

Histopathological Study Of The Fibrotic Layer Of Hydatid Cyst In Camel, Cow And Sheep

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1. Abstract

Hydatidosis or echinococcosis is one of the most important common diseases between humans and animals.

1.1. Aim: Hydatid cyst is often caused by *Echinococcus granulosus* worm. The closest host tissue to the cyst is the fibrous layer. The purpose of this study is to investigate any differences in the fibrosis layer in the lung and liver of ruminants.

1.2. Methods: For each of these three animals, 22 infected lung or liver samples were collected from Tehran slaughterhouses during the months of May to July 2022. A piece of one square centimeter was removed from the fibrotic layer and also from the healthy tissue near the cyst. In the usual process of section preparation, pathological observation was included. Sections were observed and reported by a pathologist.

1.3. Findings: In total, 138 sections were studied pathologically. The diameter of the cysts varied between 1.5-0.6 cm in camels and cattle and 0.5-0.2 cm in sheep. Most cases of sterile cysts were seen in cows. Fibrous layer was present in all (100%) of the 66 studied samples. Necrosis was observed in 20 cases (30.30%), including 12 cases in the lungs of camels, 6 cases in the lungs of cows, and only 2 cases, one in the lung and the other in the liver of sheep. Only one case (1.52%) of granuloma was observed in cow liver.

1.4. Conclusion: Differences were observed in the fibrotic layer of hydatid cyst in cows, camels and sheep. In all 3 types of animals, the rate of progression of the lesion was towards the formation of necrosis and granuloma, and also, the frequency of these lesions had a significant difference, which is due to the difference in the parasite strain, the host

type, and also the average age of these animals

2. Keywords: Fibrosis, Necrosis, Hydatid cyst, Camel, Cow, Sheep, Histopathology

3. Introduction

The larval stage of a number of species of the genus *Echinococcus* and mostly (*E. granulosus*) *Echinococcus granulosus* causes a chronic and threatening infection in herbivores, and humans are also accidentally infected with it, which leads to the creation of large vesicles containing liquid under pressure. It is called hydatid cyst [1-2].

This infection has been a problem in the history of medicine and veterinary medicine and has caused heavy biological and economic damages [3-4]. Harmful and dangerous consequences, lack of safe drug treatment, as well as problems after surgery, have provoked detailed studies in various aspects, especially in the field of histopathological structure and hydatid cyst formation [3-5, 6].

The cyst wall, from the center to the outside of the cyst, consists of two layers, one is the reproductive layer that creates protoscolices and the other is the outer layer, which is called the conformational layer. The fibrous capsule of the host origin surrounds the aforementioned vesicle and leads to the formation of a complete hydatid cyst, often in the liver and lung and then in other organs of the body (Figure 1) [7].

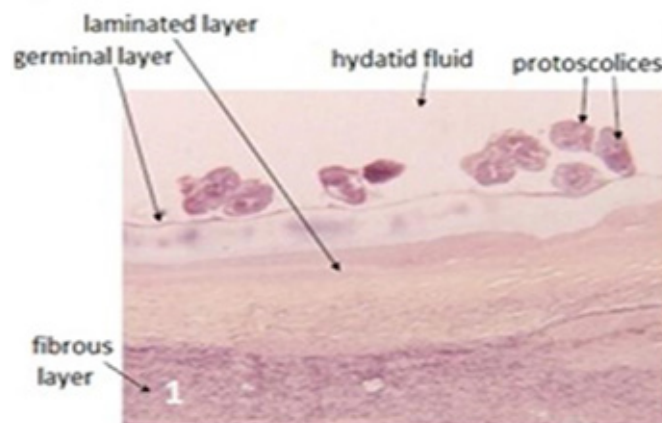


Figure 1: The normal shape of the hydatid cyst and its layers from the inside to the outside, respectively, the germ layer and the corresponding layer of the parasite origin, the paracyst layer or the fibrous capsule of the host origin

There are studies that have examined the histopathological changes of cysts in different hosts in scientific sources [8-9]. The results of these

studies indicate the mutual reaction of the parasite and the host during the process of hydatid cyst formation [10, 8, 1-2]. but with all this, there is a study gap in the field of comparative study of the fibrous layer with the origin of the host among different hosts. The purpose of this study was to investigate the possible differences of the fibrous layer in the outer part of the tissue and the inner part of the tissue of a single cyst, in liver and lung cysts, as well as in different hosts of camels, cows and sheep using histopathological methods.

4. Methods

After observing the carcasses in Isfahan slaughterhouses, only livers and lungs infected with hydatid cysts were collected. While confirming the presence of hydatid cyst by parasitological and molecular methods, sampling continued until reaching the number of 22 suitable samples for each type of animal from April to July 2015. The samples were transferred to the laboratory and the fluid inside the cysts was taken to determine the fertility or sterility of the cysts while calculating the volume of the cyst fluid. With the emptying of the cyst fluid, its remains were also removed from their place. Then, a piece with dimensions of about 1 square cm and thickness of 0.5 cm was removed from the fibrous layer as well as healthy tissue near the cyst. The samples entered the usual process of preparation of pathological sections [9].

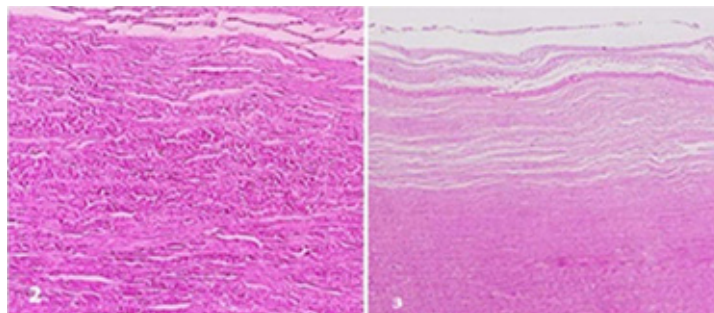
Briefly, tissue samples were placed in paraffin molds. Paraffin blocks were prepared and a 5 micron thick cross-section was prepared with a microtome, and after dehydration using ethanol with increasing concentrations, they were stained with hematoxylin-eosin dye and prepared for microscopic observation using ethanol with decreasing concentrations.

5. Findings

In total, more than 80 infected lungs or livers were collected from three types of animals and 66 numbers (22 numbers for each type of animal) were selected. The diameter range of cysts in camels and cows was between 1.5-6 cm and for sheep between 0.5-2 cm. The highest number of sterile cysts was seen in cows. The average age of camels, cows and sheep was 9, 6 and 3 years, respectively.

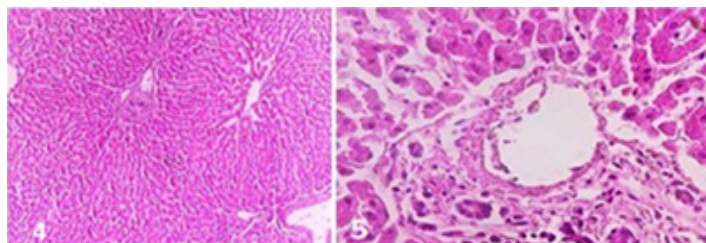
In total, 138 sections including the fibrous layer and natural tissue around the cyst were prepared from 66 samples and were observed, photographed and reported from the point of view of histopathology.

The lesions were classified into three groups: fibrosis, necrosis and granuloma. No significant difference was observed between the fibrous layer of the part of the cyst that is outside the host tissue and the part that is inside the host tissue. Although the thickness of this layer was different in different sections (Figures 2 and 3).

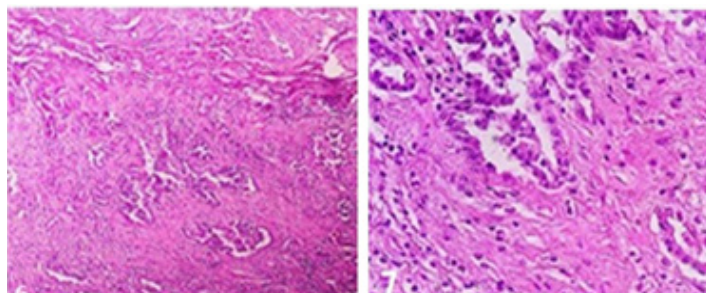


Figures 2-3: The intra-tissue part and the outer-tissue part of a single cyst did not show any significant difference in microstructure.

In liver sections, bile ducts thickening, epithelium proliferation, mucus hyperplasia and infiltration of lymphocytes and eosinophils were observed. Swollen liver lobes and hepatocytes were arranged in a rope manner, which was observed under the microscope along with pyknotic nuclei and large spaces of Disse. The stage of cyst destruction, which is often accompanied by the formation of granular tissue in the cyst space, was observed (Figures 4-5-6-7)

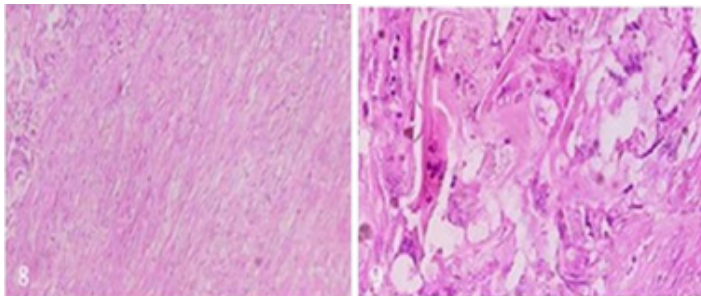


Figures 4-5: 100x and 400x normal liver tissue showing hepatocytes along with Port's triad without any pathological signs compared to Figures 2-3 (indicating inflammation, cellular infiltration and destruction)



Figures 6-7: Large liver sections, 100x and 400x, respectively, showing fibrosis with wide collagen bands

All sections of the lung showed tissue fibrosis reaction (capsule) and cellular reaction, necrosis and obstruction of the pulmonary ducts near the cyst (Figures 8-9). Cell infiltration was observed to different degrees, severe, moderate, diffuse or complex. Some sections tended to form granuloma and were seen with giant cells.



Figures 8-9: Sections of cattle liver 100x and 400x showing granuloma with multinucleated giant cells, histiocytes and lymphocytes.

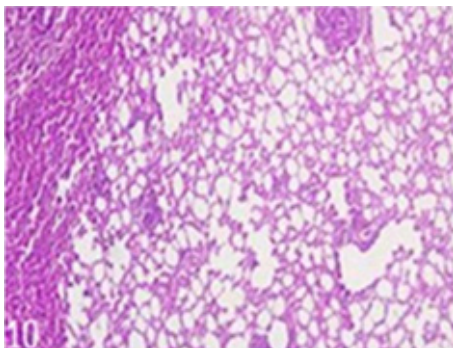
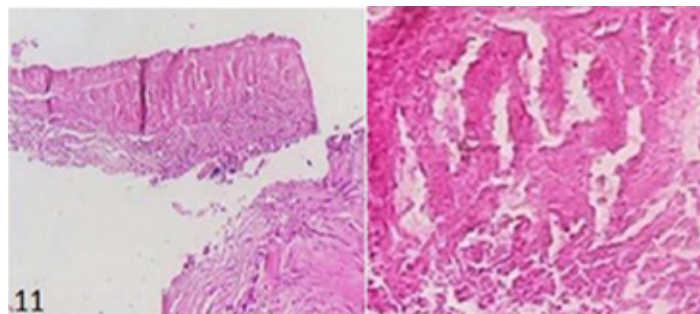


Figure 10: Cross section of a natural camel lung.

The type of infiltrated inflammatory cells were mainly mononuclear cells, macrophages, lymphocytes and plasmocytes (Figures 11 and 12).



Figures 11-12: Necrosis is noticeable in the cross-section of the camel lung with 100x and 400x magnification, nuclear particles and granular deposits, respectively. All sections are stained with hematoxylin-eosin

In total, fibroses, necrosis and granuloma were seen in 45 cases (68.20%), 20 cases (30.30%) and 1 case (1.52%) of the samples, respectively. The most cases of necrosis (12 cases or 45.55%) were seen in camels. Cows ranked second with 6 cases (27.3 percent) and sheep ranked the lowest with 2 cases (9.1 percent). Only 1 case of granuloma (1.52%) was observed, which happened in a cow (Table 1).

Total number (percentage)	Fibrosis Number (percentage)	necrosis number (percentage)	Granulom number (percentage)	Member	Animal
5(22/73)	5(22/73)	0(0)	(0) 0	Liver	camel
17(77/30)	5(22/73)	12(55/54)	(0) 0	the lung	
5(22/73)	4(18/20)	0(0)	1 (4/55)	Liver	cow
17(77/30)	11(50/00)	6(27/30)	(0) 0	the lung	
10(45/50)	9(81/80)	1(4/55)	(0) 0	Liver	sheep
12(55/54)	11(50/00)	1(4/55)	(0) 0	the lung	
66(100)	45(68/20)	20(30/30)	1(4/55)		total

Table 1: Frequency distribution of fibrosis, necrosis and granuloma in 66 samples of liver or lung infected with hydatid cyst in camel, cow and sheep.

The tissue close to the hydatid cyst may show changes depending on the location of the cyst, the age of the host and the cyst, the type of parasite and the host, and other possible factors [13-15]. Herbivores play an important role as intermediate hosts for hydatidosis. have a considerable effect on human health. Therefore, in the present study, the fibrous layer of hydatid cyst in three animals, camel, cow and sheep, was studied comparatively using histopathological techniques. Often, a part of A single-cavity hydatid cyst is placed inside the host tissue and part of it outside the host tissue, which only has a fibrous capsule of host origin [16-18].

Cases of destructive processes that turn the fibrotic layer into necrosis were observed in 30.3% of the samples. The highest number of cases of necrosis was in camels (54.54 percent) and cows ranked second (27.3 percent). The lowest number of necrosis was seen in sheep (1.9 percent). It is likely that the difference in the frequency of necrosis in the three types of animals is related to the age of the host; So that the average age of these animals in the present study was 9 years for camels, 6 years for cows and less than 3 years for sheep. This is an indication of the formation of necrosis. Only 1 case (1.52%) of granuloma was observed in hydatid cyst lesions in cow liver. Although this was an interesting case and caused the observation of all three types of lesions in the near layer of the hydatid cyst of host origin, but due to its small number, it cannot be considered as an epidemiological result. In many studies, including the present study, the rate of hydatid cyst in the lungs was higher than in the liver [19-20, 14-15, 12, 7-8].

In this study, in total, the infection rate of lungs (69.7%) was higher than liver (30.3%). This ratio was 77.3% in camels and cows and more than sheep (45.55%). This difference may be due to the higher average age of camels and cows compared to sheep or the larger body mass of camels and cows. Another reason for this difference may be the different geographical conditions of different regions. In this study and other studies conducted in Iran, the highest level of sterolitis has been reported in cattle. This rate has been reported between 30-70% [20-19, 12].

The final conclusion is that differences were observed in the fibrotic layer of hydatid cyst in camels, cows and sheep. The degree of progression of the lesion towards the formation of necrosis and granuloma in three types of animals and also, the frequency of these lesions had a significant difference, which may be due to the difference in the parasite strain, host type and also the average age of these animals.

Studies of specific methods such as histochemistry and immunopathology are suggested in future studies.

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