

EPERYTHROOON OVIS INFECTIONS OF SHEEP AND GOATS IN SHALATEEN, ABU-RAMAD AND HALAEEB, RED SEA GOVERNORATE, EGYPT

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Abstract

An outbreak occurred in sheep and goats in triangular area (Shalateen –Abu-Ramad and Halaeeb), Red Sea Governorate, Egypt was described between January to December 2013. This outbreak showed clinical signs characterized by fever, inappetence, emaciation, anemia, icterus and ill-thrift. Clinical signs are most likely to be observed in young suckling and weaner animals. It is shown to be a complex problem attributable to many causes, some of which can be eliminated by supplementary feeding, drenching, vaccination, dipping and management. However, severe prolonged anemia accompanied by the development of ill-thrift still present in 248 animals (165 sheep and 83 goats). Infection with *Eperythrozoon ovis* was demonstrated in 21.42% and 12.50% of lambs and kids, 41.37 % and 28.75% of weaner sheep and goats and 10.75 % and 8.51% of adult sheep and goats respectively. When the general prevalence was analyzed by age, it was observed that weaners were mostly affected and disease was highly observed during winter and summer. Hematological examination showed macrocytic hemolytic anemia, with anisocytosis, poikilocytosis and marked left shift in erythrocyte maturation. Infected sheep and goats showed significant reduction in values of Hb, PCV and RBC while leukocytic count appeared slightly elevated. The main pathological changes in *E. ovis* infection were an increase in size of spleen at the peak of parasitaemia and an increase in liver size. Haemosiderin was present in livers, kidneys and spleens of all infected sheep and goats at the peak and late stages of parasitaemia.

Key words: *Eperythrozoon ovis* – Incidence – Hematology – Clinical significance – Histopathology.

INTRODUCTION

Eperythrozoonosis is a parasitic disease caused by *Eperythrozoon* parasitizing on the erythrocyte surface, plasma and bone marrow of sheep, goats and cattle and transmitted by blood sucking arthropods (Sheriff et al., 1966). Sheep and goats parasitized by *Eperythrozoon* have the symptoms of anemia, icterus, fever and reproductive disorders. Deaths may occur in severely affected young sheep and goats especially if they are stressed by yarding. Losses of up to 30% of the flock have occurred in these circumstances (Brun et al., 2007). *Eperythrozoon* causes further losses when infected carcasses are condemned because of jaundice (Veale and

Nicholls, 1986). Infections are frequently unapparent and chronic. *E. ovis* in goats results in more severe disease (Mason and Statham, 1991). *E. ovis* has not been cultivated, diagnosis is made by detection of organisms on erythrocytes in Romanowsky type or Prussian blue stained blood smears and by serological methods. Parasitism of erythrocytes often occurs at a low level and is transient; detection requires examination of repeated blood smears (Gulland et al., 1987). The aim of this study is to record the incidence, clinical significance, hematological and pathological changes of *Eperythrozoon ovis* infection in sheep and goats.

MATERIALS AND METHODS

Animals:

A total of 248 animals [165 sheep (14 lambs 1-4 months, 58 weaner >4-12 months and 93 mature > 12 months) and 83 goats (8 kids, 28 weaner and 47 mature)] from the triangular area (Shalateen, Abu-Ramad and Halaeeb) were included in this study between January to December 2013. These animals were subjected to careful clinical and laboratory investigations.

Parasitological examination:

Blood samples were collected with a sterile needle and syringe from the jugular vein of each animal showed symptoms of fever, inappetence, emaciation, anemia, icterus and ill-thrift. About 2 ml of blood was collected from each animal into test tubes containing ethylene diamine tetra-acetic acid (EDTA) as anticoagulant. In all groups active infection was demonstrated by the examination of thick and thin blood films, stained with Giemsa stain. The mean value of parasitaemia was recorded according to Patricia and Harbutt, (1969). Percentage of infected RBCs was assessed by counting the number of parasites in erythrocytes present per 1000 cells at magnification of x1000 then divided by ten and expressed as percentage (Urquhart et al., 2003).

Hematological examination:

Hb, PCV, RBC and WBC were determined using the standard methods described by (Schalm et al., 1975). The MCHC, MCH and MCV were also calculated.

Necropsy examination:

Seven dead animals (5 lambs and 2 kids) and 2 live affected weaners were necropsied after slaughter. Parts from liver were fixed in 10% neutral buffered formalin, then embedded in paraffin, sectioned at 5 μ m and stained with Prussian blue according to Bancroft and Stevens (1993).

RESULTS

Table (1) showed that the percentage of infection was higher in summer (28.57%) and winter (22.72%) in sheep and goats respectively. The lower percentages were in spring (17.5%) and autumn (10%) in the same animals. The clinical signs observed were fever, inappetance, emaciation, ill thrift, anemia, jaundice and death.

Incidence of *Eperythrozoon ovis*:

Eperythrozoon ovis infection was demonstrated in 21.42% and 12.50 % in lambs and Kids, 41.37 and 28.57% in weaner sheep and goats and 10.75 % and 8.51% in adult sheep and goats respectively (Table 2).

Table 1. Seasonal distribution of *Eperythrozoon ovis* in sheep and goats.

Season	Examined sheep			Examined goats		
	Total	+ve	%	Total	+ve	%
Winter	42	10	23.80	23	5	22.72
Spring	40	7	17.5	19	2	10.25
Summer	42	12	28.57	21	4	20
Autumn	41	8	19.51	20	2	10
Total	165	37	22.42	83	13	15.66

Table 2. Incidence of *Eperythrozoon ovis* in different ages of sheep and goats.

Animals		No. of examined animals	No. of infected animals	% of infection
sheep	lambs	14	3	21.42
	weaner	58	24	41.37
	Adult	93	10	10.75
Total no. of sheep		165	37	22.42
Goats	Kids	8	1	12.5
	weaner	28	8	28.57
	adults	47	4	8.51
Total no. of goats		83	13	15.66

Morphological characters:

Plate 1 and 2 showed that *Eperythrozoon ovis* was observed on the surface of erythrocytes as small cocci, spheres clustered bodies of approximately 0.5–1.0 µm in diameter. These bodies attached to the perimeter of erythrocytes or found free

floating between erythrocytes. In heavy infections, chains of the parasites surrounding the margin of the red cell, measuring about 1-3 μm in diameter (plate 1).

Hematological examination:

Mean values of hematological parameters are presented in table (3). Infected sheep and goats showed significant reduction in the values of Hb, PCV, RBC while the leukocytic count is slightly elevated. Whereas, MCHC was in the same range. On the other hand, MCH and MCV were highly elevated.

Table 3. Mean hematological values of sheep and goats infected with *Eperythrozoon ovis*

Indices	Mean Values	Range	Reference Range
Hb (g/dl)	4.8	3.5-5.9	9-14
PCV (%)	12.6	10-17	21-39
RBC ($\times 10^6/\text{L}$)	2.8	1.4-3.5	9-15
WBC ($\times 10^3/\text{L}$)	14.1	9.7-22.1	4-11
MCHC (%)	32.5	30-35	29-35
MCH (PG)	18.9	17-23	8-11
MCV (fl)	58	40-69	28-40

Parasitaemic status:

The mean value of parasitaemia (count /1000 RBC) reached 0.48-4.52% with mean of $2.27\% \pm 0.09$ in sheep and 0.29-2.9% with mean of $1.21\% \pm 0.12$ in goats (Table 4). There was marked red cell anisocytosis and polychromatic. Red cells also showed spherocytosis and schistocytosis. These results were consistent with severe responsive macrocytic normochromic anaemia.

Table 4. Parasitaemia of *E. ovis* percent

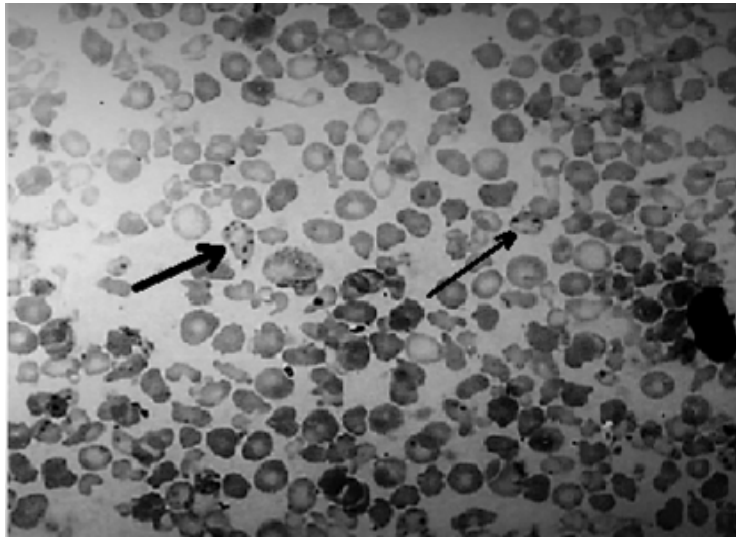
Animals	Range	Mean S.F
Sheep	0.48-4.52	2.27 ± 0.09
Goats	0.29-2.9	1.21 ± 0.12

Postmortem examination:

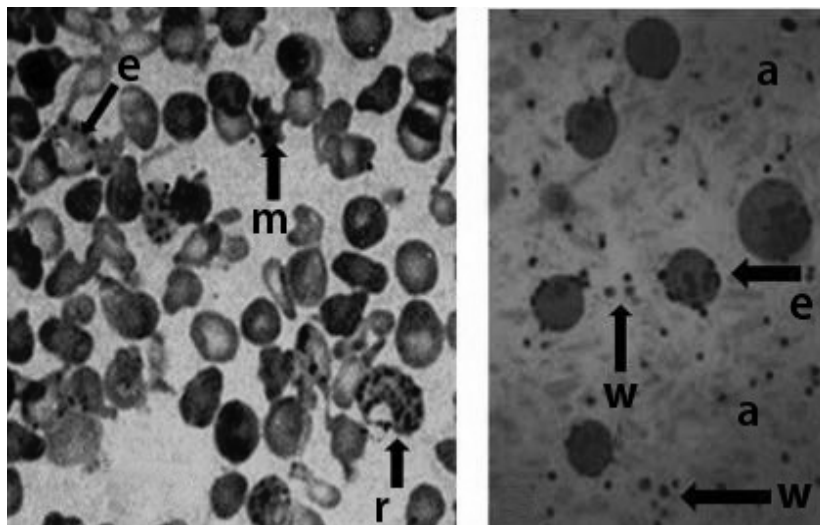
Postmortem examination of infected animals showed pale mucous membranes and various degrees of jaundice. Liver and spleen showed fine mottling and much enlarged with a prominent white pulp.

Histopathology:

The most consistent lesions were a moderate generalized peri-acinar necrosis of the liver, marked hyperplasia of the germinal centers and generalized severe haemosiderosis of liver and spleen.

**Plate 1.**

Giemsa stained blood smear from weaner goat showing *Eperythrozoon ovis* with morphological alteration in some RBCs (x1000)

**Plate 2.**

Giemsa stained blood smear from weaner sheep showing *Eperythrozoon ovis* (x1200).

Eperythrozoon organisms are attached to the perimeter of erythrocytes (e).

The large gaps between red cells indicate anemia (a).

Heavy infection with high parasitaemia between RBCs (w) and inside them (r).

Morphological alteration in RBCs (anisocytosis and poikilocytosis) (m).

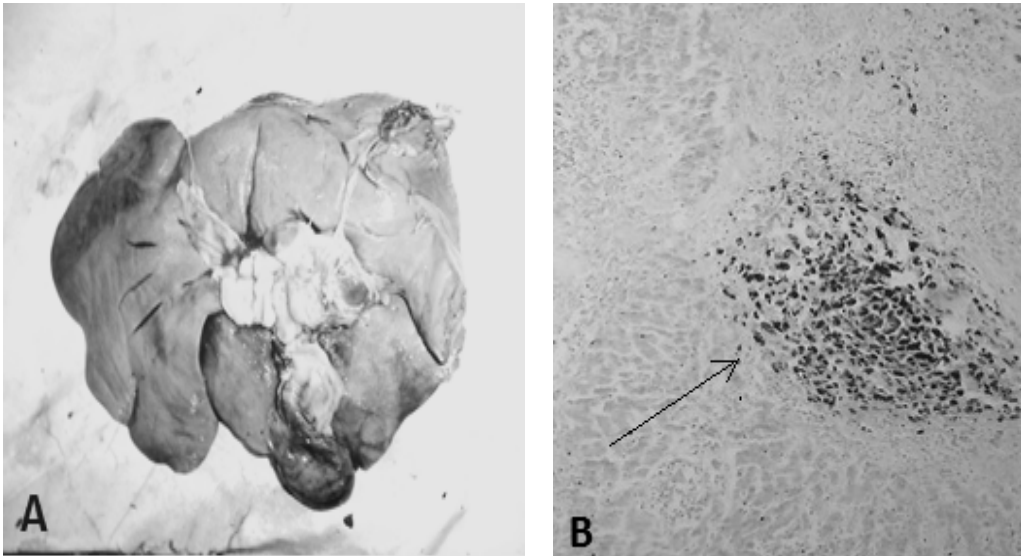


Plate 3.

(A): Liver of sheep infected with *E. ovis* showing severe jaundice.

(B) Haemosiderin in liver of sheep stained with Prussian blue (arrow) x20

DISCUSSION

Results of this investigation revealed *Eperythrozoon ovis* was demonstrated in 21.42% and 12.50% in lambs and kids, 41.37 % and 28.57% in weaner sheep and goats and 10.75 % and 8.51% in adult sheep and goats respectively. Al- Khalifa et al., (2009) recorded that infection with *E. ovis* was 2-9% in Saudi Arabia. In Australia *Eperythrozoon* infection was reported in 10% of weaner and 51% of adult sheep (Veale and Nicholls, 1986) and 40% in Iraq (Hasan, 2012). The difference in percentage of infection may be due to the latent nature and the non-specific clinical symptoms of the disease. It seems likely that the prevalence of *E. ovis* in sheep and goats populations may be considerably greater than the number of clinical cases might lead one to suppose (Overas, 1969). Clinical outbreaks are most likely to be observed in young suckling and weaner animals characterized by fever, anemia, icterus, poor weight gain and depression. These results are in agreement with the results of other studies carried out in other countries (Mason and Statham, 1991 and Robson and Kemp, 2007). The high incidence of infection was recorded in winter and late summer. Our findings are in accordance with other workers (Sheriff, 1979 and Burroughs, 1988). *Eperythrozoon ovis* observed in this study could be due to the favourable environmental conditions for the survival and proliferation of the arthropods vectors such as fleas, mosquitos and ticks. Neimark and Ganter (2004) stated that the *Eperythrozoon ovis* in sheep and goats was transmitted mechanically

by fleas and mosquitos. Ticks (*Rhipicephalus*) are vectors of *Eperythrozoon* for cattle. Also, transmission may occur via surgical procedures through blood contamination of instruments and needles during vaccination or treatment. *E. ovis* appeared as minute bluish coloured cocci, rings, spheres clustered on the surface of RBCs and found free in plasma. Brun et al., (2007) and Hasan, (2012) recorded similar results on this parasite. In the present study hematological studies showed marked decrease of total HB, PCV and RBCs concentration. Macrocytic hemolytic anemia, with anisocytosis, poikilocytosis and marked left shift in erythrocyte maturation were observed. The leukocytic count is slightly elevated and confirm the finding of other researchers. Sheriff, (1979) and Gullanad et al., (1987) reported *Eperythrozoon ovis* causes haemolytic anemia, reduced production and decreased exercise tolerance in sheep and goats. Fitzpatrick et al., (1998) reported that the infection with *E. ovis* can result in damage to red blood cells as the parasite is found attached to their surface or free in the plasma, and the consequence of a heavy infection particularly in young weaned lambs may be cause haemolytic anemia and death. Adejinmi (2004) reported that *Eperythrozoon ovis* is an organism that infect the red blood cells and may trick the body's immune system into thinking that these cells are foreign thus causing the immune system to destroy the cells. The destruction of red blood cells may lead to life-threatening anemia. The mean percent of parasitaemia of *E. ovis* were 2.27% and 1.21% in sheep and goats respectively. Wanphen (2015) mentioned that numbers of *E. ovis* ranging from 1-50, in highly dense parasitaemia. Parasitaemia develops during the pyretic stage at which time parasites can be found in great numbers on the red blood cells and in the plasma (Brun et al., 2007). The severity of anaemia and parasitaemia was variable. It is postulated that in such infection the host immune response may be expressed before infection is fully developed. It was also shown that there was a significant correlation ($p < 0.01$) between the maximum degree of anaemia and maximum degree of parasitaemia for all infected sheep and goats. For this reason the possibility of a direct effect by the parasite on the erythrocyte, contributing to the anaemia cannot be discounted. In case of *Eperythrozoon* infection, the mechanism of disease appears similar to that occurs of other blood parasites, where in immunemediated hemolysis within macrophages follows attachment of the organisms to erythrocytes (Harold et al., 2004). Gulland et al., (1987) recorded that infection of adult sheep with a single strain of *Eperythrozoon ovis* led to three different situations. First, the animal resisted the organism and no haematological changes occurred. Second, the host developed a controllable parasitaemia in which erythrocyte values fell shortly after peak parasitaemia and then returned to normal.

Third, the host failed to control the parasitaemia and chronic low grade anaemia developed. In our study the main pathological features in *E. ovis* infection are mostly similar with Sutton (2011). This investigation indicates that *E. ovis* infection once present in a flock of sheep and goats may spread easily among them and that the infection seems to be persisting within the flock.

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إصابة الضأن والمعز بالإبيريثروزون في شلاتين - أبورماد - حلايب بمحافظة البحر الأحمر، مصر

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ظهرت إصابة الضأن والمعز بالإبيريثروزون بمنطقة المثلث في شلاتين وأبورماد وحلايب بمحافظة البحر الأحمر مصر في الفترة من يناير إلى ديسمبر ٢٠١٣. لوحظ ارتفاع درجة الحرارة و فقدان الشهية وفقر الدم والهزال واليرقان وسوء التغذية خاصة على الرضع و حديثي الفطام . تبين أن المشكلة معقدة و تعزي إلي عدة أسباب منها ما أمكن التغلب عليه باستخدام التغذية التكميلية والتجريع والتطعيم والتغطيس و بعض من المعاملات الغذائية. أوضحت الدراسة أن من بين ٢٤٨ من الحيوانات (١٦٥ من الضأن و ٨٣ من المعز) كانت نسبة الإصابة بالإبيريثروزون هي ٢١.٤٢% و ١٢.٥% في الحملان وصغار المعز و ٤١.٣٧% و ٢٨.٥٧% في المفطوم من الضأن والمعز و ١٠.٧٥% و ٨.٥١% في البالغات من الضأن والمعز على التوالي وجد أن نسبة الإصابة أعلى في الحيوانات المفطومه والتي تظهر في الصيف والشتاء. تبين وجود فقر دم إنحلالي وتفاوت في شكل كريات الدم الحمر مع التحول الملحوظ جهة اليسار لكريات الدم الحمر الناضجة و تميزت الإصابة بانخفاض ملحوظ في العدد الكلي لكريات الدم الحمر ، تركيز خضاب الدم ونسبة العدد المصمت لخلايا الدم بينما كانت كريات الدم البيضاء مرتفعة قليلا. كانت الصورة المرضية للطفيل عبارة عن زيادة في حجم كلا من الطحال والكبد وكذلك وجود الهيموسيدرين في الكبد للضأن والمعز المصابة في الذروة والمراحل المتأخرة من الوجود الطفيلي في الدم.