Study of Clinical ,Hematological and serological Diagnosis of Ovine Theileriosis in Basrah province

Tariq Refaat M. and Israa Abdulwadood MA.
Collage of Veterinary Medicine /Basrah University
Basrah /Iraq

Summary

The present study was carried out to determine prevalence of ovine theileriosis in the sheep in Basrah province, with studying clinical signs and hematological parameters supported by serological diagnosis of *Theileria lestoquardi* by ELISA technique.

The survey study extended from 15 July 2010 to 30 May 2011.A total of 1466 sheep of different age ,sex and breed examined randomly from different areas in northern, southern, eastern, western and central of Basrah provinc. A total number of sheep exanimated clinically are 1466 and 460 sheep of them showed a clinical signs such as the fever, enlargement of superficial lymph nodes , respiratory signs ,tachycardia, paleness of mucus membrane . The blood examination showed the high level of parasitemia varied between 12-80% and observed all parasite phase; erythrocyte stage 64.8%, lymphocytic stage 22.62% and erythrocytes & lymphocytic stage 12.58%.

The study showed the high prevalence of ovine theileriosis in northern area with 65.82% followed by southern area with 58.94% and lower prevalence of theileriosis in the central of province with 52.98%.

The hematological parameter showed the macrocyitc hypochromic and normocytic normochromic anemia in sheep infected by *Theileria lestoquardi* with significant decreases in Hb 7.8 ± 0.4 (g/l) , PCV 26.81 ± 2.991(%), MCHC 28.99 ± 1.8 (%), RBCC 5944545± 62.38 (10/mm), WBCC 5870.0± 709.76 (10 /mm), in the lymphocytes 3602.95± 450.5 (absolute) and neutrophils 1166.8 ± 235.72 (absolute),while recorded a significant increase in MCV 4545 ± 6.27(%) while significant increase in monocytes and eosinophiles.

The serological diagnosis by indirect ELISA test showed a high prevalence of Malignant theileriosis by *Theileria lestoquardi* 93.14% ,while the lower prevalence of benign theileriosis by *Theileria ovis* 6.85%.

Introduction

Ovine theileriosis is an important hemoprotozoal disease of sheep and goats in tropical and subtropical regions (Altay *et al*, 2007a) due to at least six species of *Theileria* including *Theileria ovis*, *Theileria separate*, *Theileria recondite*, which are considered as being low or non-pathogenic to the small ruminants (Alani and Herbert, 1988). *Theileria lestoquardi* (*Theileria hirci),* and *Theileria spp*. (*China 1*) and *Theileria spp. (China 2*) that recently was reported from north of China (Ahmed *et al*., 2006 ;Niu *et al*. ,2009*).*

T. lestoquardi and *Theileria china* unlike the other three species are highly pathogenic (Altay *et al*.,2005).

Hummandi,(1978) show that high incidence of the disease is recorded in the middle and south parts of Iraq as well as the transmitted vectors which causes the malignant ovine theileriosis in sheep are hard ticks as (*Haylamma anatolicum anatolicum*),which are widely in all of Iraq.

Al-Hasnawi,(2009) showed that Basrah province was endemic area by bovine theileriosis. The infection distributed in all parts of Basrah and concentrated in the Al-Hartha and Shat- Al-Arab. there

are three clinical forms which can be observed when animal infected by ovine theileriosis especially malignant *Thleileriosis* which assumes acute, sub acute and chronic forms . The acute disease is characterized by fever. other symptoms appear which include inappetence, cessation of rumination, rapid heartbeat, dyspnea, weakness, listlessness and swollen of the superficial lymph nodes. Marked anemia and icterus develop in a few days (Guo *et al.,* 2002) and very high mortality in 3-6 days which ends with death. In subacute and chronic cases, signs are generally less marked except for anemia and emaciation (Radostits *et al*,.2007)**.**

this study conducted to study the clinical ,hematological and serological diagnosis of ovine theileriosis .

Materials and Methods

The study conducted in Basrah province ,the area of the study is divided into: northern, southern, eastern, western and central region .

A total number of (1466) sheep for different age, sex and breed were clinically examined randomly belonging to different locations in Basrah province during the period from July 2010 to May 2011. The clinical signs and hematological and serological studies represented infected group and 150 sheep as a control group.

Also Blood sample was collected from jugular vein of each animal showed clinical signs, and used for making blood film. 10 ml of blood sample was collected by puncture the jugular vein, 5 ml of blood placed in tube with EDTA for heamatological examination and 5 ml without EDTA was allowed to coagulant and spirited the serum by using centrifuge (3000 rpm) for used in ELISA test .The serum were stored at -20°C until use.(Aktas *et al*.,2005).

1-Hematological Examination.

1- Red Blood Cell Count (RBCC) done according to (Schalm *et al*., 1975) .

2-Total White Blood Cell Count (WBCC)done according to(Dasie and Lewis, 1984).

3-Differential White Blood Cells Coun**t.**

4-Hemoglobin Concentration (Hb) : according to (Coles,1986).

5- Packed Cell Volume (PCV): according to (Dasic and Lewis, 1984).

2-Blood smear:

The blood smear is made from blood collected from Jugular vein in EDTA after sterilizing animal jugular vein site with alcohol 70%. thin blood film was prepared (Chaudhri&Gupta, 2003) **.**

3-Lymph Samples

Lymph samples were collected from the enlarged superficial lymph node specially prescapular lymph node by puncture and diatheses the lymph to making lymph film according the procedure of (Al- Robayi, 1999).

4- Serological diagnosis (ELISA test):

The total number of serum samples 1466 were collected from clinically infected sheep in different ages. And 875 of 1466 serum samples were positive to microscopical examination used by ELISA test, in eight geographical areas in Basrah province , The Serum samples which previously labeled and stored at −20 ◦C **.**

Result

The total number of sheep examinant clinically are 1466 in different ages, sex and breeds. 460 of 1466 sheep in acute form showed clinical signs varied from rise of body temperature(40.4-41.8C^0), increasing respiratory rate (50-90/mint.),dyspnea, interrupted coughing, increasing heart rate (90-140/mint.) as in table (2). The size of lymph nodes specially the superficial prescapular lymph node was varied from slightly to large enlargement .The appetite of infected sheep ranged from loss appetite to anorexia, lameness, salivation, nasal discharge and lacremation; pale-yellowish and congested mucus membranes yellowish urine; some time associated with corneal opacity, semi-solid to pasty feces and yellowish diarrhea or constipation, ticks infestation . In chronic form the sheep showed emaciation and dehydration and anemia table (1). Some time the disease occurred as subclinical form (carrier) which reached 415 of 875 (47.42%).

The study showed high level of parastemia in infected sheep varied from 12-80 % and the RBC can be infected by(1-4) parasites in maximum limit; all forms of *Theileria hirci* piroplasms including rod, round, ring and anaplasmoid form with abnormalities in erythrocyte structure were observed. While in the WBCs specially lymphocytes the macroschizont and microschizont stage (koch blue body) observed in blood smear and schizont in lymph smear. Moreover the examination showed 22.62% of blood cells infected by schizont lymphocytic stage , 64.8% infected by erythrocytes stage and 12.57% showed the erythrocytes and lymphocytic form table(3).

The result of the study showed a high prevalence rate in the North area of the province 65.82% ,South area 58.95% followed by West area 56.12% followed by East area 55.47% and then Center area of the province 52.98% with significant difference in $P<0.05$ table (4)**.**

Table (1) Clinical signs related to infected sheep by Ovine theilerosis

Clinical signs	No. of sheep	Percentage of infection
Fever	219	47.60
Increase Respiratory rate	310	67.39
Increase heart rate	325	70.56
Nasal discharge	400	86.95
Salivation	72	15.65
Coughing	122	26.52
Dyspnea	75	16.3
Enlargement of prescapuler lymph node	455	96.73
Lameness	97	21.08
Anorexia	198	34.04
Loss of appetite	325	70.6
Pale-Yellowish mucosal membrane	380	82.60
Eye congested	108	23.47
Lacrimation	203	44.13
Corneal opacity	13	2.82
Dehydration	223	48.47
Emaciation	183	39.87
Anemia		

Yellowish urine	212	46.08
Constipation	91	19.78
Yellowish-Soft diarrhea	10	2.17
Semi-Solid feces	85	18.47
Ticks infestation	107	23.26
	430	93.47

Table (2) Range of body temperature ,pulse and respiratory rate of infected sheep compared with control sheep.

Signs	Infected sheep Range	Control sheep Range
Body temperature /C^0	40.4 - 41.8	38.8-40.0
Heart rate / Mint.	90 – 140	80-90
Respiratory rate/ Mint.	50 – 90	30 -40

Table (3) percentage of infected blood cells with different stages of parasite.

Phase of parasites	No. of sheep	Percentage of infected blood cell %
1- Lymphocytic stage (Koch s blue body)	198	22.62
2- Erythrocytes stage	567	64.8
3- Erythrocytes and Lymphocytic stage	110	12.58

Table (4) Prevalence rate of ovine theileriosis in Basrah province .

Location	No. of sheep exanimate	Infected sheep	
		Positive	(%)
Northern area of Basrah	553	364	(65.82)
Eastern area of Basrah	393	218	(55.47)
Central area of Basrah	134	71	(52.98)
Western area of Basrah	196	110	(56.12)
Southern area of Basrah	190	112	(58.95)
	1466	**875**	**59.68**
X^2 = 15.135 . P< 0.05 SD.			

X^2=Chi-Square SD.= significant difference & P= Probability value

The hematological analysis showed statistically a significant decreases ($P<0.05$) ; in the mean of red blood cells count (RBCC), white blood cells count (WBCC), hemoglobin concentration (Hb) ,packed cell volume there is a significant increases found at Mean corpuscular volume (MCV) ($P<0.05$) ; and abnormalities in

The differential white blood cells showed decrease in lymphocyte and neutrophils count with significant difference ($P<0.05$).While the monocyte and esonophilis count increased as statically a significant ($P<0.05$).On the other hand the basophiles showed statistically an important change ($P>0.05$) .

Table. (5) changes in some blood parameter in infected sheep with theileriosis.

Hematological parameter	Control sheep Mean ± Std. deviation	Infected sheep Mean ± Std. deviation	F value	Significant Differed
RBCC (10^6/mm)	10814000 ± 108.27	5944545± 62.38	163.39	P<0.05 S.D
WBCC (10^6/mm)	8012.5 ± 397.577	5870.0± 709.76	73.67	P<0.05 S.D
Hb (g/l)	14.4 ± 0.52	7.8 ± 0.4	1051.36	P<0.05 S.D
PCV (%)	43.3 ± 1.8	26.81 ± 2.991	227.92	P<0.05 S.D
MCV (fl)	39 ± 4.33	45 ± 6.27	4.93	P<0.05 S.D
MCH (pg)	13.40 ± 1.57	13.12 ± 1.4	0.80	P>0.05 S.D
MCHC (%)	33.48 ± 1.41	28.99 ± 1.8	32.10	P<0.05 S.D
Differential White Blood Cells Count (Absolute)				
Lymphocytes	4604.81±280.1	3602.95± 450.5	36.51	P<0.05 S.D.
Neutrophils	2560.37± 358.1	1166.8 ± 235.72	113.04	P<0.05 S.D.
Monocyte	319.92± 107.56	453.96± 93.95	9.294	P<0.05 S.D.
Eosinophils	520.95± 175.81	627.47± 106.53	2.883	P<0.05 S.D
Basophils	23.135± 37.33	19.61± 28.36	0.60	P>0.05 NS.
No. of sheep	**150**	**815**		
S.D= Significant difference & NS.= Non Significant				

•

The total Number. of serum samples are using in ELISA test 875 that positive to blood smears examined. And 815 of 875 serum samples (93.14%) were positive by ELISA test identified to genus of *Theileria hirci* and 60 (6.85%) of serum samples were negative to ELISA ,the later suggested that infected by *T. ovis*, and recorded high prevalence rate of *T. ovis* in northern region (66.66%) followed by eastern region (18.33%) and lower prevalence rate (6.66%),(5%) and (3.33%) in western, central and southern region respectively.

The result of examining 875 serum samples obtained from different geographical area are shown in table (7) The highest prevalence rate of *T. lestoquradi* 98.21 % recorded in South area of province and lower prevalence 83.16% recoded in North area of province.

Table (6) prevalence of *Theileria hirci* in sheep by using ELISA test compared with microscopical examination.

Location	No.of sheep examination	No. and percentage of microscopical examination		No. and percentage of ELISA examination	
		Positive	%	Positive	%
Northern area of Basrah	553	364	(65.82)	323	(88.73)
Eastern area of Basrah	393	218	(55.47)	207	(94.95)
Central area of Basrah	134	71	(52.98)	68	(95.77)
Western area of Basrah	196	110	(56.12)	107	(97.27)
Southern area of Basrah	190	112	(58.94)	110	(98.21)
$X^{2=}$ 19.348 P<0.05 S.D		$X^{2=}$ 20.404 P<0.05 S.D			

X^2=Chi-Square S.D= significant difference & P= Probability value

Discussion

All clinical signs showed by infected sheep agree with results studies of (;El-Azazy *et al*,2001; Yin *et al*.,2003; Haddadzadeh *et al*,2004 and Zia-ur-Rehman *et al*., 2010).

The corneal opacity was explained by (Hussine *et al.*,2004 and Osman &Al-Gaabary,2007) as a result of white blood cells infiltration .The prescapular lymph node enlargement in infected sheep could be explained by lymphoid hyperplasia in early stage of disease that occurs due to increases of proliferation of microschizonts inside the lymphocyte caused inflammatory reaction in the infected lymph node (Al-Robayi,1994). Pale-yellowish of mucus membranes was explained by (Al-Robayi,1994) as a result of increase total bilirobin level in blood due to the infection, Singh *et al.*,(2001) show that paleness of mucous membranes exhibited the development of anemia and reduction of hemoglobin concentration and the total erythrocytes count was due to the distraction and the removed of the infected erythrocytes by reticulo-endothelial system.

Diarrhea seen in 18.47% of infected sheep was explained as a result to inflammatory reaction and ulceration to abomasal and gastro intestinal tract (Al-Robayi.,1994). On the other hand ,the study showed that 415 of 875 sheep infected by ovine theileriosis are characterized by subclinical signs. These result agreed with (Heidarpour Bami, *et al*.,2010) which revealed that subclinical infections are common in their study of infected sheep by ovine theileriosis.

Generally, the disease is widely distributed in all province include Northern, Southern Eastern, Western and Central and recorded higher prevalence rate 93.14% serologically by using (ELISA) test than microscopical observation 59.68%. However, result of the study of malignant ovine theileriosis in Basrah province agree with (Al-Hasnawi.,2009) how showed that Basrah province was endemic area by bovine theileriosis infection genous *T.annulata*. The infection distributed in all parts of Basrah.

The result of the study may indicate continuous transmission of *T.lestoquardi* in Basrah. This is supported by the fact that *Hyalomma anatolicum anatolicum* adult ticks were found to be active throughout most of year in the present study area as well as in the other parts of Iraqi country .

The present study showed that ovine theileriosis is highly affected in the blood parameters which represented by decrease TRBCC, TWBCC, PCV, Hb, MCH and MCHC, that causes anemia which differed in severity from mild to severe anemia in infected sheep. These agreement with result of

(Alsaad *et al.*,2009; Al-Hasnawi,2009 and Zia-ur-Rehman *et al*., 2010). The study showed that *Theileria* infection complained from the type of anemia macrocytis hypochromic anemia, due to increasing of MCV, where, it gradually raised through mode of disease and concurrently with severity of anemia; this denoted that the bone marrow response to erythropoeisis and normocytic normochromic anemia were observed in infected sheep related to decrease of MCHC due to decrease in Hb concentration. These result were agree the with results

of (Dhar and Gautam,1979) who showed that the infected animals by theileria appeared normochromic normocytic and hypochromic machrocytic anemia .

This study showed a significant decreases in hemoglobin concentration Hb and total RBCs count these results agree with most hematological studies as (Omer *et al.*,2002; Hussine *et al.*,2004; Nazifi *et al.*,2009 and Al-Hasnawi, 2009). Moreover there exist significant decreases in PCV and MHCH and significant increases in MCV with an significant statically change in MCH these result were agreement with (Al-Robayi, 1994; Alsaad *et al.*, 2006; Nazifi *et al.*,2009 and Al-Hasnawi, 2009). All these changes occur as a result of anemia, persistent loss of blood caused by permanent blood sucking ticks which play a role as well (Mbassa *et al.*,1994 and Durrani *et al.*,2008). The other important cause to make the anemia in ovine theileriosis was destruction of RBCs by reproductive of parasite inside RBCs which caused hemolytic anemia. As well as the modern research explains the mechanism of anemia that occurs due to the activity of antioxidant enzymes such as superoxide's dismutase (SOD) was effective by parasites and results increased fragility of RBCs and thus ,acceleration of erythrocytes clearance by phagocytic cells (Grewal *et al.*,2005 and Asri & Datir 2006).

The present study showed that a significant decreases in total WBCC, this result agreed with the result of (Al-Hasnawi,2009 and Zia-ur-Rehman *et al.*, 2010). The decreased in white blood cells count might be explained by destruction of WBCs specially lymphocyte cells although occurs hyperlymphoplasia due to infection, in addition present of some lymphocyte destruction in blood smears which are related to reproductive of parasite inside lymphocyte. The differential WBCC show decrease in lymphocyte and neutrophils with a significant difference $P< 0.05$ this result agreement with (Zia-ur-Rehman *et al.*, 2010) . On the other hand there is significant increase in eosinophils and monocytes count; while basophiles don't show importing statically change these results were agree with (Singh *et al.*, 2001; Hussine *et al.*,2004 and Al-Hasnawi, 2009).

The seroprevalence by using ELISA test was highest than microscopic observation in identification and diagnosis of ovine theileriosis genus of *T. lestoquardi* .These results agree with the result of (Guo *et al.*, 2007) which showed that 824 (66.77%) sheep infected by ovine theileriosis from 1234 sheep diagnosed by ELISA , (Yin *et al.*,2003) reported that 164 of the 173 (94.79%) samples examinaned were positive to *Theileria lestoquardi* by ELISA test

In present study two species of *Theileria, T. lestoquardi* and *T. ovis* observed, this result agree with Heidarpour Bami,et al,(2010). These results are explained by difficult difference between *T. lestoqurdi* and *T. ovis* in microscopic examination due to *T. ovis* taken multi shape as *T. hirci* (Al-Robayi,1994 and Hashemi-Fesharki,1997). These findings agree with (Altay *et al.*, 2005) who reported that four *Theileria* species (*T. lestoquardi*[1], *T.ovis*[2], *T. separata*[3] and *Theileria spp. China*[4]) can cause theileriosis in sheep. It is difficult to differentiate these species on the basis of the morphology of piroplasm and schizont stages, especially in mixed infections.

The present study showed that the distribution of benign theileriosis genus *T. ovis* is restricted in northern region rather than region of province, and recorded high prevalence rate in northern region (66.66%) followed by eastern region (18.33%) and lower prevalence rate (6.66%),(5%) and (3.33%) in western, central and southern region respectively. These findings agree with the result of (Zaeemi *et al.*,2010) who reported that *T. ovis* is the dominant species in the north and north-west regions of Iran.

دراسة سريريه ودمويه والتشخيص المصلي للاصابه بالثاليرية في الاغنلم في محافظة البصره

طارق رفعت , اسراء عبد الودود

كلية الطب البيطري/ جامعة البصره

الخلاصة

أجريت هذه الدراسة لتحديد انتشار الإصابة بداء الثايليريا في أغنام محافظة البصرة , اذ تم تقيم الإصابة بواسطة تسجيل العلامات السويسرية والمعاير الدمية وكذلك استخدام اختبار الاليزا المصلي لطفيلي *Theileria lestoqurdi*.

امتدت الدراسة من تموز2010الى 2011 اذ تم جمع 1466 عينة من أغنام بأعمار وأجناس وسلالات مختلفة وبصورة عشوائية من مناطق مختلفة شمال وجنوب وغرب وشرق وكذلك مركز المحافظة.

لوحظت العلامات السريرية على 460 رأس من الأغنام التي فحصت سريريا والتي تمثلت بالحمى ,تضخم العقد اللمفاوية السطحية ,علامات تنفسية وزيادة في ضربات القلب . وأظهرت الفحوصات الدموية ارتفاع في مستوى الطفيلنمية حيث سجلت نسبتها مابين 12%- 80% ولوحظ جميع إشكال وأطوار الطفيلي اذ كانت نسبة الطور الدموي 64.8% والطور اللمفاوي 22.62% والطور الدموي واللمفاوي 12.57% .

بينت نتائج الدراسة أن اعلي نسبة حدوث لداء الثايليريا في الأغنام كانت في شمال المحافظة 65.8% تلتها جنوب البصرة وبنسبة 58.9% بينما اوطا نسبة حدوث كانت في مركز المحافظة 52.98% .

أظهرت المعاير الدمية وجود فقر دم الكبير الحجم السوي الصباغ وفقر الدم سوي الحجم سوي الصباغ في الاغنام المصابة وانخفاض معنوي في قيم خضاب الدم (g/l) 0.4±7.8. وحجم الخلايا المرصوص PCV 26.81± 2.991. ومعدل تركيز خضاب الدم الكروي 1.8 ±28.99

وعدد كريات الدم الحمر 62.38±5944545 وعدد كريات الدم البيضاء 709.76±5870 والخلايا اللمفاوية 450±3602.95 والعدلات 235.72±1166.8 بينما لوحظ ارتفاع معنوي في الخلايا الحمضية وخلايا وحيدة النواة وكذلك زيادة في معدل الحجم الكروي.

بينما التشخيص المصلي للدراسة باستخدام اختبار الاليزا أشار إلى أن أعلى نسبة للإصابة كانت لتشخيص النوع الخبيث *Theileria lestoquardi* 93.14% بينما اوطا نسبة للإصابة كانت بالنوع الحميد للمرض *Theileria ovis* 6.85% .

References

1- **Altay, K., Aktas, M.,and Dumanli, N. (2007).** *Theileria* infections in small ruminants in the East and Southeast Anatolia. Türkiye Parazitoloji Dergisi., 31:268–271.

2- **Alani, A.J. and Herbert, I.V.(1988).**Pathogenesis of infection with *Theileria recondite* (Wales) isolated from *Heamaphysalis puncutata* from North Wales. Vet. Parasitol.,4:293-301.

3- **Ahmed, J.S.; Luo, J.; Schnittger, L.; Seitzer, U.; Jongejan, F.;Yin, H. (2006)**. Phylogenetic position of small ruminant infecting piroplasms.Ann.N.Y.Acad.Sci.,1081:498-504.

4- **Niu, Q.; Luo, J.; Guan, G. Ma, M.; Liu, Z.; Liu, A.; Dang, Z.; Gao, J.; Ren, Q.; Li, Y.; Liu,J. and Hong Yin.(2009).** Detection and differentiation of ovine *Theileria* and *Babesia* by reverse line blotting in China. J. Parasitol. Res.,104:1417–1423.

5- **5-Altay, K., Dumanli, N., Holman, P.J. and Aktas, M. (2005).** Detection of *Theileria ovis* in naturally infected sheep by nested PCR. J. Vet. Parasitol., 127:99–104.

6- Humandi, A.K.J. (1978). Possibility of Gamma-ray using in control of Hayloma anatolicum anatolicum .M.Sc. Theisis collage of Vet. Med., Uni. Baghdad.(In Arabic).

7-**Al-Hasnawi, H. R. D. (2009).**Epidemiological and diagnostic study of bovine theileriosis in north Basrah province.M.Sc.Thesis.,CollegeofVeterinary Medicine, University of Basrah. Pp:49-90.

8- **Guo, S.; Yuan, Z.; Wu, G.; Wang, W.; Ma, D. and Du, H. (2002).** Epidemiology of ovine theileriosis in Ganan region, Gansu Province, China. Parasitol. Res. 88: 36-37.

9- **Radostitis, O.M.; Gay, C.C.; Hinchcliff, K.W. and Constable, P.D. (2007).** Veterinary Medicine, 10th Ed. A textbook of the disease of cattle, sheep, goats ,pigs and horses.Saunders Company Ltd.,London, New York.Pp: 1328-1329.

10- **Aktas, M.; Altay, K. and Dumanli, N.(2005).** Survey of *Theileria* parasites of sheep in eastern Turkey using polymerase chain reaction. Parasitol. of Vet. Med. Univ. of Firat, Turkey. J. of small ruminant Research., 60: 289-293.

11- **Schalm, O. W.; Jain, N. C. and Carrol, E. J. (1975).** Veterinary hematology. Leo and Febiger, ed. Philadelphia.1st Ed. Pp: 140-152 .

12- **Dasie, J. V. and Lewis, S. M. (1984).**Theileriosis. Practical hematology 6th ed. Edinburgh, Churchill., Pp: 40-55.

13- **Coles, E. H. (1986).** Anemia.Veterinary clinical physiology. 4th ed.,W.B. Saunders company, Philadelphia, London. Pp: 16.

14- **Chaudhri, S.S. and Gupta, S.K.(2003).** Manual of General Veterinary Parasitology. 1st edition . department of Vet. Parasitology College of Vet. Sci. Haryana Agicultural University India.Pp:46-47.

15- **Al-Robayi H. M**. **Ali**. (**1999)** Epidemiology of *Theileria annulata* infection in with Al Ashaiki farm . Ph. D. Thesis., Colleg of Veterinary Medicine, University of Baghdad. (In Arabic).Pp:29-74.

16- **El-Azazy, O.M.E.; El-Metenawy, T.M. and Wassef, H.Y. (2001).** *Hyalomma impeltatum* (Acari: Ixodidae) as a potential vector of malignant theileriosis in sheep in Saudi Arabia. Vet. Parasitol., 99: 305-309.

17- **Yin, H.G.; Luo, J.; Guan,G.; Man, M. and Bai, Q. (2003).**Observation on the schizont stage of an unidentified Theileria sp. In experimentally infected sheep. Parasitol. Res.,91:34-39.

18- **Haddadzadeh, H.R.,Rahbari, S., Khazraii nia, P. and Nabian, S.(2004).** New concepts on limiting factors of ovine and caprine malignant theileriosis (OCMT)in Iran. Iranian J. of Vet. Res.,Vol.5, No.2 Ser., No.10:1383.

19- **Zia-ur-Rehman,; Khan, M.S.; Avais,M.; Aleem, M.; Shabbir, M.Z. and Khan, J.A.(2010).** Prevalence of theileriosis in sheep in Okara District, Pakistan. Pakistan J.Zool.,42(5): 639-643.

20- **Hussein, A.; Mohammed, N. A. and Mohammed H. K. (2004)**.Theileriosis and Babesiosis in cattle : hemogram and some biochemical parameters. J. Vet. Parestiol., 136: 117-122.

21- **Osman, S.A. and Al-Gaabary, M.H. (2007).** Clinical, haematological and therapeutic study on tropical theileriosis in water buffaloes (Bubalus bubalis) in Egypt. Vet. Parasitol., 146: 337-340.

22- **Cheville, N.F.(1976).** Cell pathology. Lowa state University press Ames. U.S.A. Cited by **Al-Robaiy, H.M. Ali. (1994).** A study of hematological& biochemical changes insheep experimentally infected with (Theileria hirci).M. Sc. Thesis., College of Veterinary Medicine, University of Baghdad. (In Arabic).Pp:4-67.

23- **Oteng, A.K. (1969).** Bovine theileriosis in Australia. Abst. of M.V.S.C. Thesis Univ. Queensland .P:1(Vet. Bull.(1970), Abst. No.2225).

24-**Al-Robaiy, H. M. A. (1994).** A study of hematological &biochemical changes in sheep experimentally infected with (*Theileria hirci)*. M. Sc. Thesis., College of Veterinary Medicine, University of Baghdad. (In Arabic).Pp:4-67.

25- **Glass, E.J.; Craigmile, S.C.; Springbett, A. Preston, P.M.; Kirvar,E.; Wilkie, G.M.; Eckersall, P.D.; Hall, F.R.; and Brown, C.G.D.(2003).** The protozon parasite *Theileria annulata* induces a

distinct acute phase protein response in the cattle that is associated with pathology. Int. J. Parasitol., 33: 1409-1418.

26- **Singh, A.; Singh, J.; Grewal, A. S. and Brar, R. S.(2001)**.Study on some blood parameters of crossbred calves with experimental *Theileria annulata* infections. Veterinary research communications., 25: 289-300.

27- **Heidarpour Bami, M.; Khazraiinia, P.; Haddadzadeh, H. R. andKazemi, B.(2010).** Identification of *Theileria* species in sheep in the eastern half of Iran using nested PCR- RFLP and microscopic techniques. Iranian J. of Vet. Res., 11:3-32.

28- **Latif, B.M.A.; Hawa, N.J. and Bakir, F.A. (1977).** Incidence of malignant theileriosis (*Theileria hirci*) of sheep in Iraq .Iraqi J.Vet.Med.1:29-37.

29- **Latif, B.M.A.; Al-Izzi, S.A.; Majeed, M.A.H. & Sultan, A.S. (1987).** Prevalence of piroplasmosis among sheep in the central part of Iraq. Iraqi J. Vet. Med.(11): 132-137.

30- **Billiouw M**. (**2005)**. The epidemiology of bovine theileriosis in the eastern province of Zambia. Ph. D. Thesis., Parasitologieen immunologie faculteit Diergeneeskunde, University of Gent.Pp:19-56.

31- **Alsaad, K. M.; Al-obaidi, Q. T. and Esmaeel, S. A. (2009).** Hematological and biochemical study on the effect some common blood parasites innative goats in Mosularea. (In Arabic). Iraqi J. of Vet. Sci.,23(1):101-106.

32- **Dhar, S. and Gautam, O.P. (1979).** Observation on anemia in experimentally induced with Theileria annulata clinic- chemical studies . Vet. Bull.(1978), Abst.No.1901.

33- **Omer, O. H.; El-Malik, K. H.; Mahmoud, O. M.; Haroun, E. M.; Hawas, A.; Sweeney and Magzoub M. (2002).** Haematological profiles in pure breed cattle naturaly infected with *Theileria annulata* in Soudi Arabia. J. Vet. Parastol., 107: 161-168.

34- **Nazifi, S. ; Razavi, S. M. ;Hasanshahi, F. and Eamailnzhad, Z. (2009).** Eeffect of severity of *Theileria annulata* infection on some hematological parameter and antioxidant enzymes in natural infection cattle. Bulgarian. J. Vet. Med., 12(1):63-71.

35- **Al-Saad, K.M. and AL-Mola, G.M.(2006).**Clinical and pathological study of equine Babesiosis in drought horses in Mosul. Iraq. J. Vet. Sci.,20:89-101.

36- **Al-Obaidi, Q.T. (2006**).Clinical and hematological studies of single infection with some blood protozoa ,endo and ectoparasite in native goats in Mosul. Iraqi. J. Vet. Sci., 20: 283-289.

37- **Grewal, A.; Ahuja, C. S.; Singh, S. P. and Chaudhary, K. C. (2005).** Status of lipid peroxiadtion some antioxidant enzymes and erythrocytic fragility of crossbred cattle naturally infection with *Theileria annulata.* J. Vet. Res. Com., 29:387-394.

38- **Asri Rezaei, S. and Dalir-Naghaded, B. (2006)**. Evaluation of antioxidant status and oxidative stress in cattle infected with *Theileria annulata* . J. Vet. Parasitol., 142: 147-186.

39- **Guo, S.Z.; Mu, Y.J.; Liu, Z.J.; Ma, D.L.; Yang, S.M.; Ge, G.H.; Fang, B.Q.; Ga, D.J.; Ma, M.L.; Luo, J.X.; Yin, H.; Seitzer, U. and Ahmed, J.S. (2007)**. Serological investigation of ovine theileriosis by ELISA in Gannan Tibet Region of Gansu Province in China. Parasitol. Res.,101:197–200.

40- **Heidarpour Bami, M.; Khazraiinia, P.; Haddadzadeh, H. R. and Kazemi, B.(2010).** Identification of *Theileria* species in sheep in the eastern half of Iran using nested PCR-RFLP and microscopic techniques. Iranian J. of Vet. Res., 11:3-32.

41- **Hashemi-Fesharki, R.(1997).**Tick-borne disease of sheep and goats and their related vectors in Iran.Parasitol.Res.39:115-117.

42- **Zaeemi, M.; Haddadzadeh, H.; Khazraiinia, P.; Kazemi, B. and Bandehpour, M.(2010).** Identification of different *Theileria species* (*Theileria lestoquardi, Theileria ovis,* and *Theileria annulata*)in naturally infected sheep using nested PCR–RFLP. J. Parasitol. Res. DOI 10.1007/s00436-010-2119-0.