

## Prevalence of *Fasciola* spp. Among Slaughtered Livestock in Zakho City, Duhok Governorate - Iraq

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### ABSTRACT

Fascioliasis is one of the most common neglected zoonotic disease caused by *Fasciola* species. The current study was conducted during January to December 2019, in which five districts were selected in various parts of Duhok governorates which differ in their environmental and geographical conditions they included: Duhok city, Zakho, Shelidiza, Aqrah and Bardarash. Each slaughter house was visited once monthly to determine the rate of fascioliasis among domestic animals; cattle, sheep and goats. The infected livers were examined grossly and the bile duct and gall bladder were inspected for the presence of *Fasciola* species, the work was performed in Parasitology Laboratory, Biology Dept., Faculty of Science, Zakho University. Fascioliasis was diagnosed in 2.0% (195/9757) of total slaughtered livestock, with the highest rate of infection in cattle which was 3.27% (66/2021), followed by sheep 1.76% (122/6932) and the lowest in goats 0.87% (7/804). Slightly higher prevalence rate from positive cases was found in local livestock compared to imported one (2.10% versus 1.88%).

According to gender, males showed higher prevalence than females (2.08% versus 1.08%). Regarding *Fasciola* species, among sheep and goats; the prevalence of *F. hepatica* was higher than *Fasciola gigantica*. While among cattle, *F. gigantica* was the most common species. With respect to parasite burden, the heaviest burden was recorded in 38.46% (20-100 flukes/liver) of slaughtered livestock with the heaviest being in sheep (55.74%), followed by goats (71.43%) while lightest burden was recorded in cattle in 62.12% (1-10 flukes/liver). Most of the liver flukes were recovered from the bile ducts (67.69%), followed by gall bladder (21.54%) and the least (10.77%) from both the bile ducts and gall bladder.

**Keywords:** Bile ducts, Gall bladder, Duhok.

### 1. Introduction

Fascioliasis is considered as one of the most common and economically important parasitic diseases of domestic livestock, in particular sheep, goats and cattle. The disease causes a considerable economic loss in cattle industry, mainly through mortality, liver condemnation, reduction in milk and meat production in addition to the expenditure for anthelmintic (Hillyer and Apt, 1997). The disease is caused by digenean trematodes of the genus *Fasciola*, commonly known as liver flukes. The most commonly implicated, species are *Fasciola hepatica* and *Fasciola gigantica*. This disease causes a serious effect to grazing animals (Torgerson, 1999). The economic losses due to fascioliasis are estimated to be more than 3.2 billion Dollars annually (Jaja *et al.*, 2017).

Lately, fascioliasis has been well studied and concerned because of its zoonotic feature, as it is both species can infect the humans (Schweizer *et al.*, 2005). Humans are incidental hosts and can acquire the infection by ingesting watercress or drinking water containing encysted cercariae. The illness occurs worldwide, particularly in regions with intensive sheep or cattle production. Incidence of human infection has increased over the past 20 years because of the large numbers of people and animals infected worldwide, fascioliasis causes considerable morbidity (Piedrafita *et al.*, 2010). The life cycle of *Fasciola* involves the existence of snail which acts as an intermediate host that spread the disease as a consequence of infection (Mas-Coma *et al.*, 2005). *F.*

*hepatica* infects the snail *Lymnaea truncatula*, which is frequently found in cold and mild climatic regions therefore, it is common in Europe, Australia and America (Mas-Coma and Bargues, 1997). Whereas, *F. gigantica* is most common in sub-tropical and tropical regions of Asia and Africa (Thanh, 2012). Detection and diagnosis of fascioliasis in domestic animal depend on classical parasitology diagnosis technique such as coprological investigation besides identifying the morphological features of the parasite (Thanh, 2012). Although coprological examination is an easy and cheaper methods besides had a high specificity, but has low sensitivity especially between closely related species when using eggs morphology (Adedokun *et al.*, 2008).

Some studies on the prevalence of fascioliasis have been performed in Iraq and they reported variable rates. In Mosul city Al-Mahmood and Al-Sabaawy (2019), reported a rate of 4% (17/425) in cattle, while a very high rates 54.7% and 23.71% were reported among cattle and buffalo, respectively in Thi-Qar (Gatie *et al.*, 2018). In Kirkuk rates of 0.26%, 1.04 % and 1.16%, were recorded among sheep, goats and cattle, respectively (Abass *et al.*, 2019). In Karbala a rate of 3.61% was recorded among infected cows (Hussain *et al.*, 2017). Although many epidemiological studies were performed on the prevalence of *Fasciola* among domestic animals in the country, most of them dealt with one species of animals and in one area of the country. Hence, the present investigation estimated the rate of fascioliasis among cattle, sheep and goats in different districts of Duhok and also involved the local breed and the imported one to get more inside about the prevalence of fascioliasis and to correlate the subsequent data with some risk factors.

## 2. Materials and Methods

### 2.1 Study Area and Sample Collection

The study area included five districts in various parts of Duhok governorates which differ in their environmental and geographical conditions they

included: Duhok city, Zakho, Shelidiza, Aqrah and Bardarash. The study was conducted between January and December of 2019, a monthly visit was made to the slaughter house of each district of Duhok governorate. Liver flukes were isolated from the gall bladder, bile ducts and both of them of 195 infected livestock (122 sheep, 7 goats and 66 cattle/9757 examined) at different abattoirs. The infected livers were examined grossly and the bile duct and gall bladder were inspected for the presence of *Fasciola* species by applying the routine inspection procedures to internal organ. Several incisions were made in different parts of the liver for confirming the presence of liver flukes in the parenchyma of liver, liver pieces were pressed in order to squeeze the flukes out from the tissue and bile ducts. Each isolated liver fluke was examined and classified on the basis of shape and size (Soulsby, 1982). Regarding the source of imported animals, the sheep and cattle were imported mostly from neighboring countries such as Syria, Turkey, Iran and some from other countries like India, Brazil, Romania, and Vietnam. While the source of all goats was from Kurdistan region-Iraq. The numbers of total animals slaughtered and infected and non-infected animals in each district were carefully recorded on standardized data sheets designed for the study.

### 2.2 Data Analysis

The statistical package for social sciences (SPSS) (Kauffman *et al.*, 2000) software was used for analyzing the data. The relative proportions were calculated with a confidence interval of 95%. To determine the association between variables, the chi-squared ( $\chi^2$ ) test was used; a  $p$ -value < 0.05 was considered significant and more than that considered non-significant.

### 2.3 Result and Discussion

Table (1) shows the prevalence of *Fasciola* spp. among slaughtered livestock, the overall rate of infection was 2.00 % (195/9757). According to the animal species, cattle showed the highest rate of infection 3.27%

(66/2021), followed by sheep 1.76% (122/6932), while only 0.87% (7/804) of goats were infected. The overall, rate of fascioliasis was slightly higher among the local livestock as compared to imported one, which was 2.10% (111/5294) vs 1.88% (84/4463), respectively. Furthermore, local cattle showed the highest rate (11.63%), followed by local sheep (2.04%). In general, statistical analysis of the results showed the presence of significant difference ( $P < 0.05$ ) in the overall rates of infection between animals' species and source.

In this study, the highest prevalence of infection was seen among slaughtered cattle (3.27%), which is close to the rates reported in other studies such as, in Erbil a rate of 3.44% in cattle (Koyee *et al.*, 2011) and in Kirkuk (2.63%) by Rasheed and Kadir (2008). Regarding the rate of fascioliasis among infected sheep, 1.76% is much lower than the rate reported in a study by Meerkhan and Razak (2013) as they reported a rate of 16.8% in sheep in Duhok. Also, very high rates in sheep have been reported from Dewania, Babylon and Abu-Ghraib district, which were 76.6%, 72%, 35% and 12.73% (Al-Khafaj, 2011; Abass *et al.*, 2018; Abdalnabi, 2012 and Oleiwi *et al.*, 2017), respectively. Regarding the rate of fascioliasis among infected goats, in the current study the lowest rate was reported (0.87%), which somewhat near the rate reported by Abass *et al.* (2019) in Kirkuk which was 0.7%. Whereas, Abdalnabi (2012) in Babylon and Koyee *et al.* (2011) in Erbil, recorded rates of 68.4% and 3.62%, respectively in goats. Higher rates of fascioliasis than that of the present study have been reported in neighbor countries such as Iran (66.6% in cattle, 93% in sheep) (Akca *et al.*, 2014) and Turkey (31.4% in sheep) (Acici *et al.*, 2015).

These results confirm that the prevalence of fascioliasis is much lower than in the neighboring countries in spite of the lack of reliable anthelmintic controlling regimes in Iraq during the last decade. The highest rate of fascioliasis among cattle followed

by sheep and lowest among goats could be due to the difference in the grazing style of animals on grassland as goats eat leaves and heaths in mountain areas, high percentage of fascioliasis among cattle may be due to that cattle were more frequently grazing near springs, pools, streams and river where naturally snails are present, hence getting more contacts with encysted metacercariae, regarding sheep they often graze on open land where getting less chance of contact with infected metacercariae and therefore the risk of their infection with fascioliasis is reduced (Kordshooli *et al.*, 2017).

With respect to animal source, in general, the overall rate of fascioliasis among local livestock was slightly higher than imported one (2.10% vs 1.88%), furthermore, local sheep and cattle also showed higher rates than imported one (2.04% vs 1.28%) in sheep and (11.62% vs 2.70%) in cattle, respectively. In this regard the present results are consistent with the studies conducted in Egypt by Hussein and Khalifa (2010) as they reported high rate of infection (78.7%) from local cow and lower (21.3%) from imported one. In Ethiopia Abebaw *et al.* (2012) reported higher rate of infection in local sheep (72.65%) than imported sheep (44%). Also, in Nigeria Aliyu *et al.* (2014) reported high rate of fascioliasis among local cattle breed (15.6%) with no infection among the imported one.

On the other hand, in Saudi Arabia, Sanad and Al-Megrin (2005) reported higher infection rate (15.1%) with fascioliasis among imported sheep than the local ones (4.96%). In another study also in Saudi Arabia, Degheidy and Al-Malki, (2013) reported higher prevalence of fascioliasis among imported cattle which was 8.6%. While in Turkey, Yildirim *et al.* (2007) they did not find any significant association among sheep infected with *F. hepatica* and the type of breed. These differences among local and imported breed may be due to local geographic disruption and low levels of awareness of these infectious diseases

among domestic animals, or attributed to grazing in areas that could be heavily infested with intermediate host (Aliya *et al.*, 2014).

The highest rate of fasciolosis in sheep and goats was reported at Shelidiza abattoir which were 6.58% and 1.64%, respectively. While the highest rate of infection for cattle was at Zakho abattoir which was 3.91%. With significant differences between the rate of infection and types of animal's slaughtered in different slaughter houses ( $P < 0.05$ ) as shown in table (2).

**Table 1: the prevalence of *Fasciola* spp. in sheep, cattle and goats of both sources slaughtered in Duhok Governorate**

Host type	Total No. of slaughtered animals	Total No. of infected animals	% of infected animal	Source of Animal					
				Imported			Local		
				Total No. slaughtered animals	No. of infected animals	% of infected	Total No. slaughtered animals	No. of infected animals	% of infected
Sheep	6932	122	1.76	2571	33	1.28	4361	89	2.04
Goats	804	7	0.87				804	7	0.87
Cattle	2021	66	3.27	1892	51	2.70	129	15	11.63
<b>Total</b>	<b>9757</b>	<b>195</b>	<b>2.00</b>	<b>4463</b>	<b>84</b>	<b>1.88</b>	<b>5294</b>	<b>111</b>	<b>2.10</b>

P < 0.05 analyzed using the SPSS software using the chi-squared (x2) test

A clear variation in the prevalence of fasciolosis among different districts of Duhok governorate, was observed which can be attributed to many factors such as geographical variations, soil composition, climate conditions, besides the life cycle of *Fasciola* spp. in a given area depends on the availability of the snail hosts, which dramatically influenced by the temperature and the amount of Rainfall, since the development of the eggs and the intramolluscan stages of the parasite depends on the environmental conditions, in general all these factors play an important role in the prevalence of fascioliasis among definitive hosts in a given area (Yadegari *et al.*, 1990; Torgerson and Claxton, 1999; Rokni 2008; Kooshan *et al.*, 2010).

Regarding the gender of infected animal, males showed a higher rate of fascioliasis than females (Table 3), The overall rate in males was 2.08 % versus 1.08% in females. Regarding livestock species, the highest rate was reported among female cattle, which was 3.28%. followed by sheep (1.83%). Statistically significant difference ( $p < 0.05$ ) was observed in the rate of fascioliasis in relation to the gender of infected animals, Similarly, Asnake *et al.* (2018) in Ethiopia reported higher rate of fascioliasis in male sheep than females which were 17.3% vs 15.6%. Magaji *et al.* (2014) stated that the prevalence of fascioliasis in cattle revealed more common in males than in females and attributed it to the disparity in susceptibility to helminth infection between the both sexes which could be due to the differences in the host intrinsic factors

**Table 2: The prevalence of *Fasciola* splices. among slaughtered livestock in Duhok governorate slaughter houses.**

Type of livestock house name	No. slaughtered	No. infected	% of infection
Sheep			
Duhok	3443	79	2.29
Zakho	2653	19	0.72
Aqrah	676	15	2.22
Shelidiza	76	5	6.58
Bardarash	84	4	4.76
Total of sheep	6932	122	1.76
Goats			
Duhok	146	0	0
Zakho	253	1	0.40
Aqrah	323	5	1.55
Shelidiza	61	1	1.64
Bardarash	21	0	0
Total of goats	804	7	0.87
Cattle			
Duhok	370	9	2.43
Zakho	1022	40	3.91
Aqrah	181	6	3.31
Shelidiza	157	3	1.91
Bardarash	291	8	2.75
Total of cattle	2021	66	3.27
Total No	9757	195	2.00

P < 0.05 analyzed using the SPSS software using the chi-squared (x2) test

(genetics, physiology) or the number of female animal's slaughtered is lesser than males besides that the vast majority of females are kept for breeding and milk production (Magaji *et al.*, 2014; Asnake *et al.*, 2018).

**Table 3: The prevalence of *Fasciola* species according to gender**

Host	Male			Female		
	No. of slaughter	No. of infective	%	No. of slaughter	No. of infective	%
Sheep	6216	114	1.83	716	8	1.12
Goats	734	7	0.95	70	0	0.00
Cattle	1977	65	3.29	44	1	2.27
Total	8927	186	2.08	830	9	1.08

P < 0.05 analyzed using the SPSS software using the chi-squared (x2) test

*Fasciola hepatica* was recorded at a rate of 66.15% (129/195) of total infection, with the highest being in sheep 59.49% (116/195), while lower rates of 3.59% (7/195) in cattle and 3.08% (6/195) in goats. *Fasciola gigantica* was reported in cattle only at a rate of 18.97% (37/195), while single infections with *F. gigantica* was not recorded in sheep and goats. Furthermore, mixed infections with both species were recorded at a rate of 14.87% (29/195) with the highest being in cattle, in 11.28% (22/195), as shown in table (4). Hence significant difference was observed among species of *Fasciola* in sheep, cattle and goats (p<0.05).

The present results agreed with other studies, in which *F. hepatica* was most prevalent in sheep and goats (Yusuf *et al.*, 2016; Piri *et al.*, 2018) and *Fasciola gigantica* was more common among cattle (Abunna *et al.*, 2009). This indicate high susceptibility of sheep to *F. hepatica* than *F. gigantica*, this might be due to several reasons such as, the biochemical characteristics of *Fasciola* and the immune response of the host against fasciolosis. *Fasciola hepatica* showed effective defenses mechanisms against nonspecific immune response (free radical killing by macrophages or eosinophils) (Meeusen and Piedrafita, 2003; Pleasance *et al.*, 2011).

Hence non-specific immune cells such as eosinophils play a major role against *F. gigantica* than *F. hepatica* in infected sheep. Furthermore, the excretory/secretory

products (ESPs) of *F. gigantica* stimulate the immune response of the T cells, B cells, NK cells against *F. gigantica* in early stage of infection and as a consequence an early proliferation of T-helper cell against *F. gigantica* occur which may lead to macrophages activation and resulting in elimination of juvenile *F. gigantica* (Meeusen and Piedrafita, 2003; Zhang *et al.*, 2004).

Regarding the parasites burden among different types of slaughtered animals, the heaviest burden (>20-100) per host liver was recorded in 38.46% (75/195) of slaughtered livestock, with the heaviest being in sheep which was 55.74% (68/122). While 71.43% (5/7) of goats had the moderate worm burden and 62.12% (41/66) of cattle had the lightest worm burden. Statistical analysis showed the presence of significant relationship (P<0.05) between the number of *Fasciola* species and the types of infected animals.

**Table: 4 The rate of infection with *Fasciola* species according to host type.**

Species of <i>Fasciola</i>	Host	No. of infected	% of infection
<i>F. hepatica</i>	Sheep	116	59.49
	Goats	6	3.08
	Cattle	7	3.59
Total <i>F. hepatica</i> infections		129	66.15
<i>F. gigantica</i> only	Sheep	0	0
	Goats	0	0
	Cattle	37	18.97
Total <i>F. gigantica</i> infections		37	18.97
Mixed infection	Sheep	6	3.08
	Goats	1	0.51
	Cattle	22	11.28
Total of mixed infections		29	14.87
Total Infections		195	100

P < 0.05 analyzed using the SPSS software using the chi-squared (x2) test

The heavy burden of liver flukes observed in sheep in this study, may be due to that most slaughtered sheep were young aged and young animals usually are more susceptible to infection with heavy burden of parasites. while most infected cattle carry light burden of parasites with a ratio of 62.12%, this may be due to that cattle have more immune response against

invasion with liver flukes compared to sheep and goats (Mulcahy *et al.*, 1999; Phiri *et al.*, 2006).

**Table 5: Number of parasites isolated (burden) from each infected animal**

Types of animals	No. of animals Infected	1-10 Light	>10-20 Moderate	>20-100 Heavy
Sheep	122	27/122 (22.13)	32/122 (26.23)	68/122 (55.74)
Goats	7	2/7 (28.5)	5/7 (71.43)	0
Cattle	66	41/66 (62.12)	18/66 (27.27)	7/66 (10.60)
Total	195	70/195(35.90)	55/195(28.21)	75/195(38.46)

P < 0.05 analyzed using the SPSS software using the chi-squared (x2) test

With respect to the location of the parasites within the infected organ (bile ducts and gall bladder) of infected animals, most of the liver flukes were recovered from the bile ducts (67.69 %) followed by gall bladder (21.54%) and the least (10.77%) from both the bile ducts and gall bladder. Regarding animal species, in 85.71% of infected goats, the parasites were recovered from the bile ducts, followed by cattle in 72.72% and in 63.93% of sheep. Statistically non-significant difference was observed between the location of the parasites and the types of infected animals as shown in table (6).

**Table 6: Prevalence of Fasciola species according to parasite location**

Types of animals	No. of infected animals	Bile duct No. (%)	Gall bladder No. (%)	Bile ducts & Gall bladder No. (%)
Sheep	122	78 (63.93)	29 (23.77)	15 (12.30)
Goats	7	6 (85.71)	1 (14.29)	0
Cattle	66	48 (72.72)	12 (18.18)	6 (9.09)
Total	195	132 (67.69)	42 (21.54)	21 (10.77)

P value = 0.514 analyzed using the SPSS software using the chi-squared (x2) test

Most of *Fasciola* species in this study were isolated from the bile ducts of cattle, sheep and goats than other organs. Since the bile duct is the main organ for maturation of adult flukes, especially in light and moderate burden of parasites. Gall bladder of infected animals carries the parasites in case of heavy burden of liver flukes as shown in the case of infected

sheep (table 6). The percentage of *Fasciola* species recovered from bile duct of cattle was 72.7% was slightly higher than that recovered from the bile ducts of sheep 63.9 % because of grazing style of cattle which is different from that of sheep, where cattle frequently spend more of their life near rivers and pools area where there are a plenty of vegetations on which the cercariae can encyst, hence they have more chance to acquire infection (Kordshooli *et al.*, 2017; Takeuchi-Storm *et al.*, 2018). Adult flukes and their eggs may trap in the bile duct because of intensive calcification of bile duct which more commonly occurs in cattle after several week of infection, hence most infected cattle have light burden of liver flukes compared to other domestic animals (Phiri *et al.*, 2006; Bouvry & Rau 1986).

In conclusion, in Duhok governorate, both *F. hepatica* and *F. gigantica* were isolated from local and imported slaughtered livestock. Moreover, fascioliasis was more prevalent in cattle. Therefore, strategic prevention should be initiated by regular deworming of local breed before and after rainy season in order to control the disease in snail host. In addition, proper inspection at abattoirs should be intensified and quarantine regulations should address the careful inspection of live imported animals to avoid the introduction of exotic parasites.

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