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An Incidence of intestinal protozoa infection in sheep, sheep handlers and non-handlers in Wasit Governorate/ Iraq

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Abstract
Intestinal protozoa in sheep and human usually incriminated in diarrhea, when there are suitable conditions in the intestinal lumen that promote the parasite multiplication. This study aimed to investigate the cyst and trophozoites infection in sheep and handlers. One-hundred eighty fecal samples from sheep and 50 from handlers, were collected from three different areas (Al-Hafriya, Al-Suwaira, and Al-Azizia) in Wasit governorate. Sheep aged 7-36 months, while handlers were 10-40 years. Fecal samples were examined directly and by staining methods to detect intestinal protozoa cysts and trophozoites. Al-Suwaira showed highest infection rates, 91.66%, and 87.5%, in sheep and handlers, respectively. Male represented higher infection rates than female in sheep (90.69%) and handlers (75%). In conclusion, this study approved the incidence of intestinal protozoa infection in sheep and sheep handlers. The authors suggest doing another future study in different areas of the Wasit to investigate the prevalence rate of the intestinal protozoa.

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Introduction

Parasitic diseases contribute significantly to the burden of infectious diseases worldwide. They are incriminated in significant illness and deaths affecting people in developing countries, which are mostly of zoonotic nature (Ortega *et al.*, 2008). Illnesses usually brought by livestock animals, domestic pets, and open farms cause several zoonotic outbreaks (Chalmers *et al.*, 2011). Enteric protozoa inhabiting the intestinal tract of higher vertebrates are a diverse group of unicellular parasites, resulting in diarrhea, which is a relatively frequent symptom and asymptomatic colonization (Cama and Mathison, 2015). Infections usually occur through ingestion of cysts/oocysts, contaminating raw food or drinking water (Baldursson and Karanis, 2011; Torgerson *et al*., 2015). Diarrhea is not precisely attributed to an identified parasite in a patient's feces for all protozoa. Vice versa, some intestinal protozoa

such as *Giardia duodenalis*, *Entamoeba histolytica, Cryptosporidium spp., Cyclospora cayetanensis, Cystoisospora belli*, and *Microsporidia spp.* have been indeed recognized to cause diarrhea in humans (Agholi *et al.,* 2013). The parasitic infections in people regard as a major problem in the world especially in communities suffering from poor sanitation and low personal hygiene such as primary schools pupils and rural communities or in animal breeders (Garcia and Bruckner, 1993; Hussin *et al.,* 2015; Hussin *et al.,* 2016). Review of literature revealed scarce published studies regarding the incidence of intestinal protozoa infection in sheep and human in contact. Therefore, this study intends to investigate the impact of intestinal protozoa infection in sheep, sheep handlers, and non-handlers in three cities in Wasit governorate/ Iraq.

MATERIALS AND METHODS

Samples of study

Human

The study comprised of 50 subjects who had been in contact with the sheep. Their ages ranged from 10-40 years including 32 males and 18 females. In addition, 50 apparently healthy individuals including 36 males and 14 females, their ages were also 10-40 and hadn't contact with animals and acted as controls.

Sheep

One hundred eighty fecal samples collected from sheep. The criteria for their inclusion depend on their contact with persons included in this study.

Fecal samples

Single Stool samples were collected and processed for the detection of trophozoites or cyst stages of intestinal protozoa. The following methods were used for detection of protozoa: direct smear method, concentration method and modified Ziehl-Neelsen method for the identification of red-pink oocysts of acid-fast parasites according to Markell *et al.,* (1986).

Results and Discussion

Several studies on the prevalence of intestinal parasites have been done all over the world and have shown variations in the prevalence of infections depending on geographical regions and localities, sanitary environment and hygienic habits of people living there (Gleason *et al.,* 1970; Abu-Zeid *et al.,* 1989). The results of this study showed that the overall infection rate of intestinal protozoa in sheep collected from the three regions of Wasit province was 83.33% comparable with 72% in human without significant differences between the studied regions (Table.1). These results are in agreement with Abd Al-Wahab, (2003), who recorded an infection rate of intestinal protozoa reached 85% in lambs in Baghdad Province. The reported protozoa as follow: *Cryptosporidium sp*, *Entamoeba sp*, *Giardia sp*, *Eimeria sp* and *Buxtonella sulcata* in sheep. Moreover, results of this study approved that *Eimeria sp* and *Buxtonella sulcata* were not observed in human stool. Nevertheless, only *Entamoeba sp* recognized in the non-handlers stool (Table. 2). Regarding the distribution of these protozoa in sheep and human, the results of this study approved the zoonotic impact and nature of some parasites. However, some of this protoza didn't show this zoonotic impact (Al-Zubaidi and Al-Mayah, 2011; Al-Seady and Kawan, 2014).

Table (1): Shows the infection rate of intestinal protozoa according to the region of study

Region of study	kind of Samples	No. of Samples Examined	No. of Positive samples	Infection Rate %
Al-Hafriya	Sheep	60	49	81.66
	Handler	18	10	55.55
	Non Handlers	18	Nil	0
Al-Suwaira	Sheep	60	55	91.66
	Handler	16	14	87.5
	Non Handlers	16	2	12.5
Al-Azizia	Sheep	60	46	76.66
	Handler	16	12	75
	Non Handlers	16	2	12.5
Total	Sheep	180	150	83.33
	Handler	50	36	72
	Non Handlers	50	4	8

Table (2): Shows species of intestinal protozoa isolated from sheep and handlers

Parasite	Kind of samples	No. of Samples Examined	No. of Positive samples	Infection Rate %
Cryptospordum sp	Sheep	180	91	50.55
	Handler	50	26	52
	Non- Handlers	50	0	0
Giardia sp	Sheep	180	11	6.11
	Handler	50	12	24
	Non- Handlers	50	0	0
Entamoeba sp	Sheep	180	71	39.44
	Handler	50	6	12
	Non- Handlers	50	0	4
Eimeria sp	Sheep	180	42	23.33
	Handler	50	Nil	0
	Non- Handlers	50	0	0
Buxtonella sulcata	Sheep	180	23	12.77
	Handler	50	Nil	0
	Non- Handlers	50	0	0

The highest infection rate with intestinal protozoa in sheep was recorded (Table. 3) in September, November and December (100%), while the lowest infection rate was during February (36.66%). These results are in agreement with Abd Al-Wahab, (2003) and incompatible with studies performed by Al-Zubaidi (2012) and Al-Seady & Kawan (2014) in Baghdad governorate. This variation may be attributed to the suitable environmental condition, e.g. temperature, humidity, and raining season, that providing an appropriate media for the survival of the cysts (Xiao, 1994). In sheep, the infection rate was 90.69% and 81.02% for male and female, respectively. The current study revealed that both gender and age has no significant differences concerning the incidence rate of protozoal diseases (Tables. 4, 5). This results revealed the exposure of both sexes and all ages to same environmental, management and pathogenic factors (Minnat, 2014). However, younger animals show higher susceptibility to infection than adults do and this is mostly related to the lowered immunity or to crowding stress and managements in the farm, where much of sheep get in contact in or access into the infected fields without any preparation plan against the infection.

Tables (3): Shows infection rate of intestinal protozoa according to months

Months	Kind of samples	No. of Samples Examined	No. of Positive samples	Infection Rate %
September	Sheep	30	30	100
	Handler	7	6	85.71
	Non Handlers	7	Nil	0
October	Sheep	30	25	83.33
	Handler	9	7	77.77
	Non Handlers	9	Nil	0
November	Sheep	30	30	100
	Handler	9	5	55.55
	Non Handlers	9	Nil	0
December	Sheep	30	30	100%
	Handler	7	6	85.71
	Non Handlers	7	Nil	0
January	Sheep	30	24	80
	Handler	9	7	77.77
	Non Handlers	9	2	22.22
February	Sheep	30	11	36.66
	Handler	9	5	55.55
	Non Handlers	9	2	22.22
Total	Sheep	180	150	83.33
	Handler	50	36	72
	Non Handlers	50	4	8

The acquired immunity due to previous exposure could explain the low infection rate of gastrointestinal parasites in the adult population, which was approved in the previous studies (Bhat *et al.*, 2012; AL-Gelany, 2003). These results are compatible with Al-Seady and Kawan (2014) and Minnat (2014) in Baghdad and Diyala governorate / Iraq. The results of this study also showed that the sheep handlers revealed highest infection rate with intestinal protozoa (Table. 3) in September and December (85.71%), while the lowest infection rate was during November and February (55.55%)%) with no significant differences ($p>0.05$). These results are incompatible with AL-Gelany, (2003). These variations might be explained by the existence of suitable environmental factors such as temperature and moisture that facilitated spread and completing the life cycle of these parasites (Kirkpatrick and Farrell, 1984; Wallis *et al.*, 1984).

The infection rate were (75%) and (66.66%) in males and females respectively, although the males showed higher percentages, but no significant differences ($p>0.05$) was observed. Results shown by this study (Table. 4) agreed with AL-Shaheen *et al.*, (2007), and Al-Gelany (2003). Al-Hanoon and Mukhlis, (1982) argued that social customs and the abundance male activity increases the chance of exposure to sources of infection than females. The highest infection rate in the 10-30 year aged groups (Table. 5) supported the idea that acquired immunity from previous infection in older ages play a great role in defeating the protozoal infection. Vice versa, the close contact of the children to the animals and their feces made them more prone to the protozoal infection (Mahdi and Ali, 2002).

Table (4): Infection rate of intestinal protozoa in sheep, Handlers and non-handlers according to sex

Sex	Kind of samples	No. of Samples Examined	No. of Positive samples	Infection Rate %
Male	Sheep	43	39	90.69
	Handlers	32	24	75
	Non Handlers	36	2	5.55
Female	Sheep	137	111	81.02
	Handlers	18	12	66.66
	Non Handlers	14	2	14.28
Total	Sheep	180	150	83.33
	Handlers	50	36	72
	Non Handlers	50	4	8

Table (5): Infection rate of intestinal protozoa in Sheep, Handlers and Non-handlers according to age groups

Kind of samples	Age groups	No. of Samples Examined	No. of Positive Samples	Infection Rate %
Sheep (months)	7-12	70	64	91.42
	13-18	15	12	80.00
	19-24	42	32	76.19
	25-30	27	22	81.48
	31-36	26	20	76.92
Handlers (years)	10-20	30	24	80.00
	21-30	12	10	83.33
	31-40	8	2	25.00
Non handlers (years)	10-20	18	Nil	0
	21-30	20	2	10.00
	31-40	12	2	16.66

In conclusion, this study raised the zoonotic nature of *Cryptospordium sp., Giardia sp.* and *Entamoeba sp.,* which are transmitted from sheep to their handlers, in opponent to *Eimeria sp* and *Buxtonella sulcata*, the non-zoonotic.

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