages of coccidia and other protozoa (Anders and Jorgen 1998).

27.4 Qualitative techniques for faecal examinations

A large number of different procedures are available for demonstrating coccidian Oocysts in poultry faeces. The most widely used principle for concentration of parasite oocysts is flotation. Coccidian oocysts have a specific gravity, which is lower than that of plant residues in the faeces, the oocysts may be separated from other faecal particles by mixing the faeces with a fluid (saturated NaCl + glucose) in which the oocysts float, these procedures include test tube flotation and simple flotation (Anders and Jorgen 1998).

2.7.5 Quantitative techniques for faecal examinations

The qualitative flotation techniques, which are used for nematode eggs, cestode eggs and coccidia oocysts, have been elaborated to become quantitative, when the eggs are allowed to float in a special counting chamber, called the McMaster chamber. Many modifications exist, and a Simple McMaster Technique and slightly more elaborated Concentration McMaster (Anders and Jorgen 1998).

2.8 Prevention and control

Oocysts can rapidly build up in the environment whenbirds are overcrowded and use an area for a prolonged period of time. The disease risk increases significantly when these conditions result in oocyst contamination of food and drinking water. In captive situations, good husbandry and sanitation, including continual removal of contaminated feed and litter, can minimize the potential for coccidiosis. Captive birds can be treated with therapeutic agents that control, but that do not eliminate,

the level of infection. Therefore, oocyst shedding by those birds after they are removed from therapy should be considered if they are to be released or mixed with other birds.

2.9 Treatment of coccidiosis

Treatment programs may be used as an alternative to vaccination. A wide selection of drugs (coccidiostats) is available for prevention and treatment. The choice of drug will depend on the type of flock, the coccidian species and the aim of the medication program. Most coccidiostats have withholding periods and medication programs must take this into account. Outbreaks of coccidiosis may occur if the level of coccidiostat in the feed is too low, if the birds are not eating enough or if the coccidiostats is withdrawn too early (before immunity has developed). Lack of vitamins A and K will cause the outbreak to be more severe as will other diseases that reduce the general resistance of the bird.

CHAPTER THREE

3.0 MATERIALS AND METHOD

3.1 Description of Study Area

The study was carried out in poultry farms found within Minna metropolis, Niger state. Minna the capital city of Niger State is located within longitude $6^{o}33E$ and latitude $9^{o}37N$, covering a land area of $88km^2$ with an estimated human population of 1.2 million. It has a tropical climate with mean temperature, relative humidity and rainfall of $30.2^{o}C$, 61% and 1,334mm respectively. The climate presents two distinct seasons: a rainy season (between April and October, with highest mean monthly rainfall in September) and a dry season (between November and March), completely devoid of rains. The duration of the rainy season is approximately 180days. Mean maximum temperature remain high throughout the year, hovering about $37.6^{o}C$. The lowest minimum temperature of $19^{o}C$ occurs usually between December and January, when most part of the state comes under the influence of tropical continental air mass which blows from the north. Its vegetation is typically grass dominated savannah with scattered short tress.(Niger State MAAH Bulletin, 2008).

The study was conducted between June and October 2013 in twelve poultry farms found in different locations within Minna. Sample collection were taken in: Yasmin Farms(A), Abu-Turab farms(B), Talba farm(C), Wachiko farms(D), Shanun poultry(E), Adams poultry(F), Nimatullah poultry(G), Maimasa farms(H), Up-hill poultry(I), Hajiya Ayi farms(J), Zaguru farms(K), and Tunga poultry farm(L).

3.2 Sample collection

Samples were collected daily between the hours of 8.30am and 10.30am. A total of 450 fresh faecal droppings were randomly collected from poultry into sterile universal bottles and 80 carcasses were also collected into polythene bags and transported to the laboratory immediately for processing.

3.3 Data collection

Information was gathered through verbal conversation with the farm owners at the time of sampling which included farm name, address, location, flock age, flock breed and use of coccidiostats in the feed for that flock and previous infection within the year in the farms.

3.4 Laboratory examination

Laboratory examination was by wet mount smears of the faecal droppings as described by Fleck and Moody (1993). While Centrifugation method as described by (Smyth, 1994) was also used for faecal examination.

Post-mortem examination was carried out after dissecting the carcasses and the intestines removed aseptically searched for the presence of lesions. The intestine was scraped and observed under the microscope, according to Jordan and Pattison (1999).

3.4.1 Analysis of faecal samples

3g of faecal samples was weighed and emulsified in 33.9ml formalin (Smyth, 1994; Cheesebrough, 1999). The mixture was strained through double layers of fine mesh cheesecloth and then the strained mixture was transferred to a centrifuge tube and spinned at 1500 to 2000rpm for 2 minutes. The supernatant was discarded and the sediment was re-suspended in 33.9ml formalin and further suspended to 2000rpm for 1 minute. This procedure was repeated two to three times until a clear supernatant was obtained. The sediment was resuspended in 33.9ml of formalin and 11.3ml of ethyl acetate was added. A stopper was inserted and the preparation was shaken vigorously for 30seconds. The stopper was removed and the mixture was centrifuged for 1 minute at 2000rpm. The supernatant was decanted and the sediment was transferred onto a microscope slide by pipetting 0.15ml and covered with a cover slip and viewed at x10 magnification of the light microscope. The oocyst of coccidian were systematically sought for and counted. The number of oocyst was estimated as described by Cheesbrough (1999).

3.4.2 Post mortem examination

The carcasses were disinfected with formalin and the animal was rest on its back on a dissecting board and the limps were pinned to the side with a dissecting needle. A spatula was flamed and used to pierce the skin through the middle. The skin was cut open along the middle and flexed to each side holding them to the board with dissecting needle. A sterile blade was used to cut open the animal to expose the internal organs, and the surface was sterilized with hot spatula before collecting the

specimen. Samples were collected from parts of the intestine, and the caeca and were transferred into sterile universal bottles.

Wet smears of mucosa were prepared from intestinal and caecal scraping for microscopic examination of *Eimeria* spp. *Eimeria* spp.were Identified according on the site of infection and oocysts morphology including size, color presence or absence of micropyle, (Soulsby, 1982).

3.5 Statistical analysis

The data obtained were analyzed to determine significant association between prevalence and age of poultry birds using Chi-Square (x^2); while the significant difference between intensity of infection and prevalence was evaluated using Analysis of Variance (ANOVA). ($p<0.05$).

CHAPTER FOUR

4.0 RESULTS

4.1 Prevalence and intensity of coccidian infection

Table4.1 shows the overall prevalence and geometric mean intensity (GMI) of coccidial infection in the twelve poultry farms surveyed within Minna, Niger State. Out of the total 450 faecal droppings examined, 166(36.8%) were positive for coccidian oocysts, and an overall intensity of 8.12 Oocyst/3g of faeces was recorded. The birds were grouped under the age category of 2-6weeks (young) and greater than 6 weeks (adults), respectively. The result shows that the adult birds had the highest prevalence of infection (37.2%) and intensity of 8.10 Oocyst/3g of faeces and a prevalence of (36.5%) and intensity of 7.25%Oocyst/3g of faeces was recorded in younger birds, However, differences between the prevalence of coccidia by age was not significant ($p<0.05$).

4.2 prevalence and intensity of *Eimeria* species in the post-mortem study

54 out of the 80 carcasses examined were positive to oocyst of Eimeria species. Table 4.2 shows the overall prevalence of (67.5%) and geometric mean intensity of infection (GMI) of 8.33oocyst/3g of faeces in the post mortem studies. Five *Eimeria* species were encountered and isolated during the study with *E. tenella* having the highest prevalence of (42.5%), followed by *E. mitis* with (22.5%), *E. necatrix* (8.8%), *E maxima* and *E. acevulina* had the least prevalence of (7.5%). Similarly *E. tenella* also had the highest intensity although there was no significant difference ($P<0.05$) intensity of infection among the Eimeria species isolated.

TABLE 4.1: Prevalence (%) and intensity (GMI) of coccidial infection by age of poultry birds surveyed within Minna metropolis.

	YOUNG				ADULT				GRAND TOTAL			
Farms	**No exam**	**No +ve**	**prev%**	**GMI**	**No exam**	**No +ve**	**prev%**	**GMI**	**No exam**	**No +ve**	**prev%**	**GMI**
A	20	7	35.0	16.57	25	8	32.0	15.90	45	15	33.3	16.21
B	15	8	53.3	9.29	22	9	40.9	9.22	37	17	45.9	9.26
C	12	4	33.3	14.21	25	8	32.0	16.11	37	12	32.4	15.45
D	20	7	35.0	12.85	23	10	43.5	13.59	43	17	39.5	13.28
E	16	7	43.8	13.44	24	10	41.7	8.75	40	17	42.5	10.44
F	21	6	28.6	11.98	24	13	54.2	14.80	45	19	42.2	13.89
G	23	6	26.1	15.13	20	6	30.0	10.81	43	12	27.9	12.79
H	10	3	30.0	16.35	15	6	40.0	8.27	25	9	36.0	10.38
I	12	3	25.0	4.04	23	5	21.7	3.98	35	8	22.9	4,01
J	18	11	61.1	8.59	12	6	50.0	3.82	30	17	56.6	4.35
K	15	3	20.0	2.29	25	8	32.0	6.96	40	11	27.5	5.15
L	15	7	46.6	10.69	15	5	33.3	17.08	30	12	40.0	13.0
TOTAL	197	72	36.5	7.25	253	94	37.2	8.10	450	166	36.3	8.12

TABLE4.2: Prevalence (%) and Intensity (GMI) of *Eimeria spp* isolated during post-mortem examination from poultry birds in Minna.

			E. tenella			***E. maxima***			***E. necatrix***			***E. acevulina***			***E. mitis***			**Grand Total**		
Farms	**No of carcass**	**No+**	**No+**	**%**	**GMI**	**No+**	**%**	**GMI**	**No+**	**%**	**GMI**	**No+**	**%**	**GMI**	**No+**	**%**	**GMI**	**No+**	**%**	**GMI**
A	3	3	1	2.9	12.99	2	33.3	15.19	-	-	-	-	-	-	2	11.1	27.13	5	55.6	18.57
B	4	4	3	8.8	26.05	-	-	-	1	14.2	15.99	2	33.3	4.69	-	-	-	6	50.0	13.56
C	10	8	7	20.5	13.10	1	16.6	2.99	1	14.2	20.99	1	16.6	4.00	-	-	-	10	33.3	10.77
D	6	6	4	11.7	11.14	-	-	-	3	42.8	3.91	-	-	-	-	-	-	7	58.9	7.12
E	6	3	2	5.8	14.28	-	-	-	1	14.2	1.99	-	-	-	3	16.7	4.04	6	33.3	5.47
F	10	7	7	20.5	22.11	-	-	-	1	14.2	1.99	1	16.6	2.99	2	11.1	6.33	11	36.7	11.80
G	10	6	1	2.9	2.99	-	-	-	-	-	-	-	-	-	5	27.8	3.05	6	60.0	3.04
H	6	6	3	8.8	7.11	2	33.3	2.45	-	-	-	-	-	-	3	16.7	3.91	8	44.4	4.36
I	5	1	1	2.9	12.00	-	-	-	-	-	-	-	-	-	-	-	-	1	20.0	12.00
J	8	4	-	-	-	1	16.6	2.99	-	-	-	1	16.6	1.99	3	16.7	12.76	5	62.5	6.59
K	5	2	2	5.8	4.47	-	-	-	-	-	-	-	-	-	-	-	-	2	40.0	4.47

L	7	4	3	8.8	17.18	-	-	-	-	-	-	1	16.6	11.00	1	5.6	2.99	5	71.4	11.09
Total	80	54 (67.5)	34	42.5	8.45	6	7.5	4.82	7	8.8	5.02	6	7.5	4.24	18	22.5	6.22	66	82.5	8.33

4.3 prevalence (%) of single and mixed infections of *Eimeria* species.

Table 4.3 shows the overall prevalence (%) of (55.6%) single infection and (44.4%) mixed infections of *Eimeria* species isolated from the post-mortem examination. Farm I and K had the highest prevalence of (100%) for single infection while farm J and L had the highest prevalence of (75.0%) in mixed infections.

Table 4.3: prevalence of single and mixed infections of *Eimeria* species in poultry birds in Minna metropolis

Farms	No examined	No infected	Single infection		Mixed infection	
			No	%	No	%
A	3	3	1	33.3	2	66.7
B	4	4	2	50.0	2	50.0
C	10	8	5	62.5	3	37.5
D	6	6	2	33.3	4	66.7
E	6	3	1	33.3	2	66.7
F	10	7	3	42.8	4	57.1
G	10	6	5	83.3	1	16.7
H	6	6	4	66.7	2	33.3
I	5	1	1	100	-	-

J	8	4	1	25.0	3	75.0
K	5	2	2	100	-	-
L	7	4	3	75.0	1	75.0
Total	80	54	30	55.6	24	44.4

4.4 Discussion

An overall prevalence of 36.8% of coccidian infection was recorded in the faecal samples examined and a prevalence rate of 66.3% in the post mortem study carried out on the carcasses; this is in conformity with the work of (Fabiyi, 1984) who reported predominance 30% in a survey of coccidiosis in poultry in Nigeria. This rate of prevalence also indicated that coccidiosis is still an endemic disease of poultry in Nigeria. The relatively high prevalence of infection in the samples examined might be due to the period of the study which coincided with the rainy season that is an enabling factor for coccidian infection in chickens, it may also be due to the management system such as wet litter that encourages oocyst sporulation, contaminated drinkers and feeders, bad ventilation and high stocking density (Ruff, 1993; Al-Quuraishy, Abdel-Baki and Dkhil, 2009). The result of current study revealed that the prevalence of coccidiosis was almost similar in young (2-6weeks) 36.5% and adult (greater than 6 weeks) 37.2% birds, this agreed with report of Julie, (1999) who stated that all ages of poultry are susceptible to infection, but usually resolves itself around 6-8weeks of age. From the result obtained, infected birds associated with age also support the findings of Kaschula (1961) and Soulsby (1973) that younger birds are more susceptible to infection than older birds. Though there is a marked difference in the percentage of infected birds as regards to age, it cannot be considered significant because of the relatively different number of samples collected. In the present study, five species of *Eimeria* were isolated which are the causative agents of coccidiosis in chicken, The result obtained in this work associated with the species of *Eimeria* support the statement of (Khan *et al*., 2006)

which identified the *E. maxima*, *E. tenella*, *E. mitis* and *E. necatrix* from poultry litter. It is also in agreement with other findings in Nigeria, (Majaro, 1983) stated that infection with species of *Eimeria* in poultry has been shown to be due to *E. tenella, E. necatrix, E. acevulina* and *E.brunette*. This result is also in agreement with the statement of Beate and Martin (1999) which stated that the species of *E. acevulina, E.maxima* and *E. tenella* are considered to be the most important to poultry industry. The biological characteristics of coccidian of chickens are well known and variable, and can be identified on the basis of oocyst size (McDougald, & Mattiello, 1997). These results are in agreement with reports from Sweden, France, and Argentina and Jordan (Except *E. brunetti*) suggesting that those species of *Eimeria* have worldwide distribution in most countries where poultry are produced on a commercial basis (Al-Natour and Suleiman, 2002; McDougald and Mattiello, 1997). The finding that most of the infected birds harbored more than one species of *Eimeria* agrees with McDougald (2003) who reported multi species infections of *Eimeria* in chickens with up to six species occurring together.

CHAPTER FIVE

5.0 CONCLUSION AND RECOMMENDATION

5.1 Conclusion

An overall prevalence of (36.8%) and intensity of 8.12Oocyst/3g of coccidial infection was recorded in the 12 poultry farms visited; this shows that coccidiosis is still an important parasitic disease of poultry in Minna, Niger State. The study has shown the presences of pathogenic *Eimeria spp* of *E. tenella, E. acevulina, E. mitis, E. necatrix* and *E. maxima*, both single and multiple infections were also observed in the sampled birds, this shows the level of *Eimeria* infestations among poultry farms in Minna.

5.2 Recommendation

The level of *Eimeria* infestation among poultry farms needs to be monitored especially to establish their pathological influence amongst farms on health of the birds reared in the different farms. Routine check up for oocyst and its identification that will help in detecting subclinical coccidiosis and the associated Eimeria species in poultry farms is highly recommended. This will help both farmers and the clinicians in the appropriate choice of drugs and or vaccine against coccidiosis in the study area. Proper control measures must be taken in the form of strict biosecurity measures, avoiding water spillage, overcrowding and good use of prophylactic anticoccidial programmed. Poultry houses should be disinfected in the intervals between depopulation and restocking. These measures have been established to be effective in reducing the menace of coccidiosis in many countries.

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APPENDIX A

Test of significance in prevalence (%) between young and adult birds in 12 poultry farms found within Minna metropolis.

CONTINGENCY TABLE

	Prevalence % (young)			Prevalence % (Adult)			
farms	Observed (O)	Expected (E)	$\frac{(O-E)^2}{E}$	Observed (O)	Expected (E)	$\frac{(O-E)^2}{E}$	Total
A	35.0	32.9	0.13	32.0	34.0	0.12	67.0
B	53.3	46.4	1.03	40.9	47.8	0.99	94.2
C	33.3	32.2	0.04	32.0	33.2	0.04	65.3
D	35.0	38.7	0.35	43.5	39.8	0.34	78.5
E	43.8	42.1	0.07	41.7	43.4	0.07	85.5
F	28.6	40.8	3.64	54.2	42.0	3.54	82.8
G	26.1	27.6	0.08	30.0	28.5	0.09	56.1
H	30.0	34.5	0.59	40.0	35.5	0.57	70.0
I	25.0	22.9	0.19	21.7	23.7	0.17	46.7
J	61.1	54.7	0.75	50.0	56.4	0.73	111.1
K	20.0	25.6	1.23	32.0	26.4	1.19	52.0
L	46.1	39.3	1.36	33.3	40.6	1.31	79.9
Total	437.8		9.46	451.3		9.16	889.1(G)

Note

Expected value is defined by the expression

$$E = \frac{\text{column total (observed, O, only) x row total (O only)}}{\text{Grand (G) total}}$$

And it is calculated for example

Young (1) $E = \frac{437.8 \times 67.0}{889.1} = 32.9$; young (2) E=……..etc

Adult (1) $E = \frac{451.3 \times 67.0}{889.1} = 34.0$; adult (2) E=…….etc

$$X^2 = \frac{\sum(\text{observed, O, - expected, E})^2}{E}$$

$X^2_{cal} = 18.62$

DF = (column, C,-1)(row, r,-1)= (12-1)(2-1)= 11

$X^2_{tab},0.05,11 = 19.67$

Decision rule

Since $X^2_{cal} < X^2_{tab}$: there is no significant difference in prevalence of coccidian infection among poultry birds examined in the different poultry farms.($p>0.05$).

APPENDIX B

Oocyst of Coccidia isolated from post mortem study on poultry birds within Minna metropolis. – Single factor ANOVA.

Farms	*E. tenelle* GMI	*E.maxima* GMI	*E. necatrix* GMI	*E.acevulina* GMI	*E. mitis* GMI
A	12.99	15.19	-	-	27.13
B	26.05	-	15.99	4.69	-
C	13.10	2.99	20.99	4.00	-
D	11.14	-	3.91	-	-
E	14.28	-	1.99	-	4.04
F	22.11	-	1.99	2.99	6.33
G	2.99	-	-	-	3.05
H	7.11	2.45	-	-	3.91
I	12.00	-	-	-	-
J	-	2.99	-	1.99	12.76
K	4.47	-	-	-	-

L	17.18	-	-	11.00	2.99	
ni	11	4	5	5	7	∑ni=N=32
$\sum_{i=1}^{ni}$∑xii	129.14	23.62	68.49	24.67	60.21	∑(∑xii)=306.13
X	20.83	5.91	13.69	4.93	8.60	
G.S.S	1516.10	139.48	938.18	121.72	517.89 = 3233.37	
$\frac{(\sum_{ni}\sum xii^2)}{ni}$						
Correction factor	$C=\frac{(\sum^{ni}\sum xii)^2}{N} = \frac{(306.13)2}{32} = 2928.61$					
TSS	Tss-C = 4489.82 – 2928.61 = 1561.21					

Group sum of squares $GSS=\frac{\sum(\sum xii)2}{Ni} - C = 3233.37 – 2928.61 = 304.76$

Within sum of squares WSS= TSS – GSS = 1561.21- 304.76 = 1256.45.

Source of variation	Degree of freedom	Mean sum of squares (Mss)	F
Total sum of squares, TSS	N -1 = 32 – 1 = 31		

Group sum of squares, GSS	K – 1 = 5-1 = 4	GSS = 304.76 K–1 4∑£ = 76.19	F=XGSS XWSS
Within sum of squares, WSS	N – K = 32 – 4 =28	WSS = 1256.45 N-K 28 = 44.87	= 76.19 44.87

Fcal= 1.69

Decision rule:

H_o, the geometri mean intensity of oocyst of coccidian isolated from post mortem study in poultry birds within Minna are the same or no significant difference i.e Fcal<Ftab. (P>0.05).

H1: GMI of oocyst are never the same: i.e there was significant difference (p<0.05).

Since Fcal (1.69) < Ftab (2.71), we accept the H_o (Null Hypothesis) i.e. No significant difference in oocyst distributon (p>0.05).

Tabular value of F0.05, DF (4/28) (1) = 2.71

APPENDIX C

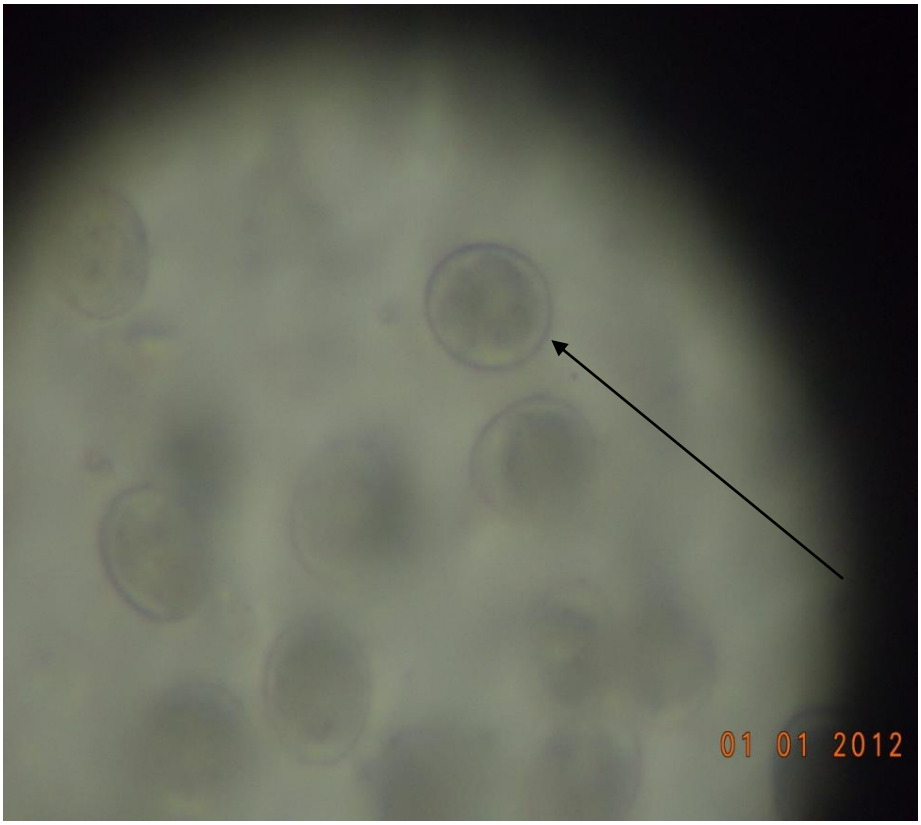

Plate I: oocyst of *Eimeria tenella* isolated from post mortem study

APPENDIX D

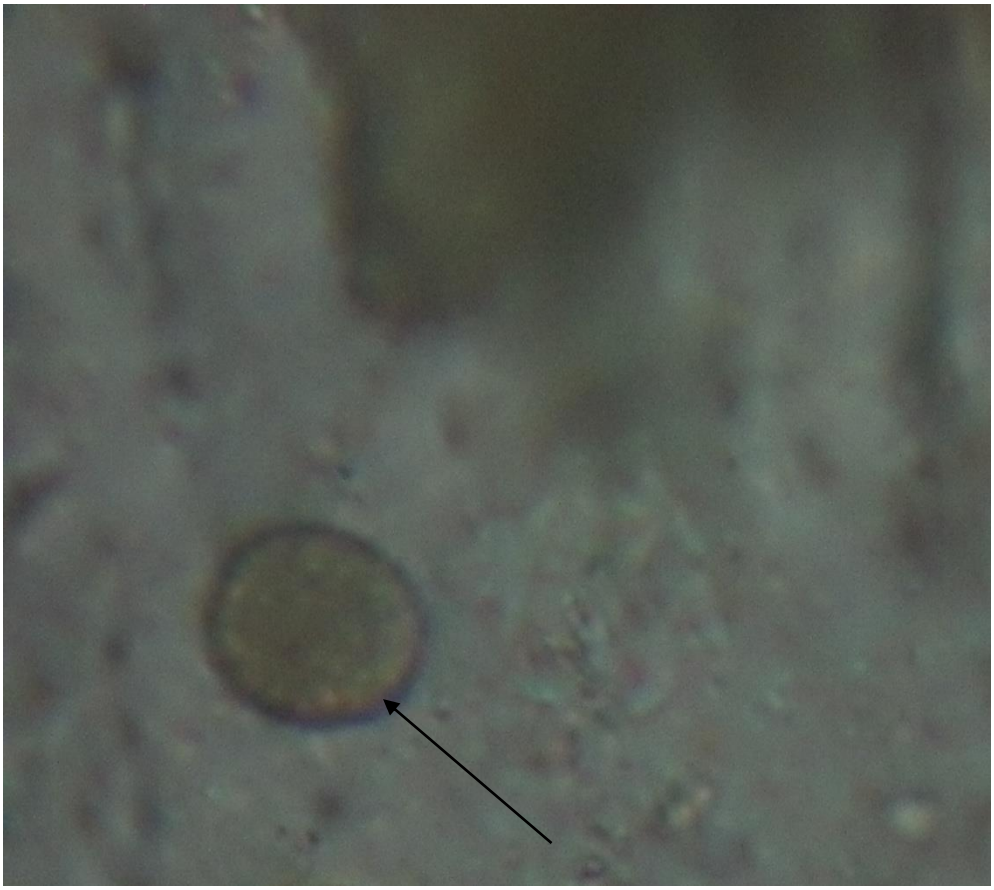

Plate II: Unpopulated oocyst of *Eimeria acevulina* isolated from post mortem study.

APPENDIX E

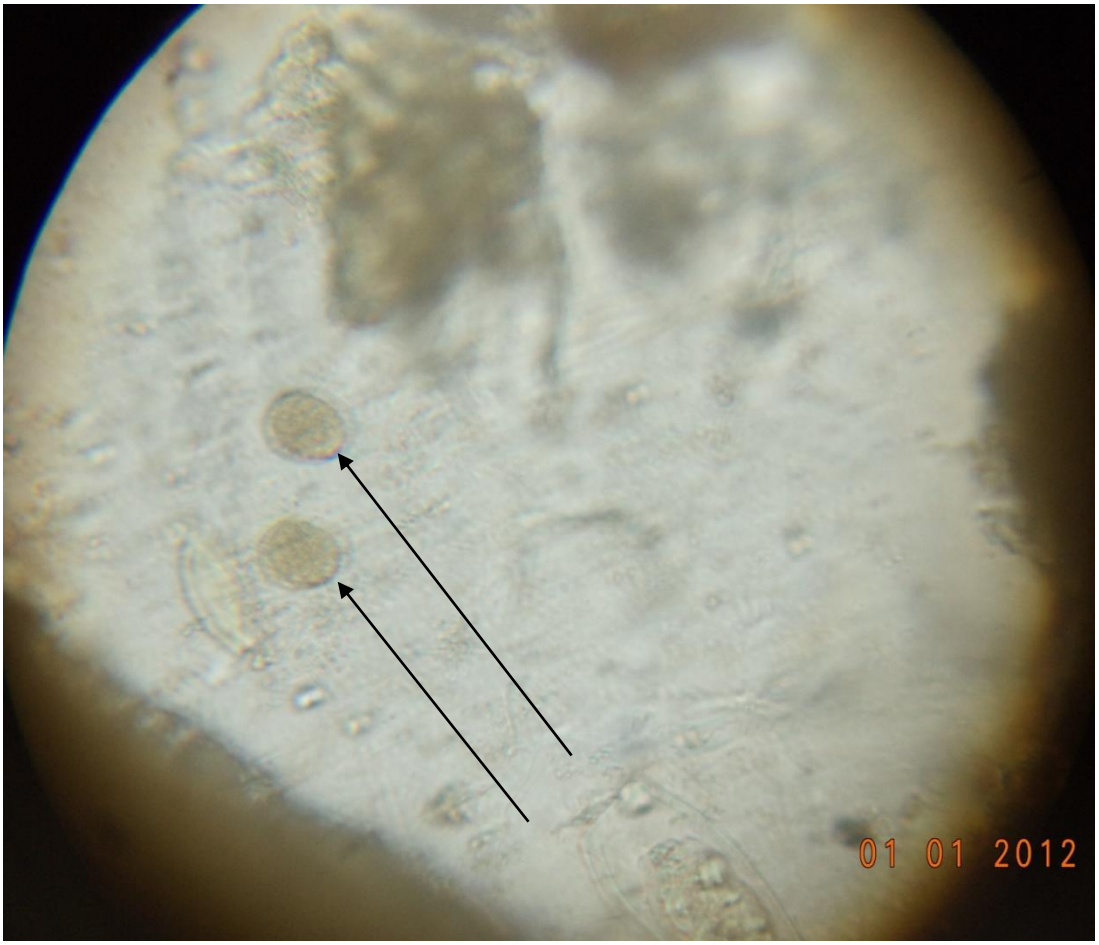

Plate III: oocyst of *Eimeria mitis* iosolated from post mortem study

APPENDIX F

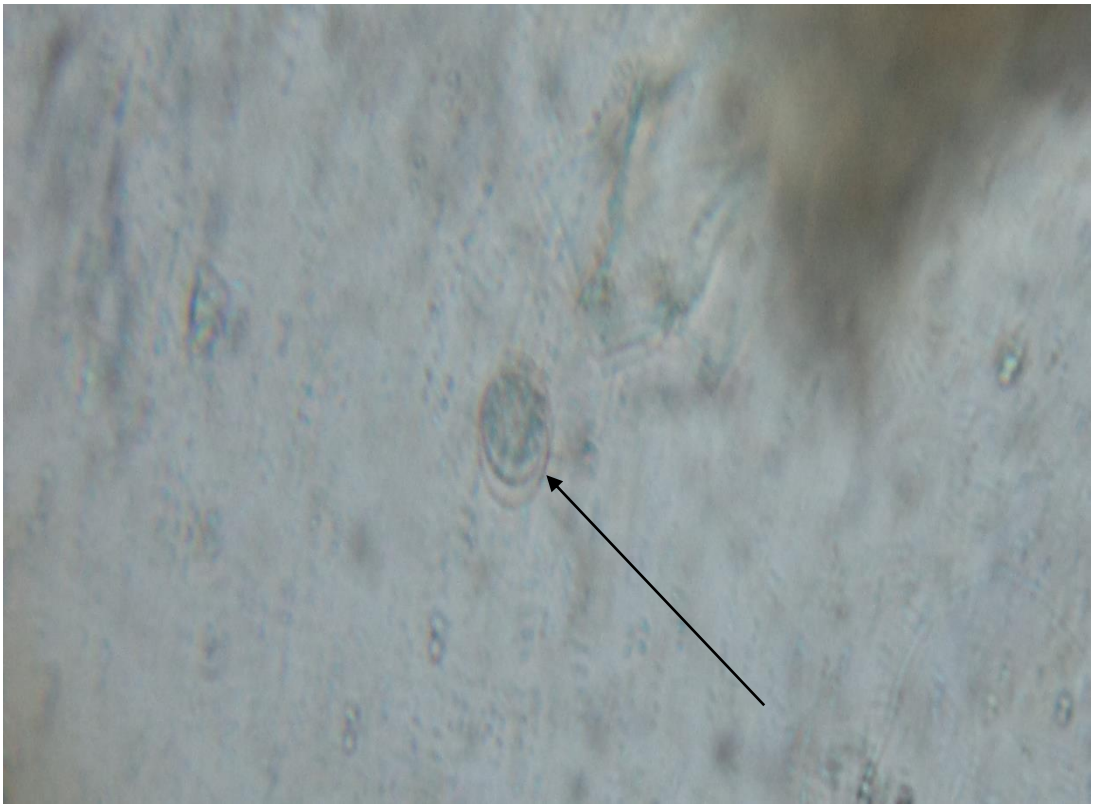

Plate IV; oocyst of *Eimeria necatrix* isolated from post mortem study

APPENDIX G

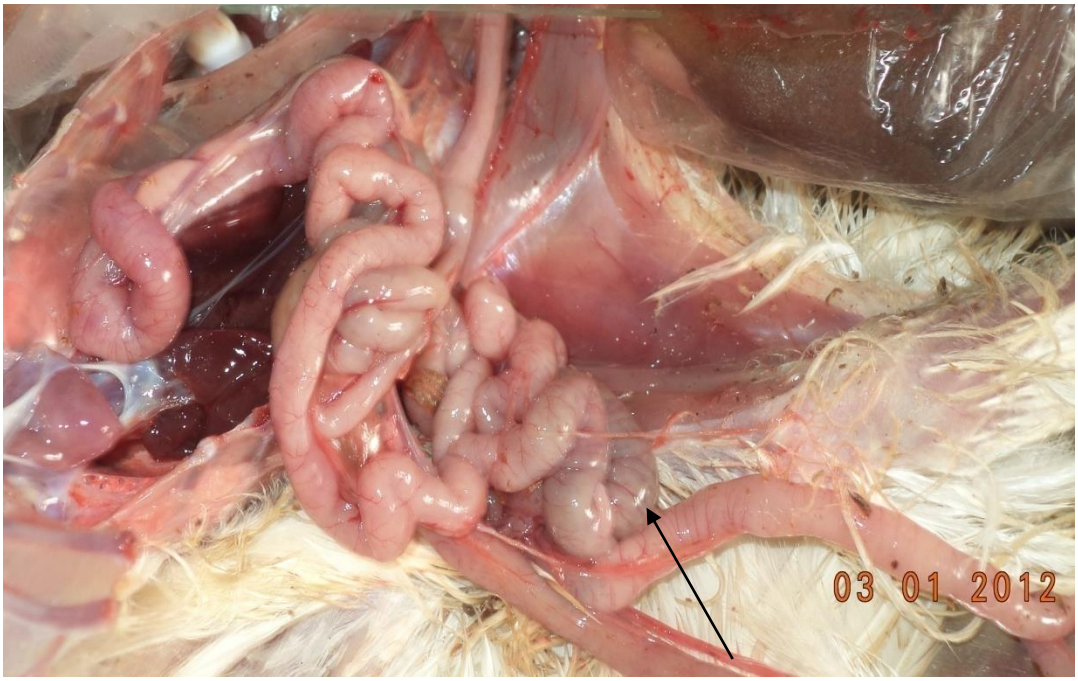

Plate V: Intestine of a young chick showing the presences of haemorrhages.

APPENDIX H

Plate VI: A poultry house

APPENDIX I

Plate VII: An infected bird with symptoms of coccidiosis.