Comp Clin Pathol (2013) 22:233–238
DOI 10.1007/s00580-011-1391-4

ORIGINAL ARTICLE

The role of cattle in the epidemiology of *Echinococcus granulosus* in Kerman area, southeast of Iran

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Received: 17 October 2011 / Accepted: 9 December 2011 / Published online: 24 December 2011

Abstract The study was carried out from September 2010 to September 2011 at Kerman Municipal abattoir to assess the status of hydatidosis in cattle. Out of the total 1,000 cattle examined visually and manually (palpation and incision), 86 (8.6%) were found harboring hydatid cysts. A significantly higher infection was detected in older cattle ($P<0.05$) than younger ones. Of the total of 86 infected, 13 (15.11%) had hydatid cysts only in the lung, 33 (38.37%) in the liver, 2 (2.32%) in the muscles, while the rest 38 (44.18%) had multiple organ infections. Of the 125 viscera harboring hydatid cysts, the highest (56.8%) was the liver followed by the lungs (40.8%) and the muscles (2.4%). From the total of 721 hydatid cysts counted, 205 (28.43%), 222 (30.79%), 45 (6.24%), and 249 (34.53%) were found to be small, medium, large, and calcified cysts, respectively. The distribution of characterized cysts in different organs based on their size was found to be statistically significant ($P<0.05$). In addition, out of the total 721 cysts collected, 5.40% were fertile, 60.05% sterile, and 34.53% calcified or purulent cysts. The rate of cyst calcification was higher in the liver than in the lung. There was a significant difference in the fertility of the cyst from different organs ($P<0.05$); fertility rate was higher among the cysts of the liver. Hydatid cyst viability rate of 61.53% was observed.

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Keywords Hydatidosis · Cattle · Fertility · Viability · Kerman

Introduction

Hydatidosis is a zoonotic disease caused by the larval stage (metacestode) of the tapeworm *Echinococcus granulosus*. Dogs and other carnivores are the definitive hosts for the parasite, while a wide range of mammalian species including domestic ungulates and man act as intermediate hosts (Daryani et al. 2007). Hydatid disease is characterized by cyst containing numerous tiny protoscolices that most often develop in the liver and lungs and also develop in the kidneys, spleen, nervous tissue, bone, and other organs (Magambo et al. 2006).

Hydatidosis has greater public health importance and economic impact in countries especially in rural communities where there is a close contact between dog (definitive host) and various domestic animals, which may act as intermediate hosts (Eckert and Deplazes 2004; Berhe 2009). Hydatidosis is the major cause of disease in Iran. So far, three distinct cycles of *E. granulosus* have been suggested in Iran: A domestic cycle between dogs and livestock, a desert cycle between dogs and camels, and a sylvatic cycle between wild carnivores and wild ruminants. In the domestic cycle, the mean prevalence of *E. granulosus* in domestic dogs is 23.45%, which vary widely from 3.3% to 63.3% depending on the local condition (Eslami and Hosseini 1998).

The prevalence of *E. granulosus* has been reported in Iran from 5.1% to 74.4% in sheep, 1.7% to 20% in goats, 3.5% to 38.3% in cattle, and 11.4% to 70% in camels (Ahmadi 2005; Rokni 2009). Sheep and camel (with 88% and 70% of fertile cysts, respectively) are the most

Table 1 Prevalence of hydatidosis in different age and sex groups in cattle slaughtered at Kerman Municipal abattoir

Age groups (years)	Male		Female		Total (%)
	No. of cattle	No. of infected cattle (%)	No. of cattle	No. of infected cattle (%)	
<2	342	10 (2.92)	100	11 (11)	21 (4.75)
2–4	204	11 (5.39)	172	23 (13.37)	34 (9.04)
>4	50	3 (6)	132	28 (21.21)	31 (17.03)
Total	596	24 (4.02)	404	62 (15.34)	86 (8.60)

important intermediate hosts, and cattle (with 19% fertile cysts) have been considered as the weakest intermediate host of *E. granulosus* in Iran (Hosseini and Eslami 1998; Rokni 2009). Slaughter survey has been recommended in studies of cystic echinococcosis for various reasons. It is an economical way of gathering information on livestock diseases, particularly subclinical conditions. Also, no satisfactory test exists for cystic echinococcosis in living ruminants (Njoroge et al. 2002). Therefore, the objectives of the current study were to determine the prevalence and status of cystic echinococcosis (CE) (hydatidosis) and to study the localization and fertility/sterility rates of hydatid cysts and the viability of their protoscoleces will also be investigated in Kerman Abattoir, southeast Iran.

Materials and methods

Study area

The study was conducted in the Kerman area, Kerman Province, Iran. Kerman is located at 30°17′13″ N and 57° 04′09″ E southeast of Iran. The mean elevation of the city is about 1,755 m above sea level. Kerman City has a hot and arid climate, and the average annual rainfall is 135 mm. Because it is located close to the Kavir-e lut, Kerman has hot summers. Based on climate, soil, and other geographical conditions, Kerman has a different vegetation and agricultural type. The density of livestock animals in this area per square kilometer is 33 over one; however, this density in cultivable lands is 505 over eight. The system of breeding in the rural region of Kerman is small herds of cattle and they are kept together with other animals like sheep and goats. Stray dogs in urban areas and free or roaming dogs in rural areas are the main definitive hosts.

Study animals and sampling methods

This cross-sectional study was carried out on 1,000 cattle in the Kerman Municipal abattoir, southeastern Iran, from September 2010 to September 2011. For this purpose, the industrial slaughterhouse was visited periodically to examine the liver, lungs, and other organs of slaughtered animals for the presence of cystic echinococcosis. During the antemortem inspection, the age and sex of each individual animal were recorded. After arrival to the abattoir, the ages of animals were estimated by observing the teeth. A total of 1,000 cattle (404 females and 596 males) in three age groups (<2, 2–4, and >4 years old) were inspected for infection with cystic hydatidosis.

Different organs of each of slaughtered animals, particularly lung, liver, spleen, kidney, heart, and the muscles, were visited. Each organ was accessed macroscopically either by visual inspection or palpation and where necessary one or more incisions were made in order to detect small hydatid cysts. The infected organs from each positive animal were collected; the total number of hydatid cysts was counted per infected organ and recorded.

Examination of cyst size

The size of the diameter of collected hydatid cysts on the affected organs was measured and classified as small (diameter less than 4 cm), medium (diameter between 4 and

Table 2 Frequencies and percentages of positive cattle by age class, mean number, and viability of hydatid cysts at Kerman Municipal abattoir

Age groups	No. of infected (%)	Data on hydatid cysts (mean number and viability)						
		Number	Mean no.	Fertile	Sterile	Calcified/caseous	Viable	Nonviable (%)
<2	21 (4.75)	233	11.09	2 (0.85)	143 (61.37)	88 (37.76)	2 (100)	0 (0)
2–4	34 (9.04)	227	6.67	19 (8.37)	143 (62.99)	65 (28.63)	14 (73.68)	5 (26.31)
>4	31 (17.03)	261	8.41	18 (6.89)	147 (56.32)	96 (36.78)	8 (44.44)	10 (55.55)
Total	86 (8.6)	721	8.38	39 (5.40)	433 (60.05)	249 (34.53)	24 (61.53)	15 (38.46)

Table 3 Distribution of hydatid cysts in different organs of positive cattle at Kerman Municipal abattoir

Organs affected number of animals				
	Examined	Number of cases	Percentage	Relative frequency (%)
Lung only	1,000	13	1.3	15.11
Liver only	1,000	33	3.3	38.37
Spleen only	1,000	0	0	0
Muscles only	1,000	2	0.2	2.32
Lung and liver	1,000	37	3.7	43.02
Lung, liver, and muscles	1,000	1	0.1	1.16

8 cm), and large (diameter greater than 8 cm) (Schantz 1990).

Examination of cyst fertility and viability of protoscoleces

The pressure of the cyst fluid was reduced by using a sterile hypodermic needle. Then cyst wall was incised with a sterile scalpel blade, and the content was transferred into a sterile container and examined microscopically (×40) for the presence of hydatid protoscolices. Similarly, the germinal layer was put in glycerin between two microscopic glass slides and examined for the presence of protoscoleces. The presence of protoscolices either attached to the germinal epithelium in the form of brood capsule or its presence in the cyst fluid was considered as indicative of fertility (Macpherson et al. 1985). Cysts which contained no protoscoleces as well as heavily supurative or calcified were considered unfertile. Fertile cysts were subjected to viability test.

A drop of the sediment containing the protoscolices was placed on the microscope glass slide and covered with cover slip and observed for amoeboid-like peristaltic movements with ×40 objective. The viability of protoscoleces was assessed by the motility of flame cells together with staining with a 0.1% aqueous eosin solution (Smyth and Barrett 1980). The viability of protoscoleces was carried out for each fertile cyst per animal species and organ. For clear vision, a drop of 0.1% aqueous eosin solution was added to an equal volume of protoscolices in hydatid fluid on microscope slide, with the principle that viable protoscolices should completely or partially exclude the dye while the dead ones take it up (Smyth and Barrett 1980; Macpherson et al. 1985). Furthermore, infertile cysts were further classified as sterile or 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 (Soulsby 1982).

Statistical analysis

Data collected from antemortem, postmortem, and laboratory findings were entered into MS Excel; the statistical package SPSS version 9.0 for Windows (SPSS Inc., Chicago, IL, USA) was employed to analyze results. Mean and percentage were used to calculate prevalence.

Results

Postmortem inspection

Of the total of 1,000 local cattle examined at the Kerman Municipal abattoir, 86 (8.6%) were found harboring hydatid cysts. The prevalence of hydatidosis in different age and sex groups in cattle slaughtered at Kerman Municipal abattoir was described in Tables 1 and 2. As many as 24 out of 596 males (4.02%) and 62 out of 404 females (15.34%) were found to be positive (Table 1). The infection rate increased with age ($P<0.05$). Also a significant difference was observed between males and females ($P<0.05$). The prevalence of hydatidosis in different seasons was not significantly different ($P>0.05$; Table 9).

Table 4 Distribution and number of organs with hydatid cysts in infected cattle slaughtered in Kerman Municipal Abattoir

Organ	No. of organs infected	Relative prevalence	Cyst count			
			Mean/organ	Range	Total	Percentage
Lung	51	40.8	5.15	1–39	263	36.47
Liver	71	56.8	6.40	1–37	455	63.10
Spleen	0	0	0	0	0	0
Muscles	3	2.4	1	1	3	0.41
Total	125	100	5.76	1–39	721	100

Table 5 Cyst size and counts in relation to organ involvement in cattle slaughtered at Kerman Municipal abattoir

Organ	Number (%) of the different cyst sizes				Total
	Small	Medium	Large	Calcified	
Lung	75 (28.51)	119 (45.24)	24 (9.12)	45 (17.11)	263 (36.47)
Liver	130 (28.57)	101 (22.19)	20 (4.39)	204 (44.83)	455 (63.10)
Spleen	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
Muscles	0 (0)	2 (66.66)	1 (33.33)	0 (0)	3 (0.41)
Total	205 (28.43)	222 (30.79)	45 (6.24)	249 (34.53)	721 (100)

Hydatid cyst characterization

Single and multiple hydatid cyst distribution was recorded in different organs (Table 3). Most of the hydatid cysts were found concentrated in great number in the livers. From the total of 86 cattle harboring hydatid cysts, 48 (55.81%) were found involving a single organ and the remaining 38 (44.18%) had a multiple organ involvement. About 97.6% (122 of 125) of all infected viscera is attributed to overall involvement of the lungs and liver. The chi-square test for the differences of location was significant ($P<0.05$). The total number, relative prevalence, and mean number of cysts harbored in each individual affected organ were described (Table 4).

A total of 721 cysts were collected from the infected cattle, 263 (36.47%) in the lungs, 455 (63.10%) in the liver, and 3 (0.41%) in the muscles (Table 4). The mean number of cysts per animal was 8.38 (minimum, one; maximum, 39). Of the 125 viscera harboring hydatid cysts, the highest (56.8%) was the liver followed by the lungs (40.8%) and the muscles (2.4%). The distributions of hydatid cyst between organs of infected animals were significantly different in cattle ($P<0.05$).

The total cyst counts with respect to cyst size in each affected organ of cattle are described in Table 5. Size systematic measurement of the cysts revealed that majority of large- and medium-sized cysts were found in lungs, while a large number of small sized and calcified cysts were found in liver. The variation in the size of cysts of different organs was significant ($P<0.05$).

Fertility and sterility of hydatid cyst was described (Table 6). Fertility status of cysts from different organs have shown a significant difference ($P<0.05$), with cysts of liver origin being highly fertile.

The viability percentage of protoscolices was higher in liver (57.14%) than in the other organs ($P>0.05$) (Table 7). The numbers of protoscoleces and viable protoscoleces are summarized in Table 8.

Discussion

The occurrence of hydatidosis in cattle at Kerman Abattoir was found to be 8.6% during the study period. There are reports of hydatidosis in cattle in different parts of Iran, that a prevalence of 38.3% was recorded in Ardabil; 15.3% and 25.9% in Hamadan; 16.4% in Western Iran; 26.9% in North Khorasan Province; and 28.02% in Kurdistan (Rokni 2009)

In general terms, throughout the world, there had been different magnitude records of hydatidosis in cattle with low medium and high rates of occurrences. Generally, the variation in prevalence rate among different geographical locations could be ascribed to the strain differences of *E. granulosus* that exists in different geographical locations (McManus 2006). Additionally, variability could be related with age factors. Other factors like difference in culture, social activities, and attitudes to dogs in different regions may contribute to variation (Arbabi and Hooshyr 2006). The prevalence of CE differed significantly by host sex in cattle. Females were more likely to have CE infection than males in the cattle examined. Females are more susceptible to the infection by metacestode of *E. granulosus* than the males and the parasite may cause hormonal imbalance especially in testosterone and estradol in chronic stages, to be able to

Table 6 Fertility/sterility of cysts collected from different organs of cattle slaughtered at Kerman Municipal abattoir

Organ	Fertile cyst (%)	Sterile cyst (%)	Calcified (%)
Lung	7 (2.66)	211 (80.22)	45 (17.11)
Liver	32 (7.03)	219 (48.13)	204 (44.83)
Spleen	0 (0)	0 (0)	0 (0)
Muscles	0 (0)	3 (100)	0 (0)
Total	39 (5.40)	433 (60.05)	249 (34.53)

Table 7 Viability statuses of fertile cysts collected from organs of cattle slaughtered at Kerman Municipal abattoir

Organ	Viable cyst (%)	Nonviable cysts (%)	Total
Lung	4 (57.14)	3 (42.85)	7
Liver	20 (62.50)	12 (37.50)	32
Spleen	0 (0)	0 (0)	0
Muscles	0 (0)	0 (0)	0
Total	24 (61.53)	15 (38.46)	39

Table 8 The numbers of protoscoleces and viable protoscoleces

Infected organ	Average number of protoscoleces/ml	Average number of live protoscoleces/ml (%)
Lung	11	5 (45.45)
Liver	7	4 (57.14)
Spleen	0	0 (0)
Muscle	0	0 (0)

remain for long periods in its host (Blancas et al. 2007) (Table 9).

A significant variation was observed in the rates of infections between age groups where animals >4 years of age were highly infected. In this region, females remain longer than males for reproductive purposes, therefore the probability of having more infective prevalence and mean intensity is higher in females than in males. Similarly, old animals have a chance to be infected or more time for cyst to develop than young animals. The results showed that the infection prevalence and mean intensity were higher in the older age classes. The age-dependent increase in infection rate among examined animals is in accordance with Azlaf and Dakkak (2006) and Rinaldi et al. (2008). This age variation can be translated into differential exposure to infection because older livestock may have been exposed to more infective stages (Ibrahim et al. 2008).

Livers and lungs were the most frequently infected visceral organs examined. This is explained by the fact that livers and lungs possess the first great capillary sites 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 (Kebede et al. 2009; Fathi et al. 2011).

The finding that the livers of cattle were found to be more commonly infected with hydatid cysts than the lungs is in agreement with the previous findings of Gusbi et al. (1990) and Ibrahim (2010). The reason for the higher percentage of medium and large cysts in the lungs is due to the softer consistency of the lungs, while the higher yield of calcified cysts in the liver could be attributed to the relatively higher reticuloendothelial cells and abundant connective tissue reaction of the organ. Likewise, the high proportion of small cysts may be due to the immunological response of the host which might preclude the expansion of cyst size (Lahmar et al. 1999).

Data on the prevalence and fertility of cysts in various domestic herbivores provide reliable indicators of the importance of each type of animal as a potential source of infection to dogs. Cysts, depending on the geographical situation, host, site, size, and type of cyst may have different rates of fertility (Dalimi et al. 2002). With regard to fertility and viability of cysts, the finding of 60.05% sterile, 5.40% fertile, and 34.53% calcified cysts in cattle may generally imply that most of the cysts in cattle are infertile. Cattle have been considered as the weakest intermediate host of *E. granulosus* in Iran (Hosseini and Eslami 1998; Rokni 2009). The variation in fertility rate in different geographical zone could be due to the difference in the strain of *E. granulosus* (Arene 1985; McManus 2006). In comparison of the fertility rate among the organs, it was higher in liver (7.03%) than lungs (2.66%) and spleen (0.0%) ($P<0.05$). Cystic echinococcosis is widespread in this study area. It was documented that a prevalence of 7.2% was recorded in cattle, 9.2% in sheep, and 6.8 in goats slaughtered in Kerman slaughterhouse, southeast Iran (Sharifi 1996), while the present study shows that 8.6% of cattle were infected with hydatid cysts.

Backyard slaughtering is a common practice in the study districts. Almost all livestock owners and urban dwellers keep at least one dog for the purpose of safeguarding their properties from wild carnivores and thieves. In general, the widespread practice of offering dogs with uncooked infected offal, the absence of well-constructed abattoir, and the habit of leaving the dead unburied are important factors that favor the maintenance and widespread existence of the disease in the study areas. The communities in the study areas depend on livestock husbandry, which is an important economic activity. In such areas, bovine hydatidosis in domestic animals can result in significant production losses, including reduction in live weight gain, yield of milk, fertility rates, and the value of hide and skin and in decreased edible offals.

Table 9 The structure of the sampled host populations by the season

Season	Male		Female		Total (%)
	No. of cattle	No. of infected cattle (%)	No. of cattle	No. of infected cattle (%)	
Summer	108	4 (3.70)	67	11 (16.41)	15 (8.57)
Autumn	101	4 (3.96)	68	12 (17.64)	16 (9.46)
Winter	175	7 (4.00)	122	18 (14.75)	25 (8.41)
Spring	212	9 (4.24)	147	21 (14.28)	30 (8.35)
Total	596	24 (4.02)	404	62 (15.34)	86 (8.60)

Acknowledgments This project was funded by the Shahid Bahonar University of Kerman. We also thank Mr. Mansour Aminzadeh for his technical support.

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