## CLINICAL AND HEMATOLOGICAL STUDY ON OVINE ANAPLASMOSIS IN SULAIMANI PROVINCE- IRAQ.

Bahzad. H. salih Mustafa.

Department of Animal Production, Collage of Agriculture, University of Sulaimani. Sulaimani, Iraq.

Keyword; Anaplasmosis, blood smears, sheep

(Received 24 April 2011, Accepted 7 December 2011)

### **ABSTRACT**

A study was carried out on Ovine Anaplasmosis in the Sulaimani province of Iraq from September to December2007. *Five hundred* thin blood smears were prepared form Peripheral blood of sheep in 10 different localities and stained by Giemsa. The prevalence of *Anaplasma Ovis* was 24 (4.8%) and the rate was highest 8.8% was among animals above 3 years. The infected animal showed mild fever, hyperpnoea and tachycardia. Hematological values of the infected sheep was as follows: Packed cell volume  $22 \pm 4.75$  %, erythrocyte count  $6.42 \pm 1.35 \times 10^6$ /  $\mu$ l, hemoglobin concentration  $6.95 \pm 1.28$  g/dl, mean corpuscular volume  $35.5 \pm 5.85$ / fl, mean corpuscular Hemoglobin concentration  $11..04 \pm 2.42$ / pg, mean corpuscular hemoglobin concentration  $31.62 \pm 6.62$  g/dl and WBCs  $11.15 \pm 2.18 \times 10^3$  / $\mu$ l, serum protein value  $5.875 \pm 0.801$  mg/dl, serum calcium  $8.341 \pm 1.029$  mg/dl, conjugate serum bilirubin  $0.85 \pm 0.314$  mg/dl and serum cholesterol  $57.291 \pm 6.153$  mg/dl.

#### INTRODUCTION

Anaplasmosis is also transmitted in a variety of mechanical ways for example through vaccination, tattooing or castration tools. Geography and climate are the two important factors determining what tick species are responsible for local cases of Anaplasmosis (1). Anaplasmosis is generally a benign rickettsiosis of sheep and goats, which is endemic in tropical and sub-tropical Africa, Asia, part of southern and central Europe and the western USA (2). It is caused by intraerthrocytic rickettsiae of genus Anaplasma and infection with *Anaplasma ovis* is the most widely recognized cause such infection may result in severe anemia and jaundice (3). The disease is transmitted by ticks, although iatrogenic transmission by biting arthropods (4, 2, 5). The detection of *A. ovis* in blood (erythrocyte) films together with evidence of anemia

#### Bas.J.Vet.Res.Vol.10,No.2,2011.

and haematological reactions is considering diagnosis. Giemsa stain is most commonly used and organism appears as homogenous dense bodies 0.5-1 microns in size (6). Factual information on prevalence and distribution of anaplasmosis has not been available in Sulaimani province. The present survey was conducted to study ovine anaplasmosis and some related hematological changes in Suliamani province.

#### MATERIALS AND METHODS

Field studies were conducted in 10 areas of Suliamani province from September to December 2007. Five hundred thin blood smears were prepared from external ear vein of sheep of different ages. These smears were normally stained with Giemsa stain as methanol described by (7). From cases confirmed to be positive to A. ovis, 5ml of blood was drawn from Jugular vein in vacutioners test tube that contained disodium ethylene diamine tetra acetic acid (Disod. EDTA). This blood is used for determining the following hematological parameters: packed cell volume (PCV %), hemoglobin concentration (Hb g/dl) total erythrocytes count (RBCs × 10<sup>6</sup>/µl), total leukocytic count (WBCs  $\times$  10<sup>3</sup>/  $\mu$  l) and calculated mean corpuscular volume (MCV/fl), Mean corpuscular hemoglobin (MCH/pg) and mean corpuscular hemoglobin concentration (MCHC g/dl). Also 10 ml of blood was drawn in vacutioner test tube without anticoagulant. The blood samples were allowed to coagulate and then serum was separated and used for determined calcium, bilirubin, cholesterol and total protein values in plasma. These biochemical parameters determined according to the instructions of the manufacturer of the complex Kits which was received from Biomerieux and Boilable, France. Data were statistically analysis using Chi-Square procedure (8).

#### RESULTS

The prevalence of Anaplasmosis (Anaplasma reaction) is shown in Table (1). Out of 500sheep, 24(4.8%) were positive reactors by blood smears, and the rates rang from 2% in Ranya ,Qaradakh and Halabja to 10% in Maydan.

Table1: Prevalence of *Anaplasma ovis* in sheep in Different Districts and Sub districts in Sulaimani province (%).

011501100	districts in Sulaimani province (70).		
Area	Number (%) of + ve reactions		
Ranya	1 (2)		
Qaladzea	3 (6)		
Mawat	0 (0)		
Aghjalar	4 (8)		
Chamchamal	4 (8)		
Qaradakh	1 (2)		
Penjwen	2 (4)		
Halabja	1 (2)		
Maydan	5 (10)		
Kalar	3 (6)		
Total	24 (4.8)		

The prevalence of reactors according to age of animal is shown in Table (2). Statistical analysis revealed an associated between age and No. of +ve sample such associated is due to the high incidence of such samples among the older groups. Difference between the other two age groups was not significant.

Table: 2 Prevalence of Anaplasma ovis according to age.

Age	Number of Sample	Number of (%) of +ve sample
Under one year	135	5 (3.7)
1 – 3 year	240	8 (3.3)
Above 3 years	125	11 (8.8)
Total	500	24 (4.8)

In table (3) showed the mild fever  $(40.63 \pm 1.44 \text{C}^{\circ})$  in infected animals, hyperpnoea  $(32\pm6.13 \text{ breath /minute})$  and laboured respiration and tachycardia  $(82.21\pm5.68 \text{ beat/minute})$ , pale and icterus of mucous membranes, emaciation and weakness and ruminal stasis. The *A. ovis* was detected in the stained blood smears as inclusion body at periphery of the erythrocyte with parasitemia rate  $(4.30\pm2.26 \%)$ .

Table: 3- Systemic reactions in sheep due to Anaplasma ovis infection

Systemic Reaction	(Mean ± SD)
Rectal Body Temperature (c)	40.629± 1.444
Respiratory Rate breath/minute	$32 \pm 6.129$
Pulse Rate beat/ minute	82.208±5.679
Parasitemia	4.291± 2.255

Table (4) shown various clinical sign in infected sheep including, pale and icterus of mucous membranes, emaciation and weakness and ruminal stasis.

Table 4: Clinical signs in sheep infected with Anaplasma ovis.

Enlargement of Lymph node  Haemoglobinurea  Icterus of elastic tissue (mm and sclera of eyes) +	
Icterus of elastic tissue (mm and sclera of eyes) +	
receits of elastic tissue (finit and selecta of eyes)	
Paleness of mucous membrane	
Emaciation	
Lacrimation	
Nasal discharge	
Diarrhea	
Present the tick in the body rare	ly
Decumbency	
Ruminal stasis +	

---- = no reactions , + = Mild reactions , ++=

Sever reactions

Hematological parameters of 20 cases confirmed Anaplasmosis is presented in Table (5). The blood picture is seen to be typically that of anemia with low PCV% (22  $\pm$  4.7%), hemoglobin (6.95  $\pm$ 1.2 g/dl) and red blood cell count (6.42  $\pm$  1.3  $\times$ 10<sup>6</sup>  $\mu$ l); there was a mild leukocyte (11.15  $\pm$  2.18  $\times$ 10<sup>3</sup> $\mu$ l). The mean values of the mean corpuscular volume in fl were (35.50  $\pm$  5.85) and infected sheep had average corpuscular hemoglobin (11.04 $\pm$ 2.42 pg) and the mean corpuscular hemoglobin concentration (31.62  $\pm$  6.59 g/dl). Those figures indicated evidence of Normocytic hypochromic anemia.

Table5: Hematological parameters in infected sheep with Anaplasma ovis.

Parameters	units	(Mean ± SD)
PCV	%	$22 \pm 4.75$
Hb	g/dl	$6.95 \pm 1.28$
Total RBC	$10^{6}/\mu l$	$6.42 \pm 1.35$
Total WBC	$10^3/\mu$ l	11.15 ±2.18
MCV	fl	$35.50 \pm 5.85$
МСН	pg	$11.04 \pm 2.42$
МСНС	g/dl	$31.62 \pm 6.59$

Table (6) shows the mean value of smear chemistry. The infected sheep had average  $5.875 \pm 0.801$  mg/dl of serum protein,  $8.341 \pm 1.029$  mg/dl of serum calcium,  $0.85 \pm 0.314$  mg/dl of serum bilirubin and  $57.291 \pm 6.153$  mg/dl of serum cholesterol.

Table: 6 - Biochemical Analysis in sheep infected with Anaplasma ovis.

Parameters	(Mean±SD)
serum protein mg/dl	$5.875 \pm 0.801$
serum calcium mg/dl	8.341 ± 1.029
serum bilirubin mg/dl (conjugate)	$0.85 \pm 0.314$
serum cholesterol mg/dl	57.291 ± 6.153

#### **DISCUSSION**

In most countries, little attention is paid to *A. ovis* infected, regions with a large important goat population being an exception. Hence, there are only few recent reports an anaplasmosis of sheep. Some distributions of Ovine and Caprine anaplasmosis in Africa, American, Asia and Europe was reported (9). Preliminary present result indicated that the prevalence of *A. ovis* among sheep was low, 4.8%. In Sulaimani province-Iraq, this study agreement with (10) who revealed the prevalence rate was under 5% in both sheep and goats on hematological examination in Turkey. This study differing with (11, 12 and 13) who showed the prevalence rate of *Anaplasma ovis* in goat (32.19%) in Baghdad province, 8.6% in Jordan and 14.61% in

#### Bas.J.Vet.Res.Vol.10,No.2,2011.

Israel respectively. The differences may be due to infections occurrence by ixodid ticks in Sulaimani province, while the infection in other countries transmitted by another vectors than ixodid ticks (mechanically transmitted like flies).

Parasitemia was low and not associated with disease. The distribution of A .ovis in sheep according to age of animals revealed lowest prevalence in animal under one year, this might be due to material antibodies obtained by the lambs to resist the infection .For the oldest group the chance for exposing to vector was more the younger animals. Similar finding was record in goats (11). The ages less than eight months have lowest rate than the age above four years old (14). Reported the highest rate (45.34%) in age four years this explained due to previous infections because it considered the anaplasmosis is subclinical, mild febrile disease of apparently little economic importance, and A. ovis appears to be more pathogenic for goats than for sheep (15). And the organism generally causes subclinical disease (4, 2, 5) in sheep and does not cause economically significant losses. Clinical signs are more likely to be seen in goats than in sheep (16). And anaplasmosis can be a serious disease also in sheep when naïve animals under severe stress are exposed to heavy tick infestation (17). There have been relatively few reports on the haematological and biochemical changes of the anaplasmosis in sheep. The pathogenesis of anemia could be directly correlated to the cell injury due to invasion of erythrocytes by the anaplasma, but loss of red blood cells was closely correlated to the level of infection in severely anemic animal (18).

# دراسة سريرية ودمية لداءالانابلازما في منطقة السليمانية - العراق بهزاد صالح مصطفى الخلاصة

أجريت الدراسة على مرض داء الانابلازموس Ovine Anaplasmosis محافظة السليمانية- العراق الغنمية الفترة من أيلول الى كانون الأول لسنة 1.00 حيث تم تخضير 1.00 مسحة دموية (خفيفة) من الأغنام ومن عشر مناطق وتم صبغها بصبغة كمزا. حيث أظهرت أن معدل أنتشار المرض في الأغنام حوالي 1.00 ونسبة الأصابة كانت عالية في الأعمار التي تزيد عن 1.00 سنوات وهي 1.00 ، والرتفاع طفيف في درجة الحرارة ومعدل التنفس ومعدل سرعة النبض. كما واظهرت تغيرات في المقادير الدموية والكيمياوية حيث كانت 1.00 عدد كريات الدم الحمراء 1.00 عدد كريات الدم الحمراء الهيموكلوبين 1.00 عدد عرام / ديسي ليتر وحجم الكرية الجزيئي 1.00 فيمتوليتر وهيموكلوبين الخلية الجزيئي 1.00 فيمتوليتر وهيموكلوبين الخلية الجزيئي 1.00 غيرام / ديسي

#### **REFERENCES**

- 1- de Silva, R..B. and Nadine, V. 2008. Surveillance of vector-borne diseases in cattle
  - with special emphasis on bluetongue disease in Switzerland. *PhD Thesis*, University of Basel, Faculty of Science
- 2- **Stoltsz,W.H.** 1994. Ovine and Caprine anaplasmosis Coatzer,J.A.W;Thomson,G.E; Tustin, R.C, eds, Infectious disease of livestock, Vol. In 1.Oxford University Press,Cape Town, Oxford, New York. pp 431-438.
- 3- Martin, W.B and Aitken. 2000. Diseaseases of sheep, 3<sup>rd</sup> Edition. Blackwell science Ltd.pp 310-311.
- 4- Yeruhams, I.; Handani, A.; Galker, F; Rosen, S. and Schlien, J. 1997. Astudy of haemoparasites In two flock sof sheep in Israel. Israel. J. Vet. Med. 47(3):107-111
- 5- **Hashemi- Fesharki. 1997.** Tick borne disease of sheep and goats and their 107 111 related vectors in Iran. Parasitologica Roma 39(2): **115-117.**
- 6- Losos, J.G. 1986. Infectious Topical Disease of Domestic Animals part IV Rickettsial Diseases. Longman Scientific and Technical pp. 741-795.
- 7- Coles, E.H. 1986. Veterinary Clinical pathology .4<sup>th</sup> edition.W.B.Saunder Company. Pp: 12-20.
- 8 Zar, J.H. 1999. Biostatistical Analysis, Fourth Edition, Prontice Hall.
- 9- **Neitz,W.O.1986.** Veterinary clinical Pathology.4<sup>th</sup> edition .W.B.Saunders Company. 12-20.
- 10- Goksu, k. 1965. A review of anaplasmosis and observation of infectious in sheep and goats, Turk Veteriner Heklimeri dernigi Dergisis. 35: 399-417.
- 11-Al.Amery,M.A.Y and Hasso,S.A.2002. An epidemiological survey for some Blood and intestinal protozoa in the goats in Baghdad province. *Bas J.Res.* 1 (2): 41-48 (in Arabic).
- 12- Sherkov, S.N., El Rabie, Y. and Kokash ,L. 1976. A survey of parasitic blood diseases" tick borne fever" in domestic animals in Jordan. *Egypt. J.Vet. Sci.* 13: 29-35.
- 13- **Pipano, E. 1991**. Observations on the seasonal distribution of blood parasites in Israel. *J. Vet Med.* **46:37-38.**

- 14- Shompole, S.; Waghela, S.D.; Rurangirwa, F.R. and McGurire, T.C. 1989. Cloned DNA probes identify Anaplasma ovis in goate and reveal a high prevalence of Infection *J.Clin. Micro* . 2-7(12): 2730-2735.
- 15-Yousif, Y.A.; Dimitri,R.A.; Dwivedi,S.k. and Ahmed,N.J. 1983. Anaemia due to Anaplasmosis in Iraqi goats. I. Clinical and haemological features under field Conditions. *Indian Vet.J.* 60: 576-578.
- 16 Lu, W.S.; Lu, W.X.; Zhang, Q.C.; Dou, H.F.; Yin, H.; Ji, Z.D.; Hu, T.Y.; Sun, B.G.; Han, J.S.; Gao, D.F. and Bao, Y.J. 1987. Study on anaplasmosis in goats. Identification of the pathogen and observation of the clinical signs (in Chinese). Chinese. J. Vet. Sci Technol. 3: 5-8.
- 17- **Manickam, R. 1987**. Epidemiological and clinical observations of acute Anaplasmosis in sheep. Indian. *J.Vet*. *Med*. **7:159-160**.
- 18- Jones, E.W.; Kliewer, I.O.; Norman, B, B and Brock, W.E 1968. Anaplasma marginate infection in young and aged cattle, Am. Vet. Res. 29(3):535-44.