EPIDEMIOLOGICAL STUDY OF CRYPTOSPORIDIOSIS AND DISEASE - ASSOCIATED FACTORS IN SHEEP IN A STEPPE REGION OF ALGERIA (AIN OUSSERA)

Dahmani Hichem[1]*, Ahcen Hakem[2], Mustapha Oumouna[3]

*[1]*University Saad Dahleb Blida 1, Algeria Research Laboratory for Biotechnology Related to Animal Reproduction; [2]University Ziane Achour Djelfa, Algeria, [3]University Doctor Yahia Faress Medea, Algeria; *Corresponding author: dahmanivet@gmail.com*

Abstract: This work has focused on the search for *Cryptosporidium* sp. carried out on a sample of 1 605 animals (1 235 lambs, 300 ewes, and 70 rams) from sixty-two flocks distributed in the commune of Ain Oussera one of the steppe regions of Algeria between 2014-2016., fifty-two flocks were positive for *Cryptosporidium* sp screening, with a flock prevalence 84% (CI95 % 72,8 % - 91 %).The total individual prevalence was 14.6%, the prevalence of lamb infestation was 15,4 %, for ewes and rams were 13,7 %, 4,3 % respectively ($p < 0,05$).

Key words: *Cryptosporidium*, Lamb, prevalence, Ain Oussera, Algeria

INTRODUCTION

Cryptosporidium is a ubiquitous enteric protozoan pathogen of vertebrates, and although recognised as a cause of disease in humans and domestic animals for over 50 years (2).

The importance of cryptosporidiosis in veterinary medicine stems from the fact that it is a difficult disease to control in many livestock, leading to significant economic losses (18, 3). In addition, infected animals pose a risk to humans given the zoonotic potential of this parasite (19). The sheep population is about 97,500 head, and no one can dispute the importance of sheep farming in steppe. It is a first-class resource for livestock farmers in the region because meat (fattening) and d lambs (breeding) are the main activity of breeders.This ovine population is confronted with several diseases that despoil the economy of this rural population which has no other subsistence resource than livestock, Risk factors such as flock size, farm hygiene, litter type, diet, season, prior existence of diarrhea, age, play a role in the transmission of species of *Cryptosporidium* sp in domestic animals (16, 5,4).

In Algeria, infestation with cryptosporidia has been the subject of few epidemiological studies in the ovine species, and studies relating to cryptosporidiosis are limited. Most of the recent publications related to the study of cryptosporidiosis and its prevalence concern the calf (17, 1,11). In sheep, so far no overall work has been done to study the prevalence and factors associated with the disease. These considerations prompted us to undertake a survey of cryptosporidiosis in sheep aimed at assessing the prevalence and relationship with certain factors known to predispose to cryptosporidian infestation in this steppe area of Algeria.

MATERIAL AND METHODS

1-Samples

The target population was flocks with more than 70 heads (lambs and sheep), who agreed to participate in the survey, the number of flocks required in these conditions was

62. In each flock we took a sample of all the lambs of one to six months, whether they were diarrhea or not and, 4 or 5 ewes and 1 or 2 rams were chosen empirically.

2- **Technique**

The modified Ziehl-Neelsen staining technique (9) was carried out after prior enrichment by the Ritchie method for the detection of *Cryptosporidium* sp oocysts.

3- **Statistical Treatments**

All the data collected was entered in a Microsoft Excel file. The chi-square test, with the correction of Yates, or even the exact test of Fisher, were used according to their indications, using a threshold of significance for the risk α of 0.05 to study the association of known factors to be associated with the prevalence of the parasitosis studied. Percentage confidence intervals were calculated for a 95 % probability.

RESULTS AND DISCUSSION

The total number of flocks randomly selected and able to participate in the study was 62. Out of a total of 1 605 samples, 1 235 were from lambs and 370 from adult animals (300 ewes and 70 rams). The prevalence of *Cryptosporidium* spp at the flock level was 84% (95% CI: 72,8% - 91%), which is identical to the 84% prevalence flock recorded in Spain by (5) and very far than 3% reported in Iran by (14).

The prevalence individual was 14,6%, comparable to that reported in Spain 15% by (12) and upper than11,3% in Iran (9), but lower than 85 % in the United States of America (20). This can be explained by the difference in climate, the breeding behavior and choice of the population studied. These results show that *Cryptosporidium* spp. is widespread in the studied farms. The prevalence of infestation in lambs was 15.4%, lower than (84,6 %) in Spain (5) respectively ; in ewes, it was 13,4 %, higher than that observed in rams 4,3%. Ewes and rams are asymptomatic immune-competent animals that can be a source of contamination, which is why they should be diagnosed early to limit infestation in the flock (Table 1).

Table1

Prevalence of *Cryptosporidium* sp on a sample of sheep in the Aïn Oussera region (2014-2016)

Sheep population	Number of samples	Number of positive samples	%
Ewes	300	41	13,6
Rams	70	3	4,3
Lambs	1 235	190	15,4
Total	1 605	234	14,6

P=0,03

Cryptosporidium sp was more frequently isolated in diarrheal than non-diarrheal lambs (32,1 % vs 5,8 %, respectively), (Table 2) aged essentially two weeks (28 %) with a higher degree of infestation (2 and 3) than the other age groups (table 3).

Cryptosporidium sp, was detected in all age groups but at different percentages (p = 0, Table 3). At the first week of life 16% of the lambs were infested with *Cryptosporidium* sp. The 2-week-old lamb showed the highest rate of parasite infection (28%), this rate was lower in lambs between 1 and 3 months 7% with an equal infestation level 1, (Table 3, 4) which agrees with other observations (15,8).

Table 2

Prevalence of *Cryptosporidium* sp in lambs (n = 1 235) according to their clinical status (Ain Oussera region, 2014-2016)

	Search result oocysts		
Lambs	Total	Positif	(%)
Diarrhoeics	448	144	32,1
No Diarrhoeics	787	46	5,8
Total	1 235	190	15,4

P=0

Table 3

Percentage of positive feces by age of lambs

Age	**Number of samples**	**Positive +**	**Prevalence (%)**
< 1 Week	175	28	16
2 weeks	300	83	28
3 weeks	290	40	14
4 weeks	220	25	11
1 à 3 months	160	11	7
3 à 6 months	90	3	3
Total	1 235	190	15,4

P=0

The higher percentage of two-week-old lambs with a positive result (28%) is associated with significantly higher levels of infestation (2 and 3) than for lambs in other age groups (p = 0; table 4).

Table 4

Distribution of lambs according to their degree of fecal infestation by *cryptospridium sp* and age until four weeks (n = 985)

Age	**Number of samples**		**Degree of infestation**			**Total samples**
	Negatives (%)	Positifs (%)	1 (%)	2 (%)	3 (%)	
1 week	147 (84)	28 (16)	20 (11)	2 (1)	6 (3)	175
2 weeks	217 (72)	83 (28)	32 (11)	31 (10)	20 (7)	300
3 weeks	250 (86)	40 (14)	26 (9)	9 (3)	5 (2)	290
4 weeks	195 (89)	25 (11)	18 (8)	5 (2)	2 (1)	220
Total	809 (82)	176 (18)	96 (10)	47 (5)	33 (3)	985

P=0

The distribution of the parasite does not seem to be related to the pregnancy state of the ewe, 16% in the pregnant ewes vs 11 % in the ewes after the part,(Table 6) (Naciri, 1994; Ye et al.,2013) observed a peak of oocysts excretion in sheep at lambing time between one week before and one week after lambing , this may be explained by the excretion of oocytes by the pregnant ewes that increase during this period.

Table 6

Percentage of positive fecal specimens by *cryptospridium sp* in the ewe before and after parturition

Befor parturition				**After parturition**			
Weeks	Number of samples	Cas positifs	Prevalence %	Weeks	Number of samples	Cas positifs	**Prevalence %**
3	40	5	12	1	30	5	**17**
2	70	11	16	2	80	9	**11**
1	40	8	20	3	40	3	**7**
Total	150	24	16	Total	150	17	**11**

P=0,29

The rate of infestation was lower in flocks with less than 100 heads (67%) compared to large flocks up than 100 (95 %),(Table 7) which corresponds to the study of (7), which showed that a too high density of animals favors massive contamination of the environment.

No significant differences were found between flocks receiving concentrated or non-concentrated feeding (91 % vs. 67 %, respectively), or whether the lambs were attached to the mother or away from the sheep (85 vs 83 %, respectively) since lambs are probably infected just after birth (5)

However, flock infestation rates were high during the lambing period (97% vs 71%, respectively), which is consistent with data from the literature (16) that show that the excretion of oocysts increases in ewes during lambing; this could be linked either to the sheep carrying or to massive contamination of the premises by the lambs belonging to the previous cohort. The intensity of the infestation by *Cryptosporidium* sp. was significantly high in intensive livestock farms, 94%, where 30 to 100 heads are clustered in a small, unhygienic building, unlike extensive livestock farming, 71%, where animals graze in the large rangelands, which reduces the pressure of the infestation; (92% vs 68%), which is consistent with other observations (8) that hygiene plays an important role in the rate of contamination of the environment in which the animal lives.

Table 7

Frequency of *Cryptosporidium* sp positive flocks according to factors associated with the disease. (n = 52 flocks)

Factors associated with the disease		**Total of flocks**	**Number of positifs flocks**	**Nombre of negatifs flocks**	**% Des of positifs flocks**	**P**
Size of herd	<100	24	16	8	(67)	0,01*
	>100	38	36	2	(95)	
Flock receiving a concentrate ration	Yes	44	40	4	(91)	0,05**
	No	18	12	6	(67)	
lambing period	(Sept-Janv)	31	30	1	(97)	0,005
	No	31	22	9	(71)	
Attached to mother	Yes	33	28	5	(85)	0,9*
	No	29	24	5	(83)	
Type of reaning	Extensif	28	20	8	(71)	0,04*
	Intensif	34	32	2	(94)	
Hygiene in the stable	Yes	22	15	7	(68)	0,03*
	No	40	37	3	(92)	

*Correction of Yates. ** Exact test of Fisher.

CONCLUSION

At the end of this first survey in Algeria, we can say that sheep cryptosporidisoe is widely present in the farms of the commune of Ain Oussera with a flock prevalence of 84% (95% CI 72.8% - 91%), which exposes lambs to the risk of diarrhea due to this disease. Factors associated with the disease are numerous, but the most implicated are stable hygiene, flock size, lambing period and type of rearing. In addition to the study that we have just conducted, other studies can be envisaged in a longitudinal survey on a limited number of animals chosen wisely to have a better knowledge of the evolution of the disease in order to establish more the impact of this disease on livestock.

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