

FASCIOLA GIGANTICA PSEUDO-INFECTION IN GERMAN SHEPHERD DOGS

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SUMMARY

Fasciola gigantica is liver parasite of herbivorous animals and man. It cause great economic losses in livestock animals. Tracing the literature, few reports about *Fasciola gigantica* infection in dogs were recorded. In this study, five German Shepherd dogs were routinely examined before vaccination by stool analysis, it revealed presence of *Fasciola* eggs that were counted. Our investigation depend upon counting *Fasciola* eggs in dogs stool daily, history of food, measuring liver enzymes and bilirubin and examining dogs liver, gall bladders and bile ducts by ultrasonography. The results confirmed that it is *Fasciola gigantica* pseudo-infection where stool eggs count declined daily with stopping feeding on the *Fasciola* infected bovine liver and the *Fasciola* eggs completely disappear from stool at the 4th day. The liver enzymes and bilirubin are within the normal range. The ultrasound images of the liver, gall bladders and bile ducts of the five dogs were normal. It is concluded that a pseudo-infection with *Fasciola gigantica* could be happened in dogs that may confuse the Vets.

Keywords: *Fasciola*, pseudo-infection, stool analysis, liver enzymes, bilirubin, ultrasonography, dogs.

INTRODUCTION

Infection of dog and human with *Fasciola* is most common in villages and larger towns within rural areas, especially sheep- and cattle-grazing areas. Levels of infection depend on the frequency of eating plants from water bodies contaminated with animal faeces. Infection might also occur from contaminated drinking water or cooking utensils. The real fasciolosis could be happened in dogs that were fed raw liver infected with immature liver flukes as recorded by Dalton (1998).

False fascioliasis (pseudo-fascioliasis) refers to the presence of eggs in the stool resulting not from an actual infection but from recent ingestion of infected livers containing eggs. This situation can be avoided by having the patient follow a liver-free diet several days before a repeated stool examination as previously reported by **Demerdash et al. (2011)** and **Espino and Finlay(1994)**. Extrahepatobiliary infection of *Fasciola* is not common. extrahepatic fascioliasis has been reported in the subcutaneous tissue (**Aguirre et al.,1981**), brain (**Ruggieri et al.,1967**),, lungs (**Couraud et al.,1975**), epididymis (**Aguirre et al.,1981**), inguinal lymph nodes (**Arjona et al.,1995**) and in gastrointestinal system organs like the stomach(**Acosta-Ferreira et al.,1979**) and the cecum (**Park et al.,1984**).

Other disease problems can be associated with *Fasciola* such as pharyngeal infection (halzoun) in people who ate raw sheep or goat liver that contained immature flukes. In this problem, the larval flukes emerge from the liver and attach to the pharyngeal mucosa, it is most common in the Middle East (**Stephen and Richard, 2001**). It is clinically characterized by dysphagia, dyspnea, bleeding, and airway obstruction. The false fasciolosis increases the percentages of false positive diagnosis that depends on detection of *Fasciola* antigen in animal stool. In this study, we investigated the problem of pseudo-fascioliasis in German Shepherd Dogs by stool examination, liver enzymes and bilirubin estimation and ultrasonography.

MATERIALS AND METHODS

Animals

Four three months old German Shepherd puppies and their dam were brought to the teaching hospital of Medicine and Infectious Diseases in the Department of Medicine and Infectious Diseases at the Faculty of Veterinary Medicine, Cairo university, for the vaccination with Vanguard[®] (Pfizer company). They exposed to the routine clinical examination including stool analysis. They were kept as inpatient for 5 days and they were fed on liver free diets. The case history were recorded focusing on type of food and previous medication. The owner informed that he fed the dogs raw bovine liver slices and brought liver slices for examination.

Samples

Stool were collected from the five dogs daily for analysis. Blood were collected from the five dogs and sera were separated to measure liver enzymes and bilirubin. The rest of liver slices used as a diet for dogs were examined to detect both *Fasciola* flukes and eggs .

Stool analysis

The stool of the examined dogs were examined by concentration sedimentation techniques according to **Denham and Suswillo (1995)**. The *Fasciola* eggs were counted by using method of **Happich and Boray (1969)**, the actually counted *Fasciola* eggs were considered eggs per gram stool.

Blood chemistry

Liver enzymes (ALT and AST) and bilirubin (total and direct) were estimated according to **Sodikoff(2001)**.

Ultrasonography

The five dogs were examined by ultrasonography by the ultrasound device at department of surgery, anesthesiology and radiology. The liver tissues, gall bladders and bile ducts were examined to detect any abnormal changes.

Examination of the frozen bovine liver slices

The rest liver slices that were used for feeding of dogs, were examined for detection of *Fasciola* flukes or eggs. The slices were examined for detection of whole fluke or head of fluke that were found inside the bile ducts. The slices were also washed with tap water and the collected water was centrifuged, then the sediment was microscopically examined to detect *Fasciola* egg.

RESULTS

Stool analysis

Presence of the golden yellow oval operculated eggs measuring up to 170 μm long by 80 μm wide, that were identified as *Fasciola* eggs according to (**Aitken,2007; Stephen and Richard, 2001 ; Dalton,1998**) as shown in figure-1. The number of *Fasciola* eggs per gram stool declined daily as illustrated in table (1).

Ultrasonography

It showed normal liver tissue architecture, common bile ducts, bile ductules and gall bladder as illustrated in table-1 and figures (2 and 3).

Liver enzymes and bilirubin

levels of ALT, AST, direct and total bilirubin in sera of the examined dogs were within the normal range as presented in table-1.

Examination of the frozen bovine liver slices

Examination of the liver slices revealed presence of leaf like flukes in bile ductules, it measures up to 7.5cm long by approximately 1.2 cm wide. Characteristically, it has less well-defined shoulders and long straight sides that were identified as *Fasciola gigantica* worms according to (Stephen and Richard, 2001 and Aitken,2007). The microscopical examination of the sediment of the collected water used to wash liver slices showed presence of *Fasciola* eggs.

Table.1. Results of stool, liver enzymes, bilirubin and ultrasound examinations of dogs

	AGE in month	SEX	(number of Fasciola egg per gram stool					Liver enzymes		Bilirubin		Ultraso und of organs
			1 st Day	2 nd	3 rd	4 th	5 th	ALT	AST	T	D	
Puppy-1	3	F	7	2	0	0	0	37	29	0.5	0.1	Normal
Puppy-2	3	F	9	1	0	0	0	42	45	0.4	0.06	Normal
Puppy-3	3	M	5	3	2	0	0	56	38	0.45	0.07	Normal
Puppy-4	3	F	4	2	0	0	0	29	13	0.15	0.11	Normal
Dam	24	F	11	4	1	0	0	60	54	0.55	0.17	Normal

Normal values: Billirubin: T:Total (0-0.6 mg/dl), D:Direct (0-0.2 mg/dl)

Figure-1: *Fasciola gigantica* egg (X400)

Fasciola Gigantica Pseudo-Infection..

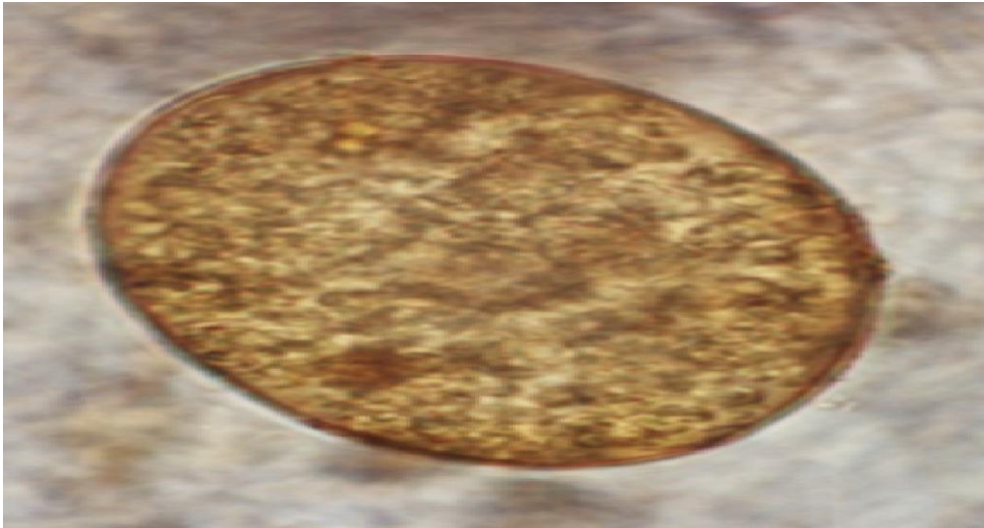


Figure-1: Fasciola gigantica egg (X400)



Figure-2: Ultrasonographic image showed normal liver architecture and gall bladder of German shepherd dog.

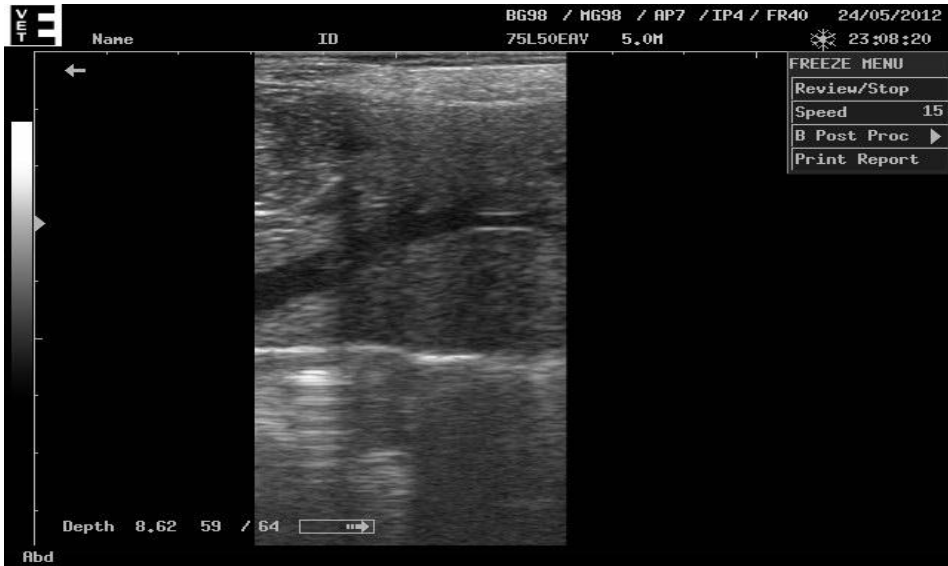


Figure-3: Ultrasonographic image showed normal liver, bile duct and ductules of German shepherd dog.

DISCUSSION

In the real clinical fascioliasis, encysted metacercariae ingested with grasses or water are the infective stage. Post-excystment, the metacercariae penetrate the intestine and enter the peritoneal cavity as immature flukes. They migrate to the liver after several days and penetrate Glisson's capsule during the acute hepatic phase of infection. After migration through the liver for 5–6 weeks, the worms enter the bile ducts to begin the chronic biliary phase. The flukes mature after 3–4 months and begin to lay eggs. *Fasciola* fluke secretes an average of 9000–25 000 eggs/day. *Fasciola* has a number of immune evasion mechanisms that allow for chronic fascioliasis to happen that are (i) rapid turnover of the glycocalyx, (ii) cleavage of surface-bound immunoglobulins by secreted proteases in order to prevent antibody-dependent cellular cytotoxicity, (iii) These proteases have also been shown to be directly toxic to host cells and to be capable of degrading extracellular matrix components, which may assist in tissue migration and (iv) proteases may have host cytokine mimicry, which enables the fluke to control the immune response (Stephen and Richard, 2001).

While in the pseudo-fascioliasis, nothing of the previous changes were happened. That was proved by (i) stool analysis and stool egg count for the examined dogs showed that the *Fasciola* egg count were declined until disappeared at the 4th day from the beginning of stool analysis and stopping feeding on liver. (ii) liver damage or bile duct obstruction were excluded by estimation the levels of ALT, AST, total bilirubin and direct bilirubin in sera of the examined dogs, the estimated levels were within the normal range as presented in table-1. That agrees with **Aitken (2007)**, who mentioned that ALT, AST, total and direct bilirubin levels can indicate chronic disease once *Fasciola* adult flukes are present in the biliary tree and may provide useful prognostic indicators. (iii) normal architecture of liver and the patency of gall bladder and bile ducts were also confirmed by ultrasonography. That showed no damage nor inflammation in the liver and no flukes nor fibrosis in the gall bladder and bile ducts as presented in figures (2&3).

In the pseudo-fascioliasis, the *Fasciola* eggs are passed through the digestive tract of the examined dogs without any change because it is highly resistant to the digestive enzymes and secretions. Conclusively, the pseudo-fascioliasis could happen in dogs eating liver infected with *Fasciola* adult flukes and misdiagnosed during the stool analysis of dogs and may confuse Vets. It could be differentiated from the real clinical fascioliasis by stopping feeding on liver, daily stool analysis at least for 5 days, measuring liver enzymes (ALT and AST) and bilirubin (direct and total) and finally ultrasonography of liver, gall bladder and bile duct that can confirmed the case of pseudo-fascioliasis.

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