Prevalence of Bovine Hydatidosis and Its Cyst Characterization in Debre Zeit Elfora Export Abattoir, Oromia Regional State, Ethiopia

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Abstract: Cystic hydatidosis is one of the important zoonotic parasitic diseases caused by infection with the larval stage of the dog tapeworm *Echinococcus granulosus*. The disease has both public health and economic importance worldwide. A cross-sectional study was conducted from December 2015 to March 2016 to assess prevalence, identify risk factors and cyst characterization of bovine hydatidosis in cattle slaughtered at Debre Zeit Elfora export abattoir. Ante mortem examination, post mortem examination (visualization, palpation and incision) and cyst characterization were conducted. As a result of 384 examined animal, 69 (17.97%) were harbouring a single or multiple hydatid cysts. Out of 69 viscera harboring hydatid cysts, the highest 46(66.67%) was lung followed by liver 13 (18.84%), heart 3(4.35%), kidney 3(4.35%) and spleen 1(1.45%) and the rest 3 (4.35%) infections involved multiple organs. Out of the total 69 cysts collected, 32 (46.37%) were fertile, 19 (27.53%) calcified and 18 (26.08%) sterile. Hence, 32 fertile cysts subjected to viability test, 25 (78.13%) were viable and 7(21.87%) nonviable. Cyst size assessment was made on 69 hydatid cysts indicated that 25 (36.23%) were small, 21 (30.3%) were medium and 23(33.33%) were large size cysts. The highest numbers of large size cysts were recovered on the lung (45.83%) than in the liver (7.14%), while the liver was found harboring large number of small size cysts (71.42%). There was highly statistically significance variation between the age (P=0.000, x^2=28.972, CI=0.014-0.049), body condition score (P=0.000, x^2=46.498, CI=0.024-0.065) and prevalence of cyst recovered. But no significant variation was observed with regard to origin of animals (p=0.733, x^2=19.352, CI=0.765-0.844) and the prevalence of the disease. High prevalence of the disease and fertile cysts was recovered from the current study. Especially, liver and lung were the most affected organs than other visceral organs. Therefore, public awareness about the zoonotic importance of the disease and application of the conventional preventive and control measures like detail meat inspection and proper disposal of infected organs at abattoirs should be practiced.

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1. Introduction

Echinococcosis is one of the most important and commonly found parasitic zoonoses in both humans and different animals, which is caused by the cestode helminthes *Echinococcus granulosus* and causes considerable economic losses and public health problems in many countries including Europe, Asia, Africa, South America, Canada and Australia (Budke *et al.*, 2006; Moro and Schantz, 2006).

Echinococcus is a genus of parasitic zoonotic cestodes (tapeworms) found worldwide in which the adult stages occur in the small intestines of canids and felids, and the larval stages in tissues of various organs of other mammalian hosts, including humans. Four species of *Echinococcus* are currently considered taxonomically valid: *E. granulosus, E. multilocularis*, *E. oligarthrus* and *E. vogeli* and *E. shiquicus* (Eckert *et al.,* 2002). The cestode *E. granulosus* is the causative agent of cystic hydatid disease, or hydatidosis, which is recognized as one of the major zoonoses, affecting both animals and humans in the world (Gottstein and Hemphill, 1997). Although *E. granulosus* penetrates deep between the villi of the small intestine of the definitive host , there are no pathogenic effects even in heavy chronic zoonotic disease condition associated to infections, suggesting that infected definitive hosts infection with the larval stage (hydatid cysts) of the dog are asymptomatic carriers of the parasite (Elmahdi *et al.*, 2004*).*

Transmission of parasites from this genus occurs in a predator/prey interaction between canids and less commonly to felids (definitive hosts) and a range of domestic and wildlife species of herbivores (intermediate hosts). Two hosts are involved in the completion of the life cycle of *E. granulosus*. The definitive hosts are carnivores which harbour mature tape worms in the intestine (Khuroo, 2002; Zhang *et al*., 2003). The intermediate hosts of *E. granulosus*

include ungulates both domestic and wild animals and humans. The adult worm lives in the small intestine of carnivores (definitive host), and the intermediate larval stage (Hydatid cyst or Hydatid) develops in the internal organs of a wide range of mammalian species such as goats, sheep and cattle, including humans, which acquire the infection through accidental ingestion of the tapeworm eggs (Eckert and Deplazes, 2004).

In Ethiopia, hydatidosis is the major cause of organ condemnation in most abattoirs and leads to huge economic losses from treatment costs and livestock associated production losses. It is also a disease of public health importance in Ethiopia (Daniel *et al*., 2012*)*. However, there is no current information regarding the prevalence and associated risk factors of hydatidosis in livestock in Bishoftu Elfora export abattoir. Hence, it would be essential to have information on the status of hydatidosis with regard to its magnitude of occurrence and risk factors of this disease in the study abattoir. Therefore, the aim of this study was:

- ✓ To determine the prevalence of bovine hydatidosis in Debre Zeit Elfora export abattoir
- ✓ To assess associated risk factors of bovine hydatidosis in study area
- ✓ To assess the fertility and viability of hydatid cysts in cattle slaughtered in the abattoir

2. Materials and Methods

2.1. Study Area:

The study was conducted starting from December 2015 to March 2016 at Debre Zeit Elfora abattoir, East Shewa zone, Oromia National Regional State. Debre Zeit is located in Central Oromia regional state, at a distance of 47 km of the South Eastern part of Addis Ababa. It is the main city of Ada'a-Liban district, situated on two international trade routes, which are connected by Franco-Ethiopia Djibouti railway and Addis-Moyale Nairobi, intercontinental asphalted road transport route. This is the only gate for import/export of livestock and agricultural commodities besides other commercial goods, from or to international market (CACC, 2003).

Debre Zeit Town lies between 9° N latitude and 39° E longitude and an altitude of 1860 meter above sea level (m.a.s.l). It has three agro-ecological locations, 6% Dega greater than 2000 m.a.s.l, 89% Woinadega greater than 1600 meter above.sea.level, and 5% Kola greater than 1000 m.a.s.l. It gets an annual rainfall of 871 mm of which 80% is received during long rainy season starting from June to September and the remaining in short rainy season extending from March to May, and the dry season from October to February. The mean annual maximum and minimum temperature are 26 and 14°c, respectively with a minimum relative humidity of 63.8% (NMSA, 2005).

Debre Zeit Elfora export abattoir is located in Debre Zeit town. The abattoir is established far away from the town and located away from rivers and deep wells, factories, houses, church, mosque and farm. It has good accessibility to roads as it is found beside highway (Located 2 Km from Addis Ababa to Adama highway). The abattoir has sufficient supply of potable and powerful water and electricity. The abattoir which exports mutton, lamb, goat meat, and edible offal/organs like liver, kidney to Middle East countries (Dubai and Saudi) and they have various facilities to carry out a range of slaughter house procedures(watering system, hygienic condition, Carcass Storage and cooling Facilities, Waste disposal system and ways of condemnation etc).

2.2. Study Population:

The study population was Cattle brought for slaughter from different areas of the country (South Wollo, East Wollega, East Gojjam and North Gondar) to Debre Zeit Elfora export abattoir.

2.3. Study Design:

A cross sectional study design was used to examine the prevalence of bovine hydatidosis and cyst characterization for those positive cases from December to March in Debre Zeit Elfora export abattoir.

2.4. Study Methodology

2.4.1. Ante mortem examination: Regular visits (4 days per week) were made on Elfora export abattoir during the period from December 2015 to March 2016. During ante mortem examination, each study animal was given identification by number and breed; origin, sex, age, and body condition of animals were recorded. The age of the animals was determined on the basis of the dentitions as described by De Lahunta and Habel (1986) and two age groups were considered; less or equal to five years and above five years. The body condition scoring was done according to Nicholson and Butter Worth (1986) and classified in to three categories as poor, medium and good. Finally, apparently healthy animals were passed for slaughtered during period of study.

2.4.2. Post mortem examination: From animals that passed for slaughter 384 cattle were randomly selected and postmortem examination was carried out through visual inspection, palpation and incision of visceral organs during period of study. The liver, lung, spleen, kidney, and heart of slaughtered cattle were examined for the presence of hydatid cysts. Each organ was accessed macroscopically either by visual inspection or palpation and where necessary, one or more incisions were made in order to detect hydatid cysts.

2.4.3. Cyst characterization: The infected organs from each positive animal were collected and recorded for fertility test, viability test and size of cysts. The total number of hydatid cysts were collected and recorded for each infected organs. To undergone fertility test, the fluid of cysts was aseptically aspirated and transferred separately into sterile tubes, then the cyst fluid was centrifuged at 500 rpm for 5 min and after centrifugation the supernatant was discarded carefully and a drop of the sediment was placed on the clean microscopic slide and covered by cover slip and examined under 40% microscope for the presence of protoscolices, which looks like white dots on the germinal epithelium, in hydatid fluid so as to classify cysts as fertile or infertile. Furthermore, infertile cysts were further classified as sterile or calcified. Sterile cysts (fluid filled cyst without protoscoleces) were further classified as calcified or none calcified. Sterile hydatid cysts were characterized by their smooth inner lining usually with slightly turbid fluid in its content. Typical calcified cysts produce a gritty sound feeling up on incision (Mojtaba *et al.,* 2013; Soulsby, 1982).

Fertile cysts was subjected to viability test, drop of the sediment containing protoscolices was placed on the microscopic glass slide and covered with a cover slip and observed for amoebioed like peristaltic movements with 40x objective. The viability of protoscoleces was tested for each fertile cyst per randomly selected hydatid cysts. For clear vision a drop of 0.1% aqueous eosin solution was added to equal volume of protoscoleces, of hydatid fluid on slide with the Principle that viable protoscoleces should completely or partially exclude the dye, while the dead ones taken up (Mojtaba *et al.,* 2013).

According to their size, the size of the diameter of hydatid cyst was measured by caliper and classified as large (diameter >10 cm), medium (5 to 10 cm) and small (diameter < 5 cm) (Oostburg *et al.,* 2000; Kebede *et al.,* 2009b).

2.5. Sample size Determination and Sampling Methods:

Simple random sampling technique was used daily to identify the study animals for inspection during the study period. The sample size was calculated using the formula given by Thrusfield (2005) with 50% expected prevalence due to there was no information about the prevalence of the disease in the study abattoir previously and 5% absolute precision at 95% confidence interval. Therefore,

$$N = \frac{1.96^2 \times P\,exp\,(1-Pexp)}{d^2}$$

Where: N = required sample size

Pexp = expected prevalence

d = desire absolute precision

$$N = \frac{1.96^2\,(0.5)*(1-0.5)}{0.05^2} = 384$$

Therefore, by substituting the value in the given formula n = 384 were used as representative animal for this study.

2.6. Data Management and Analysis:

Data obtained from ante mortem, post mortem findings and cyst characterization in the abattoir were coded and stored in to Microsoft excel and analyzed by using SPSS version 16. Descriptive statistics were computed. The prevalence was calculated as the number of positive samples divided by the total number of examined samples. Chi-square ($\chi 2$) test was used to evaluate the association of different host related factors such as age, body condition, origin and breed of cattle with prevalence of hydatidosis and *P* value which is < 0.05 was considered as significant.

3. Results

3.1. Ante mortem Examination Results:

Up on regular visit done on animals brought for slaughter, individual animals were assessed with regard to their place of origin, age, sex, breed and body conditions. Since almost all the cattle presented to slaughter house were male and local breed, infection rate regarding sex and breed variation were not included. Origin did not show significant difference with regard to cyst detection (p=0.733), but body condition score and age shows significant difference (p=0.000 and p=0.000, respectively). Prevalence of hydatidosis based on age, origin of animal and body conditions in the abattoir is shown in Table 1.

3.2. Postmortem Examination Results:

From apparently healthy animals, 384 cattle were randomly selected and postmortem examination was carried out through visual inspection, palpation and incision of visceral organs during period of study, 69(17.97%) were found infected with hydatid cysts in the abattoir, harboring one or more cysts in different visceral organs (liver, lung, kidney, spleen, and heart). The total of 69 cattle found positive, 46 (66.67%) had cysts merely in lungs, 13 (18.84%) in liver, 3 (4.35%) in heart, 3 (4.35%) in kidney and 1 (1.45%) in spleen, whereas, the rest of 3 (4.35%) infections involved multiple organs. Prevalence of hydatidosis in different organs of cattle slaughtered in study area is described below (Table 2).

Table 1: Prevalence of hydatidosis based on age, origin of animal and body conditions in Bishoftu Elfora export abattoir

Risk factors	No examined	No-positive	Prevalence	X^2	P value	95% CI
Age						
>5 year	359	60	16.7	28.977	0.000	0.014-0.049
≤5 year	25	9	36			
Total	**384**	**69**	**17.97**			
Origin						
Wollega	51	6	11.8			
Wollo	137	26	19	19.352	0.733	0.765-0.844
Gondar	123	22	17.9			
Gojjam	73	15	20			
Total	**384**	**69**	**17.97**			
BCs						
Poor	51	20	39.2	46.498	0.000	0.024-0.065
Medium	331	48	14.5			
Good	2	1	0.5			
Total	**384**	**69**	**17.97**			

***BCs = body conditions**

Table 2: Prevalence of hydatidosis in different organs of cattle slaughtered in study area

Organs infected	No examined	No positive	Prevalence	Proportion (%)
Lung	384	46	11.98	66.67
Liver	384	13	3.39	18.84
Heart	384	3	0.78	4.35
Kidney	384	3	0.78	4.35
Spleen	384	1	0.26	1.45
Liver, spleen	384	1	0.26	1.45
Lung,liver, spleen	384	1	0.26	1.45
Lung, spleen	384	1	0.26	1.45
Total	**384**	**69**	**17.97**	**100.0**

3.3. Cyst Characterization Results:

Of the 69 hydatid cysts those subjected to fertility and viability test end measuring of size of cysts, 32 (46.37%) were fertile, 18 (26.07%) sterile, and 19 (27.54%) calcified and out of 32 fertile cysts, 25 (78.13%) were viable and 7(21.87%) were nonviable and as shown (Table 3 below). Out of the total hydatid cysts (69), 25(36.23%), 21 (30.3%), and 23(33.33%) were small, medium, and large cysts in size respectively, as shown in Table 4 below.

Table 3: cyst fertility and viability test in different organs slaughtered in study area

Examined organ	No of cysts examined	Fertility test			Viability test	
		fertile(%)	sterile(%)	Calcified(%)	Viable(%)	Nonviable(%)
Lung	48	23(47.92)	13(27.08)	12(25.0)	18(78.26)	5(21.74)
Liver	14	6(42.86)	4(28.57)	4(28.57)	4(66.67)	2(33.33)
Heart	3	1(33.33)	0(0.0)	2(66.67)	1(100)	0(0.0)
Kidney	3	1(33.33)	1(33.33)	1(33.33)	1(100)	0(0.0)
Spleen	1	1(100)	0(0.0)	(0.0)	1(100)	(0.0)
Total	**69**	**32(46.37)**	**18(26.08)**	**19(27.53)**	**25(78.13)**	**7(21.87)**

Table 4. Cyst size and counts in relation with organ involvements in infected cattle slaughtered in study area

Examined organ	No of cysts examined	Size of cysts		
		Small (<5 cm) (%)	Medium (5-10cm (%)	Large (>10cm) (%)
Lung	48	8(16.67)	18(37.5)	22(45.83)
Liver	14	10(71.42)	3(21.42)	1(7.14)
Heart	3	3(100)	0(0.0)	0(0.0)
Kidney	3	3(100)	0(0.0)	0(0.0)
Spleen	1	1(100)	0(0.0)	0(0.0)
Total	**69**	**25(36.23)**	**21(30.43)**	**23(33.33)**

4. Discussion

The current study revealed that the prevalence of bovine hydatidosis in cattle slaughtered at Debre zeit Elfora export abattoir was found to be 17.97%. The prevalence of bovine hydatidosis in the abattoir inline with the prevalence of bovine hydatidosis 22.4% in Jimma (Moges, 2003), 22.6 % in Konso (Fikre, 1994) and in 17.95% south Wollo (Degefu and Damet, 2013) and also with the findings reported from Morocco and Kenya in which the prevalence of 23.0 % (Azlaf and Dakkak, 2006) and 19.4% (Njoroge *et al*., 2002) were recorded, respectively. Relatively the current prevalence finding was higher than the 7.5% prevalence report in Harrargie region (Woubet, 1987) and 6.8% in Modjo luna export abattoir by Daniel *et al*. (2012). However, it was lower than 52.6 % in Hawassa (Alemante, 2009), 46.5% in Debre Zeit (Jobre, *et al*., 1996) and 34.05% in Bahir Dar (kebede *et al*., 2009a). The variation in prevalence from different area and a country might be attributed to the strain difference of *E. granulosus* that exist in different geographical situations and other factors like difference in culture, social activity and attitude to dog in different regions (Arene, 1995).

In the present study, among the factors considered (age, body condition and origin), age and body condition of the cattle was statistically associated (P=0.000) with the occurrence of the disease. Cattle having poor and medium body conditions were found to have higher cysts burden which may be explained due to the retarded growth, weight loss and moderate to severe infection in such animals described by Polydorous (1981). A significant variation was observed in the rates of infections between age groups where animal's equals or less than 5 years of age were highly infected. This might be due to the fact that cattle slaughtered at Elfora abattoir were mostly greater than or equals five years, so their ≤5 years of age with which they have greater chance of being infected with *E. granulosus* and the growth of the hydatid is slow,maturity being reached in 6 - 12 months (Urquhart *et al*., 1996). The origin of cattle was not stastically associated with the prevalence of the diseases (P=0.733). This might be due to the similarity in environmental factors, religious factors and animal's husbandry practices of the community of animal origin.

In the present study, the lung and liver (66.67% and 18.84%) were the most commonly infected organs. The kidney, heart and spleen (4.35%, 4.35% and 1.45%) were the least affected organs in the study animal. This might be mainly due to the lung and liver possess first great capillaries encountered by the migrating echinococcus oncosphere (hexacanth embryo), which adopt the portal vein route and primarily negotiate hepatic and pulmonary filtering system sequentially before any other peripheral organ is involved (Urquhart *et a*l., 1996). From the organ prevalence study, the lung was found to be the most commonly affected organ followed by liver may be due to the presence of greater capillary beds in the lungs than other organs (Getaw *et al*,. 2009).

Prevalence and fertility of hydatid cysts in various organs of cattle are important indicators of potential sources of infection to perpetuate the disease in dogs. In current study, the percentage of fertile cysts recovered was 46.37% (69/384) and 25 (36.23%) of the fertile cysts were proved to be viable. This was relatively higher as compared to the report of fertility rates 1.76% around Wolayita Soddo (Kebede *et al*., 2009c) and 9.85% in Nekemte (Berssisa, 1994.). Fertility rate among the organs was higher in the lungs (47.92%) than in livers (42.86%). This may be lung has relatively softer consistency which allows easier development of the pressure cysts and fertility of hydatid cyst may show a tendency to increase in advanced age of host. This may be related with reduced immunological compatibility of the hosts at their old age of infections (Getaw *et al.,* 2010; Ibrahim, 2010). The variation in fertility rate in different geographical zones might be due to the difference in strain of *E. granulosus* (Arene, 1995).

Out of the total 69 hydatid cysts, 25(36.23%) were small, 21 (30.3%) medium, and 23(33.33%) large cysts. A greater frequency of medium size and large size cysts was found in the lung than in the liver, while the liver harbored a large number of small size cysts. The reason for the higher percentage of medium

and large cysts in the lungs might be due to the softer consistency which allows easier development of the pressure cysts, while the higher number of calcified cysts in liver might be attributed to relatively higher reticuloendothelial cells and abundant connective tissue reaction of the organ and the higher proportion of small cysts may be due to immunological response of host which might preclude expansion of cyst size (Haftu and Kebede, 2014).

5. Conclusion and Recommendations

From the present study, it can be concluded that bovine hydatidosis was one of the most important parasitic diseases in cattle slaughtered at Debre Zeit Elfora export abattoir. The current study revealed that high prevalence of the disease and fertile cysts. Lung and liver were the most frequently affected body organs than other visceral organs. Fertile cysts mostly in lung in cattle were important indicators of potential sources of infection to perpetuate the disease in dogs. Among the important risk factors in prevalence of bovine hydatidosis recorded, age and body conditions of the animals were found to be the risk factors for hydatidosis infections in study area.

In line with above facts the following recommendations are forwarded:

- ❖ Further studies should be conducted to isolate the strains of *Echinococcus granulosus* and to know the economic and public health importance of the disease in study area
- ❖ Public awareness about the application of the conventional preventive and control measures like detail meat inspection and proper disposal of infected organs should be done
- ❖ Further promoting the construction of abattoirs with their appropriate disposal pits to control stray dogs

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