INTRODUCTION

Background

Liver fluke is a common trematode (flat worm) that causes a parasitic disease called fascioliosis or fluke disease. It has a pathogenic effect on ruminants (cattle, sheep and goats) as well as other farm animals like horses, donkeys, pigs and to a lesser extent poultry. Species of liver fluke include *Fasciola gigantiga* and F. *hepatica*, with the former being more prevalent in cattle and the latter sheep and goats (Ozung *et al*., 2011)

Fascioliosis caused by *Fasciola hepatica* and *Fasciola gigantica*is regarded as one of the most important parasitic diseases in the world and as meat consumption, Fasciolosis is an important parasitic food borne disease, responsible for significant public health problems and substantial economic losses to the livestock industry.(Odigie *et al* ., 2013).

Both *F.*hepatica and *F.gigantica* are transmitted by the snails of the family *L.ymnaesida* .Infestation with fasciolosis is usually associated with grazing wet land and drinking from the snail infesting watering places**.** (Dechasa *et al*., 2012).

Fasciola gigantica is a parasite of cattle, sheep and wild animal in the tropic and sub **-**tropics, and are more pathogenic than *fasciola hepatica* this is very similar parasite to *fasciola hepatica* and it is found more commonly in tropical regions of the world. Areas affected include Africa, Asia, many pacific island including Hawaii, the Middle East, Southern Europe, and south of USA. *Fasciola hepatica* has become increasing wide spread in New Zeland in recent years following the colonization of alarge area of the country y by the exotic snail. (Elhaj., 2001).

The development of fasciolosis involves the presence of an intermediate host (*Lymnaea sp*.), suitable habitats for mollusks and environmental factors such as high humidity, adequate temperature and rainfall. Furthermore, when infecting the definitive host, mature flukes lay eggs that spread in the environment and cause pasture recontamination (Silva *et al*., 2007).

Fascioliosis or liver fluke is worldwide distributed. In the Sudan the disease is highly endemic and reported in many areas of the country such as Upper Nile, Blue Nile, White Nile, as well as Northern states **(**Koko et al., 2003).

The incidence of fascioliosis in the Sudan has probably increased during recent years as result of agricultural extension and introduction of canalization and pump. Also fascioliosis in Sudan have economic losses a result of livestock mortality and more often due to loss in production and condemnation of infected tissue in the slaughter house ,besides, large amount of money is spent annually on treating infected cases (Altahir.,1975).

Liver fluke infections caused by *Fasciola hepatica*, Fasciola gigantica, are major public health problems in East Asia, east Europe, Africa and Latin America. Currently, more than 780 million people are at risk of infection with fasciolosis. (Marcos et al., 2008)

Among many parasitic problems of farm animals, fasciolosis is a major disease, which imposes direct and indirect economic impact on livestock production, particularly of sheep and cattle**,** through mortality, liver condemnation, reduced production of meat, milk, wool and cost for anthelmintics and apart from its veterinary and economic importance Through the world fasciolosis has recently been shown to be are emerging and widespread zoonosis affecting many people **(**Belay *et al.*, 2012**)**

Considering the worldwide spread, occurrence and zoonotic nature, fasciolosis has emerged as a major global and regional concern affecting all domestic animals and infection is most prevalent in regions with intensive sheep and cattle production (WHO, 2007)

Surveys in some Asian countries have shown that among domestic animals, cattle are the most suffering animals from fasciolosis (Kuchi. 2011).

Several reports exist on how variable climatic factors and patterns determine major period and level of fasciolosis transmission in the divergent agro -ecologic zone in the world. Apart from climatic factors, other factors including the sex of the animals have been suggested as a variable that could influence the prevalence of fasciolosis in cattle (Abedokun et al., 2008).

Sever out- break with heavy mortalities were en countered in the White Nile .province among cattle, sheep and goat, in 1959 and outbreak near in Bahr El ghazal province in which 60 cows perished(Saeed.,1992). In 1973 a high number of sheep near Kosti died from acute fascioliasis .*fasciola* gigantica was first mentioned in 1914 in the annual report of the Sudan veterinary service (s.v.s).In 1938 fascioliasis was again reported in Malakal.The report of 1953-1954 stated the incidence of fascioliosis increased in Kosti and Eldueim.The annual reports since 1955 indicate that highest percentages of liver condemnation due fasciolosis are as follows: Upper Nile province (27-58%), Baher El gazal province (16-18%), Equatorial province (13-40%).in all area in Sudan fascioliasis has been reported and several place with high prevalence (Saeed.,1992).

The objective of the study:

(1) To Estimate the prevalence of bovine fasciolosis in North Darfur state.

(2) To investigate potential risk factors which are associated with bovine fasciolosis in North Darfur.

CHAPTER ONE

1. LITERATURE REVIEW

1.1. Definition:

asciolosis is an economically important parasitic disease, which caused by trematodes of the genus *Fasciola* that migrate in the hepatic parenchyma, and establish and develop in the bile ducts .*Fasciola* is commonly recognized as liver flukes and they are responsible for wide spread of morbidity and mortality in cattle characterized by weight loss, anemia and hypo proteinemia. The two most important species, *Fasciola hepatica* found in temperate area and in cooler areas of high altitude in the tropics and subtropics and *Fasciola gigantica*, which predominates in tropical area **(**Mulugeta *et al*., 2011).

The distribution of fasciolosis is linked to climatic factors, management of reservoir animals, topographic factors and presence in the environment of molluscs of the genus *Lymnaea,* which are needed for the parasite's life cycle to be completed (Bernardo *et al* 2011).

The snail of the genus *Lymnae natalensis* and Lymnae truncatula are known as intermediate host in life cycle of fasciolosis.Infection with *Lymnea truncatula* is usually associated with herds and flocks grazing wet marshy land. On the other hand, *Fasciola gigantica* is a fresh water snail and infection with this species is associated with livestock drinking from snails infected watering places as well as with grazing wetland **(**Mulugeta *et al*., 2011).

. It is estimated that 17 million people are infected in the world and

91.1 million Area at risk of infection, both species overlap in many areas of Africa and Asia, whereas *F. hepatica* is a major concern in the

Americas, Europe and Oceania. Geographical pattern of these flukes is not uniform. The rural areas of the Andean Region of Peru and Bolivia are the most constantly affected regions in the world with prevalence rates between 6 and 68% The proximity of these rural areas to big industrialized cities creates a potential source of infection to non endemic area (Marcos *et al* .;2008).

In tropical regions, fasciolosis is considered as the single most important helminthes infection of cattle with prevalence rates of 30-90% in Africa, 25-100% in India and 25-90% in Indonesia (Fekadu *et al.*;2012).

1.2. Epidemiology:

A total of 208 cattle were randomly selected among slaughter houses, household and livestock farms to determine the prevalence offascioliasis. Epidemiological studies on fascioliasis of cattle were undertaken in such localities under different climatic conditions existing in Ladakh region of Jammu and Kashmir State. Infection rate was 51.42%, 27.69% and 21.91% in slaughtered, livestock farm and household cattle, respectively. Significant variations were observed in the prevalence with respect to various host factors and the climate of the study area. Overall, the highest seasonal prevalence (45.19%) in all types of cattle was recorded during wet season while as only (24.40%) was recorded during the dry season. It was noticed that a higher infection rate was recorded in young cattle ages (0- 2 years) (40.02%) than in adult ones (28.04%) (3- 8 years). Moreover, the prevalence of infection in females was more (38.07%) than males (29.09%). It was also observed that the infection rate was high in comparatively low land areas (37.14%) as compared to high altitudes (30.09%). This study will provide necessary information regarding fascioliasis in cattle of Ladakh for their effective Control and hence for a better production which will be beneficial resource to poor people where live stock rearing is one of the important sources of livelihood (*Kuchai et al., 2001)*.

Also a cross-sectional study was carried out from November, 2011 to March, 2012 at Nekemte municipal abattoir to assess prevalence and economic significance of bovine fasciolosis. Out of 384 cattle examined at post mortem, 21.9% (84) were positive for fasciolosis. The prevalence of bovine fasciolosis was found to be significantly affected ($P < 0.05$) by the age of animal, in which young animals were affected than adult animals. The prevalence of bovine fasciolosis was also higher ($P < 0.05$) in poor body conditioned animals than medium and good body conditioned animals. Sex of the animal was not found as a significant factor ($p > 0.05$) affecting the prevalence of disease. The prevalence of *Fasciola hepatica* was 14.1% (54), which was predominant among *Fasciola* species, causing bovine fasciolosis in the study areas. Whereas, the prevalence of *Fasciola gigantica* was 5.2% (20), and 2.6% (10) animals were mixed infected. The economic significance of bovine fasciolosis was also assessed based on condemned livers. Thus, based on retail value of bovine liver, the direct economic loss from fasciolosis during the study time was estimated to be 63072 ETB annually**.** (Petros *et al*., 2013)

Another study was conducted to evaluate the economic losses and temporal distribution of the prevalence of liver condemnation due to bovine fasciolosis. The abattoir in Atilio Vivacqua, in the South of the State of Espirito Santo, which is under state inspection by the veterinary service of the Livestock and Forest Protection Institute of Espirito Santo, was used as the data source. The prevalence of liver condemnation due to Fasciolosis over the period 2006-2009 was calculated. The $\chi2$ test, simple linear regression analysis and $\chi2$ for trend were used, with significance Level of $p \leq 0.05$. Over the period analyzed, 110,956 cattle were slaughtered and the prevalence of liver condemnation due to *Fasciola hepatica* was 15.24% in 2006, 23.93% in 2007, 28.57% in 2008 and 28.24% in 2009. The historical trend of liver condemnation is an increasing trend, thus indicating that this parasitism has become established in the herd as a problem in this region, with prevalence similar to that of traditionally endemic regions. Condemnations occurred throughout the year, with the highest prevalence in April and May and with significant differences between the dry and wet seasons. The economic losses from liver condemnation can be considered high. (Bernardo*et al*., 2011).

Also a total of 239 cattle were sampled across eight locations ranging in elevation from 1112-2072 m. Fecal material was examined for presence of *Fasciola* eggs and sera were tested by ELISA for antibodies against *Fasciola* antigens. Bolstering this, 38 cattle at slaughter from 2 abattoir sites at 1150 m and 1947 m were inspected; in addition, wild buffalo stool (n = 10) opportunistically picked within Mount Elgon National Park (MENP) at 3640 m was examined. By fecal egg detection, prevalence of *Fasciola gigantica* at low (<1500 m) and high (>1500 m) altitude sites was 43.7% (95% CI 35.4-52.2) and 1.1% (95% CI 0.0-6.0), respectively, while by ELISA was much higher, low altitude - 77.9% (95% CI 69.7-85.4) and high altitude - 64.5% (95% CI 51.3-76.3). The decline in prevalence with increasing altitude was corroborated by abattoir sampling. Thirty seven aquatic habitats, ranging from 1139-3937 m in altitude were inspected for freshwater snails, 12 of which were within

MENP. At lower altitudes, *Lymnaea* (Radix) n*atalensis* was common, and often abundant, but at higher altitudes became much rarer ceasing to be found above 1800 m. On the other hand, *Lymnaea (*Galba) *truncatula* was found only at altitudes above 3000 m and within MENP alone. The snail identifications were confirmed by DNA analysis of the ribosomal 18S gene. **(**Howell et *al*., 2012).

In the winter of 1998/1999, sheep on a farm in the province of North Holland, The Netherlands, died from sub acute and chronic liver fluke disease despite four previous treatments with triclabendazole (TCBZ). Fecal examinations of sheep and cattle on the farm showed high number of liver fluke eggs. In a randomized clinical trial, the fluke egg output was monitored weekly for 3 weeks in sheep which were treated with TCBZ or with closantel; in dairy cows treated with TCBZ or with clorsulon; and in heifers treated with TCBZ or clorsulon. The results showed a significant reduction of 99.7, 98%.1% and 99.2%, respectively, in fluke egg output at 21 days in all non-TCBZ treated animals. TCBZ treatment produced percentage decreases of 15.3, 4.3 and 36.6%, respectively. These results are highly indicative of the presence of TCBZ-resistant *Fasciola hepatica* in sheep and cattle on this farm. (Lammer *et al.*, 2000).

Another Report on the species of *Fasciola* present in the Nile Delta, Egypt, appears controversial. Some authors reported the presence of both *Fasciola gigantica* and *Fasciola hepatica*; others reported *F. gigantica* only and mentioned that *F. hepatica* was found only in imported animals. This study was an attempt to identify the species of *Fasciola flukes* collected from locally bred animals. Morphologic, morph anatomic, morph metric, and chemotaxonomic criteria of the fluke isolates were studied. Speciation based on morphologic and morphometric data was not decisive due to overlap in the values of most measurements. Morph anatomic data proved the presence of both the species, and isoelectric focusing (IEF) of fluke soluble protein confirmed the presence of both *F. gigantica* and *F. hepatica* in Egypt (Lotfy *et al.,* 2001).

Also in another study was conducted in the northeastern areas of Punjab province (Pakistan) to analyze the monthly and seasonal pattern of fasciolosis in buffaloes and its relation to some climatic factors (temperature, humidity, rainfall and pan-evaporation) was also worked out. The fecal samples of buffaloes were collected from April 2003 to March 2005 on a monthly basis from randomly selected areas and analyzed for the presence of *Fasciola* egg. From 7200 samples, 1058 (14.69%) were found positive. Seasonal data showed the highest prevalence and egg count (EPG) in autumn and the lowest in spring. Monthly results showed the highest prevalence in September (32.33%) and the lowest in May (4.83%), while mean EPG was highest in October (567 ± 95.5) and the lowest in June (3.2 ± 0.48). Statistically, significant difference ($P<0.05$) was noted within seasonal and monthly prevalence's. Impact of humidity was found significant ($P<0.05$) on disease as compared to other climatic factors. **(**Qureshi *et al*., 2012).

In a cross-sectional study was conducted to determine the prevalence of *Fasciola hepatica* and to investigate the related risk factors in cattle from Kayseri, Turkey. Fecal and blood samples were collected from 282 cattle from May 2004 to April 2005 and were examined by modified McMaster sedimentation and ELISA techniques to detect *Fasciola* sp. eggs and anti-*F. hepatica* antibodies, respectively. Of the total of 282 cattle, 184 were seropositive for *F. hepatica* with a prevalence of 65.2%. In addition

24.5% of seropositive cattle had fluke eggs in the fecal examination. The mean number of EPG in infected cattle was 42.8%, 4.4%. The highest prevalence was observed in ≥6 age group (87.2%) followed by 3-5 (79.5%) and ≤2 age groups (51.6%). The differences between ≤2 and other age groups were found significant ($p<0.001$), whereas no statistically significant difference ($p>0.05$) was observed between 3-5 and ≥6 age groups. The infection was more prevalent in females (70.7%) than males (47.8%) and in cattle from the traditional farms (76.5%) than the small-scale dairy farms (37.2%). No statistically significant difference ($p>0.05$) was observed related to breed**.** (Yildrim *et al* ., 2007).

The aim of previous study was to establish the prevalence and economic significance of fasciolosis in cattle slaughtered at Arusha abattoir in Tanzania. A 3-year database (2005-2008) from the abattoir was retrieved and analyzed. In addition, meat inspection was carried out for one month (July 2008) with focus on *Fasciola* infection and its associated economic loss due to liver condemnation. Results from the retrospective study revealed that 8302 (6.7%) livers out of 123790 examined livers were condemned due to fasciolosis. Analysis of primary data (meat inspection) showed that 150 of 469 cattle livers condemned were due to fasciolosis, a relative condemnation rate of 32.0% per month. Based on the current local price of liver, the economic loss per month due to liver condemnation was estimated at Tanzania shillings (TZS) 1,800,000/- (approximately US $1,500), which summed to TZS 21,600,000/- (US $18,000) per annum. The specific cause of liver fasciolosis was *Fasciola gigantica*. These results indicate that *F. gigantica* infection is an important condition that leads to high liver condemnation rates in cattle slaughtered, resulting into high financial loss. These merits for more extensive epidemiological investigations to better determine the prevalence, economic impact and public health importance of the disease (Mwabonimana *et al.*, 2009).

Another study was conducted to determine the prevalence of bovine fasciolosis and to assess the effectiveness of commonly used anthelmenthics in Ginner district south-eastern, Ethiopia from September to December 2011. For the determination of the prevalence, 384 fecal samples were collected and examined by sedimentation technique. The result revealed that 121 (31.51%) animals were positive for *Fasciola* eggs. The infection rate was 30.81 and 32.16% in animals less than four years and greater or equal to 4 years of age, respectively while it was 29.70 and 33.51% in male and female animals, respectively. However, the differences either in age or sex groups were not statistically significant (Fekadu *et al.*, 2012).

Also in another study aimed at determining the prevalence of *Fascioliasis* in abattoirs located within some selected Local Government Areas of Benin City, Nigeria. a total of 180 cattle found within the confines of 3 abattoirs in the respective LGAs were examined. Consequently, 9 abattoirs were investigated for liver flukes (*Fasciola hepatica* and *Fasciola gigantica*), bringing the overall number of cattle examined to 540. The screening exercise was carried out between the 15th day of August and 2nd of December, 2012, using standard histo chemical techniques. The results showed that of the 540 Cattle examined, 11.5% were infected. The distribution shows that cattle slaughtered at Ikpoba Okha LGA abattoirs had the highest infection rate of 5.74%, followed by abattoirs from Egor (3.33%) and Oredo (2.44%), suggesting that there exist differences in the hygienic status of abattoirs, as well as the mode of feeding and water consumption.(Odigie,et *al.*,2013).

Another study was conducted during the period between January 1999 and December 2000, the distribution and seasonal patterns of *Fasciola gigantica* infections in cattle in the Highveld and lowveld communal grazing areas of Zimbabwe was determined through monthly coprological examination. Cattle fecal samples were collected from 12 and nine dipping sites in the Highveld and lowveld communal grazing areas respectively. Patterns of distribution and seasonal fluctuations of the intermediate host-snail populations and the climatic factors influencing the distribution were also determined by sampling at monthly intervals for a period of 24 months (November 1998 to October 2000) in six dams and six streams in the Highveld and in nine dams in the lowveld communal grazing areas. Each site was sampled for relative snail density and the vegetation cover and type, physical and chemical properties of water, and mean Monthly rainfall and temperature were recorded. Aquatic vegetation and grass samples 0–1 m from the edges of the snail habitats were collected monthly to determine the presence or absence of *F. gigantica* Metacercariae. Snails collected at the same time were individually checked for the emergence of larval stages of *F. gigantica*. A total of 16 264 (calves 5 418; wieners 5 461 and adults 5 385) fecal samples were collected during the entire period of the study and 2 500 (15.4 %) of the samples were positive for *F. gigantica* eggs. Significantly higher prevalence's were found in the Highveld compared to the lowveld ($P < 0.001$), for adult cattle than calves ($P < 0.01$) and in the wet season over the dry season ($P < 0.01$). Fecal egg output peaked from August/September to March/April for both years of the study. (Pfukeny *et al.*, 2006).

Another cross sectional study to determine the prevalence of *Fasciola gigantica* in cattle was carried out in 9 randomly selected farms and 1 slaughter House between February and May 2012. Fecal and blood samples were collected from 186 cattle in the farms and 200 cattle at slaughter. The fecal samples were analyzed using the formol ether sedimentation technique and the blood by Indirect ELISA kit (Bio-X-Diagnostic, ID VET Jemelle-Belgium) to detect *F. gigantica* eggs and antibodies to *F. Gigantica* antigens respectively. Of the 200 fecal samples collected at slaughter, 39(19.5%) had *F. gigantica* eggs; as compared to 27 (14.5%) positives out of the 186 samples collected from the farms; giving an overall prevalence rate of 66 (17.1%). There was no significant difference (P>0.05) between prevalence of infection of cattle sampled in the farms and slaughter house. 23 (11.5%) of the sera prepared from the 200 blood samples obtained at slaughter had antibodies to *F. hepatica* antigens, as against 5(2.6%) for sera from 186 blood samples collected in the farms; giving an overall sero prevalence of 28(7.3%). There was significant difference (P< 0.05) between infection at slaughter and on farms. Out Of the 200 cattle from slaughter, 20(10.0%) had *F. gigantica* eggs and also were sero positive for *F. hepatica* antigens, and of the 186 cattle from farms only 5(2.7%) that had *Fasciola* eggs and were also seropositive for *F. hepatica* antigens. Both at slaughter and on farms, infection was more prevalent in females than in males. The overall prevalence for Females using coprology and ELISA were 19.3% (41/212) and 7.5 %(16/212) respectively. The respective values for males were 13.7% (24/174) and 6.89% (12/174). However, the difference in the prevalence of females and males obtained was not statistically significant (P>0.05). No statistical difference was observed in breed prevalence. This study has established *F. gigantica* prevalence of 17.1% and 7.3% by coprological and serological examinations of faces and blood of cattle in Zaria (Aliyu et al., 2014).

Also another study was designed with the aims of determining the prevalence and risk factors of fasciolosis in cattle, sheep and goats slaughtered from October, 2010 to April, 2011 at Hashim Nur's Ethiopian Livestock and Meat Export industrialized abattoir in Debre Zeit, Ethiopia. One thousand one hundred fifty two ruminants comprising of cattle, sheep and goats (n=384 each) were subjected to routine post mortem examination for fasciolosis. The overall prevalence of fasciolosis in the study was proved to be 21% (242/1152). The prevalence of fasciolosis in adult cattle, sheep and goats' were confirmed to be 39.8%, 28.7% and 13.9%, respectively and the prevalence of fasciolosis in young cattle, sheep and goats were proved to be 23.3%, 12.7% and 7.0%, respectively. Significantly higher ($p<0.05$) prevalence of fasciolosis was seen in adult cattle, sheep and goats when compared to young ones. The prevalence of fasciolosis in poor body conditioned cattle, sheep and goats were known to be 38.1%, 28.8% and 13.6%, respectively and prevalence of fasciolosis in medium body conditioned cattle, sheep and goats were known to be 30.0%, 20.5% and 11%, respectively. The prevalence of fasciolosis in good body conditioned cattle, sheep and goats were proved to be 24.2%, 14.3% and 7.2%, respectively. Statistical analysis of the data showed the presence of significant difference ($p<0.05$) on the prevalence of fasciolosis in cattle, sheep and goats on the basis of body condition score. The high level of fasciolosis in cattle, sheep and goats in the present study represents high rate of infection and immense economic losses to the country, Ethiopia. In line with this finding it is recommended that farmers who rear cattle, sheep and goats should improve provision of feeds to their animals so that the animal can have good body condition that confers some level of resistance against fasciolosis. Besides, they should be able to regularly treat their animals with the appropriate anthelementics and awareness should be created on the prevention and control methods of fasciolosis. (Abdulhakim *et al*., 2012**).**

1.3. Taxonomy and classification:-

Kingdom: Animalia

Phylum: Platyhelminthes

Class: Trematoda

Order: Digenea

Family: Fasciola

Species: *Fasciola hepatica* and *Fasciola gigantica* (Saria. 2011)

1.4. Morphology:-

The adult mature and gravid fluke is flat with it is body shape like leaf. The size range is 25 to 30 mm and 8 to 15mm in length and width respectively, depending upon species. The adult inhabits the bile duct in the liver or gallbladder of the final host.the Fluke has an elongated anterior end known as a cephalic con that contains the oral and ventral sucker. The intestines are highly branched and present throughout the body. The male and female reproductive organs are present near the posterior sucker in the center of the body. The female reproductive tract is dense ovary and is located just above the testes and is linked. To short convoluted uterus that opens to genital pore above the ventral suckers. The vitellaria are highly dispersed and divided in the lateral and posterior region of the body.

Fasciola hepatica is very similar to each other, varying in length and width. In addition, the cephalic con of the *Fasciola hepatica* is shorter than *Fasciola gigantica*. The shape of the eggs of the tow flukes is also very similar with the measure meant of the *Fasciola hepatica* and *Fasciola* gigantica being approximately 150 um*90um and 200um *100um, respectively (saria., 2011).

1.5. Transmission:-

Both *F.hepatica* and *F.gigantica* are transmitted by the snails of the family *lymnasidae*.infection with fasciolosis is usually associated with grazing wet land and drinking from the snail infesting watering places (Dechasa et al., 2012).there are many ecology factors affecting snail population include temperature, light, hydrogen ion concentration (PH), vegetation, depth of water, chemical composition of soil and snail population competition. The most important intermediate hosts of F.gigantica in Sudan is *L.natalensis*, *L.aurcularia*; however *L.rufescens* and *L.acuminate* are the host snails in the Indian subcontinent; *L.rubigiosa* and *L.natalensis* are the hosts in Malaysia and in Africa respectively. The most important and widespread (Europe, Asia, and North America) intermediate host of *F.hepatica* is *L.truncatula*. (Soliman., 2008)

1.6. Life cycle:-

The life cycle of *Fasciola* species occurring in any particular area the following conditions must be satisfied. There must be an initial presence of infected final hosts, the intermediate snail host must be present and there must be an opportunity for transmission of the parasite from the final host to the snail habitat (Mahato et al.; 2000).

Adult liver flukes reside in the bile ducts of host animals, and eggs are passed onto the pasture in the feces. After a short period of development (usually 2 to 3 weeks), a miracidium hatches from the egg and attempts to find and penetrate a snail intermediate host. The parasite develops and replicates asexually in the snail over many weeks. Under optimal conditions, parasite maturation within the snail to the cercarial stage takes approximately 5 to 7 weeks, and a single miracidium can develop into several hundred cercariae. Under wet conditions, cercariae emerge from the snail and swim until they find and attach to vegetation. The cercariae then shed their tails and secrete a protective coat, forming the encysted infective stage called metacercariae. Cattle become infected primarily by ingesting the metacercarial cysts on forage, but they also can become infected by ingesting cysts suspended on soil and detritus while drinking contaminated water.[2] The length of time that metacercariae survive on pasture primarily depends on available moisture. Under the hot and dry pasture conditions of coastal Texas during the summer, metacercariae were rapidly killed[3]; however, under conditions of high humidity, during the summer, metacercariae may survive for extended periods.[4] Once ingested by a ruminant host, the metacercariae excyst, releasing juvenile flukes. The juvenile flukes penetrate the wall of the small intestine, migrate through the peritoneal cavity over a week's time, and then penetrate through the liver capsule. Juvenile flukes migrate through the hepatic parenchyma for approximately 6 to 8 weeks before entering the bile ducts where they mature. Egg production can begin as early as 8 weeks after infection[5] however, most infections do not become patent until after approximately 11 to 12 weeks.[6] Thus, completion of the entire parasite life cycle, from the time an egg is shed on to pasture until a newly infected animal re infects the pasture with the next generation of fluke eggs, generally requires 18 to 24 weeks (4.5 to 6 months (Kaplan., 2001).

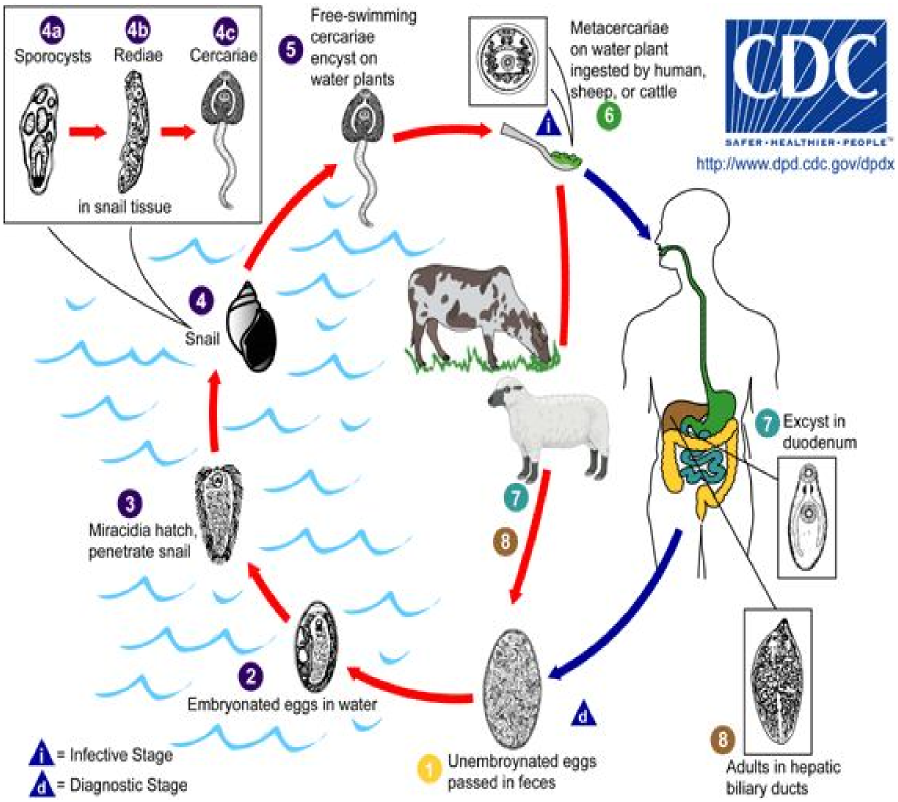

Figure1: Life cycle of *Fasciola* (http://www.dpd.cdc.gov/dpd)

1.7. Pathogenesis and pathology:-

Once ingested by a ruminant host, the metacercariae release, releasing juvenile flukes. The juvenile flukes penetrate the wall of the small intestine and migrate through the peritoneal cavity over a week's time, and then penetrate through the liver capsule. Juvenile flukes migrate through the hepatic parenchyma for approximately 6 to 8 weeks before entering the bile ducts,where they mature. Egg production can begin as early as 8 weeks after infection. Completion of the entire parasite life cycle, from the time an egg is shed onto until a newly infected animal reinfects pasture with the next generation of fluke eggs, generally requires 18 to 24 weeks (4. to 6 month). **(**Kaplan *et al.*, 2001).

Pathology associated with disease are caused by the inflammation of the bile duct which causes thickening of the lining and eventually leads to fibrosis that results in reduced flow of the bile and back pressure builds leading to atrophy and necrosis of the liver parenchyma, causing liver abscesses (Michael., 2004**)**.

Immature wandering flukes destroy liver tissue and cause hemorrhage. In acute fasciolosis damage is extensive the liver. Is an enlarged and friable with fibrinus deposit on the capsule .migratory tract can be seen, and the surface has an uneven appearance. In chronic cases cirrhosis develops. Mature fluke damage the bile duct witch become enlarged or even cystic, and have thickened, fibroses wall. In acute the duct wall become grating thickened and often calcified .fluke maybe found in aberrant sites e.g. lung (Veterinary Manual, 2005).

The principal pathogenic effects of flukes are anemia and hyper albuminaemia. More than 0.5 ml blood per fluke per day can be lost within the bile duct. In acute form, there is massive invasion due to immature flukes into the liver which cause sudden death while in chronic from, there is liver cirrhosis caused by the wandering flukes which when mature. Calcification of bile ducts and enlargement of gall bladder has been noticed in chronic cases and sub mandibular edema frequently occurs (Salam et *al*., 2009)**.**

Traumatic injury caused by the migrating flukes, tracts of coagulative necrosis develop, which result in a diffusely fibrotic hepatic parenchyma containing hemorrhagic streaks and foci. These lesions can predispose cattle to black disease (infectious necrotic hepatitis) and bacillary hemoglobinuria due to *clostridium novyi* and *c.haemolyticum* respectively. (Kaplan *et al*., 2001)).

Also chronic inflammation, chronic injuries and regenerative hyperplasia of the bile duct epithelium may be related to malignant transformation. Recent reports have identified *Fasciola hepatica* as a neoplastic risk agent, primarily in animals, which could cause the genetic damage of the surrounding host tissue. (Azra *et al* .;2004).

1.8. Clinical finding:-

Acute fascioliasis is common in sheep and goats while the chronic form is found mostly in cattle. Symptoms of fascioliosis include anemia, emaciation and reproductive dysfunction in animals with the chronic form. While in acute fascioliasis; the animals usually show signs of anorexia, dullness, diarrhea, muscular atrophy, subcutaneous edema and impaired immune systems. Hepatic fascioliasis is often characterized by a swollen. (Ozung *et al*., 2011).

Sub- acute fasciolosis is characterized by jaundice, some ill thrift and anemia. The burrowing fluke causes extensive tissue damage, leading to hemorrhae and liver damage. The outcome is severe anemia, liver failure and death in 8–10 weeks (Boray.2007).

Chronic fasciolosis is the most common form of liver fluke infection in sheep, goats and cattle – and particularly in more resistant hosts, such as horses and pigs. It occurs when the parasites reach the bile ducts in the liver. The fluke ingests blood, which produces severe anemia and chronic inflammation and enlargement of the bile ducts. The clinical signs develop slowly. The animals become increasingly anemic, appetite is lowered, the mucous membranes of the mouth and eyes become pale and some animals develop edema under the jaw (‘bottle jaw’). Affected animals are reluctant to travel.

Fasciolosis may not show any obvious symptoms. Some animals may show abdominal pain and may become jaundiced. Death is usually due to blood loss resulting from hemorrhage in the liver. The liver hemorrhage is the result of the immature fluke burrowing through the liver. (Boray., 2007).

1.9. Diagnosis:-

Fasciolosis should be considered when there are deaths, anemial or ill. Diagnosis in dead animals relies on seeing mature or immature fluke in the liver.. (Bory., 2007).

Chronic fasciolosis is indicated by fluke eggs in fecal samples. The sampling technique is generally reliable in sheep but much less so in cattle. Fecal examination for *F. gigantica* egg was carried out using the sedimentation method described by Bile was examined for eggs using a modification of the method described. This was carried out by mixing equal volumes of bile and water, straining through a tea strainer before centrifuging at 3000 rpm. After obtaining a clear supernatant by repeated mixing of sediment with water and Centrifuging, the sediment was examined under the microscope. (Adodokum *et al*., 2008).

Another method is a serological test (ELISA) is also available for diagnosis of fasciolosis. It detects infection with both immature and adult fluke in a flock or herd, but it is not sensitive enough for diagnosis in individual animals .There are evidences to show that sero diagnosis can detect the presence of infection as early as 2 weeks after infection. Furthermore, serological methods like Enzyme Linked Immune sorbent Assay (ELISA) can detect serum antibodies to specific antigens of *Fasciola sp*. using adult fluke extracts, or excretory/secretory (ES) materials. Also Agar Gel Diffusion Test (AGDT) has also been demonstrated to be simple and valuable for detection of *Fasciola sp*. (Adodkum *et al*., 2008).

1.10. Public health significant:-

Fascioliosis is a parasitic disease caused by the fluke *Fasciola hepatica* the disease is acquired for the most part by eating watercress, other vegetables or by drinking water contaminated with metacercariae. Human fascioliosis is a serious health problem in many countries, the number of reported cases have increased significantly worldwide .One of the remarkable clinical characteristics of human fascioliosis in endemic areas is the relative absence of parasites cause bile duct obstruction or hepatic dysfunction . Early diagnosis and treatment might avoid expensive and risky procedures. The early detection gastrointestinal symptoms1n *some* cases are diagnosed during surgery**.** (Marcos *et al.*, 2005).

Fasciola may give rise to condition known as "halzoan" This is characterized by an acute irritation of the throat, which is due to the transient attachment of the fluke ingested in low liver. Wide variety of other mammals' could by infected by ingestion of contaminated vegetation. (Elhaj., 2001).

Man is seldom infected by the common liver fluke and number reported probably amount to not more than 300 cases .Man usually infects himself by eating water grass grown in water in which the snail are living and which is contaminated by the faces of the fluke infested cattle or sheep. During migration of immature fluke, eosinophilia is astage which may aid in diagnosis of human fasciolosis .And the symptoms in human patient including urticaria, jaundice, enlarged tender liver and eosinophilia. (Elhaj., 2001).

The prevention of human fascioliosis may be achieved through strict control of watercress and other metacercariae-carrying aquatic plants for human consumption, especially in endemic zones. Among vegetables incriminated freshwater plant species which may differ according to geographic zones and human dietary habits (Mas coma *et al*., 2005).

1.11. Economic importance:-

Fasciolosis also known as fascioliasis, distomatosis and liver rot, is an important helminthes disease caused by trematode species, *Fasciola hepatica* (the common live fluke) and *Fasciola gigantica. f. Hepatica* infects more than 300 million cattle and 250 million sheep worldwide and together with *F. gigantica*, causes significant economic losses to global agriculture; estimated at more than US$3 billion annually (Aliyu,*et al.*,2014*).*

Economic losses from liver flukes may result directly from increased liver condemnations at slaughter and indirectly from decreased livestock productivity. More economically important, beef producers are affected by increased culling of cows, reduced sale weights of culled cows, lowered reproductive performance in the brood cow herd, reduced Calf weaning weights, and reduced rates of growth in stockers, fluke-infected dairy cows produce less milk. Also liver flukes reduce animal fertility in addition liver flukes do affect sex hormone balance and metabolism (Kaplan *et al.*, 2001).

Fasciolosis causes a substantial economic loss which includes; death, loss in carcass weight, reduction in milk yield, predisposes animal to other disease and cost of treatment expenses (Mult *et al*., 2012)

1.12. Prevention and control:

The methods to control fasciolosis generally include strategic application of anthelmintics to eliminate the parasite from the host at the most convenient time for effective prevention of pasture contamination, reduction of the number of intermediate host snails through drainage and other practices and reduction of the chances of infection by efficient farm and grazing management. In fact control of fasciolosis requires intervention of relationships between the environment, ruminant hosts, snail hosts, the parasite life cycle, agricultural cycles and animal husbandry procedures. Therefore, good understanding of the environment-host-parasite inter relationships are essential for formulating the control measures suitable for an area (Mahato *et al.*; 2000)

The types of control measures depend on the setting (such as epidemiologic, ecologic, and cultural factors). Strict control of the growth and sale of watercress and other edible water plants is important (Rapsch *et al.,* 2008).

No vaccine is available to protect people against *Fasciola* infection. Individual people can protect themselves by not eating raw watercress and other water plants, especially from endemic grazing areas. As always, travelers to areas with poor sanitation should avoid food and water that might be contaminated, Vegetables grown in fields that might have been irrigated with polluted water should be thoroughly cooked .another Suitable control strategies, such as pasture management strategies could help to avoid some of these losses. Geographical information systems such as risk maps could help identify areas where disease monitoring should be established. Since F. hepatica transmission is linked to its intermediate host L. truncatula, information on suitable environmental conditions can help locate possible areas with enhanced infection risk by means of cartography.(Rapsch*etal*.,2008).

Destruction of inter- mediate host snail host by suitable molluscicides is adopted in many part of the world to use of molluscicide in defined water bodies such as dam, stream and lakes. Is often very expensive in term of labor and material dangerous the regular application of molluscicides in Three month sufficiently controls the disease since snail is killed before emitting cercaries. (Altahir., 1975.)

The anthelminthic drugs recommended by the World Health Organization (WHO) as essential drugs to treat these diseases, namely praziquantel and triclabendazole (TCZ), WHO recommends the inclusion of FBTs in the group of helminthic diseases whose control relies on the preventive chemotherapy concept, i.e. early administration of anthelminthic drugs, either alone or in combination, to infected individuals to prevent overt morbidity in later stages of life. Life Triclabendazole (TCZ) is the treatment of choice for fascioliosis and is effective at a single dose of 10 mg/kg body weight against the adult parasites in the bile ducts and immature flukes migrating through the liver (WHO, 2006).

Strategic liver fluke treatment of all cattle and buffaloes which are older than 8 months should be carried out once a year. In addition, animals in poor condition should be treated to prevent severe losses, especially in high prevalence areas or where strategic treatment was Problems of liver fluke control include the lack of knowledge about the parasite at farmer's level and the lack of availability of drug supplies at the village level, both of both]. Which are important to allow strategic treatment and control of animals (*Kuchai,et al.*,2011).

Various drug such as bithionol and praziguantel are used for treatment of human fasciolosis, but with variable result. Emetine and dehydroemetine are used frequently with significant efficacy in the treatment of human fasciolosis, but cause variety of serious and toxic side-effect. Preliminary studies on therapeutic effect of triclobendazole have demonstrated that this drug is highly effective in human chronic fasciolosis (Pedros *et al.*, 2000)

CHAPTER TWO

MATERIALS AND METHODS

2.1 Study area:

The study was conducted in Elfashir abattoir. Elfashir is the capital city of North Darfur state and it is one of the 17states of Sudan. North Darfur state is one of the 5 states composing the Darfur region (figure 1). It has an area of 296,420 km² and an estimated population of approximately 1,583,000. Elfasher town is located in northwestern Sudan, 120 miles (195 km) northeast of Nyala. A historical caravan centre, it is located at an elevation of about 2,400 feet (700 meters). (Department of statistics, Advance tabulation of fourth population Census of Sudan, Darfur, 1994).

2.2 The climate:

Northern Darfur State is characterized by desert and semi-desert climate. The rainy season usually starts in July and ends in October. July and August are the wettest months of the year with a total rainfall above 29% of the total annual rainfall (398 – 402 mm). The temperature reaches its maximum (37 to 41^{o}C) in the summer from March to June and drops to lowest readings (15 to 17^{o}C) in the winter during November to February. Annual measure relative humidity was estimated as 35.6% and reaches its peak (81.3%) in July and August (Department of statistics, Advance tabulation of fourth population Census of Sudan Darfur, 1994).

North Darfur located between 8^{o}: 15 and 20^{o}: 00 north and 22^{o}: 00 and 27^{o}: 30 East. The state area is about 500,000 km2 approximately. The desert part extend from north of parallel 16^{o}: 00 to 20^{o}: 00 and is about

145000 km2 and this represent 29% of the total area of region, (Department of statistics, Advance tabulation of fourth population Census of Sudan Darfur, 1994).

Elfashir town is located in north Darfur state between the parallel 12^{O}: 45 and14^{O}: 15 North and 29^{O}: 30 and 27^{O}: 15 East with total area of all, 2005).33275 km2 (North Darfur live stock work shop, Darfur services and information DSI, 2005).

2.3. The soil:-

Three types of soil are recognized in Darfur region. The continental sand, the soil of low fertility which spreads over much of Northern Darfur State and parts of Southern and Western Darfur States and the silt, which is the most fertile of all Darfur soils, is found along the banks of the valleys, km^2 (North Darfur live stock work shop, Darfur Development services and information DSI, 2005).

2.4. Sampling methods**: -** Systematic random sampling techniques was used the prevalence was calculated using formula described by (Martinet al., 1987) as follow:

$$\textbf{Prevalence rate} = \frac{\text{No. of Cattle with fasciolosis} * 100}{\text{Total no. of cattle at a particular point in time}}$$

Sample size determination:-

The sample size was calculated by the formula:-

$$N = \frac{4 * P * Q}{L2}$$

N= sample size

P= expected prevalence

L= desired absolute precision

Q= (1-P).(Martin et al.,1987)

The expected prevalence was estimated according to the study prevalence of bovine fasciolosis and related risk factors in Dessie Municipal abattoir , south Wollo Zone, Ethiopia (Belay *et al* ., 2012),which was estimated as 25.2%,The sample size was calculated as follow:-

$$N = \frac{4*(,252)*(0.748)}{(0.0025)} = 302 \text{ animals}$$

2.5. The study design:-

A cross-sectional epidemiological study was conducted at North Darfur state abattoir. The abattoir is located in the North El fasher .small ruminant and cattle were slaughtered, the target populations were cattle

2.5.1. Ante-mortem and post- mortem inspection:-

Regular visits were made by the investigator to conduct ante-mortem examination of slaughter animals. The cattle were selected by systematic random method and enumerate marks on it is body (tagged before slaughter). Total of 302 cattle were examined in the North Darfur abattoir. During the survey Period which extended from May2013 to August 2013. On the hand questionnaire was designed from animal and from owners. The information in questionnaire include: animal grazing (indoor/out door), source of animal, using drug, rain fall, water bodies and vegetation. During the ante-mortem inspection the ages, sex, breed and body condition of each individual animal were assessed and recorded. The age was divided to three categories: ≤ 3 years, 4-6 years and >6 years; sex to male and female; the cattle breed in two categories: local and cross breed.

Post-mortem examinations of liver and associated bile duct were carefully performed by visualization and palpation of the entire organ followed by transverse incision of the organ across the thin left lobe.

2.5.2 Fecal samples collection and examination

Fecal sample examination was carried out at the central laboratory in Elfasher at the Ministry of Animal Resource, North Darfur state. Fecal samples were collected directly from the rectum during ante mortem examination using plastic gloves. Specimens carried in a plastic container and transported to the laboratory for microscopic examination using sedimentation technique to detect the presence of *fasciola spp* eggs and identification was done on the basis of morphology of the egg. Water was added to feces into a container, mixed thoroughly then filtered through a tea strainer , the filtered material was poured into 15 ml centrifuge tube and centrifuged at 3000 rpm for 3minuts then removed from the centrifuge the tube had sediment, then the supernatant was discarded. Mixing of water and centrifuging until clear supernatant was obtained then a small drop of the sediment was transferred to slide microscope slide. Covered with cover slip and examined under the microscope at 10*10 magnifications.

2.5.3. Statistical analysis:

All the data were collected during the study period from the May 2013 to August 2013. analysis of the data was carried out using SPSS version16.Descriptive statistical analysis was used and illustrated in frequency tables., Unavailable analysis was performed by Chi square and illustrated in tables showing the risk factors, number of animal tested, percentage of the positive number, degree of freedom ,x^2 value and p-value. Risk factor with a p-value less than or equal 0.25 were entered in a multi- variate analysis using logistic regression to investigate association between bovine fasciolosis and potential risk factors. Result was illustrated in tables showing Exp B, 95% confidence interval and p-value. A p-value of 0.05 or less indicated significant association between bovine fasciolosis and the risk factors.

CHAPTER THREE

RESULTS

A total of 302 cattle were investigated for fasciolosis, 30 cattle were found to be infected with fasciolosis giving a prevalence rate of 9.9% (30/302) through meat inspection and 6 cattle through fecal examination giving prevalence rate of 1.9%(6/302).

Table 3.1: Prevalence of fasciolosis in 302 cattle examined by post-mortem examination.

Postmortem examination

		Frequency	Percent	Valid Percent	Cumulative Percent
Valid	negative	272	90.1	90.1	90.1
	positive	30	9.9	9.9	100.0
	Total	302	100.0	100.0	

Table 3.2: Prevalence of fasciolosis in 302 cattle examined by fecal samples for eggs.

Fecal examination

		Frequency	Percent	Valid Percent	Cumulative Percent
Valid	negative	296	98.1	98.1	98.1
	positive	6	1.9	1.9	100.0
	Total	302	100.0	100.0	

Table 3.3: Over- all prevalence of fasciolosis in 302 cattle examined by post-mortem and fecal sedimentation in North Darfur state.

Result	frequency	percent	Valid percent	Cumulative percent
negative	266	88.0%	88.0%	88.0%
postmortem	30	9.9%	9.9%	97.9%
Fecal examination	6	1.9%	1.9%	100.0%
total	302	100.0%	100.0%	

3.4. Post-mortem examination results:-

3.4.1. The effect of Age on fasciolosis:

The result of the study showed the distribution of 302 cattle examined for fasciolosis by age. Table 3.4 showed that 14.9% of examined cattle were young, 51.0% were middle age and 34.1% were adult. Total number of young animals was 45 animals. Among these no animal was found infected, the prevalence of fasciolosis was 0%. Total number of middle age animals was 145 animals .Among these 6 animals were found infected, the prevalence was 3.9% and total number of old animal of age was 103 .Among these, 24 animals were found infected, and the prevalence was 23.3% table (3.5).In the Chi-square test the results showed that there was significant association (p-value 0.000) between age and fasciolosis, table (3.7).

3.4.2. The effect of Sex on fasciolosis:

The result of study showed the distribution of 302 cattle's examined for fasciolosis by sex.Table3.4 showed that 22.2% of examined cattle were female and 77.8% were male .total number of female 67 animals' .Among these 14 animals was found infected. The prevalence of fasciolosis was 20.9%. Total number of male was 235 animals .Among these, 16 animals were found infected .the prevalence was 6.8% table (3.5) .In the Chi-square test, the results showed that significant association (p-value 0.001) between sex and fasciolosis, table (3.7).

3.4.3the effect of Breed on fasciolosis:

The result of the study showed that the distribution of 302 cattle examined for fasciolosis by breed. Table 3.4 showed that 92.4% of examined cattle were local, and 7.6% were cross. Total number of local breed was 279 animals. Among these 30 animals were found infected .the prevalence was 10.8%. Total number of cross breed was 23 animals. Among these, no animal was found infected, the prevalence rate was 0%table (3.5) .In the Chi-square test the results showed no significant association (p-value .098) between breed and fasciolosis, table (3.7).

3.4.4. The effect of Body condition on fasciolosis:

The result of the study showed that the distribution of 302 cattle examined by body condition. Table 3.4 showed that 97.4% of examined cattle were good body condition and 2.6% were poor body condition. Total number of good body condition was 294 animals .Among these, 30 animals were found infected, the prevalence was 10.7%. Total number of poor body condition was 8 animals .Among these no animal was found infected .the prevalence was 0% table (3.5).In the Chi-square test the result showed that, there was not significant association (p-value.341) between body condition and fasciolosis, table (3.7).

3.4.5. The effect of Source of animal on fasciolosis:

The result of the study showed that the distribution of 302 cattle examined by source of animal. Table 3.4 showed that 63.2% of examined cattle were from North Darfur and 36.8% from south Darfur. Total number from North Darfur was 191 animals .Among these, 11 animals was infected, prevalence was 5.7%. Total number of animal from South Darfur was 111 animals. Among these, 19 animals was infected, the prevalence was 17.2% table (3.5). In the Chi-square test the results showed significant association between source of animal and fasciolosis,(p-value 0.001),table (3.7).

3.4.6. The effect of Grazing on fasciolosis:

The result of the study showed that the distribution of302 cattle examined by grazing. Table 3.4 showed that 36.8% of examined cattle were indoor and 63.2% were out door. Total number of indoor was 191 animals. Among these, 12 animals was found infected, the prevalence of fasciolosis was 6.3%.Total number of outdoor was 111 animals. Among these, 18 animals was infected, the prevalence was 16.8% table (3.5). In the Chi-square test there result s was significant association (p-value 0.005) between grazing and fasciolosiss table (3.7).

3.4.7. The effect of use anthlemintics on fasciolosis:

The result of the study showed that the distribution of 302 cattle examined by using drug .Table 3.4 showed that 61.6% of examined cattle were animals used drug and 38.4%were animals not used drug .Total number of animals used drug was 186 animals. Among these, 9 animals were found infected, prevalence was 4.8%. Total number of animals not used drug was 161 animals. Among these, 12 animals was found infected, prevalence of fasciolosis was (18.1%) table (3.5) .In the Chi-square test the results showed that the significant association (p-value 0.007) between using drug and fasciolosiss table (3.7).

3.4.8. The effect of Rain falls on fasciolosis:

The result of the study showed that the distribution of 302 cattle examined by rain fall. Table 3.4 showed that 57.3% of examined cattle were in rain fall and 42.7% animals in no rain fall .Total number of animals in no rain fall were 173 animals. Among these, 8 animals was infected, the prevalence of fasciolosis was 57.3% .Total number ofanimals in rain fall was 192 animals. Among these, 22 animals was found infected .the prevalence of fasciolosis was 42.7%table (3.5) In the Chi-square test the results showed significant association (p-value 0.000)between rain fall and fasciolosiss table (3.7).

4.3.9. The effect Water body on fasciolosis:

The result of the study showed that the distribution of 302 cattle examined by water body. Table 3.4 showed that 57.3% of examined cattle were found in water body and 42.7% were in no water body. Total number of animals in no water body was 173 animals .Among these, 8 animals was infected, prevalence of fasciolosis was 4.6%.Total number of animals in water body was 192 animals. Among these 22 animals was infected,the prevalence of fasciolosis was 17.1% table (3.5) In the Chi-square test the results showed that there was significant association (p-value 0.000) between water body and fasciolosiss table (3.7).

3.4.10. The effect of Vegetation on fasciolosis:

The result of the study showed that the distribution of 302 cattle examined by vegetation. Table 3.4 showed that 57.3% of examined cattle were in vegetation and 43.0%were in no vegetation .Total number of animals in no vegetation was 172 animals. Among these 7 animals was infected, prevalence of fasciolosis was 4.1% Total number of animals in vegetation was 130 animals Among these, 23 animals were infected, the prevalence of fasciolosis was 17.6% table (3.5) In the Chi-square test the results showed that there was significant association between vegetation and fasciolosiss(p-value 0.000) table (3.7).

3.5. Fecal examination result:-

3.5.1. The effect of Age on fasciolosis:

The result of the study showed the distribution of 302 cattle examined for fasciolosis by age. Table 3.4 showed that 14.9% of examined cattle were young, 51.0% were middle and 34.1%were adult table. Total number of young animals was 45 animals. Among these, no animals was found infected, the prevalence of fasciolosis was 0%.Total number of middle age was 154 animals. Among these, no animal was found infected .the prevalence of fasciolosis was 0% and Total number of adult age was103 animals. Among these, 6 animals was infected, the prevalence of fasciolosis was 5.8% table (3.6). In the Chi-square test the results showed that there was significant association (p-value 0.003) between age and fasciolosis, table (3.8).

3.5.2. The effect of Sex on fasciolosis:

The result of study showed the distribution of 302 cattle examined for fasciolosis by sex Table 3.4 showed that 22.2%of examined cattle were female and 77.8% were male. Total number of female was 67 animals .Among these, 3 animals was found infected, prevalence of fasciolosis was 4.5% .Total number of male was 235 animals. Among these, 3 animals were infected. The prevalence of fasciolosis 1.3% table (3.6) .In the Chi-square test, there was no significant association (p-value 098) between sex and fasciolosis, table (3.8).

3.5.3. The effect of Breed on fasciolosis:

The result of the study showed that the distribution of 302 cattle examined for fasciolosis by breed. Table 3.4 showed that 92.4% of examined cattle were local, and 7.6% were cross. Total number of local breed was 279 animals .Among these, 6 animals were infected, prevalence of fasciolosis was 2.2% .Total of number of cross breed was 23 animals. Among these, no animal was found infected, prevalence of fasciolosis 0% table 3.6 .In the Chi-square test, there was no significant association (p-value .477) between breed and fasciolosis, table (3.8).

3.5.4. The effect of Body condition on fasciolosis:

The result of the study showed that the distribution of 302 cattle examined by body condition. Table 3.2 showed that 97.4% of examined cattle were good body condition and 2.6%were poor body condition. Total numbers of 294 animals were found good body condition. Among these, 6 animals were infected, prevalence of fasciolosis was 2.1% .Total number of poor body condition were 8 animals. among these, no animal was found infected, The prevalence of fasciolosis was 0% .table(3.6).In the Chi-square test, there was no significant association (p-value.683) between body condition and fasciolosis, table (3.7).

3.5.5. The effect of Source of animal on fasciolosis:

The result of the study showed that the distribution of 302 cattle examined by source of animal. Table 3.4 showed that 63.2% of examined cattle were from North Darfur and 36.2% were from south Darfur. Total numbers of animals from North Darfur were 191animals. Among these, 1 animal was infected; prevalence of fasciolosis was 0.6%. Total numbers of animals from South Darfur were 111 animals. Among these, 5 animals were infected; the prevalence of fasciolosis was 4.5%. Table (3.6). In the chi-square there was significant association (p-value= 0.017) between source of animal and fasciolosistable (3.8).

3.5.6. The effect of Grazing on fasciolosis:

The result of the study showed that the distribution of 302 cattle examined by grazing .Table 3.4 showed that 36.8% of examined cattle were from indoor and 63.2% were from out door. Total numbers of animals from indoor were 191animals. Among these, 2 animals were infected, prevalence of fasciolosis was 1.1% .Total number of animals from outdoor were 111 animals. Among these, 4 animals were infected; the prevalence of fasciolosis was 3.6%. Table (3.6) In the Chi-square test there was not significant association (p-value =.125) between grazing and fasciolosiss table (3.8).

3.5.7. The effect of use of anthlemintics on fasciolosis:

The result of the study showed that the distribution of 302 cattle examined by using drug. Table 3.4 showed that 61.6%of examined cattle were used drug and 38.4% not used drug. Total of 186 animals were used drug .Among that 1 animal was infected, prevalence of fasciolosis was 0.5%.Total of 116 animals were not used drug .Among these, 5 animals were infected, the prevalence of fasciolosis was 4.4%. Table (3.6) In the Chi-square test there was significant association (p-value 0.024) between using drug and fasciolosiss table (3.8).

3.5.8. The effect of Rain falls on fasciolosis:

The result of the study showed that the distribution 302 cattle examined by rain fall .Table 3.4 showed that 57.3% of examined cattle were in rain fall and 42.7%were in no rain fall. Total number of 173 animals were in no rain fall .Among these 1 animal was infected, prevalence of fasciolosis was 0.6%.Total of 129 animals were in the rain fall .Among these 5 animals were infected, prevalence of fasciolosis was 3.9% . Table (3.6) In the Chi-square test there was significant association (p-value 0.024) between rain fall and fasciolosiss) table (3.8).

3.5.9. The effect of Water body on fasciolosis:

The result of the study showed that the distribution of 302 cattle examined by water body. Table 3.4 showed that 57.3% of examined cattle were in water body and 42.7% were in no water body. total of 173 animals were in no water body .Among these 1 animal was infected, prevalence of fasciolosis was 0.6% .total of 129 animals were in water body .Among these, 5 animals was infected . Prevalence of fasciolosis was 3.9%. Table (3.5) In the Chi-square test there was significant association (p-value 0.042) between water body and fasciolosiss table (3.8).

3.5.10. The effect of Vegetation on fasciolosis:

The result of the study showed that the distribution of 302 cattle examined by vegetation .Table 3.4 showed that 57.3% of examined cattle were in vegetation and 43.0 %were in no vegetation. Total of 172 animals were in no vegetation. Among these, 1 animal was infected; prevalence of fasciolosis was 0.6%.Total of 130 animals was in vegetation .Among these, 5 animals were infected, the prevalence of fasciolosis was 3.8%. Table (3.6) In the Chi-square test there was significant association (p-value 0.044) between vegetation and fasciolosiss table (3.8).

Table 3.4 `: Frequency distribution of 302 cattle examined for bovine fasciolosis in North Darfur state according to potential risk factors investigated

Risk factor	frequency	percent	Cumulative percent
Age (years)			
Young(≤ 3years)	45	14.9%	14.9%
Middle (4-6years)	153	51.0%	65.9%
Old (> 6 years)	103	34.1%	100.0%
Sex			
Female	67	22.2%	22.2%
male	235	77.8%	100.0%
Breed			
Local	279	92.4%	92.4%
Cross	23	7.6%	100.0%
Body condition			
Good	294	97.4%	97.4%
poor	8	2.6%	100.0%
Source of animal			
North Darfur	191	63.2%	63.2%
South Darfur	111	36.8%	100.0%
Grazing			
Indoor	191	63.2%	63.2%
outdoor	111	36.8%	100.0%
Use anthlemintics			
Yes	186	61.6%	61.6%
No	116	38.8%	100.0%
Rain fall			
low	173	57.3%	57.3%
high	129	43.7%	100.0%
Water body			
No	173	57.3%	57.3%
Yes	129	43.7%	100.0%
Vegetation			
poor	172	57.0%	57.0%
good	130	43.0%	100.0%

Table 3.5: Cross tabulation of bovine fasciolosis diagnosed by post-mortem with potential risk factors in 302 cattle slaughtered at Elfasher abattoir, North Darfur

Risk factors	No tested	No positive	percent
Age(years)			
Young(≤3years)	45	0	0%
Middle(4-6yars)	154	6	3.9%
Old(>6years)	103	24	23.3%
Sex			
Female	67	14	20.9%
Male	235	16	6.8%
Breed			
Local	279	30	10.8%
Cross	23	0	0%
Body condition			
Good	294	30	10.7%
Poor	8	0	0%
Source of animal			
North Darfur	191	11	5.7%
South Darfur	111	19	17.2%
Grazing			
Indoor	191	12	6.3%
Outdoor	111	18	16.8%

Table 3.5: continued

Risk factors	No tested	No positive	percent
Use of anthlemintics			
Yes	186	9	4.8%
No	161	21	17.1%
Rain fall			
low	173	8	4.6%
high	129	22	17.1%
Vegetation			
poor	173	8	4.6%
good	129	22	17.1%
Water body			
No	172	7	4.1%
Yes	130	23	17.6%

Table 3.6: Cross tabulation of bovine fasciolosis diagnosed by fecal sedimentation with potential risk factors in 302 cattle slaughtered at Elfasher abattoir, North Darfur

Risk factor	No tested	No positive	Percent%
Age(years)			
Young(≤3years)	45	0	0%
Middle(4-6years)	154	0	0%
Old(>6years)	103	6	5.8%
Sex			
Female	67	3	4.5%
Male	235	3	1.3%
Breed			
Local	279	6	2.2%
Cross	23	0	0%
Body condition			
Good	294	6	2.1%
poor	8	0	0%
Source of animal			
North Darfur	191	1	0.6%
South Darfur	111	5	4.5%
Grazing			
Indoor	191	2	1.1%
outdoor	111	4	3.6%

Table 3.6: continued

Risk factor	No. tested	No. Positive	Percent (%)
Use of anthelmintics			
Yes	186	1	0.5%
No	116	5	4.4%
Rain fall			
low	173	1	0.6%
high	129	5	3.9%
Water body			
No	173	1	0.6%
Yes	129	5	3.9%
Vegetation			
poor	172	1	0.6%
good	129	5	3.8%

Table 3.7: Univariate analysis for the association between fasciolosis diagnosed by post-mortem examination and potential risk factors in 302 cattle examined in North Darfur state using the Chi-square test

Risk factor	Total No	No. positive	Percent (%)	df	X^2	p-value
Age(years)				2	31.808	0.000
Young(≤3years)	45	0	0%			
Middle(4-6)	154	6	3.9%			
Old(>6years)	103	24	23.3%			
Sex				1	11.564	0.001
Female	67	14	20.8%			
Male	235	16	6.8%			
Breed				1	2.746	.098
Local	279	30	10.7%			
Cross	23	0	0%			
Body condition				1	.906	.341
Good	294	30	10.2%			
Poor	8	0	0%			
Source of animal				1	10.122	0.001
North Darfur						
South Darfur	191	11	5.7%			
	111	19	17.1%			
Grazing				1	7.742	0.005
Indoor	191	12	6.3%			
Outdoor	111	18	16.2%			

Table 3.7: continued

Risk factor	Total No	No. positive	Percent (%)	df	X^2	p-value
Use of anthelmintics				1	7.254	0.007
yes	189	12	6.4%			
no	113	18	15.9%			
Rain fall				1	12.761	0.000
low	172	8	4.6%			
high	130	22	16.9%			
Water body				1	12.761	0.000
no	172	8	2.3%			
yes	130	22	16.9%			
Vegetation				1	15.357	0.000
poor	172	7	5.5%			
good	130	23	17.6%			

Table 3.8: Univeriate analysis for the association between fasciolosis diagnosed by fecal sedimentation and potential risk factors in 302 cattle examined in North Darfur state using the Chi-sugars test

Risk factor	Total No	No. positive	Percent (%)	d.f.	X^2	p-value
Age(years)				2	11.827	0.003
Young (≤ 3years)	45	0	0%			
Middle(4-6years)	154	0	0%			
Old> 6years)	103	6	5.8%			
Sex				1	2.743	.098
Female	67	3	4.5%			
Male	235	3	1.3%			
Breed				1	.505	.477
Local	279	6	2.3%			
Cross	23	0	0%			
Body condition				1	.167	.683
good						
poor	294	6	2.0%			
	8	0	0%			
Source of animal				1	5.713	0.017
north Darfur	191	1	0.5%			
South Darfur	111	5	4.5%			
Grazing				1	2.356	.125
Indoor	191	2	1.04%			
Outdoor	111	4	3.6%			

Table 3.8: continued

risk factor	Total No	No. positive	Percent (%)	d.f.	X	p-value
Grazing				1	2.356	.125
Indoor	191	2	1.04%			
Outdoor	111	4	3.6%			
Use of anthelmintics				1	5.222	0.022
Yes	186	1	0.5%			
No	116	5	4.3%			
Rain fall				1	4.127	0.042
low	173	1	0.6%			
high	129	5	3.8%			
Water body				1	4.127	0.042
No	173	1	0.5%			
Yes	129	5	3.8%			
Vegetation				1	4.053	0.044
poor	172	1	0.7%			
good	130	5	3.8%			

3.9. Results of multivariate analysis:

3.9.1Post mortem examination: potential risk factors found to be significantly(p-value ≤ 0.25) associated with fasciolosis in the unvariate analysis (table3.7) were entered to the multivariate analysis using logistic regression, in the final model .seven risk factors were found to be significantly associated(p-value<0.05) with fasciolosis. These risk factors included Age (p-value 0.00), sex (p-value=0 .011), source of animal (p-value =0.001), grazing (p-value=0 .005), using drug (p-value=0 .007), rain fall (p-value 0.000), vegetation (p-value0.000)

3.9.2. Fecal examination:

Potential risk factors found to be significantly associated (p-value ≤ 0.25) with fasciolosis . in the univariate analysis (table3.8) were entered to logistic regression In the final model five risk factor were found significantly associated (p-value≤0.05) with fasciolosis in the multivariate analysis. These risk factors included age (p-value =0.003), source of animal (p-value =0.017), using drug (p-value=0 .022), rain fall (p-value=0 .042), and vegetation (p-value =0.044)

Table 3.9: Multivariate analysis for the association between fasciolosis diagnosed by post-mortem examination and potential risk factors in 302 cattle examined in North Darfur state using logistic regression.

Risk factor	No. positive	No. positive (%)	Exp(B)	p-value	95% CI
Age(years)				.000	
Young(≤ 3years)	0	0(0%)	ref		
Middle(4-6years)	6	6(3.9%)	.94		1.20 - 4.30
Old(> 6years)	24	24(23.3%)	30.301		0 -5.17
Sex				0.001	
Male	16	16(6.8%)	ref		
Female	14	14(20.9%)	.465		.182-1.189
Breed				.098	
Cross	0	0 (0%)	ref		
Local	30	30 (10.8%)	.000		
Source of animal				0.001	
North Darfur	11	11(5.7%)	ref		
South Darfur	19	19(17.2%)	.890		.240-3.300
Grazing				0.005	
Indoor	12	12(6.3%)	ref		
Out door	18	18(16.8%)	.390		.078-1.956

Table 3.9: continued

Risk factor	No. positive	No. Positive (%)	Exp(B)	p-value	95%CL
Use of anthelmintics				0.007	
Yes	12	12(4.8%)	ref		
No	18	18(13.1%)	2.174		.455-10.4
Rainfall				0.000	
low	8	8(4.6%)	ref		
high	22	22(17.1%)	.000		
Vegetation				0.000	
poor	7	7(4.1%)	ref		
good	23	23(17.6%)	52.155		

Table 3.10: Multivariate analysis of the association between fasciolosis diagnosed by fecal sedimentation and potential risk factor association in 302 castles examined in North Darfur state using logistic regression

Risk factor	No. positive	No. positive (%)	Exp(B)	p-value	95% CL
Age(years)				0.003	
Young(≤3years)	0	0(0%)	ref		
Middle(4-6years)	0	0(0%)	.725		.
Old(>6)	6	6(5.8%)	42.264		
Sex				.098	
Male	3	3(1.3%)	ref		
Female	3	3(4.5%)	.561		.099-3.198
Source of animal				0.017	
North Darfur					
South Darfur	1	1(0.6%)	ref		
	5	5(4.5%)	2.765		.186-41.20
Grazing				.125	
Indoor	2	2(1.1%)	ref		
Outdoor	4	4(3.6%)	.46		.099-2.468
Use of anthelmintics				0.022	.216-92.002
Yes	1	1(0.5%)	ref		
No	5	5(4.4%)	4.462		

Table 3.10: continued

Risk factor	No. positive	No. positive (%)	Exp(B)	p-value	95%CL
Rainfall				0.042	
low	1	1(0.6%)	Ref		
high	5	5(3.9%)	67.594		
Vegetation				.044	
poor	1	1(0.6%)	Ref		
good	5	5(3.8%)	.000		

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CHAPTER FOUR

DISCUSSION

Liver fluke is a common trematode (flat worm) that causes a parasitic disease called fascioliasis or fluke disease. It has a pathogenic effect on ruminants (cattle, sheep and goats) as well as other farm animals like horses, donkeys, pigs and to a lesser extent poultry Species of liver fluke include *Fasciola gigantiga* and F. *hepatica*, with the former being more prevalent in cattle and the latter in sheep and goats (Ozung *et al*., 2011).

Previous studies on animal fasciolosis revealed that the disease is prevalent in the Sudan, such as Darfur, upper Nile, Khartoum, Bahr ElGazal, Equatorial Blue Nile, Kassala as well as North State .(Koko *et al* .,2003). The annual reports since1955 indicate that highest percentages of liver condemnation due fascioliases are follows:

Upper Nile province (27-58%),Baher El Gazal province (16-18%),Equatorial province(13-40%) .In all area in Sudan fascioliasis has been reported in several places with high prevalence (Intisar, 1992).

This study was carried out in North Darfur state to investigate risk factors and to estimate prevalence of bovine fasciolosis in North Darfur state .The prevalence rate was 9.9% by post mortem examination and1.9% by fecal examination for f*asciola* agg using sedimentation techniques.

The prevalence rate of post -mortem examination9.9% is higher than prevalence rate of fecal examination 1.9%,because some infected animal might have been missed by sedimentation technique which is characteristically apoor in detection of fluke (Pfukeny *et a*l.,2006)

Also this variation could probably be due to the fact that in abattoir study the liver is usually damaged by immature fluke infected which cannot be detected by fecal examination. In addition, most cattle that are infected with flukes shed relatively few eggs and the recently infection are found only from 8 week after infection (Pfukeny *et al*., 2006).

However the prevalence rate in this study (9.9%) was lower than many other studies from different abattoirs n in Gezira state, Central Sudan by Koko *et al*., (2003) where the prevalence was12.5% or in Africa, such as from Nekemte municipal abattoirs the prevalence rate was 21.9% (Petros *et al*., 2013) and from Jimma municipality abattoirs where the prevalence rate was 48.19% (Fromsa et *al*., 2011).

This variation in result could be due to the geographical location and ecological condition such as altitude, rain fall and temperature, *fasciola spp* prevalence has been reported to vary over the years mainly due to variation in amount and pattern of rain fall. The Bahr el Ghazal province of Sudan was also reported to have a high prevalence of fasciolosis than in the more open and dry savannah Darfur province (Pfkeny *et al*., 2006).

In this study, the age was found significantly associated with fasciolosis the p-value is (0.000) in post mortem examination. The prevalence rate is higher in old animals and middle age than young animals. (23.3%, 3.9%, 0%) respectively. this result is in agreement with previous study in Debre Zait town, Ethiopia by Abdulhkim *et a*l., (2012).

However, the result does not agree with another study in Ladakh by Kuchai *et al*., (2011) who found higher prevalence in young animals, In this study, the age was found significantly associated with fasciolosis the (p-value is 0.003) in fecal examination. The prevalence rate was higher in old animals and middle age than young animals, The prevalence rate of fecal examination was (5.8%,0%, 0%) respectively, These results are in agreement with previous study in Zimbabwe by pfukeny *et al* (2006) who found high infection rate in old animal ,the(p-value is.<0.001) , The higher infection rate in older animal was reported to be probably due to longer exposure time, or due to management system with longer exposure of old animals outdoor while young animals are kept indoor (Pfkeny *et al*., 2006).

Also this study is in agreement with another study in Kayseri province, Turkey by Yildrim et al., (2007) who found high infection rate in old animals. However, our results do not agree with another study in Ginnir District by Fekadu *et al*., (2012), where a significant difference was not observed between age groups (p-value > 0.05).

In this study, sex was found significantly associated with fasciolosis in post mortem examination (p- value =0.001). This is in agreement with aprevious study in Assela, Ethiopia by Mulugeta .,*et al* (2011) who found significant association with fasciolosis (p-value <0.05). However, this study does not agree with study in Nekemte Municipal abattoir by Petros *et al*., (2013) who did not find significant association with fasciolosis (p-value > 0.05).

The prevalence rate of fasciolosis in female was higher (%20.9) than the prevalence rate of male (%6.8). This variation in our opinion may be due to the fact that female exposed to stress after parturition and this reason decrease immunity.

Also in this study, the sex was not found significantly associated with fasciolosis (p-value=0.098), in fecal examination .This finding is not in agreement with a study carried out around Assela by Mulugeta *et al., (*2011) who found significant association between sex and fasciolosis

.also in this study, source of animal was found significant association with fasciolosis (p-value=0.001), in post- mortem examination .This finding is in agreement with a study carried out in southern Espirato Santo by Bernardo *et al.*(2011).However, this study does not agree with a previous study in Sowth Wollo zone, Ethiopia by Bely *et al.,* (2012).

The prevalence rate of fasciolosis in south Darfur (%17.2) was higher than the prevalence rate of North Darfur (%5.7). This variation can be attributed mainly to the variation in the climate .south Darfur has higher rain fall than North Darfur and a higher presence of inter mediate host.

. Also in thisstudy revealed that grazing type was significant association with fasciolosis the p-value =0.005), in post-mortem examination. A higher prevalence of fasciolosis in outdoor cattle (%16.2) as compared with animals in indoor (%6.2) this variation in our opinion may be due to the fact that animal grazes outdoor is more exposed to the disease from pasture, dams, ponds than animal kept indoor.

In this study, use of anthelmenticswas found significant association with fasciolosis (p-value 0.007), (0.022) in post- mortem examination and fecal examination respectively. The prevalence is higher in animals whose owner did not used drug (%18.1) than in animals whose owner used drug4.4%, and the prevalence in fecal examination was 4.4% and 0.5%respectively. This variation in our opinion may be due to the fact that animals treated routinely with anthlementics are less affected by bovine fasciolosis.

In this study there was highly significant association between rain fall and fasciolosis (p-value=0.000), in post-mortem examination. The prevalence of animal in area of high rain fall was 17.1% and 4.6% in area of low rain fall.

Also in this study, there was significant association between rain fall and fasciolosis (p-value=0.042), in fecal examination. This is in agreement with a previous study in Zimbabwe by Pfukeny *et al.*, (2006).The prevalence of animal in area of high rain fall was 3.9% and 0.6% in area of low rain fall. This variation may be due to the fact that high rain fall is favorable ecology factor for the presence of snail intermediate host, (Pfukny et al., 2006).

In this study there was significant association between vegetation and fasciolosis (p-value=0.000), in post mortem examination. The prevalence of animal in area with good vegetation was 17.6% and 4.1% in area with poor vegetation; this variation in our opinion may be due to the fact that good vegetation was favorable condition for snail intermediate host

In this study there was significant association between vegetation and fasciolosis (p-value=0.042), in fecal examination for *fasciola* agg. The prevalence of animal infested with fasciolosis in area with good vegetation was 3.8% and 0.6% in area with poor vegetation; this is in agreement with other study in Zimbabwe by Pfukeny *et a*l, (2006).

In this study there was seven risk factors found to be significantly associated (p-value <o.o5) with bovine fasciolosis in the multivariate analysis. These seven risk factors included: age, sex, source of animal, grazing, using drug, rain fall and vegetation.

4.1. Conclusion

In view of the findings fasciolosiss is prevalent in North Darfur state. the study further confirmed that fasciolosis diagnosed through post-mortem examination is more prevalent than by fecal sedimentation. The overall prevalence was 12.1%. Old Animals were more affected compared to young animals. The prevalence of fasciolosis was higher in high rain fall and good vegetation area compared to animals in low rain fall and poor vegetation. The prevalence of fasciolosis was higher in animals which they graze outdoor than animal which graze indoor. Prevalence of fasciolosis was higher in animals whose owners did not use anthelmintics compared to those their owners used anthelmintics.

4.2. Recommendations:

- Knowledge about the disease epidemiology in the country; should be varios from place to another according to the agricultural and ecological variation.

- Awareness of animal owner's about the disease, treatment and the control strategy is vital.

Destruction of inter mediate- host by suitable molluscicides

-Vegetables grown in fields should be thoroughly cooked to reduce infection of fasciolosis in human.

- Prophylactic use of anthelmintics during the rainy season seen to be important

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Appendix 1

Frequency table of 302 cattle diagnosed from North Darfur state according to risk factors.

Appendix1.1: age

age

		Frequency	Percent	Valid Percent	Cumulative Percent
Valid	young	45	14.9	14.9	14.9
	middle	154	51.0	51.0	65.9
	old	103	34.1	34.1	100.0
	Total	302	100.0	100.0	

Appendix1.2: sex

sex

		Frequency	Percent	Valid Percent	Cumulative Percent
Valid	female	67	22.2	22.2	22.2
	male	235	77.8	77.8	100.0
	Total	302	100.0	100.0	

Appendix1.3: breed

breed

		Frequency	Percent	Valid Percent	Cumulative Percent
Valid	local	279	92.4	92.4	92.4
	cross	23	7.6	7.6	100.0
	Total	302	100.0	100.0	

Appendix1:4

Body condition

		Frequency	Percent	Valid Percent	Cumulative Percent
Valid	good	294	97.4	97.4	97.4
	poor	8	2.6	2.6	100.0
	Total	302	100.0	100.0	

Appendix1. 5: source of animal.

Source of animal

		Frequency	Percent	Valid Percent	Cumulative Percent
Valid	north Darfur	191	63.2	63.2	63.2
	south Darfur	111	36.8	36.8	100.0
	Total	302	100.0	100.0	

Appendix 1.6: grazing.

grazing

		Frequency	Percent	Valid Percent	Cumulative Percent
Valid	indoor	191	63.2	63.2	63.2
	out door	111	36.8	36.8	100.0
	Total	302	100.0	100.0	

Appendix1.7: use of anthelmintics

Use of anthelmintics

		Frequency	Percent	Valid Percent	Cumulative Percent
Valid	yes	186	61.6	61.6	61.6
	no	116	38.4	38.4	100.0
	Total	302	100.0	100.0	

Appendix1.8: rain fall

rainfall

		Frequency	Percent	Valid Percent	Cumulative Percent
Valid	low	173	57.3	57.3	57.3
	high	129	42.7	42.7	100.0
	Total	302	100.0	100.0	

Appendix 1.9: water body.

Water body

		Frequency	Percent	Valid Percent	Cumulative Percent
Valid	no	173	57.3	57.3	57.3
	yes	129	42.7	42.7	100.0
	Total	302	100.0	100.0	

Appendix 1.10: vegetation.

vegetation

		Frequency	Percent	Valid Percent	Cumulative Percent
Valid	poor	172	57.0	57.0	57.0
	good	130	43.0	43.0	100.0
	Total	302	100.0	100.0	

Appendix 2

Cross tabulation table of 302 cattle diagnosed by post- mortem examination in North Darfur state for fasciolosis and risk factors.

Appendix 2.1: age

		age			Total
		Young	middle	old	
result	negative	45 100%	148 96.1%	79 76.7%	272 90.0%
	positive	0 0%	6 3.9%	24 23.3%	30 9.9%
Total		45	154	103	302

Appendix 2.2: sex

		sex		Total
		Female	male	
result	negative	53 79.1%	219 93.1%	272 90.1%
	positive	14 20.9%	16 6.8%	30 9.9%
Total		67	235	302

Appendix 2.3:breed

		breed		Total
		Local	cross	
result	negative	249 89.2%	23 100%	272 90.1%
	positive	30 10.8%	0 0%	30 9.9%
Total		279	23	302

Appendix 2.4: body condition.

		Body condition		Total
		Good	poor	
result	negative	264 94.6%	8 100%	272 90.1%
	Positive	30 10.7%	0 0%	30 9.9%
Total		279	8	302

Appendix 2.5:source of animal

		Source of animal		Total
		north Darfur	south Darfur	
result	negative	180 94.3%	92 82.8%	272 90.1%
	positive	11 5.7%	19 17.2%	30 9.9%
Total		191	111	302

Appendix 2.6:Grazing

		grazing		Total
		Indoor	out door	
Result	negative	179 93.7%	93 83.7%	272 90.1%
	positive	12 6.3%	18 16.2%	30 9.9%
Total		191	111	302

Appendix 2.7:use of anthelmintics

		using anthelmintics		
		Yes	no	Total
result	negative	177 95.2%	95 81.8%	272 90.1%
	positive	9 4.8%	21 18.1%	30 9.9%
Total		186	116	302

Appendix2.8: Rain fall.

		rainfall		
		Low	high	Total
Result	negative	165 95.4%	107 82.9%	272 90.1%
	positive	8 4.6%	22 17.1%	30 9.9%
Total		173	129	302

Appendix 2.9:water body

		water body		
		No	yes	Total
result	negative	165 95.4%	107 82.9%	272 90.1%
	positive	8 4.6%	22 17.1%	30 9.9%
Total		173	129	302

Appendix 2.10:vegetation

		vegetation		
		Poor	good	Total
pr	negative	165 95.9%	107 82.3%	272 90.1%
	positive	7 4.1%	23 17.6%	30 9.9%
Total		172	130	302

Appendix 3.

Cross tab table of 302 cattle diagnosed by fecal examination in North Darfur state according to risk factors

Appendix 3.1:age

		age			Total
		Young	middle	old	
Result	negative	45 100%	154 100%	97 94.2%	296 98.1%
	positive	0 0%	0 0%	6 5.8%	6 1.9%
Total		45	154	103	302

Appendix 3.2:sex

		sex		Total
		Female	male	
Result	negative	64 95.5%	232 98.7%	296 98.1%
	positive	3 4.5%	3 1.3%	6 1.9%
Total		67	235	302

Appendix 3.3: breed.

		breed		
		Local	cross	Total
Result	negative	273 97.8%	23 100%	296 98.1%
	positive	6 2.2%	0 0%	6 1.9%
Total		279	23	302

Appendix 3.4: body condition**.**

		Body condition		
		Good	poor	Total
Result	negative	288 97.9%	8 100%	296 98.1%
	positive	6 2.1%	0 0%	6 1.9%
Total		294	8	302

Appendix 3.5: source of animal.

		source of animal		
		north Darfur	south Darfur	Total
Result	negative	190 99.4%	106 95.5%	296 98.1%
	positive	1 0.6%	5 4.5%	6 1.9%
Total		191	111	302

Appendix 3.6: grazing.

		grazing		
		Indoor	out door	Total
Result	negative	189 98.9%	107 96.4%	296 98.1%
	positive	2 1.1%	4 3.6%	6 1.9%
Total		191	111	302

Appendix 3.7: use of anthelmintics

		using anthelmintics		
		Yes	no	Total
Result	negative	185 99.5%	111 95.6%	296 98.1%
	positive	1 0.5%	5 4.4%	6 1.9%
Total		186	116	302

Appendix 3.8: Rain fall.

		rainfall		
		low	high	Total
Result	negative	172 99.4%	124 96.1%	296 98.1%
	positive	1 0.6%	5 3.9%	6 1.9%
Total		173	129	302

Appendix 3.9: water body.

		water body		Total
		No	yes	
result	negative	172 99.4%	124 96.1%	296 98.1%
	positive	1 0.6%	5 3.9%	6 1.9%
Total		173	129	302

. Appendix 3.10: vegetation.

		vegetation		Total
		Poor	good	
Result	negative	171 99.4%	125 96.2%	296 98.1%
	positive	1 0.6%	5 3.8%	6 1.9%
Total		172	130	302

Appendix 4

Association of different potential risk factors with fasciolosis diagnosed by post-mortem examination using chi-square test$(x)^{2}$.

Appendix 4.1: Age.

	X^2	df	p-value
Pearson Chi-Square	31.808[a]	2	.000
Likelihood Ratio	32.929	2	.000
N of Valid Cases	302		

Appendix 4.2:sex

	Value	df	p-value
Pearson Chi-Square	11.564[a]	1	.001
Continuity Correction[b]	10.043	1	.002
Likelihood Ratio	9.917	1	.002
Fisher's Exact Test			
N of Valid Cases	302		

Appendix 4.3: breed.

	X^2	df	p-value
Pearson Chi-Square	2.746[a]	1	.098
Continuity Correction[b]	1.676	1	.196
Likelihood Ratio	5.017	1	.025
Fisher's Exact Test			
N of Valid Cases	302		

Appendix 4.4:Body condition

	X^2	df	p-vale
Pearson Chi-Square	.906[a]	1	.341
Continuity Correction[b]	.125	1	.724
Likelihood Ratio	1.698	1	.193
Fisher's Exact Test			
N of Valid Cases	302		

Appendix 4.5: source of animal.

	X^2	df	p-value
Pearson Chi-Square	10.122[a]	1	.001
Continuity Correction[b]	8.892	1	.003
Likelihood Ratio	9.702	1	.002
Fisher's Exact Test			
N of Valid Cases	302		

Appendix 4.6: grazing.

	x2	df	p-value
Pearson Chi-Square	7.742[a]	1	.005
Continuity Correction	6.672	1	.010
Likelihood Ratio	7.424	1	.006
Fisher's Exact Test			
N of Valid Cases	302		

Appendix 47: using anthlemintics.

	X^2	df	p-vale
Pearson Chi-Square	14.050[a]	1	.007
Continuity Correction	12.607	1	.000
Likelihood Ratio	13.673	1	.000
Fisher's Exact Test			
N of Valid Cases	302		

Appendix 4.8: Rain fall.

	X^2	df	p-value
Pearson Chi-Square	12.761[a]	1	.000
Continuity Correction[b]	11.410	1	.001
Likelihood Ratio	12.824	1	.000
Fisher's Exact Test			
N of Valid Cases	302		

Appendix 4.9:water body:

	X^2	df	p-vale
Pearson Chi-Square	12.761[a]	1	.000
Continuity Correction[b]	11.410	1	.001
Likelihood Ratio	12.824	1	.000
Fisher's Exact Test			
N of Valid Cases	302		

Appendix 4.10:vegetation

	X^2	df	p-vale
Pearson Chi-Square	12.761[a]	1	.000
Continuity Correction[b]	11.410	1	.001
Likelihood Ratio	12.824	1	.000
Fisher's Exact Test			
N of Valid Cases	302		

Appendix5

Association of different potential risk factors with fasciolosis diagnosed by fecal examination using chi-square test(x) $^{2.}$

Appendix 5.1:age

	X^2	df	p-vale)
Pearson Chi-Square	11.827[a]	2	.003
Likelihood Ratio	13.145	2	.001
N of Valid Cases	302		

Appendix 5.2: sex

	X^2	df	P-value
Pearson Chi-Square	2.743[a]	1	.098
Continuity Correction[b]	1.346	1	.246
Likelihood Ratio	2.277	1	.131
Fisher's Exact Test			
N of Valid Cases	302		

Appendix 5.3: breed

	X^2	df	p-value
Pearson Chi-Square	.505[a]	1	.477
Continuity Correction[b]	.000	1	1.000
Likelihood Ratio	.961	1	.327
Fisher's Exact Test			
N of Valid Cases	302		

Appendix 5.4: body condition

	X^2	df	p-value
Pearson Chi-Square	.167[a]	1	.683
Continuity Correction[b]	.000	1	1.000
Likelihood Ratio	.325	1	.568
Fisher's Exact Test			
N of Valid Cases	302		

Appendix 5.5: source of animal

	X^2	df	p-value
Pearson Chi-Square	5.713[a]	1	.017
Continuity Correction[b]	3.852	1	.050
Likelihood Ratio	5.632	1	.018
Fisher's Exact Test			
N of Valid Cases	302		

Appendix 5.6: grazing

	X^2	df	p-value
Pearson Chi-Square	2.356[a]	1	.125
Continuity Correction[b]	1.226	1	.268
Likelihood Ratio	2.249	1	.134
Fisher's Exact Test			
N of Valid Cases	302		

Appendix 5.7: use of anthlemintics

	X^2	df	p-value
Pearson Chi-Square	5.222[a]	1	.022
Continuity Correction[b]	3.464	1	.063
Likelihood Ratio	5.235	1	.022
Fisher's Exact Test			
N of Valid Cases	302		

Appendix5.8: rain fall

	X^2	df	p-value
Pearson Chi-Square	4.127[a]	1	.042
Continuity Correction[b]	2.608	1	.106
Likelihood Ratio	4.296	1	.038
Fisher's Exact Test			
N of Valid Cases	302		

Appendix 5.9: water body

	X^2	df	p-value
Pearson Chi-Square	4.127[a]	1	.042
Continuity Correction[b]	2.608	1	.106
Likelihood Ratio	4.296	1	.038
Fisher's Exact Test			
N of Valid Cases	302		

Appendix 5.10: vegetation

	X^2	df	p-value
Pearson Chi-Square	4.053[a]	1	.044
Continuity Correction[b]	2.549	1	.110
Likelihood Ratio	4.229	1	.040
Fisher's Exact Test			
N of Valid Cases	302		

Appendix 6

Appendix 6:1

Figure 2: Sudan map describes North Darfur state location (red color), www.help sudan.international.org.

Appendix 6:2

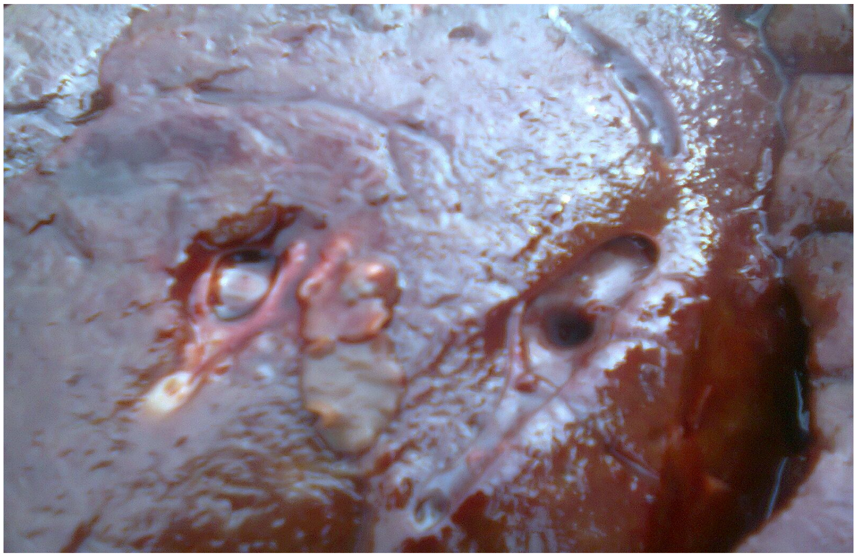

Figure 3: The adult parasite in the liver surface.

Appendix 6:3

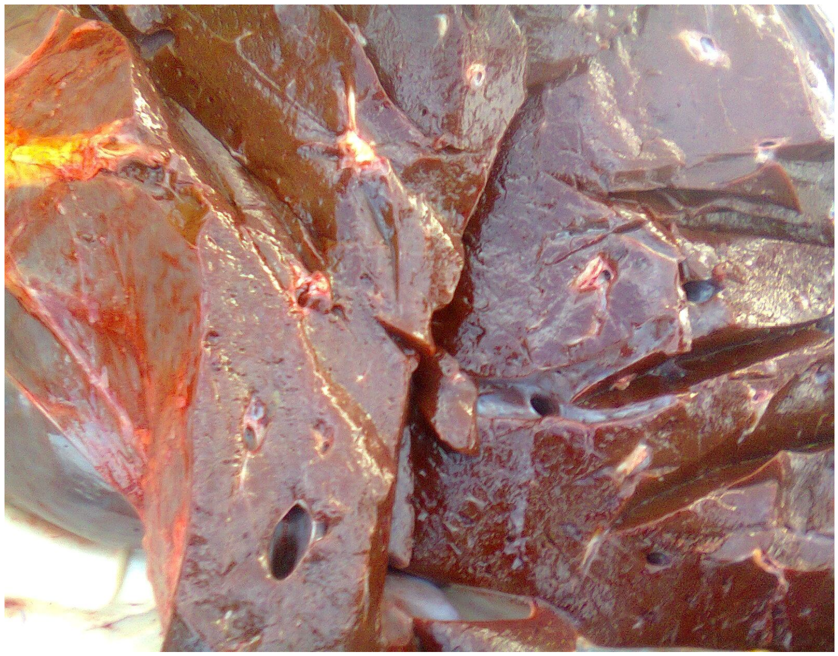

Figure 4: Infected liver of fasciolosis

Appendix 6:4

Figure 5: Fecal examination at the Ministry of Animal Resource and Fisheries- Elfasher.

Appendixs 7

Questionnaire

To obtain information about fasciolosis in cattle and information regarding geographical localization and risk factors influencing the presence of the disease.

General information:

1. Gender: ………………………………………………………………………….
2. Level of education:…………………………………………………………………

Information about the fasciolosis risk factors.

1. Age:

Young (   )

Middle (   )

Old   (   )

2. Sex:

Male   (   )

Female   (   )

3. Breed:

Local   (   )

Cross   (   )

4. Body condition:

Good   (   )

Poor   (   )

5. Source of animal

South Darfur (   )

North Darfur ( )

6. Grazing.

Indoor ( )

Outdoor ( )

7.use of anthlemintics

Yes ( )

No ( )

8. Rainfall

Low

high ( )

9. Water body.

Yes ( )

No ( )

10. Vegetation

Poor

good