

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

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ABSTRACT

A study was carried out on Ovine Anaplasmosis in the Sulaimani province of Iraq from September to December 2007. *Five hundred* thin blood smears were prepared from 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 ± 4.75 % , erythrocyte count $6.42 \pm 1.35 \times 10^6 / \mu\text{l}$, hemoglobin concentration 6.95 ± 1.28 g/dl , mean corpuscular volume 35.5 ± 5.85 fl , mean corpuscular Hemoglobin concentration 11.04 ± 2.42 pg, mean corpuscular hemoglobin concentration 31.62 ± 6.62 g/dl and WBCs $11.15 \pm 2.18 \times 10^3 / \mu\text{l}$, serum protein value 5.875 ± 0.801 mg/dl, serum calcium 8.341 ± 1.029 mg/dl, conjugate serum bilirubin 0.85 ± 0.314 mg/dl and serum cholesterol 57.291 ± 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 intraerythrocytic 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

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 vacuoners 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 $\times 10^6/\mu\text{l}$), total leukocytic count (WBCs $\times 10^3/ \mu\text{l}$) 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 vacuonier 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 (%).

| 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 ($40.63 \pm 1.44C^{\circ}$) in infected animals, hyperpnoea (32 ± 6.13 breath /minute) and laboured respiration and tachycardia (82.21 ± 5.68 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 ± 2.26 %).

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 ± 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*.

| Clinical Signs | Cases |
|---------------------------------------------------|--------|
| Enlargement of Lymph node. | ----- |
| Haemoglobinurea | ----- |
| Icterus of elastic tissue (mm and sclera of eyes) | + |
| Paleness of mucous membrane | ++ |
| Emaciation | ++ |
| Lacrimation | ----- |
| Nasal discharge | ----- |
| Diarrhea | ----- |
| Present the tick in the body | rarely |
| 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 ± 1.2 g/dl) and red blood cell count ($6.42 \pm 1.3 \times 10^6 \mu\text{l}$); there was a mild leukocyte ($11.15 \pm 2.18 \times 10^3 \mu\text{l}$). The mean values of the mean corpuscular volume in fl were (35.50 ± 5.85) and infected sheep had average corpuscular hemoglobin (11.04 ± 2.42 pg) and the mean corpuscular hemoglobin concentration (31.62 ± 6.59 g/dl). Those figures indicated evidence of Normocytic hypochromic anemia.

Table5: Hematological parameters in infected sheep with *Anaplasma ovis*.

| Parameters | units | (Mean \pm SD) |
|------------|---------------------------|------------------|
| PCV | % | 22 \pm 4.75 |
| Hb | g/dl | 6.95 \pm 1.28 |
| Total RBC | 10 ⁶ / μ l | 6.42 \pm 1.35 |
| Total WBC | 10 ³ / μ l | 11.15 \pm 2.18 |
| MCV | fl | 35.50 \pm 5.85 |
| MCH | pg | 11.04 \pm 2.42 |
| MCHC | 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 \pm SD) |
|-----------------------------------|--------------------|
| serum protein mg/dl | 5.875 \pm 0.801 |
| serum calcium mg/dl | 8.341 \pm 1.029 |
| serum bilirubin mg/dl (conjugate) | 0.85 \pm 0.314 |
| serum cholesterol mg/dl | 57.291 \pm 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 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 maternal 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 recorded 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 لداء الانابلازما في الاغنام في محافظة السليمانية-العراق

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الخلاصة

أجريت الدراسة على مرض داء الانابلازما *Ovine Anaplasmosis* في محافظة السليمانية-العراق للفترة من أيلول الى كانون الأول لسنة 2007 حيث تم تخضير 500 مسحة دموية (خفيفة) من الأغنام ومن عشر مناطق وتم صبغها بصبغة كمزا. حيث أظهرت أن معدل انتشار المرض في الأغنام حوالي 4.8% ونسبة الإصابة كانت عالية في الأعمار التي تزيد عن 3 سنوات وهي 8.8% ، والارتفاع طفيف في درجة الحرارة ومعدل التنفس ومعدل سرعة النبض. كما وظهرت تغيرات في المقادير الدموية والكيميائية حيث كانت حجم 1.35 ± 6.42 وتركيز $10^6/\mu l$ الكريات المرصوفة $4.75 \pm 22\%$ و عدد كريات الدم الحمراء الهيموكلوبين 1.28 ± 6.95 غرام / ديسي ليتر وحجم الكرية الجزيئي 1.58 ± 35.5 فيمتوليتر وهيموكلوبين

الخلية الجزيئي 2.42 ± 11.04 بيكوغرام وتركيز هيموكلوبين الخلية الجزيئي 6.62 ± 31.62 غرام /
والبروتين في مصل الدم 11.04 ± 2.18 و $10^3 / \mu l \pm 0.801$ ×ديسي ليتر وعدد الكريات الدم البيضاء
غرام/ديسي ليتر والكالسيوم في مصل الدم 8.341 ± 1.029 غرام /ديسي ليتر ومستوى الصفراء
في مصل الدم 0.85 ± 0.314 غرام /ديسي ليتر والكوليسترول الكلي 57.291 ± 6.153 غرام /ديسي
ليتر.

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