DETECTION OF ANAEMIA CAUSED BY PARASITIC INFECTIONS AND ESTIMATION OF HAEMATOLOGICAL VARIABLES AND BLOOD PICTURE IN SHEEP

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ABSTRACT

This study was conducted to detect anemia caused by parasitic infection, by ways of determining changes in some blood values, and blood picture to evaluate the histological change. Blood samples were collected from Awassi sheep (150 animals as anaemic group and 75 as control group), in Salahudeen city. The samples were collected with ages varying from (1-5 years). The results showed a significant reduce in (PCV, (Hb), MCV, MCH, and MCHC). The results of the current study showed the presence of parasites of the following types *Babesia, Thileria, Anaplasma*, which caused anaemia of sheep's.

Key words: Anaemia, parasitic infection, Awassi sheep, blood parasite, Hb

Introduction

Sheep are economically important animals because their nutritional and administrative requirements are inexpensive, as well as the diversity of their products and ways to benefit from them by humans, as selection in farm animals depends on the formal characteristics of any quantitative characteristic measured on the animal is a function of its phenotypic value, as the phenotypic value Hereditary factors and the extent to which they are affected by non-genetic factors and the interaction between them (Falconer and Mackay, 1996).

The area between the River Tigris and the Euphrates is considered the original home of the Awassi sheep, and it is one of the most important sheep veins in the Middle East. It is spread in Syria, Lebanon, Palestine, Jordan, northern Saudi Arabia, Iraq and southern Turkey (Talal, 2011). Also, Awassi sheep distinctive of their ability of meat and milk production, so its play economic role among Livestock (Bakunzi *et al*., 2013; Zvinorova *et al*., 2016).

Anaemia is a condition that occurs because the haemoglobin concentration is lower than the normal level. Because of the low haemoglobin level, the body's systems suffer from not getting enough oxygen. Consequently, patients complain of signs and symptoms such as fatigue, headache, lack of focus, lethargy, and others (Ježek *et al*., 2009), There are three main types of anaemia: anaemia due to blood loss (haemorrhagic), anaemia due to abnormal production of red blood cells, and hemolytic anaemia. Anaemia is the most common haematology condition. In the United States, this disease affects more than three and a half million Americans. Women and people with chronic diseases may be at increased risk of developing anaemia (Matsuda, 2010). Anaemia is also characterized by a decrease in the amount of erythrocytes, often accompanied by a decrease in haemoglobin levels or a change in erythrocytes morphology. Anaemia is a diverse and often multifactorial disease (Kassebaum *et al*., 2014) resulting from complete loss, deficiency or malabsorption of a number of elements and vitamins such as vitamin B12, iron, zinc and cobalt (trace elements) (Rizk *et al*., 2017).

Materials and Methods

Study design and Animals

Animals were divided to anaemic group included (150 sheep's) and control group included (75 healthy sheep's), the anaemic animals diagnosed by clinical symptoms that "anorexia, pale mucus membranes and poor coat".

Sampling

The samples of blood were collected, to estimation the blood test, haematological variables were observed (HB, PCV, MCV, MCH, and MCHC) and blood smears were preparation to detect blood parasites.

Blood samples were obtained from the jugular vein using methyl alcohol 70%, for sterilization, 10 ml of blood was taken, 2.5 ml placed in EDTA tubes for haematological tests and blood smear.

Results and Discussion

Parasites and abnormal forms of red blood cells:

The results of the current study showed the presence of parasites of the following types *Babesia, Theileria* and *Anaplasma* (Barcellini and Fattizzo, 2017; Al-Ammari, 2008; Razmi *et al*., 2006).

The incidence of *Babesia* was 25%, which is consistent with the findings of (Giadinis *et al*., 2011) and (Duzgun *et al*., 1991) and the rate of infection with *Anaplasma* recorded was 20%, which is in line with (Mohammed and Salman, 2016) and contrary to what was mentioned (Al- Rubaii, 2008). The

Table 1: The ratio % of single and double parasitic infection

Infection parasite	Single infection %	Infection parasite	Double infection %
Theilaria	27%	*Theilaria+ Babesia*	6%
Babesia	25 %	*Theilaria+Anaplasma*	6.4 %
Anaplasma	20 %	*Babesia+ Anaplasma*	3.2 %

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incidence of double infections was 6% for *Theilaria* and *Babesia* and 6.4% for *Theilaria* and *Anaplasma* whereas 3.2% was recorded for *Babesia* and *Anaplasma.*

The current study showed that the type of anaemia associated with blood parasite infection was macrocytic normochromic anaemia, and this result was in agreement with Stockham *et al.* (2000) and macrocytic hypochromic anaemia, and microcytic hypochromic, was also recorded by Mohammed and Salman (2016).

The results of the current study showed the presence of a number of abnormal forms of red blood cells, which are hypochromic RBC with the presence of target cells, this finding agrees with Weiss and Wardrop (2011).

The blood cells also showed various shapes like Poikilocytosis (acanthocytes and echinocytes), which was similar to Jones and Allison (2007) and Stockham and Scott (2008). Dacryocyte Elliptocyte, Howell-Jolly bodies, Basophilic stippling, Schitocyts also appeared in blood smear. The present study findings agree with previous study of Jones and Allison (2007), Weiss and Wardrop (2011) and Harvey (2011). The findings indicate variation in the size of red blood cells and anisocytosis.

Torocyte and Leptocyte corpuscles also appear in blood smear; these results are consistent with what observed by (Weiss and Wardrop, 2011) and (Stockham and Scott, 2008).

The results showed that there were 33 cases with Microcytic normochromic anaemia, as they were 13 cases at the age of one year, 5 cases at the age of 2-3 years, and 15 cases at the age of 3-5 years. A significant decrease ($P < 0.01$) was observed in the cases of anaemia at the age of one year, 3- 5 years, and age of 2-3 year in mean of MCV and MCH haemoglobin, and no significant difference in the rate of MCHC haemoglobin concentration compared with healthy sheep. Results of this study agreement with a previous study (Henniger *et al*., 2013). The causes is attributed to iron deficiency and liver failure and is in agreement with what found by (Stockham and Scott, 2008) anaemia associated with inflammations, copper deficiency, and myeloma (Harvey, 2011).

The analyzes also showed the presence of 87 cases with macrocytic normochromic anaemia, as 10 cases were at the age of one year, 20 at the age of 2-3 years and 57 cases at the age of 3-5 years, and a significant increase ($P<0.01$) was observed in anemic animals in average of the MCV and the rate of haemoglobin MCH and the absence of a significant difference in the rate of MCHC haemoglobin concentration compared with the normal sheep. The cause of this type of anaemia may be due to infection with internal parasites and blood parasites and is identical to what observed by (Al-Jumaili, 2015). Also folic acid deficiency, genetic defects in the process of manufacturing red blood cells, especially in calves of the genus Hereford, and also occurs when red blood cells are coagulated and in bone marrow disorders such as erythroleukemia and myelodysplastic syndromes (Aktas *et al*., 2006).

The analysis showed the presence of 9 cases of normocytic normochromic anaemia, as they were 9 cases of 3-5 years of age, and no significant difference was observed in the cases with anaemia in the rate of the MCV and the rate of MCH and the rate of MCHC compared with healthy sheep. Most previous sources mentioned that this type of anaemia

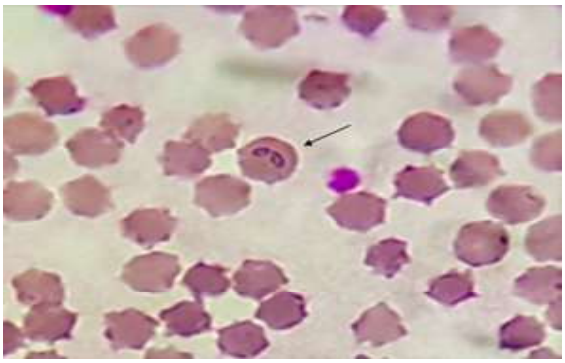

Fig. 1: Blood swab showing the infection with *Babesia spp*. (Giemsa, 1000X)

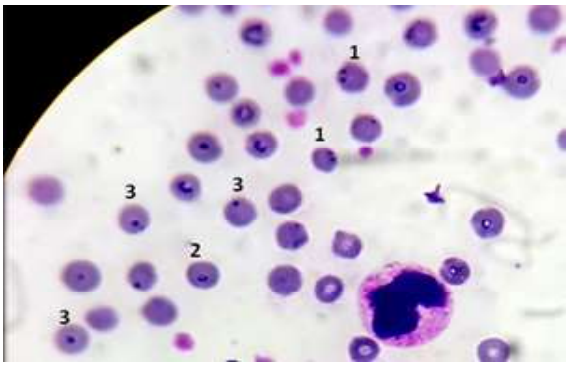

Fig. 2: Blood smear showing infestation with *Babesia spp*. (1); *Theilaria spp.* (2); *Anaplasma spp.* (3)(Geimsa, 1000*X)*

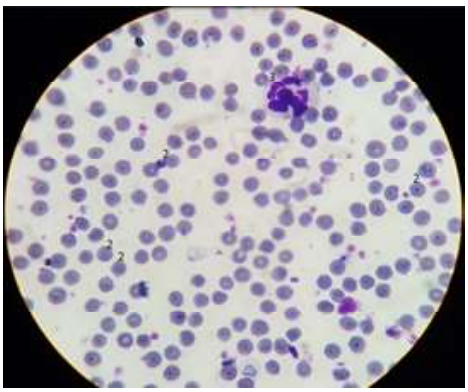

Fig. 3: Blood smear showing the infection with parasites; *Theilaria spp.* (2)*; Anaplasma spp.* (3) (Giemsa, 1000X)

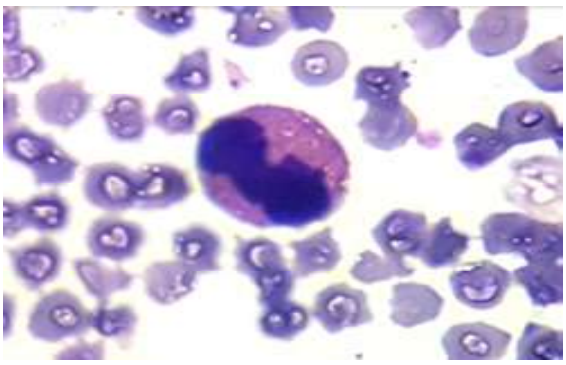

Fig.4: Blood smear showing the acid white blood cell (Eosinophil) (Giemsa, 1000X)

may be due to several reasons, including parasites infection in large animals and the effort they are exposed to during pregnancy and childbirth, and it is consistent with finding of (Weiss and Wardrop, 2011; Hussein and Al-jeburii, 2012) and so on chronic diseases (Warner and Harris, 2000). This is in agreement with what Al-Obaidi (2010), found it is also associated with haemolytic anaemia, haemorrhage, early cases of iron deficiency anaemia, tumors, chronic urinary system diseases, endocrinopathy, lead poisoning and vitamin B_{12} deficiency and is identical to the results of the current study (Harvey, 2011).

Macrocytic hypochromic anaemia was shown by 18 infected cases, 2 cases of 1 year old,(6) cases in 2-3 years old, 10 cases 3-5 years old, and a significant increase ($P < 0.01$) in anemic cases in mean corpuscle size. MCV and MCH haemoglobin rate, and a significant decrease ($P<0.01$) in MCHC concentration compared to normal sheep. This type of anaemia is caused by a defect in the generation of erythrocytes (erythropoiesis) in the bone marrow of. Infection with intestinal parasites is consistent with what has been mentioned (Hassan and El_Said, 2000), and infection with the, *Babesia, Thileria, Anaplasma*. as noted through the current study.

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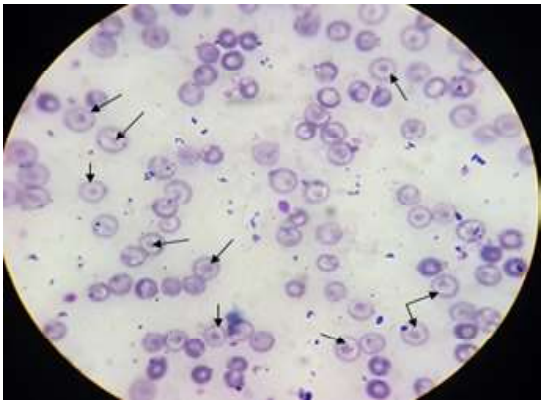

Fig. 5: Hypochromic erythrocytes with target cells (Gimsa, 1000X). Blood smear shows Hypochromic erythrocytes with the presence of target blood cells

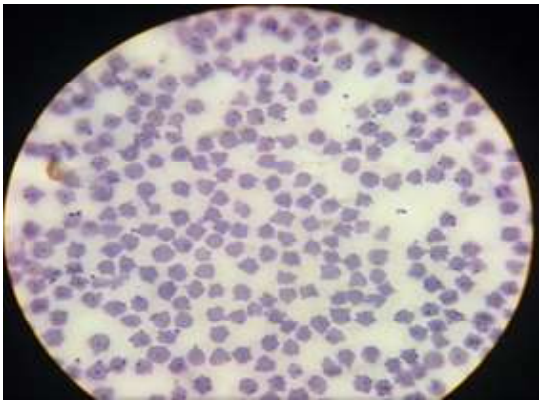

Fig. 6: Poikilocytosis (acanthocytes and echinocytes) (Gimsa, 1000X). A blood smear showing differently shaped blood cells with spinal cells

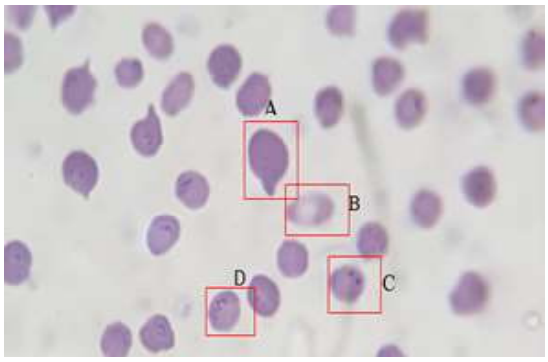

Fig. 7: Dacryocyte); (B) Elliptocyte; Howell-Jolly bodies (C); Basophilic stippling (D) (Gimsa, 1000X)

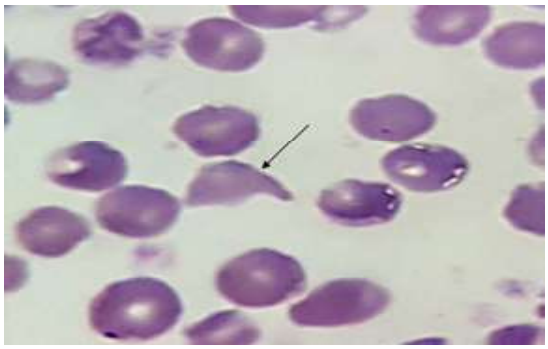

Fig. 8: Schitocyts (Gimsa, 1000X)

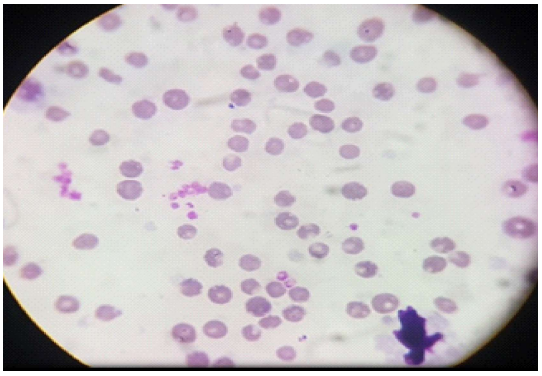

Fig. 9: Anisocytosis (Gimsa, 1000X) A blood smear showing the difference in RBC size

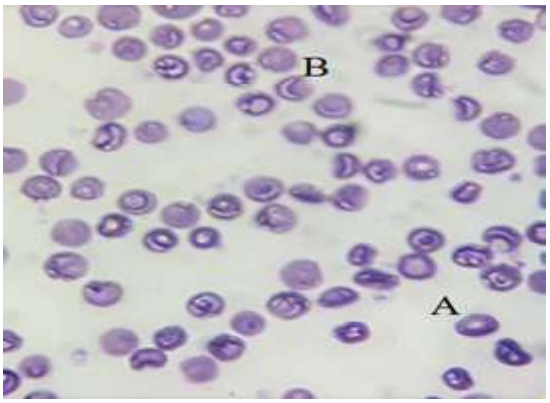

Fig. 10: Blood smear showing the presence of Torocyte (A); (B) Leptocyte (Gimsa, 1000X)

Table 1: Blood values of one-year-old anaemic sheep

Hemato-logical variables	Anaemia types					Control group
	Microcytic normochromic	Macrocytic normochromic	Normocytic normochromic	Macrocytic hypochromic	Microcytic hypochromic	
PCV%	17.84±1.14	22.2±0. 904	-	24± 2	20	33±0.757
HBg/dl	6± 0.385	7.43± 0.259	-	7.1± 0. 1	5.3	11.22±0.315
RBC×10^6 /µL	6.7±0.650	3.84±0.516	-	5.15±1.150	6.2	10.95±0.212
MCV fl	26.46±0.671**	67.9± 10.05**	-	48±7**	32	29.76±0.551
MCH pg	8.7±0.291**	22. 55± 3.24**	-	14±3**	8	9.86±0.221
MCHC g/dl	33.3± 0.33	33.36± 0.576	-	29± 2**	26	33.47±0.287
NO	13	10	-	2	1	25

Table 2: Blood values of 2-3 year old anaemic sheep

Hemato-logical variables	Anaemia types					Control group
	Microcytic normochromic	Microcytic normochromic	Microcytic normochromic	Microcytic normochromic	Microcytic normochromic	
PCV%	20.80±2.00	21.35± 0.48	-	24.20±1.15	23	34.68±0.590
HBg/dl	6.800± 0.656	7.07± 0.147	-	6.64±0.206	7	11.71±0.313
RBC ×10^6 /µL	7.6±0.871	4.05±0.305	-	3.87±0.741	6.2	10.78±0.181
MCV fl	26.40±0.671**	59.4±5.08**	-	70.8± 11.02**	37	32.28±0.808
MCH pg	8.88±0.406**	19.55±1.70**	-	19.6±3.66**	11	10.63±0.344
MCHC g/dl	32.700± 0.435	32.9± 0.173	-	27.44±1.98**	30	32.83±0.518
NO	5	20	-	6	1	25

Table 3: Blood values of 3-5 year old anaemic sheep

Hemato-logical variables	Anaemia types					Control group
	Microcytic normochromic	Macrocytic normochromic	Normocytic normochromic	Macrocytic hypochromic	Microcytic hypochromic	
PCV%	19.80±0 .97	21.34±0.31	20.33± 1.09	22.7±1.011	26	34.24±0.536
HBg/dl	6.70± 0.317	7.13±0.101	6.82± 0.361	6.28±0.209	7.2	11.40±0.224
RBC×10^6 /µL	7.48± 0.421	4.07±0.188	6.25±0. 356	4.4±0.351	6.3	10.81±0.163
MCV fl	26.5± 0.703**	61.21±3.97**	32.33±0.336	53.8±5.076**	41	31.34±0.598
MCH pg	8.70±0.25**	20.3± 1.329**	10.75±0. 214	14. 2±0.866**	11	10.22±0.22
MCHC g/dl	33.62±0.357	33.24±0.109	33.31± 0. 358	27.5±1.147**	27	32. 8±0.264
NO	15	57	9	10	1	25

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