بسم الله الرحمن الرحيم

Sudan University of Science and Technology

College of Graduate studies

Prevalence and Risk Factors of Caprine Coccidiosis in Atbara Locality-River Nile State

نسبه الإصابة وعوامل الخطر لمرض الكوكسيديا في الماعز

في محلية عطبرة - ولاية نهر النيل

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BY

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بسم الله الرحمن الرحيم
قُلْ لَوْ كَانَ الْبَحْرُ مِدَادًا لِكَلِمَاتِ رَبِّي
لَنَفِدَ الْبَحْرُ قَبْلَ أَنْ تَنْفَدَ كَلِمَاتُ رَبِّي وَلَوْ
جِئْنَا بِمِثْلِهِ مَدَدًا
الكهف - الآية 109-

Dedication

To the soul of my father, for my Kind unfailing mother,

To my lovely family, my husband, my daughter and my son, and those who help.

With love and gratitude.

Acknowledgements

Firstly , my thanks are due to Almighty ALLAH for blessing and guidance for this work to be concluded . I would like to express my deep thanks to my Supervisor: Prof . Galal Eldin Mohammed Elazhari for his keen supervision ,direction and continuous interest and constructive criticism in reviewing the dissertation.

My thanks extend to Atbara Clinic and staff for their assistance during the period of sample collection and Atbara Veterinary Research laboratory, for their help in running the bench work. Finally , I express my appreciations to the animal owners in Atbara for Sampling allowance.

Abstract

A descriptive study conducted in Atbara locality in the River Nile State the climate was dry hot(desert annual) in summer and hot wet in autumn rain fall varies from zero to 150 mm. The survey period extend three months and started in May ,June and jully2020.fecal samples were collected from total of 400 goats selected randomly. and were analyzed by two diagnostic tests, Floatation test and direct test technique, the result in two testes were equal, The positive samples was 135 out of 400 goat, The rate of coccidiosis was (33.8%) in tow testes the prevalence of coccidiosis species infection significantly ($p<0.05$) with six risk factors (sex, breed, age, housing, hygiene, season) according to result this study revealed the infection increased in female compared with male, also in small ages than adult and close system than open and in bad management compared with good mangement, high association between breed and the disease (P-value 0.007) the prevalence of coccidiosis in local breed lower than saanin. In conclusion the prevalence of coccidiosis was relatively low compared to others study . Through the field survey, we did not find any animal with symptoms of coccidiosis, but through the laboratory, we found some animals carryinthe parasite. Therefore, additional studies are required to clarify the relationship between the appearance of symptoms and the amount of the parasite in the animal's body.

المستخلص

يهدف هذا البحث إلى تحديد العوامل التي تساعد على انتشار الكوكسيديا في الماعز والتحقيق من الأسباب التي من المحتمل أن تكون قد أثرت على انتشاره. أجريت دراسة وصفية بمحلية عطبرة بولاية نهر النيل حيث كان المناخ جافاً (صحراوي سنوياً) صيفاً وحار ممطر في الخريف تتراوح الأمطار من صفر إلى 150 ملم. امتدت فترة المسح لثلاثة أشهر وبدأت في مايو واستمرت حتى يوليو 2020. تم جمع عينات من البراز من إجمالي 400 ماعز تم اختيارها عشوائياً. وتم تحليلها عن طريق اختبارين تشخيصيين اختبار الطفو وتقنية الاختبار المباشر، وكانت النتيجة في التشخيصين واحدة، وكانت العينات الإيجابية مائة وخمسة وثلاثين من أصل أربعمائة ماعز، وكانت نسبة الكوكسيديا (33.8٪) في التشخيصين. انتشار عدوى أنواع الكوكسيديا معنويا ($P < 0.05$) مع ستة عوامل مؤثرة (الجنس، السلالة، العمر، السكن، النظافة، الموسم) وفقا لنتائج هذه الدراسة أظهرت زيادة الإصابة في الإناث مقارنة بالذكور، أيضاً في الأعمار الصغيرة عن البالغين والنظام المكثف من النظام المفتوح وفي الإدارة السيئة مقارنةً بالأفضل، أظهرت الدراسة وجود ارتباط كبير بين السلالة والمرض (حيث بلغت القيمة الاحتمالية = 0.007) انتشار الكوكسيديا في سلالة السعانين أعلى من المحلية. في الختام، كان انتشار الكوكسيديا منخفضا نسبيا مقارنة بالدراسات الأخرى.

من خلال المسح الميداني لم نعثر على أي حيوان به أعراض الكوكسيديا ولكن من خلال المعمل وجدنا بعض الحيوانات تحمل الطفيل لذلك لابد من دراسات إضافية توضح العلاقة بين ظهور الأعراض وكمية الطفيل في جسم الحيوان.

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Introduction:

The coccidia are intracellular parasites of the protozoan phylum Apicomplexa. They show a wide zoological distribution affecting all vertebrates and higher invertebrates, and within the phylum Chordata, the coccidia are found in all classes including humans (Levine 1973, Rangel *etal*., 2013). It is assumed that many of domestic ruminants became infected with coccidia during their life (Paraud and Chartier, 2012,Viney and Graham, 2013, Hussin,2016, Sharma etal., 2009, Khodakaram. etal.,2013,Vihol et al.,2017). Coccidia belonging to the genus Eimeria and Cryptosporidium show wide distribution, they were reported in a large number of regions and countries (Lassen and Jonis,2009, Paraud and Chartier, 2012, Viney and Graham 2013).

Coccidiosis is diseases of major economic importance in domestic animals. Major losses come from inadequate weight gain , poor productivity, the cost of anticoccidial drugs and from death (Majewska *et al.,* 2000; Jawasreh *etal.*, 2013). The Major routes of transmission of these diseases to animals include contaminated foods, drinking and recreational water (Ruiz, *et al* 2006).

The incidence of coccidiosis diseases varies greatly between areas depending on many factors such as level of a agriculture, pasture, management, grazing habits, climate of the environment, immunological and nutritional status of the host, and the numbers of infected oocysts in the environment (Koko et al, 2003, Ruiz *et al*., 2006.).

Coccidia infect goats, sheep and cattle of various groups but they are most prevalent in young lambs and kids animals in which clinical signs are observed (Paraud and Chartier, 2012, Andrew, 2013).

Coccidiosis, an important stress induced enteric protozoan parasitic infection affecting several animal species including small ruminants worldwide (Daugschies and Najdrowski, 2005). It is caused by different species of enteropathogenic Eimeria, an apicomplexan protozoan which develops in the small and large intestine, causing more pathogenic effects in young animals.

Though the disease is present in clinical form mostly in younger animals; however, adults may also be affected severely, at times (Taylor *et al*. 2007).

Coccidiosis is a gastrointestinal disease of farm animals. it is costly to livestock farmers due to high treatment expenses and a high mortality rate among animals after contracting this disease. Coccidiosis is caused by Eimeria spp, also called Coccidia spp, and like E. arloingi, E. christenseni, and E. ovinoidalis, is highly pathogenic in kids. Eimeria are protozoa, a unicellular microorganism naturally found in the soil. Coccidia are host-specific, which means that Coccidia of cattle and chicken are specific to these species and do not cause disease in goats or vice versa.(Usal et al, 2016)

However, Coccidia of goats can affect sheep. There are numerous species of Coccidia that are naturally found in the goat intestine. The infection occurs naturally by ingesting occystes, a resistant form of the parasite, when grazing. Another form of infection is acquired with poor goat management practices that occur when feed and water supplies are contaminated with goat feces.

Although the infection can occur in any goat herd raised under semi and intensive management practices, it is most frequently observed in kids 2 to 4 weeks post weaning. (Knox and Steel, 1996)

Goats are born without Coccidia in the intestine. The infection occurs by ingesting the pathogenic sporulated oocyst (sporulated is a form of resistance of the Coccidia). Oocysts can be found in the water or in feed supplies contaminated with feces. Once ingested, oocysts penetrate the cells lining the intestine where they go through several stages of development and cause inflammation and destruction of intestinal cells. Occasionally, oocysts reach a mature stage of development. They can multiply, generating thousands of new oocysts that will be passed through the feces to the environment in about 2 to 3 weeks. However, they must undergo a period of sporulation in order to be contagious to another host. This sporulation period occurs when there is adequate moisture and warm temperatures. Unfortunately, sporulated forms

are highly resistant to ordinary disinfectants. Direct sunlight is the best disinfectant; therefore, goat housing should be dry and exposed to sunlight.

Infective oocysts can be found in a contaminated environment for a long period of time. Stress is the predisposing factor in kids during the post weaning period. Animals may die suddenly during this phase and without any warning. Outbreaks can occur during stressful conditions such as after shipping or when animals are relocated. Outbreaks can also occur during sudden weather changes, after a change in concentrated feed practices, when animals are recovering from a disease, or in worm burden cases.

Although coccidiosis can occur year around, a higher incidence occurs during post weaning. It is common to find animals naturally resistant to coccidiosis. (The Merck Veterinary Manual. 2008).

Coccidial infection is widespread in goat and sheep, and coccidiosis can be a substantial problem in the young of both species(Gregory.,etal 1980)

Objectives of the Study:

1. To determine the prevalence of coccidiosis in caprine in Atbara locality.
2. To investigate some risk factors that increasing coccidiosis infestation .
3. To obtain additional data on coccidiosis in Atbara locality.

Chapter One

Literature Review

Chapter One

Literature Review

1.1 Coccidian of goat and sheep

Coccidiosis are types of the protistan phylum Apicomplexa, subclass Coccidiasina, intracellular parasites, characterized by a typical apical complex of organelles at some stage of the life cycle at one end of the organism. Members of the genus Eimeria and Isospora are homoxenous with sexual and asexual development occur in a single host. The cause of coccidiosis in goat is complicated by the morphologic similarity of the coccidia infecting these species (Gelberg, 2012; Uzal, *et al*., 2016).

Most species of coccidian look similarly in goats and sheep, but do not cross-infect. Only three species including E. pallida, E. caprovina and E. punctata may occur in both goat and sheep, although the validity of E. punctata as a species is questioned (Uzal *et al*., 2016).

fourteen species of coccidia have been described in goat, of which E. ahsata, E. ovinoidalis, and E. bakuensis are considered serious pathogens (Reeg *et al*., 2005).

The economic cost of coccidiosis is considerable, in terms of low growth performance, decrease in productivity, mortality, morbidity, clinical and subclinical disease, and the cost of prevention and treatment. Clinical coccidiosis occurs mainly in 4-6-month old kids and lambs and has a higher prevalence under conditions of intensive husbandry. Stress factors such as weaning, dietary changes, inclement weather, poor nutrition and sanitation, overcrowding, travel and regrouping predispose animals to the disease (Gul, 2007; Chartier and Paraud, 2012; Uzal *et al*., 2016).

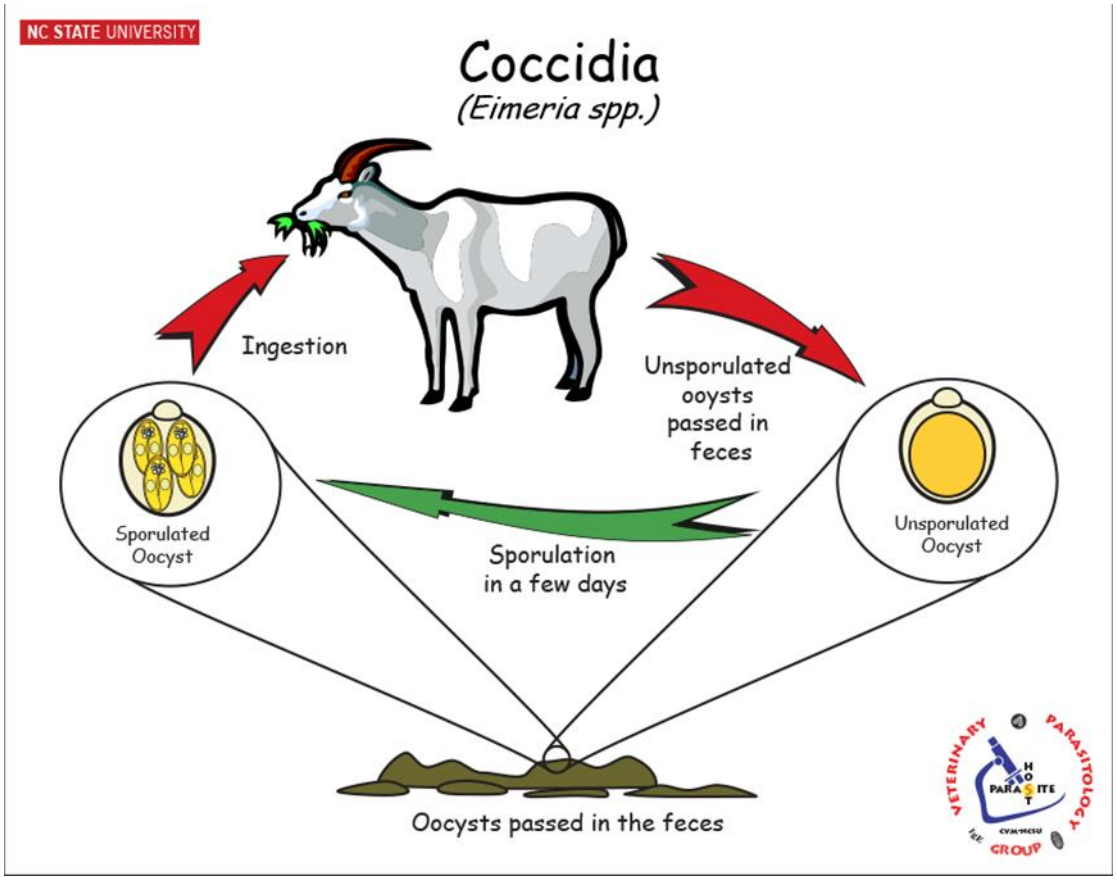
Coccidia can invade and destroy intestinal cells of the hosts, causing anemia, electrolyte loss and poor absorption of nutrients. The most common clinical sign of infection is diarrhea, and affected goat can show a rough hair coat, poor weight gain and weakness (Wang *et al.*, 2010).

Pathologic lesions vary from proliferative in goat and sheep to necrotic-hemorrhagic in cattle, avian species, dogs and cat (Gelberg, 2012; Uzal *et al*., 2016).

1.2. Epidemiology:

Coccidia are highly prolific because each sporulated oocyst has the potential to produce 23 million oocysts during the endogenous phase after just 21 days (Dendrickson, 2017). This ability leads to high levels of environmental contamination. Sporulated oocysts are resistant in the environment and can survive for weeks to months, especially in favorable conditions of moderate heat and moisture.

Buildup of high levels of contamination are most common in areas where animals congregate or are crowded and feces are more concentrated in the environment. Feedlots, drylots, and barns are common types of housing associated with coccidiosis. It can also be a problem in heavily stocked pastures, especially around watering and feeding area (Ruiz, Gonzalez, *et al*.,) Healthy ruminants are generally immune to disease by 1 year of age but serve as a reservoir to younger animals. The magnitude of infection, clinical signs, and oocyte shedding are affected by the species of Eimeria involved, level of environmental exposure, and animal immunity. Age, other stressors (weaning, weather, transportation, other diseases, and so forth), nutrition, and genetic susceptibility all contribute to animal immunity and susceptibility to coccidiosis (Morien, *et al*., 2002).



Scheme (1.1): Transmition of coccidiosis in goat

1.3. Geographical distribution:

Coccidia of small ruminants are present worldwide and it seems difficult to say that there is any particular geographical distribution for one or the other species of coccidia. Although, a dozen species of coccidia are found in each of goats and sheep; of these a few are potentially highly pathogenic, whereas several others have little pathogenic effect under normal circumstances.

In the literature review of mostly recent decade, there are various reports on prevalence rate of Eimeria spp in goats(Gul,2007).

According to these studies, the variation in prevalence and distribution of coccidiosis may be attributed to the differences in management and hygienic conditions, temperature, agro ecology, climate, weather conditions, the immune state of the host, sample size, sampling period and breed susceptibility to coccidia in different areas(Degers,Gul.,etal 2003).

Table(1.1): Eimeria species of goats (Razavi and Hassanvand**,**2007)

Animals	Eimeria species	References
Goats	*E. arloingi, E. ninakohlyakimovae, E. christenseni, E. caprovina, E. caprina, E. hick, E.apsheronica, E. alijevi, E. jolchijevi, E. africiensis, E. punctate, E. kocharli, E. capralis, E.masseyensis, E. charlestoni, E. minasensis*	(Levine, 1985), (Silva, 1998), (Soe & Pomroy, et al., 1992)

Table(1.2): Prevalence and Geographical distribution of Eimeria spp in goats (Keirandish**,** Nourollahetal etal., 2014)

Country	Goat (Species/ Prevalence)	References
Southern Portugal	*E. ninakohlyakimovae(88%), E. arloingi(85%), E. alijevi(63%), E. caprovina(63%)*	(Silva & Vila, 2014)
Czech Republic	*E. arloingi (84%), E. hirci (63%), E. ninakohlyakimovae(56%), E. christenseni (55%)*	(Koudela & Bokova, 1998)
Poland	*E. arloingi (80%), E. christenseni (70%), E. ninakohlyakimovae (40%), E. caprina (20%)*	(Balick & Ramisz, 1999)
Gran Canary (Spain)	*E. ninakohlyakimovae (30.0%), E. arloingi (28.6%), E. alijevi (20.5%), E. caprina (9.1%)*	(Ruiz & Gonzalez, 2006)
Nigeria	*E. jolchievi (24%), E. pallida (22%), E. arloingi (18%), E. apsheronica (16%)*	(Woji & Little, 1994)
Zimbabwe	*E. alijevi (99%), ninakohlyakimovae, (99%), E. hirci, (83.5%), E. arloingi, (80.6%)*	(Chhabra & Pandey, 1991)
Tanzania	*E. alijevi (63%), E. arloingi (55%), E. caprina (26%), E. ninakohlyakimovae (26%)*	(Kambarage, Kusiluk, 1996)
South Africa	*E. arloingi (97.47%), E. hirci (84.34%), E. caprovina (61.11%), E. ninakohlyakimovae (45.95%)*	(Harper, Penzhorn, et al., 1999)
Southeastern Iran	*E. arloingi (68.26 %), E. christenseni (50.9 %), E. ninakohlyakimovae (41.8 %), E. caprina (31.7 %)*	(Knox & Steel, 1996)
Sri Lanka	*E. ninakohlyakimovae (31%), E. alijevi (29%), E. arloingi (21%), E. christenseni (7%)*	(Faizal, Raja Pakse, et al., 2001)
Malaysia	*E. arloingi (71%), E. ninakohlyakimovae (67%) E. christenseni (63%) and E. alijevi (61%)*	(Abo-Shehada & Abo-Farieha, 2003)
North of Jordan	*E. carping (13.5%), E. ninakohlyakimovae(12.5%), E. arloingi (11%), E. apsheronica (10%)*	(Wang & Chena, 2010)
Northeastern China	*E. christenseni (78.3%), E. alijevi (73.7%), E. caprina (62.3%), E. arloingi (44.6%)*	(Degers, Gul, et al., 2003)
Turkey (Van province)	*E. arloingi (40.9%), E. christensini (34.3%), E. alijevi (32.6%), E. pallida (31.0%)*	(Gul, 2007)
Turkey (Igdir province)	*E. arloingi (47.43%), E. christenseni (45.14%), E. ninakohlyakimovae (36%), E. alijevi (26.85%)*	(Kambarage, Kusiluka, 1996)

1.4. Clinical signs

Coccidia can invade and destroy intestinal cells of the hosts, causing anemia, electrolyte loss and poor absorption of nutrients. The most common clinical sign of infection is diarrhea, and affected goat and sheep can show a rough hair coat, poor weight gain and weakness (Wang *et al.*, 2010).

Coccidiosis occurs mainly in 4-6-month old kids and lambs an age, genetic susceptibility, physical condition, the degree of immunity and stress factors, such as inclement weather, weaning, dietary changes, traveling, and regrouping have important roles in clinical coccidiosis (Cox, 1998; Gul, 2007).

Review of experimental studies shows that the advanced clinical signs of the infected lambs or kids have no prominent difference with the used inoculated doses, but the beginning and the severity of symptoms are dose dependent (Dai *et al.*, 2006; Hashemnia *et al.*, 2012). The main symptom is diarrhea, which can be hemorrhagic in goat. The change in the feces appearance is coincided with the first appearance of oocysts which varies according to the prepatent period of each species. In many cases, the feces are watery with clumps of mucus and color changes from brown to yellow or dark tarry (Foreyt, 1990; Koudela and Bokova, 1998; Uzal *et al.*, 2016).

During the period of diarrhea, the affected animals rapidly show marked dehydration, paleness of conjunctiva, listlessness, abdominal pain, tenesmus, and weight loss. The general condition of the animal is worsened because of decreased appetite. In certain conditions, coccidiosis can be characterized by sudden mortality without preceding digestive signs, in particular amongst young animals between 2 and 4 months old. Impairment of growth is the main sign of subclinical coccidiosis (Chartier and Paraud, 2012).

1.5. Pathology:

Gross lesions of coccidiosis are variable by host species, parasite species, and intestinal location. Lesions vary from proliferative in goat and sheep to necrotic-hemorrhagic in cattle, avian species, dogs and cat (Gelberg, 2012; Uzal *et al*., 2016). In small ruminants, coccidial-induced enterocyte hyperplasia results in nodule formation and thickening of the intestinal wall can cause reduction in food absorption, emaciation, serous atrophy of fat, diarrhea, and dehydration. Gross lesions, even in heavily infested animals, may be minimal to absent. The minimal to moderate changes are observed as

thickening of the intestinal mucosa associated with a few scattered whitish 1-2 mm in diameter non-pedunculated plaques or nodules. The large schizonts of some species are sometimes grossly visible as well.

The most common lesions of clinical coccidiosis in young goat and sheep are scattered whitish non-pedunculated plaques to nodules on mucosa of small intestines. In advanced cases, multifocal to coalescent progressive thickening, folding or corrugating to pseudoadenomatosis of the intestinal mucosa associated with numerous well-raised whitish nodules are seen. These nodules, sometimes pedunculate, and about 0.3-1.5 cm in diameter are comprised of hypertrophic crypt-villus units, in which virtually every epithelial cell is infected by mainly gametocytic stages of coccidia, which, in goat, are probably E. bakuensis and E. ahsahta and in goats, is E. arloingi (Gregory and Catchpole, 1990; Khodakaram-Tafti and Mansourian, 2008; Hashemnia *et al*., 2012; Gelberg, 2012; Hashemnia et al., 2015; Uzal *et al*., 2016).

The term pseudo adenomatous has been used to describe the polypoid lesions and the oocyst patches or plaques in coccidiosis of small ruminants and may be the result of mitogenic stimuli from progamonts of the parasite (Uzal *et al*., 2016). In some cases, a particular pattern of projections are visible from the serosa of affected intestines. Recently, the term of cerebriform or gyrate pattern was used as a diagnostic gross lesion of advanced naturally occurring coccidiosis in kids and lambs (Khodakaram-Tafti and Mansourian, 2008).

1.6. Host Specificity and Immunity:

The oocyst breaks down and the sporocysts are released. The sporozoites penetrate into an epithelial cell of the small intestine to transform into a first generation schizont. The schizont produce motile merozoites, which may initiate another generation of schizonts in other intestinal cells or become gamont, gametes and then non sporulated oocysts that are released with the fecal matter. The second generation schizogony occurs usually in the large intestines followed by the release of another generation of merozoites,

which invade epithelial cells and produce the sexual stages, the macrogametocyte and the microgametocyte. The second generation schizogony and fertilization of the macrogametocyte by the microgametocyte (gametogony) are the stages of the life cycle that cause functional and structural lesions of the large intestine (Foreyt, 1990).

Coccidia of domestic animals are relatively host, organ and tissue specific. Rarely does one species of *Eimeria* complete an infectious cycle in more than one host species. Exceptions have been noted under experimental conditions. The underlying mechanisms of host specificity are not well understood, but most likely include genetic, nutritional/biochemical, and immune factors. Virulence reflects a number of factors such as the location and type of cell infected by various stages of the organism, the function of infected cells, and the degree of host reaction stimulated by infection (Uzal *et al.*, 2016).

Immune responses to coccidia are extremely complex and different effector mechanisms may be involved depending on the stage of parasite development, prior host exposure to parasites, the nutritional status of infected animals, and the genetic makeup of the host.

Life cycle of Eimeria comprises several stages including intracellular, extracellular, asexual, and sexual, hence the host immunity is complex and involves many facets of non-specific and specific immunity, the latter encompassing both cellular and humoral immune mechanisms (Lillehoj, 1998). Non-specific factors include physical barriers, phagocytes and leukocytes, and the complement system. Specific host immunity is mediated by lymphocytes and their secretions such as antibodies and cytokines (Yun et al., 2000).

Specific immunity to each coccidial species develops after infection, so that young animals exposed for the first time are often more susceptible to a severe infection and clinical disease than other animals. The immunity

induced by the first infection seems to protect most kids from reinfections later in the grazing season (Catchpole *et al*., 1993).

It seems that, the age of animals plays an important role in the immune responses. Very young kids are relatively resistant to infection with a mixture of pathogenic species of coccidia, but susceptibility increases progressively up to at least 4 weeks of age. Young animal inoculated at 4-6 weeks of age, develop severe diarrhea, whereas the same inoculum given at 1 day of age causes no clinical disease (Gregory and Catchpole, 1989).

In the natural host, immunity is species specific such that kids immune to one species of Eimeria are nevertheless susceptible to others. Additionally, different species of Eimeria demonstrate different tissue and organ specificity in the infected host (Yun *et al*., 2000).

Immune inflammatory reactions may be incited by coccidial infection. In experimental systems, resistance to coccidial infection is thymus dependent, and is largely mediated by T-cell-promoted intracellular killing directed mainly against asexual stages in the life cycle. In mammals, acute inflammatory reactions in intestinal coccidiosis are most commonly associated with heavy infection and destruction of cells by the sexual stages and oocysts, rather than in response to asexual stages. Generally, cellular immunity is more important in resistance against reinfection than humoral immunity (Uzal *et al*., 2016).

1.7. Diagnosis:

Large schizonts are often encountered incidentally in submucosal lymphatics, or in the subcortical or medullary sinusoids of mesenteric lymph nodes in goat and sheep infected with E. apsheronica, E. ninakohlyakimovae and E. arloingi. Sometimes they may be visible grossly in these locations as pinpoint white foci. The presence of schizonts or other stages in lymph nodes are the result from establishment of sporozoites or merozoites migrating from the lacteals into the lymphatic drainage in early infection (Dai *et al*., 2006; Uzal *et al*., 2016).

The diagnosis of coccidiosis depends on the clinical findings (diarrhea, dehydration, and progressive emaciation), the presence of large numbers of oocysts in the feces, appropriate signalment and intestinal lesions at necropsy and history (Kusiluka, and Kambarage, 1996). Diagnosis is not easy because clinically normal goat often shed coccidial oocyst. Furthermore, there may be considerable intestinal damage and scouring even before oocysts appear in the faeces.

The developmental stages of Eimeria spp.in the intestinal cells can be demonstrated in Giemsa-stained intestinal smears or scrapings and, in haematoxylin eosin stained histological sections. The demonstration of various developmental stages of Eimeria spp. and the denudation of the intestinal epithelium in dead or sacrificed animals is considered to be a positive diagnosis for coccidiosis. Postmortem examination is the best means of confirming the diagnosis, providing is performed immediately after death. The intestines may be slightly reddened and full of fluid, but the most characteristic feature of coccidial infection is the presence of multiple small pale nodule 1-2 mm in diameter in the intestinal wall (Kusiluka and Kambarage, 1996;Chartier and Paraud, 2012, Gelberg, 2012; Uzal *et al*., 2016).

Microscopic analysis of fecal samples makes it possible to quantify the rate of infestation by counting the number of oocytes excreted per gram of feces (OPG). Although this method can support the diagnosis, but it is usually not very reliable because most animals will excrete the oocysts in the absence of the disease and acute coccidiosis may occur before the oocysts are demonstrable in faeces. Furthermore, Eimeria species vary in their pathogenicity. However, the presence of very high numbers of oocysts in faeces together with clinical signs may be highly suggestive of the disease (Radostits *et al*., 2007).

Fecal oocyst counts on about 10 kids are needed in order to obtain a correct estimation of the average excretion of a group of animals. Despite the

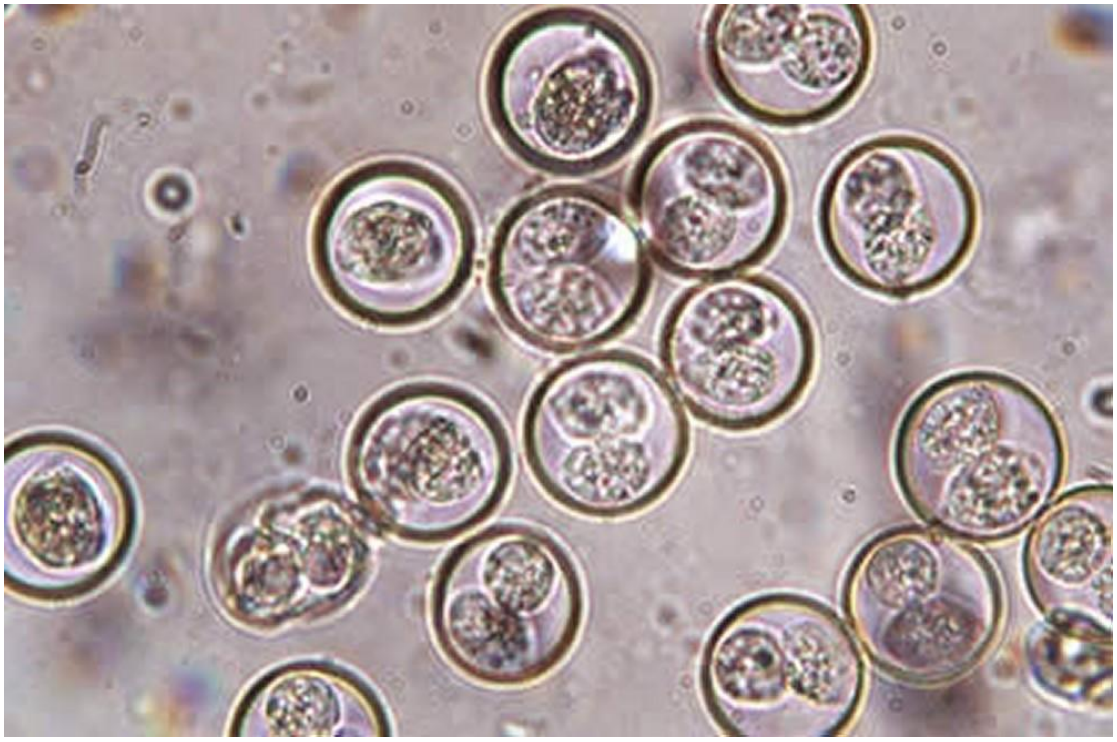
general relationship between clinical coccidiosis and high excretion of oocysts, a clear-cut threshold for coproscopical values is difficult to assess.

Identification of Eimeria species has to be made on the basis of the morphological criteria of the oocysts, most often after sporulation (fecal matter at room temperature for 2 or 3 days, or after dilution in 2% bichromate potassium and incubation at 25◦C). According to the species, there are variations in the size of the oocyst, its wall, the presence or absence of a membrane surrounding the wall, the appearance of the sporocysts (4 per oocyst) and the sporozoites (2 per sporocyst, comma or sausage shaped), polar cap, micropyle, color, oocystal and sporocystal residues, etc. (Eckert *et al*., 1995).

Accurate diagnosis of the causative agent of coccidia infection is very important and helps us to have a better understanding of the biology and life cycle of this parasite. The traditional methods are not only very subjective and time consuming but are also unreliable since the different species have overlapping properties (Haug *et al*., 2007). Furthermore, morphological observations combined with fecal examination are very labor-intensive and require skillful technique. It is essential to develop a more rapid, convenient, and accurate diagnostic method. Thus, molecular tools have recently been proven useful for the species identification or classification of this genus to overcome the limitations of traditional methods (Woods *et al.*, 2000) .
and have furthermore demonstrated the phylogenetic position of each Eimeria spp. and host–parasite relationship by forming some phylogenetic clades (Zhao *et al*., 2001; Lee *et al*., 2001; Morrison *et al*., 2004).
Recently, 18S rDNA gene sequences and the internal transcribed spacer1 (ITS1) region derived from the ribosomal RNA (rRNA) genes have been used effectively to define the genetic characterization and phylogenetic analysis of some Eimeria species in goats and cattle (Kawahara *et al.*, 2010; Khodakaram-Tafti *et al.*, 2013).

The use of acute phase proteins (APPs) and inflammatory mediators such as haptoglobin (Hp), serum amyloid A (SAA), TNF-α and IFN-γ have

been suggested as a non-specific marker for disease as well as for monitoring the response to treatment in coccidiosis. Measuring APPs along with clinical signs and oocysts per gram (OPG) can be useful for providing information about the stages of clinical and subclinical coccidiosis (Hashemnia *et al.*, 2011). Mucosal scrapings or tissue sections of mucosa containing large numbers of asexual and gametogenous coccidial forms in association with diarrhea and perhaps some hemorrhage into the intestine, support the diagnosis, in the absence of other syndromes such as gastrointestinal helminthosis (Uzal *et al.*, 2016). Accurate diagnosis of clinical and subclinical infections and prompt treatment, control and prevention is quite necessary for preventing of great economical losses of this disease in the herds.



Scheme (1.2) Coccidiosis under microscope (Merk Veternary Manual)

1.8. Treatment:

It must be done as early as possible and concern the whole group of animals(age, paddock) as animals showing no obvious signs may contaminate the environment. Treatment has to be associated with a move of the animals to a another environment (Chartier & Paraud, 2012).

Coccidiosis can be treated using decoquinate (0.5 mg/ kg BW) and lasalocid at a dose of 25-100mg/kg feed from weaning until market (Radostits, *et al*., 2007).

Sulfonamides at dosage rates of 25 to 35 mg/kg BW for at least 15 days are effective against coccidiosis in small ruminants. A combination of chlortetracycline and sulfonamide has provided protection in kids. Amprolium in feed is also used to treat the disease in goats (100 mg/kg BW for 21 days) and sheep (50 mg/kg BW for 21 days) (Radostits, *et al*., 2007).

Other drugs include monensin (20 and 16 g/ton of feed for sheep and goats, respectively), toltrazuril (20 mg/kg BW as a single oral dose) and dicrlazuril (2 mg/ kg BW as a double oral dose) (Radostits, 2007; Gelberg, 2012; Ruiz, Guedes, *et al*., 2012).

Decoquinate, toltrazuril and diclazuril are molecules which act on the whole cycle of the coccidia and this allows both a curative and a preventative effect (Radostits, 2007; Taylor & Marshal, 2003).

1.9. Preventive Measures:

The control of cocccidiosis relies on management practices (Ruizetal.,2012).

• Improve hygiene of facilities, pastures, pens, and feeding and water sources. Avoid moist areas without direct sunlight, such as under feed bunks and near water troughs.

• Avoid crowded pens and pastures.

• Quarantine before introducing new animals to existing herd.

• Minimize weaning stress. If needed, creep feed to adjust the kids to a new diet prior to weaning (Harper and Penzhorn,1999).

• Predict possible outbreaks during severe weather and post weaning (Brain, 2011)

• Add coccidiostat to concentrate as a feed additive. Coccidiostat suppresses the full development of the life cycle of the Coccidia and allows immunity to develop. Monensin acts as a coccidiostat and can enhance production performance (Cambpella,2008).

• As an alternative, Decoxquinate (Decox) can be used as a feed additive in the feeding or mineral salt in a concentration. Producers must follow feeding instructions as recommended by the feeding company on the feed tag. However, the herd manager should assess his/her feeding plan to determine if the regular feeding program will result in goats consuming enough feed to get the recommended daily dosage. You do not want to overfeed just to meet the daily mediation rate(Merck and Company incorporated,2008).

Good nutrition is essential to maintaining high level of immunity in the flock or herd. Balanced ration, with proper vitamin and mineral supplementation, should be fed. Colostrum will give immunity to coccidiosis for the first several weeks of neonates life. (Catchpole etal.,1993)

Chapter Two

Material and Methods

Chapter Two

Material and Methods

2.1 Study area:

River Nile State lies approximately between 22-35 longitude, east 16-22 latitude North ,and extend from Elsabuloga near River Nil south toward Bayoda desert to the Northern state North (Mohammed *et al*.,1996). Atbara is a town located in Nahr EL Neel State in North Eastern Sudan (Atbara, 2007). It is located at the junction of the Nile and Atbara rivers .It is an important railway junction and rail road manufacturing centre ,and most employment in Atbara is related to the rail lines .It is known as the Railway city ,and The National Railway Company's Headquarters are actually located in Atbara. The city is also a home to one of Sudan's largest cement factories(Atbara cement Corporation). The state surface area is about 124,000Km2and the climate is that of a dry hot desert annual rainfall varies from zero in Northern part to150 mm in the Southern .Trees and grasses such as Acacia, Cherenbergiana and Aristida spp. represent natural pastures beside the irrigation land around the river bank which is considerably small (Mohammed *et al* .,1996).

2.2 Study design:

Animal were selected randomly to determine the prevalence of coccidiosis spp in goats based on coprological examination ..

Across sectional study was conducted on Veterinary Hospital, Goats Farms located in (Elamn Elgzay) at Atbara City, Goats reared in houses.

Prevalence of goat was calculated by the following equation (Faroog et al., 2012).

Prevalence Rate of coccidiosis =

$$\frac{\text{No of goat with coccidiosis}}{\text{Total No of goat tested at particuler point in time}} \times 100$$

2.3 Sample Size:

Regular visits were made by investigator in live-stock farms, Veterinary Hospital cases and goats reared in houses, back yard.

Atotal of 400 goat fecal samples were examined, The survey period extended 3 months from May-June and July.

Risk factors were age, breed, species, sex, housing, hygiene and season. They were recorded during collection of samples. Age of animals was determined for both sexes based on dentition, those animals with the age less than one year were considered small while those greater than one year or equal were considered adult.

Hygiene: meaning management supply of water and feeds – fecal contamination, maintaining clear sources of feeds and water tank also clean or dry beddings.

Housing: was considered close system those goats reared in close place in and intensive care, and consider open system in which goats reared in open place.

The sample size was calculated according to Formula by using 50% expected prevalence with 5% absolute precision at 95% confidence interval. (Thrus Field, 2005).

$$N = \frac{z^{2} * p\exp(1 - p\exp ected)}{d^{2}}$$

N = sample size.

P exp = expected prevalence (o.5)

d = desired absolute precision (usually 5%).

Z = required confidence level

Z = 1.96 for 95% confidence level.

Therefore by substituting values of variables in the formula the sample size was determined to be 400 which is used as represented animal on which study was done to know the prevalence of coccidiosis.

2.4 Sample Collection and Laboratory Diagnosis:

2.4.1 Fecal collection:

A total of 400 fecal samples were collected directly from the rectum of goat using hand gloves and carried in a clean plastic container (Troncy.1989). All samples were transported to Atbara veterinary lab rotary for examination of coccidiosis spp. Each sample were clearly labeled with animal identification. Date and place of collection. all samples were stored at 4^{o} c in refrigerator until examined within 48 hours.

2.4.2 Floatation test:

Detection of coccidiosis eggs in faces was carried out by flotation. About 3g of feces were weighted by calibrated teaspoon and placed in a container with saturated solution of sodium chloride (Na Cl) was added. Mixed and filtered through a tea sieve and strained through sieve to remove coarse fecal materials. The mixture was put in tube (15ml volume). Then the cover slip was placed on top of the tube for 10 to15 minutes on the bench. The egg were fluted to the surface and touched with a cover slip. The cover slip was placed on a clean slide and examined under the compound microscope at 10×10 magnification (Soulsby.1982).

2.4.3 Direct test technique:

* It was simplest and easiest technique to facilitate detection of oocyte which passed in the feces (soulsby.1982).

*A small amount of fresh feces was placed in the center of microscopic slide which labeled with goat number, and mixed with 1-2 drop of water to form the mixture.

* We need wooden applicator stich match to mix the fresh feces with water.

* Covered the mixture with cove slip.

* Put the slide under alight microscope 10 X10 magnification.

In the Hospital lab we must:

* Registered the result.

* Recorded if the result positive or negative

2.4.4 Material required:

- Wooden applicator stick match.
- Coverslip.
- Object glass.
- Microscope Slide.
- Light microscope.
- Normal saline.
- Marker for labeling.

2.5 Data Collection :

The data were collected through observation structured questionnaire that targeted the key persons in farms and clinic selected .Moreover the samples (400) were collected using probability random sampling techniques.

2.6 Statistical Analysis:

Frequency table of the distribution according to potential risk factor was constructed. Cross tabulation of Coccidiosis according to potential risk factor was made. Univarate analysis by the Chi –square test using statistical packets for Social Sciences (SPSS). Unviarate model, the significant level in the analysis was be ≤ 0.05.

Chapter Three

Result

Chapter Three

Results

The results indicated natural coccidiosis infection was prevalent among Sudanese goat in Atbara locality at River Nile State with an overall prevalence (33.8%) among 400 goat fecal samples examined 135 were found positive but 265 animals were found to be negative for goat coccidiosis.

Table(3.1): Prevalence of coccidiosis infection among 400 goat examined in Atbara Locality

	Frequency	Percent	Valid Percent	Cumulative Percent
Positive	135	33.8	33.8	33.8
Negative	265	66.2	66.2	100.0
Total	400	100.0	100.0	

Table(3.2): Summary of frequency of all animals(n = 400) were examined for Coccidiosis in Atbara locality.

Risk factor	Frequency	Relative Frequency%	Cumulative Frequency %
Sex			
Male	129	32.2	32.3
Female	271	67.8	100.0
Age			
Adult	251	62.8	62.8
Small	149	37.2	100.0
Housing			
Open	212	53.0	53.0
Close	188	47.0	100.0
Breed			
Nobi	320	80.0	80.0
Sanin	80	20.0	100.0
Hygiene			
Good	271	67.8	67.8
Bad	129	32.2	100.0
Season			
Summer	370	92.5	92.5
Autumn	30	7.5	100.0

Sex of animals:

Among 271 female animals examined 77 were found positive for coccidiosis indicating prevalence of 57%. Among 129 male animals examined 58 were found positive for coccidiosis indicating prevalence of 43 % .Table (3.3)

The Chi- square test showed that there was significant association between coccidiosis infection and sex of animals (p- value = 0.000) Table (3.4)

Age of animals:

The result of age showed that the total number of young animals were 149 the animal infected were 90 and the rate of infection was 66.7% the total of the old animals 251 the animals infected were 45 the rate of infection was 33.3% . Table(3.3).

The Chi- square test showed that there was significant association between coccidiosis infection and Age of animals (p- value=0.002) (table 3.4)

Housing of animals:

The result of age showed that the total number of open Housing of animals were 212. The animal infected were 70 and the rate of infection was 44.3%

The total of the close Housing animals 188. The animals infected were 88 the rate of infection was 55.6%00

The Chi-square test showed that there was significant association between coccidiosis infection and Housing of animals (p-value = 0.002) (table 3.4)

Breed of animals:

The total number of cross examined was 80 the positive was 75 and the rate of infection was 55.6% the local goat examined was 320. The animals infected were 60 the rate of infection was 44.4% .Table(3.3)

The Chi- square test showed that there was significant association between coccidiosis infection and breed of animals (p- value = 0.000) (table 3.4)

Hygiene of animals:

The total number of animals were reared in good hygiene was 226 the positive was 45 and the rate of infection was 33.3%. The total number of animals were reared in bad hygiene was 174. The animals infected was 90 the rate of infection was 66.7%.Table(3.3)

The Chi-square test showed that there was significant association between coccidiosis infection and hygiene(p- value = 0.007) (table 3.4).

Season:

The total number of animals were examined in summer were 370 the positive was 105 and the rate of infection was 84%. The total number of animals were examined in autumn was 30. The animals infected was 20 the rate of infection was. Table(3.3)

The Chi- square test showed that there was significant association between coccidiosis infection and season of animals (p- value 0.000). Table(3-4)

Table (3.3): Summary cross – tabulation of Coccidiosis in 400 goat were examined in Atbara Locality:

Risk factor	Animals tested	Animals affected	Rate of infection %
Sex			
Male	129	58	43.0
Female	271	77	57.0
Age			
Adult	251	45	33.3
Small	149	90	66.7
Housing			
Open	212	70	44.3
Close	188	88	55.7
Breed			
Nobi	320	60	44.4
Sanin	80	75	55.6
Hygiene			
Good	271	45	33.3
Bad	129	90	66.7
Season			
Summer	370	105	84.0
Autumn	30	20	16.0

Table (3.4): Summarized of Univarate analysis for potential risk factor of *Coccidiosis* in 400 goat examined inAtbara locality State using Chi-square test:

Risk factor	No. inspected	No. affected %	DF	X^2	P-value
Sex					
Male	129	58(43.0)	1	57.300	0.000
Female	271	77(57.0)			
Age					
Adult	251	45(33.3)	1	75.439	0.000
Small	149	90(66.7)			
Housing					
Open	212	70(44.3)	1	22.108	0.002
Close	188	88(55.7)			
Breed					
Nobi	320	60(44.4)	1	161.006	0.000
Sanin	80	75(55.6)			
Hygiene					
Good	271	45(33.3)	1	35.109	0.007
Bad	129	90(66.7)			
Season					
Summer	370	105(84.0)	1	16.522	0.000
Autumn	30	20(16.0)			

Means significant value , P-value $\leq$ 0.05

Chapter Four

Discussion

Chapter Four

Discussion

One hundred thirty five out of 400 goat fecal samples were found positive with overall prevalence of rate (33.8%) in goats in Atbara locality in the River Nile State.

This finding was lower than those reported in Edamer Province River Nile State it was (82%) (Elrabie,1999) and lower than those reported in Khartoum State in Sudan of prevalence (86.2%) (Yousif *et al*.,2009) during some period of the years.

These difference in prevalence due to difference ecological condition or different in the management programs or various sanitation effort.The immune state of the host, sample size, sampling period and breed susceptibility to coccidia in different areas.(Degers, Gul., *et al* 2003, Morein., *et al*, 2002).

In this current study the prevalence was lower because May, June, July were hot dry months which reduce the prevalence of coccidiosis.

According to age the prevalence of coccidiosis in small age categories (less than 1 year) was significantly higher (66%) than the prevalence in adult (33%) that has been attributed to lower resistance or less immunity to Eimeria in young animals compared to the adult (Gregory et al .,1980 Kanyari, 1988). These findings same in agreement with previous study reported that coccidiosis were pathogenic effect in young animals (Taylor., et al 2007) and agreement with researches who reported coccidiosis is a disease of young animals (Soulsby, 1986).

In male the prevalence of coccidiosis was (43%) and the prevalence of coccidiosis in female was (57%) higher than male, This finding is agreement with previous researches reported that female were more susceptible than male this might revert to sex related factors such as pregnancy-lactation – giving birth (Yakhchaki, *et al* 2008).

According to the housing factor the prevalence in open and close system was (44%) and (55%) that means it was higher in close system than open with strong association between housing and disease with P-value (0.002) it was similar to the previous study reported that under modern production system kids was born into potentially heavy contaminated environment often result into severe disease (Taylor, 2007) also agree with author reported that goat a serious problem specially in intensive care (Gull,2007,Uzal,2016) and agree with those who reported that coccidiosis occurs higher prevalence under condition of intensive husbandry stress factors overcrowding (Paraud and Chartier, 2012).

The prevalence of coccidiosis in bad management was (66%) significantly higher than in good management with prevalence (33%) and there was strong association between bad management and coccidiosis the significant was (P-value = 0.007) and this study agree with previous study reported that the infection was acquired with poor management practice when the feed and water supply were contaminated with goat feces and intensive management practice (Gull, 2007, Ruiez and Gozalez., *et al* 2006).

Saanin prevalence in this study was (55%) and the prevalence of coccidiosis in Nobi goat was (44%) less than Saanin goat and there was strong association between breed and coccidiosis with significant (P-value = 0.000) and this disagree with researchers reported that Saanin were less susceptible to coccidiosis than others (kanyari, 1988).

The prevalence of coccidiosis in dry season was (84%) significantly higher than the prevalence in wet season was (16%) and the result disagree with previous observation who found Eimeria oocyte were significantly higher in wet season (Alyousif, *et al*.1992, wang, *et al*. 2010)

Conclusion:

The output of this study indicated that coccidiosis was low prevalent in goat in Atbara Locality in this period of year (May, June and Jully)with over all prevalence of (33.8%).

A high prevalence of infection was in females as compared to males .Young animals were highly effected as compared to adult animals .A high prevalence of infection was in saanin compared to local . A high prevalence of infection was in bad hygiene as compared to good hygiene . A high prevalence of infection in summer compared to autumn. A high prevalence of infection was in close housing system compared to open system.

Recommendation:

1. Proper sanitation and management of goats this include:

a) Pen should be kept dry and cleaned out frequently to prevent sporulation of Eimeria.

b) All measures which reduce the amount of fecal contamination (e.g. hair shearing) should be practiced regularly.

c) Feed and water troughs should be high enough to avoid heavy fecal contamination and feeding on the ground should be avoided if possible whenever crowding is aprope.

2. More care must directed to the animals owner so as to keep his heath and animals.

3. Absence of clinical signs lead to a lack of attention to the disease in spite of the great possibilities of its existence, therefore integrated strategics should be utilized the prevent and control Eimeria spp infections

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Appendix 1

Frequency table for the distribution of infection among 400 goat examined at Atbara locality to potential risk factor.

Frequency distribution of goat coccidiosis in Atbara Locality according to sex

Risk factor	Frequency	percent	Valid percent	Cumulative percent
Sex				
Male	129	32.2	32.2	32.2
Female	271	67.8	67.8	100.0

Frequency distribution of Age:

Risk factor	Frequency	percent	Valid percent	Cumulative percent
Age				
Adult	251	62.8	62.8	66.8
Small	149	37.2	37.2	100.0

Frequency distribution of Breed :

Risk factor	Frequency	percent	Valid percent	Cumulative percent
Breed				
Nobi	320	80	80	80
Sanin	80	20	20	100.0

Frequency distribution of Housing:

Risk factor	Frequency	percent	Valid Percent	Cumulative Percent
Housing				
Open	212	53.0	53.0	53.0
Close	188	47.0	47.0	100.0

Frequency distribution of Hygiene

Risk factor	Frequency	Percent	Valid Percent	Cumulative Percent
Hygiene				
Good	271	33.3	33.3	33,3
Bad	129	66.7	66.7	100.0

Frequency distribution of Season

Risk factor	Frequency	Percent	Valid Percent	Cumulative Percent
Season				
Summer	370	84	84	84
Autumn	30	16	16	100.0

Appendix

Cross tabulation for the distribution of infection among 400 goat examined at Atbara locality to potential risk factor

A. Goat *coccidiosis* and Sex Cross tabulation

		Sex		Total
		Male	Female	
	Positive	58	77	135
	Negative	71	194	265
Total		129	271	400

B. Goat *coccidiosis* and Age Cross tabulation

		Age		Total
		Adult	Small	
	Positive	45	90	135
	Negative	206	59	265
Total		251	149	400

C. Goat *coccidiosis* and Housing Cross tabulation

		Housing		Total
		Open	Close	
	Positive	70	88	158
	Negative	142	100	265
Total		212	188	400

D. Goat coccidiosis and Breed Cross tabulation

		Breed		
		Nobi	Sanin	Total
	Positive	60	75	135
	Negative	260	5	265
Total		320	80	400

E. Goat coccidiosis and Hygiene Cross tabulation

		Hygiene		
		Good	Bad	Total
	Positive	45	90	135
	Negative	181	84	265
Total		226	174	400

F. Goat coccidiosis and Season Cross tabulation

		Season		
		Summer	Autumn	Total
	Positive	105	20	125
	Negative	265	10	275
Total		370	30	400

Appendix III

Univar ate analysis for the association of goat coccidiosis in goat with potential risk factor using Chi-Square Tests:

A. Sex

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	57.300[a]	1	0.000
Likelihood Ratio	56.108	1	0.000
Linear-by-Linear Association	57.157	1	0.000
N of Valid Cases	400		

B. Age

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	75.439[a]	1	0.000
Likelihood Ratio	75.337	1	0.000
Linear-by-Linear Association	75.250	1	0.000
N of Valid Cases	400		

C. Housing

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	.108[a]	1	0.043
Likelihood Ratio	.108	1	0.003
Linear-by-Linear Association	.108	1	0.oo3
N of Valid Cases	400		

D. Breed

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	161.006[a]	1	0.000
Likelihood Ratio	165.234	1	0.000
Linear-by-Linear Association	160.604	1	0.000
N of Valid Cases	400		

E. Hygiene

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	.109[a]	1	0.003
Likelihood Ratio	.109	1	0.006
Linear-by-Linear Association	.109	1	0.001
N of Valid Cases	400		

H. Hygiene

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	16.522[a]	1	0.000
Likelihood Ratio	25.928	1	0.000
Linear-by-Linear Association	16.481	1	0.000
N of Valid Cases	400		

Appendix

Questionnaire for data collection to investigate the risk factors which associated with Coccidiosis in Atbara Locality.

*Animal No ( )

1. Breed of animal

0- Saanin ( ) 1- Local ( )

2. Age of animal

0- less than one years (young) 1-Equal or more than one years (old)

3.Sex of animal

0- Male ( ) 1- Female ( )

4.Housing:-

Close system1-( ) open system − 0( )

5 .Management

Bad management0- ( ) 0Good management( )

6.Season

0-Summer( ) 1-Autom( )