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Microscopic Detection on Some Bovine Haemoparasites in Al-Qadisiyah governorate

A thesis

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بسم الله الرحمن الرحيم

{ فَتَعَالَىٰ اللَّهُ الْمَلِكُ الْحَقُّ وَلا تَعْجَلْ بِالْقُرْآنِ مِنْ قَبْلِ أَنْ يُقْضَى إِلَيْكَ وَحْيُهُ وَقُلْ رَبِّ زِدْنِي عِلْمًا }

صدق الله العلي العظيم

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وزارة التعليم العالي والبحث العلمي
جامعة القادسية
كلية الطب البيطري

التحري المجهري عن طفيليات الدم
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رسالة مقدمة الى

مجلس كلية الطب البيطري بجامعة القادسية كجزء من متطلبات الحصول على درجة البكالوريوس في علوم الطب البيطري

من قبل

معتز محسن نعمة

بيداء كامل عرموط

٢٠١٧

بأشراف

م.د. يحيى اسماعيل خضير الطفيلي

١٤٣٨هـ **2017م**

Dedication

To

My mother, for he is first who established in me

the love of knowledge .

My father the pillar of strength .

Those who represent real love and brotherhood,

My Brothers

Acknowledgement

I would like to thank merciful God for enabling me to perform this work.

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Muatiz and Baydaa

Abstract

This survey is the first study in AL-Diwaniyah governorate, which conducted to investigate the occurrence of intra-erythrocytic pathogens. Amongst its Members identifies protozoan diseases (Theileriosis and Babiesiosis), Rickettsial (Anaplasmosis and Cowdriosis) and hemoplasmosis (Eperthrozonosis). Each of the diseases listed above had a variable economic impact in the responding countries.

One hundred blood samples from two different breeds of cattle were collected from various regions in AL-Diwaniyah governorate during September 2016 until March 2017. Some clinical signs were observed on the affected animals as pal mucus membrane, Emaciuation, red urine, fever and loss of appetite. Duff-quick staining blood smears determine the percentage of parasites was a total of 57 % were positive for all haemoparasites. Anaplasma and Babesia for the same group of cattle indicates that this area is endemic for these diseases but with a stable disease situation.

This survey focuses on the state of genuses and specieses of most blood parasitic diseases, and points to the wealth of information available for three protozoan diseases. The objective of this study was to detection of both *B. bovis* and *B. bigemina* in Al-Qadisiyah province by microscopically examination in naturally infected cattle. Out of 100 cattle, only 30(30%) was diagnosed by Microscopical Examination as infected by *Theileria spp* and 33%, *Babesia* spp., 9%, infected by *Anaplasma centralii* 8%, *Anaplasma marginalli* 11% , Haemobartonella 9%and Eperythrozoon(H. wenyonii**)** 4%. most of these cases were in chronic form and the highest rate (60%) of infection was among adult cow with age more than two years with no significant differences (p<001).

The study refers to there was no effects of the sex on the percentage of infection according to Microscopical Examination between males and females examined animals where both gave nearly the same results (39.9% male and 60% females) while the season revealed a considerable effect on the distribution of infection, where the hot months recorded the highest

percentage (32.14%) with a significant differences ($p<0.05$) when compared with other study months.

The present study referred to that infection with babesiosis according to age revealed that high percentage (78.12% and 62.5%) were seen in animal with equal or more than one year, whereas the lowest result in all species were recorded in animal with age under six months (≤6mons).

List of Contents

List of Figures

List of Tables

1-1- Introduction

Tick-borne diseases (TBDs) are widely distributed throughout the world especially in tropical and subtropical regions including Middle East (Jongejan and Uilenberg, 2004; Khan *et al*., 2004). It has been estimated that TBDs cause US $ 13.9 to 18.7 billion loss per annum and world's 80% cattle population are at risk of ticks and TBDs (de Castro, 1997). Anaplasmosis is the most prevalent haemo-rickettsial disease of cattle in world (Khan *et al.,* 2004; Afridi *et al.,* 2005). The prevalence of *Anaplasma marginale* has been reported to range from 4 to 75.5%, *Anaplasma centrale* 3.5 to 23%, *Babesia bigemina* 1.75 to 7.2%, *Theileria annulata* 0.7 to 24% (Khan *et al.,* 2004; Afridi *et al.*, 2005; Rajput *et al.*, 2005; Ahmad and Hashmi, 2007; Niazi *et al*., 2008; and Al-Khalifa *et al.*, 2009). The prevalence varies from region to region and various factors determine the occurrence of the TBDs including age, sex, breed, tick density, season, geographical area and management (Kjemtrup and Conrad, 2000)

It was concluded that bovine TBDs are prevalent in the southern regions, Iraq. There is a need for further investigations using modern serological and molecular techniques for the identification of the carriers of the tick-borne pathogens.

There is little information concerning differences of prevalence of TBDs based on age of animals, gender, breed, season and herd size, which may be of significance. In order to assess the presence and prevalence of various intra-erythrocytes pathogens of cattle in southern Iraq, we analyzed cattle blood samples collected for one haderd using microscopic examination,

1-2- Aims of study:

The study had been aimed to:_

- Investigate the all intra-erythrocytic parasites.

- Determined the relationship of some of epidemiological factors with the infections.

2- Literatures review

Bovine piroplasmosis, caused by *Babesia* (Piroplasmida; Babesiidae) and *Theileria* (Piroplasmida; Theileriidae) species, and bovine anaplasmosis, caused by *Anaplasma* (Rickettsiales; Anaplasmataceae) species, are tick-borne diseases (TBDs) that are common in both tropical and temperate regions. They have a considerable impact on the health and productivity of cattle and are responsible for remarkable economic losses (Wagner, *et al.* 2002).

2-1- Piroplasms

Piroplasms are a tick – transmitted parasitic protozoa divided into two genera *Theileria* and *Babesia*. They are the causative agents of Theileriosis and Babesiosis, respectively (Homer *et al*.,2000; Bishop *et al*., 2004). In general, Piroplasms are worldwide distributed diseases which affect many species of mammals including Bovine. Bovine babesiosis, Theileriosis and Anaplasmosis lead to a huge economic effect due to loss of beef production and meat of death and infected animals in addition to the costs used to tick control, disease detection, prevention and treatment.

2-1-1- Babesiosis

Babesiosis is a tick-borne, intraerythrocytic protozoan parasitic infection that causes significant morbidity and mortality in wide range of domestic and wild animals and occasionally man. It is the most important disease of cattle worldwide and transmitted by blood-sucking ticks of the Ixodidae family (hard ticks). Many *Babesia* species have been described since Victor Babes first recognized *Babesia* in the red blood cells of cattle in 1888 (Adl *et al*., 2005)

The most prevalent species, Babesia bovis and B. bigemina, are found throughout most tropical and subtropical regions, By means of the universal distribution of the ixodid tick, Babesiosis is the second most widespread blood-borne disease of animals (Hunfeld et al., 2008, Gohil et al., 2013) and, prominently, is gaining increasing interest as an emerging zoonosis of humans (Kjemtrup and Conrad, 2000; Zintl et al., 2003). The economic losses from these two

Babesia parasites vary in their virulence to the definitive host. This virulence is, in certain cases, related to the geographical origin. For example, *Babesia bigemina* from Australia induces mild clinical signs, while the South African strains are virulent, eliciting peracute and acute cases. Peracute cases exhibit sudden and severe anaemia culminating in jaundice and death, while the acute cases tend to develop high fever, haemoglobinuria with the subsequent appearance of anaemia and jaundice (Kahn and Line, 2010). *Babesia bovis* also causes a virulent form of bovine babesiosis characterised by fever, anaemia, anorexia, cachexia, low parasitaemia, generalised circulatory disturbance and often results in high mortality rates among naive cattle. Unlike *B. bigemina*, parasitised erythrocytes sequester in the microvasculature of the brain and lung resulting in cerebral babesiosis and respiratory distress (Altay, *et al.*, 2008). *Babesia divergens* causes European bovine babesiosis with clinical manifestations similar to *B. bovis*. *Babesia equi* causes a severe disease in horses characterised by fever, anaemia, icterus and haemoglobinuria. *Babesia equi*, unlike the rest of the *Babesia* species, replicates in lymphocytes to produce schizonts (Aktas, *et al.*, 2011). Thereafter, the later stages invade erythrocytes. In this respect it resembles *Theileria*.

2-1-2- Theileriosis

Theileriosis, caused by various intra erythrocytic protozoan parasites of the genus Theileria, is a tick – borne disease of domestic and wild animals. An estimated 250 million domestic cattle are at risk from the disease (Tait and Hall, 1990). Theileriosis is inducing a variety of clinical manifestations ranging from a subclinical presentation to a fatal disease depending, in part, on the animal species, host, age and the species of the microorganism. The disease is widely distributed in tropical and subtropical zones where it is known as East Coast Fever (*Theileria parva*), Corridor disease (*T. lawrencei*) and Tropical (Mediterranean) bovine theileriosis (*T. annulata*) or Mediterranean Coast Fever. East Coast Fever is of major economic importance throughout eastern, central and southern Africa where losses have been estimated at US$168 million per annum (Cossio-Bayugar, 2002). Tropical theileriosis has a wider distribution extending from north Africa to China. There are no recent figures for losses due to this disease; nevertheless it has been estimated that 200 million cattle are at risk of contracting it. The two parasites (*T. parva* and *T. annulata*) cause a lymphoproliferative disease of cattle characterised by fever, lymph node enlargement, petechial haemorrhages on mucosae and respiratory distress, and often culminates in death. In infections caused by *T. annulata* there is also an erythro-destructive stage.

The *Theileria* parasites are transmitted by ixodid ticks (*Rhipicephalus*, *Hyaloma* and *Haemophysalis*), in which a sexual cycle occurs leading to production of sporozoites infective to mammalian lymphocytes. In cattle, the sporozoites first invade the lymphocytes, develop into schizonts and then merozoites, which subsequently infect erythrocytes. In acute cases, death occurs at the macroschizont stage of infection, partly due to destruction of the lymphocytes (Gomes, *et al.,* 2013).

The Eperythrozoon were seen free in the plasma in countarary to Haemobartonella sp. which were not free in the plasma. Morphological alterations of the erythrocytes in the blood smears of the affected animals were observed. Trypanosoma congolense appeared in blood smears of cattle is monomorphic in form ranging from 8.0-10μ. (Cringoli, *et al.*, 2002).

2-2-1- Aanaplasmosis

Anaplasmosis is an arthropod-borne rickettsial disease of cattle, sheep and goats, and has a wide distribution. The disease in cattle is caused by *Anaplasma marginale* and *A. centrale.* Infection caused by *A. marginale* is characterized by severe anaemia, cachexia, abortion and death (Aktas, *et al.*, 2011), while infection with *A. centrale* induces subclinical to mild disease. *Anaplasma marginale* can be distinguished from *A. centrale* by the location and the characteristics of the inclusion bodies in the erythrocytes (CECI, *et al.*, 2014). *Anaplasma marginale* is transmitted to cattle either cyclically or mechanically by ticks of the genus *Boophilus* and *Dermacentor* and biting flies, respectively. In the tick midgut epithelial and muscular tissue, the parasite undergoes a complex developmental process involving multiplication of large reticulated forms, by binary fission, giving rise to dense bodies (26). Thereafter the parasites gain access to salivary glands where they mature into forms infective to cattle. The infection established in the gut muscle and salivary glands of the male

Dermacentor ticks have been shown to persist, rendering them reservoirs of anaplasmosis. Once the infective *Anaplasma* organisms are introduced into mammalian blood circulation, the initial bodies enter the erythrocytes by invagination of the cytoplasmic membrane with subsequent formation of a vacuole. The infective forms multiply by binary fission to produce approximately 16 initial bodies that are frequently observed during the acute phase of the disease. The number of parasitised erythrocytes diminishes as the disease becomes chronic.

Anaplasma phagocytophilum (formerly *Ehrlichia phagocytophila*) is a gram-negative bacterium, again transmitted largely by *I. ricinus* in the UK. It is the causative agent of tick-borne fever (TBF) in cattle. Infection may be characterized by fever and general immunosuppression, occasionally resulting in more severe secondary infections. However, in the UK, infection with *Anaplasma* in cattle is usually mild, the main losses being due to abortion when pregnant cows become infected.

Ahmed *et al*., (2007) clarified that the conditions of climate are determine the dynamics of tick borne diseases through affecting the take place of their seasonal occurrence.

Technique for detection of these haemoparasites has been developed separately for use in each species. The traditional method of detection of infection was making by microscopic examination of blood smears stained with Giemsa stain. This technique is usually adequate for detection of acute infection, but not for detection of carrier animals, where Parasiteamia may be low (Friedhoff and Bose, 1994).

2-2-2- Cowdriosis

Cowdriosis is a Rickettsial disease of cattle, sheep and goats caused by *Cowdria ruminantium* transmitted by the *Amblyomma* genus of ticks. Cowdriosis is endemic in sub-Saharan Africa and the Caribbean. The disease, which in some cases is peracute, is characterised by high fever, ascites, hydrothorax, hydropericardium and central nervous system signs. The development of these varies depending on the severity of the disease. Infection of wild animals such as bushbuck (*Tragelaphus scriptus*), black wildebeest (*Connochaetes gnou*) and springbok (*Antidorcas marsupialis*) by *C. ruminantium* often induces subclinical disease.

The red cell parasites formerly known as *Haemobartomlla* and *Eperythrozoon* spp have been reclassified as hemotrophic mycoplasmas (hemoplasmas) based on strong phylogenetic evidence and 16S ribosomal RNA gene sequences.

Animal babesiosis is a zoonotic disease caused by protozoan parasites of the *Babesia* genus, primarily the bovine pathogen *Babesia divergens* in Europe (Kreier, et al., 1984 and Mc Gavin, 2001) and the rodent-borne parasite *B. microti* in the United States (Montes,1995 and Smith, 1990). Parasites are transmitted by the bite of the ixodid tick when the insect takes a blood meal, and the current understanding of human babesiosis epidemiology is that *B. divergens* causes acute illness, usually in immunocompromised patients, whereas *B. microti* can also infect normosplenic immunocompetent individuals, resulting in infections that range from being asymptomatic to chronic. While many infections remain asymptomatic, especially in younger and immunocompetent individuals, the burden of severe pathology is in newborn infants and older or immunocompromised individuals, and fatality rates average 30% to 45% in these susceptible hosts (Tait, 1990). Transfusiontransmitted babesiosis is an emerging threat to public health, as

asymptomatic carriers donate blood and there are no approved or regulated tests to screen blood products for this pathogen. As a consequence, since 2011, babesiosis has been a nationally notifiable disease in 18 states in the United States (Wagner,2013).

Blood parasitic diseases have severely hindered development of livestock production in many developing countries, particularly those in sub-Saharan Africa. The bulk of these diseases are caused by vector-borne Protozoa and Rickettsia.

It is important to note that trypanosomes, Babesia and Theileria have complicated life cycles. Each developmental stage may have a set of distinctive antigens requiring a specific type of immune response. Control of protozoan diseases by the host is hindered primarily by poor immunogenicity of protozoan antigens and antigenic variation. Furthermore, immune response to one developmental stage may not confer protection to subseqsuent stage.

Rickettsial infections are a serious problem in sub-Saharan Africa and to a lesser extent in Asia and Europe. The rickettsial diseases of economic importance are anaplasmosis, cowdriosis and ehrlichiosis. These diseases have been encountered in 36 out of 59 OIE Member Countries. The immune response to these organisms involves both the humeral and cellular immune systems. Studies carried out sofar in cowdriosis suggest a major role for cellular immune response. The current state of knowledge of specific immune responses to Theileria, trypanosomes, Babesia, Anaplasma, Cowdria and Echrichia, primarily in ruminants, will be discussed

2-4- Clinical signs

Babesiosis, anaplasmosis and theileriosis are characterized by anemia, jaundice, decreased production, abortion and death (Kocan, *et al.*, 2000 Wagner, *et al*., 2001). Animals surviving infection became carriers and serve as reservoirs.

Nondescriptive cow aged around three to four years was brought with history of high fever, restlessness, dull and decreased appetite and red colored urine. On clinical examination of animals, mucous membrane was pale and animal was voiding red coloured urine which was collected in bottle. Animal was very depressed and dull. On physical examination of cow ticks were found on body of animal.

The bovine Babesiosis and Theileriosis characterized by fever (40 – 41) C^o, bloody urea (hemoglobin urea),ectric mucous membranes, muscles trembling, grinding of teeth, The feces are dry and bloody stained, dehydration causes the eye to become sunken in their sockets, falls of body temperature to subnormal level before a few hours of death , anemia , anorexia , death of untreated cases . (Wagner *et al*., 2002).

Anaemia was the major clinical sign in the affected animals, agreeing with Adejinmi et al., (2004) who reported anaemia as a reliable indicator for the severity of haemoparasitic infections. The results from the study also suggest that haemoparasitic infection may be the cause of anaemia seen in cattle reared extensively in Nigeria. Thus livestock farmers are advised to adopt the option of routine check up for their animals especially for haemoparasites to avoid problems.

Diseases caused by infection with hemoplasmas range from overt life-threatening hemolytic anemia to subtle chronic anemia, ill-thrift, and infertility. In addition, the organisms may act as cofactors in the progression of retroviral, neoplastic, and immune-mediated diseases

Blood smears were prepared from all samples to monitor microscopically the prevalence of prolozoan parasites were for babesia and theilaria, while the anaplasma were for ana. Marginale and ana. Central . The anaplasmosis showed a higher occurance in female than male

Anaplasma phagocytophilum (formerly *Ehrlichia phagocytophila*) is a gram-negative bacterium, again transmitted largely by *I. ricinus* in the UK. It is the causative agent of tick-borne fever (TBF) in cattle. Infection may be characterized by fever and general immunosuppression, occasionally resulting in more severe secondary infections. However, in the UK, infection with *Anaplasma* in cattle is usually mild, the main losses being due to abortion when pregnant cows become infected.

Haemoparasites have generally been shown to cause destruction of red blood cells resulting in anaemia, jaundice, anorexia, weight loss and infertility. Parasitic diseases have debilitating impact on human and animal health worldwide particularly in developing countries (Ellis et al., 2003).

Eperythrozoon ovis is an uncultivated, wall-less bacterium that parasitizes the surface of sheep erythrocytes. This organism is transmitted by blood-feeding arthropods and parasitizes animals in sheep-rearing regions throughout the world. Signs of infection include mild to severe haemolytic anaemia, as well as icterus; animals may exhibit poor weight gain and depression, but death is rare in adults. Infections are frequently unapparent and chronic. E. ovis also infects goats, in which it results in more severe disease (Splitter et al., 1956; Kreier & Ristic, 1963; Daddow, 1979; Mason et al., 1989; Mason & Statham, 1991). owadays Piroplasmosis is a disease with a world-wide distribution affecting many species of mammals with a major impact on cattle and human. It has increasingly been recognized throughout the world as public health problems (Schorn et al., 2011; Zanet et al., 2014). Babesia spp. and Theileria spp. are protozoan parasites transmitted mainly by Parasitism of erythrocytes often occurs at a low level and is transient; detection requires examination of repeated blood smears. However, erythrocyte parasitaemia can be as high as 100% in our experience; a high level of parasitaemia can occur even in subclinical infection and can persist for months (Overas, 1969; Brun-Hansen et al., 1997). E. ovis is completely resistant to penicillin and other antimicrobial agents that target the cell wall. Also, tetracycline treatment does not eliminate this agent from chronically infected animals. This organism has been thought to be a rickettsia because of its obligate parasitism, erythrocyte localization, small size, staining properties and transmission by arthropod vectors. E. ovis and a number of similar haemotrophic bacteria have been classified in the order Rickettsiales, family Anaplasmataceae, in the genera Haemobartonella and Eperythrozoon (Kreier & Ristic, 1984; Kreier et al., 1992). Recently, phylogenetic analysis of 16S rRNA gene sequences from four of these bacteria, Haemobartonella felis, Haemobartonella muris, Eperythrozoon suis and Eperythrozoon wenyonii, has demonstrated that these wallless bacteria are not rickettsiae, but that they are actually mycoplasmas (Neimark & Kocan, 1997; Rikihisa et al., 1997; Neimark et al., 2001, 2002b). Here, the results of a phylogenetic study of E. ovis are reported Description of Mycoplasma ovis comb. nov. (basonym Eperythrozoon ovis Neitz, Alexander and du Toit 1934)

Mycoplasma ovis (o9vis. L. fem. n. ovis a sheep). Epierythrocytic parasite of sheep and goats. Giemsa stain shows small, round bodies attached to the surface of erythrocytes. Erythrocytes can reach 100% infection. Organisms also are observed free in the blood during periods of fever in heavily parasitized animals. By using phase-contrast microscopy, M. ovis appears as a round cell. Cells seen in thin sections by EM are round or oval bodies of 0?3–0?4 mm diameter and are applied closely to the erythrocyte membrane. A 20–30 nm wide electron-dense layer partially surrounds many of the organisms. M. ovis can produce mild to severe anaemia in normal sheep. Infections are frequently unapparent. Mortality from infection is rare, but anaemia and failure to gain weight occur in lambs. M. ovis is reported to share antigens with M. wenyonii (Kreier & Ristic, 1963).

Theileriosis caused by Theileria lestoquardi (malignant ovine theileriosis) in sheep and Theileria annulata (tropical theileriosis) in cattle is an important hemoprotozoal tick-borne disease in Iran. Due to major biologic and phylogenic similarities of these two species, this study was carried out to investigate the occurrence of natural infections with T.lestoquardi and T.annulata in cattle with clinical theileriosis in Ahvaz, southwest Iran. Fifty one cattle were selected based on clinical signs of theileriosis and confirmation by microscopic examination of blood smears. Blood samples were collected from each animal and hematologic and microscopic examinations were performed. Theileria piroplasmic forms were detected in all affected cattle. Pale mucous membranes (43.14%), icterus (11.76%) and fever (70.6%) were also observed. PCR-RFLP analysis revealed T. annulata infection in all tested cattle while coinfections with T. lestoquardi were found in two samples (3.92%). All sampled cattle including the two with mixed species Theileria infection were anemic. This is the first report of Theileria species cross infections in cattle with clinical theileriosis in Iran. It can be concluded that cattle can be infected with both pathogenic Theileria species, T. lestoquardi and T. annulata which can be an important issue in the epidemiology and spread of ovine malignant theileriosis.

2-5- Transmission:

1- Mechanical Transmission:

a- Iatrogenic transmission :

Iatrogenic transmission by blood contaminated fomites has been demonstrated to occur during routine animal husbandry procedure , theblood-contaminated fomites that may spread *A. marginale*s are needles, dehorning saw, nose tongs, ear- tagging devices , castrating knives , tattoo instruments or other surgical instruments (De Wall , 2000) .

b- Biting insect transmission:

Mechanical transmission can also occur through the contaminated mouthparts of blood sucking diptera of the genera Tabanus, Stomoxy and mosquitoes (Potgierter *et al*. ,1981). Mechanical transmission of infected red blood cells must occur within a few minutes of the blood leaving the infected animal, as the blood parasite does not survive more than a few minutes outside the animal (Ewing , 1981) .This form of mechanical transmission is considered to be the major route of dissemination of *A. marginale* in areas of Central and South America and Africa where tick vectors do not occure and where *Boophilus microplus* (the tropical cattle tick) does not appear to be a biological vector of *A. marginale* (Figueroa *et al*. , 1998). In areas of the United States where geographic isolates of *A. marginale* are not infective for ticks or where ticks have been eradicated by fire ants , mechanical transmission by biting insects appears to be the major mode of *A. marginale* transmission (De la Fuente *et al*., 2001) .

2 -Transplacental Transmission: (Vertical transmission)

Vertical transmission of anaplasmosis in cattle is the transfer of disease from the dam to her gestating calf in utero , infected erythrocytes move across the placenta in uterus from infected cows to their offspring during gestation without amplification of *A. marginale* (Aubry and Geale , 2011). *Anaplasma marginale* causes abortion in acutely infected cattle (Radostits *et al*., 2000).

However, abortion is neither absolute in acutely infected cows nor in carrier cows,the relationship between utero transmission and trimester of pregnancy was described by Zaugg (1985) , six pregnant susceptible cows were inoculated with a Virginia isolate of *A. marginale* during each trimester of pregnancy and it was determined that the utero transmission occurs during the second and third trimesters of pregnancy. Potgieter and Van Rensburg (1987) reported an uterus transmission rate of 7.7 % in calves born to both acutely infected and carrier cows , therefore ; acute infection of the dam is not necessary for vertical transmission to the fetus .

3 -Biological transmission : (Horizontal transmission)

Biological transmission of *A. marginale* occurs between anaplasmosis carriers to susceptible animals , the foremost recognized mode of horizontal transmission of bovine anaplasmosis is mediated by tick vectors and approximately 20 species of ticks have been incriminated as vectors world wide (Kocan *et al*., 2004) .Tick transmission can occur from stage to stage (transstadial) or within a stage (intrastadial) , while transovarial transmission from one tick generation to the next does not appear to occur (Stich *et al*. , 1989) . Infected erythrocytes are ingested by ticks; *A. marginale* replicates within the tick's gut and salivary glands and is subsequently transmitted via tick saliva into uninfected ruminants (Ge *et al*., 1996) . Biological transmission by ticks is the most efficient means of spread for *A. marginale* because of replication and persistence capabilities within ticks (Kocan *et al*., 1992 a ,b).

Aktas *et al*., (2006) said that the ticks transmit a wide range of pathogenic agents than any other arthropod vector and these include tick-borne protozoa and tick-borne bacteria of both medical and veterinary important. Several species of ticks such as *Hyalomma* spp., *Haemaphysalis* spp., *Amblyomma* spp., and *Rhipicephalus* spp. transtadially transmit sporozoites to mammalian hosts.

Climatic conditions are play an important role in the dynamics of tick borne diseases through affecting the ticks' distribution and their seasonal occurrence (Ahmed *et al*., 2007). The increase in prevalence during hot seasons could be attributed to tick infestation rate which is influenced by temperature, rainfall and relative humidity (Gosh *et al*., 2007) .

Horak *et al*., (2002) explained that Babesiosis, caused by *B. bigemina* and *B. bovis*, is transmitted vertically by one-host ticks of the genus *Boophilus*. Although the five species of this genus have recently been placed within the genus *Rhipicephalus*.

Bock *et al*., (2004) dogmatized that the main vectors of bovine babesiosis are *Bo. microplus*, *Bo. annulatus* and *Bo. geigyi* which transmit both *B. bovis* and *B. bigemina*, and *Bo. decoloratus* which transmit only *B. bigemina*. Only *Bo. microplus* and *Bo. decoloratus* are found in Eastern, Central and Southern Africa. *Rhipicephalus evertsi* also transmits *B. bigemina*.

Climate, soil and cattle biotypes modulate the capacity of the land to support *Boophilus* tick populations and in turn, influence the babesial infection rate in cattle and ticks (Guglielmone, 1995).

2-6 Diagnosis

Microscopic examination by Giemsa stained blood smears, which is used to confirm acute anaplasmosis, can only detect levels of .106 infected erythrocytes per ml (1, 15). Subinoculation of *A. marginale*-infected erythrocytes into susceptible, splenectomized calves has been considered the “gold Based on the history, spot observation, nature of clinical symptoms and presence of ticks on body of animal, it was tentatively diagnosed as babesiois and after giving treatment to animal, on the basis of response showed by animal to treatment, it was finally diagnosed as babesiois.

2-4-1- Direct Microscopic Examination

The traditional way of determining the causative agent of infection is by microscopic examination of thin or and thick blood smears stained with Giemsa, a Romanowsky type stain (10% Giemsa in phosphate buffered saline (PBS) or Sorenson's buffer at pH 7.4). The sensitivity of thick smear can describe by its ability to detect 1 parasite among 10^6 red blood cells (RBCs) (Bose *et al*., 1995). The differentiation of species is good in thin smears when compare with the more sensitive thick smears. This method is usually sufficient in detection of acute infections, rather than in carrier's cases where the parasitaemias are mainly very low. The identification and differentiation of parasite's species can be amended by using a fluorescent dye, such as acridine orange, instead of Giemsa (Bose *et al*., 1995).

Babesia bovis is a small protozoa, located usually in erythrocyte center. It measures approximately 1-1.5µm long and is mostly present as pairs that are at an obtuse angle to each other. *Babesia divergens* is also a small parasite and very identically to *B. bovis*, but the obtuse angled pairs are mostly located at the edge of the erythrocyte. *Babesia bigemina* is pear form, but numerous varied single forms are found. It is usually a much bigger parasite (3-3.5µm long and 1-1.5 µm wide), and is mostly present at an acute angle to each other or almost parallel. In acute cases, the parasitaemia of *B. bovis* rarely arrives 1% (measured in general rather than capillary blood), but usually the parasitaemia is much higher with *B. bigemina* and *B. divergens*

2-7 Treatment

1- Berenil 5 % (Diminazene Aceturate) injection at the dose rate of 1 ml/20 kg, intramuscularly. Berenil is an antiparasitic drug for treatment and control of protozoa infection in cattle, sheep, horses and dogs. Along with berenil, oxytetracycline LA 25 ml, meloxyplus 25 ml were also given and butorphosphon plus cyanocobamin 20 ml given intramuscularly as a supplement for iron and vitamin to increase RBC formation.

Homer *et al*., (2000) referred to that the *Babesia* spp. have been difficult to eradicate from infected individuals. Kuttler, (1982) said that several treatment strategies have been employed including complete elimination of the organism or alternatively incomplete clearance leading to a state of premonition. Premunition is referred to as the immunity of infection, and is a state in which the animal remains infected, but has very few clinical signs. In many countries where re-infection is nearly impossible to prevent, the goal is to induce a state of premunition through drug treatments that reduce the severity of clinical signs, but do not result in complete sterilization of the *Babesia* infection (Kuttler, 1982; Penzhorn *et al*., 1995).

For many years three babesiacides, quinuronuim sulphate (Ludobal®), amicarbalide isothionate (Diampron®) and diminazine aceturate (Berenil®) were available in many countries for the treatment of bovine babesiosis (Vial and Gorenflot 2006). In the 1970s, a fourth drug, imidocarb dipropionate was introduced (Imizol®), and it rapidly became the drug of choice for the treatment of babesiosis (McHardy and Simpson, 1974; Vial and Gorenflot, 2006).

2- Imidocarb dipropionate is the only babesiacide that consistently clears the host of parasites when used at dosages above 1 mg/kg (McHardy and Simpson, 1974; Lewis *et al*., 1981). Treatment of *B. bigemina* infections with imidocarb can completely cure the animal from the infection but leave it susceptible to re-infection (McHardy and Simpson, 1974; Vial and Gorenflot, 2006). It can also induce complete clearance of *B. bovis*, but in some cases a second dose or a slightly increased dose may be desirable. Imidocarb is an effective chemoprophylactic that will prevent clinical infection for up to two months, but allows mild sub-clinical infection to occur as the blood drug levels decrease, resulting in an element of chemotherapeutic protection against disease while immunity develops (Kuttler, 1975; Kuttler and Johnson, 1986). Imidocarb has also been reported to affect the *Babesia* parasites in the tick vector.

3- Oxytetracycline does not have a good chemotherapeutic activity against babesiosis. However when given 1 to 2 days before exposure, the drug has been reported to lengthen the incubation period of *B. bigemina* (Kuttler and Johnson, 1986). The chemotherapeutic activity of diminazine is less than that of imidocarb although both drugs have an inhibitory effect on frozen *B. bovis* and *B. bigemina* attenuated vaccines (Kuttler and Johnson, 1986; Combrink *et al*., 2002)

2-8- Control

1. Tick control

Tick control is difficult because the ticks spend most of their life-cycle away from the host, sheltered at the base of thick damp vegetation. Attempts to reduce tick populations by environmental treatment with acaricide would be unacceptable because of effects on other invertebrates.(Radostits *et al*., 2008) explained the control of babesiosis concentrated on many approaches including, using of genetically resistant animals breed, acaricides treatment and vaccines.

Biological control to ticks by using carnivorous fungus, bacteria, virus and nematode (Samish *et al*., 2000). *Iletarhizium anisopliae* and *Beauveria bassiana* which is important fungus cause high mortality in ticks (Kaaya, 2000), Bee fiery red and house mouse which are important carnivorous to ticks control (Sutherst, 2001). In endemic areas, the tick burden can be decreased with acaricides and other methods of ticks control such as rotational grazing, the most used tick control is the use of acaricides, dipping or pour on throughout the year, mainly with flumetrin has shown to decrease the incidence of new infections in different countries (Shimizu *et al*., 2000; Jongejan and Uilenberg, 2004; Yokoyama *et al*., 2011).

Willadsen, (2004) mentioned that the genetically resistant animals, animals which show a heritable ability to become immunologically resistant to tick infestation, are a vital component of many tick control strategies. They are particularly important in the control of *Boophilus* ticks on cattle in Australia. However, this approach is not without its difficulties.

The feasibility of controlling tick infestations through immunization of hosts with selected tick antigens was first demonstrated by Allen and Humphreys, (1979). They based their trials on the crude concept that ticks feeding on an appropriately immunized host might ingest antibodies specific for antigens within the digestive tract and reproductive organs of the tick, producing deleterious effects on the feeding and reproductive behavior. By vaccinating cattle using crude antigens extracted from the midgut and reproductive organs of partially feed female *D. andersoni* ticks, they found that ticks fed on immunized cattle weighed significantly less, laid fewer eggs and very few eggs hatched compared to the control group . Since then, a number of tick antigens have been discovered in several tick species (Mulenga *et al*., 1999; Willadsen, 2004; Labuda *et al*., 2006; de la Fuente *et al*., 2007; Imamura *et al*., 2008).

Treatment with long-acting oxytetracycline following vaccination significantly reduces parasitaemia and red blood cell destruction without inhibiting the development of immunity (Pipano *et al*., 1987; Jorgensen *et al*., 1993). Usually oxytetracycline is not able to control virulent field infections.

Control of ticks by vaccination has the advantages of being cost effective, reducing environmental contamination and prevents the selection of drug-resistant ticks that results from repeated a caricide application. In addition, development of vaccines against ticks using multiple antigens that could target a broad range of tick species may prevent or reduce transmission of other pathogens (De la Fuente and Kocan, 2006 ; De la Fuente *et al*., 2006).

3- Materials and Methods

3-1-Materials:

3-1-1-The equipments, chemical and solutions that were used in study are shown in Table (3- 1).

Table (3- 1): - The equipments, chemical and solutions used in the study.

List.	Materials	Manufacture/ Origin
1	Digital Camera	Sony/Korea
2	EDTA-Vacuum blood collection tubes 10 ml	AFCO/Jordon
3	Gel vacuum blood collection tubes 5 ml	AFCO/Jordon
4	Icebox	Qmex/India
5	Light Microscope	Olympus /Japan
6	Refrigerator	Concord/Lobnan
7	Slides and cover slip	Sail brand/China
8	Stethoscope	MDF instrument/USA
9	Thermometer	Hamilton/China
10	Vacutainer Systems (needle 0.8*38mm, holder)	Belliver/industrial estate/UK
11	70% ethanol alcohol as disinfectant	Sigma-Aldrich/Germany
12	Distilled water (DW)	Prepared in laboratory

3-1-2-The Kit that was used in deferential WBCs counts is listed in Table (3- 2):

Table (3- 2): The kit of deferential WBCs counts.

List	Materials and kits	Manufacturer / origin
1	Diff Quick Stain contain	Syrbio / Switzerland
A	Fixative Solution (R1)	

B	**Eosin (R2)**	
C	**Methylene blue(R3)**	

3-1-3- Animals:

All tests done on the animals involved in this study were done under the agreements of the Ethical Committee of Veterinary Medicine. The study done in Al Diwaniyah governorate, it is the main agricultural and animal industrial province located in the Middle Euphrates in Iraq. Study animals consisted of one hundred divided 50 animals of cross breed (Holstein with native cattle) and 50 animals of local breed (native cattle), all animals were older than 6 months and randomly selected, animal's age was divided in two categories: up to two years and more than two years. The project was started on December 2016 and finished on March 2017. All cattle were examined regardless to age, sex and breed.

Table (3-3):- The animals of the study on location and their breed, age and sex.

Distribution of cattle according to breed			
Cattle breeds	**Cross breed**	**Local breed**	**Total**
numbers	**50**	**50**	**100**
Distribution of cattle according to age			
Cattle age	**≥ 2 year**	**<2 year**	**Total**
Total	**35**	**65**	**100**
Distribution of cattle according to sex			
Cattle Sex	**Male**	**Female**	**Total**
Total	**14**	**86**	**100**

3-2- Methods

I. 3-2-1- Animals Examination

II. Clinical examination of each animal was recorded clinical examination of farm animals protocol, General examinations of the cattle were taken to find heart rate, heart sounds, mucous membrane, capillaries, respiratory sounds, ruminal movements. Other signs obtained during history of animals which obtained from owners compliant, these clinical signs involved decrease milk production in dairy cattle and appetite status. In the slaughterhouse, the detected signs of any disease or health problems were described as it possible.

3-2-2- Blood Samples Collection:

Peripheral blood samples were aseptically collected. The farm owners consent was obtained before animal sampling from the jugular vein with and without anticoagulant by vacutainer system in two sterile vacuum tubes and stored in icebox until arrived to the laboratory.

3-2-3- Deferential White Blood Cells (WBCs) Count:

Hematological Examination: Fresh peripheral blood samples collected for examination of differential white blood cells count. differential leukocytes counts were done according to (Hendrix and Sirois, 2007). Differential Leukocyte counts: Blood smear was done with Wedge film blood smear and stained with Diff-Quik stains kits as following:

- A small drop of blood Placed near one end of clean glass slide.
- Other slide (spreader slide) was Placed in the narrow edge with first slide at 30 degree angle.
- The spreader slide draws back into drop to allow the blood drop to spread along of the edge of spreader slide.
- Pushed the spreader slide with raped motion forward.
- Gently waved the slide to air dried.
- The slides dripped in fixative solution for 2 sec., the allowed it to drain
- The slides dripped in R1 solution for 10 sec.,
- The slides dripped in R2 solution for 10-20 sec.

- The slides rinsed with distilled water, the allowed to dry and examined.

4. RESULTS:

4.1. Result of Microscopic examination:

Although, the random collection of blood samples, some of examined animals were showed some clinical findings, included pale mucous membrane, jaundice, enlargement of peripheral lymph nodes and emaciation. The examination of 100 blood smears of cattle in different breed, age and sex revealed the overall percentage of haemoparasitic infections were 57% (Table 4-1) depended on diff quick staining blood smear.

Table (4-1) The percentages of overall hemoparasites infections results by microscopic examination of 100 cattle samples.

Animals	Numbers	%
Infected	57	57%
Non infected	43	43%
Total	100	-

This above percentage distributed as follow, highest percentage infection for theilleria spp. was 33 %, fallow by 11 %, 8 %, 9 %, 9 %, anaplasma margenali, anaplamsa centrali, babesia spp., Haemobartonella and the lowest infectivity percentage was for Eperythrozoon (Hemoplasma wenyoni) was 4 % , (Table4-2).

Table (4-2) Shows the percentages of different intraerthrocytes pathogens in cattle.

Haemoparasites	**Numbers**	**%**
Thiellerae spp.	33	33 %,
Anaplasma marginale	11	11 %
Anaplamsa centrale	8	8 %
Babesia spp.	9	9 %
Haemobartonella	9	9 %
Eperythrozoon(H. wenyonii**)**	4	4 %
Total	57	57%

The parasite of the genus Eperythrozoon appeared in blood smear as cocci, spheres clustered on the surface of the red cells (extracellular) and are found in the intercellular space. In heavy infections, chains of the parasites surrounding the margin of the red cell (Fig 4-1).

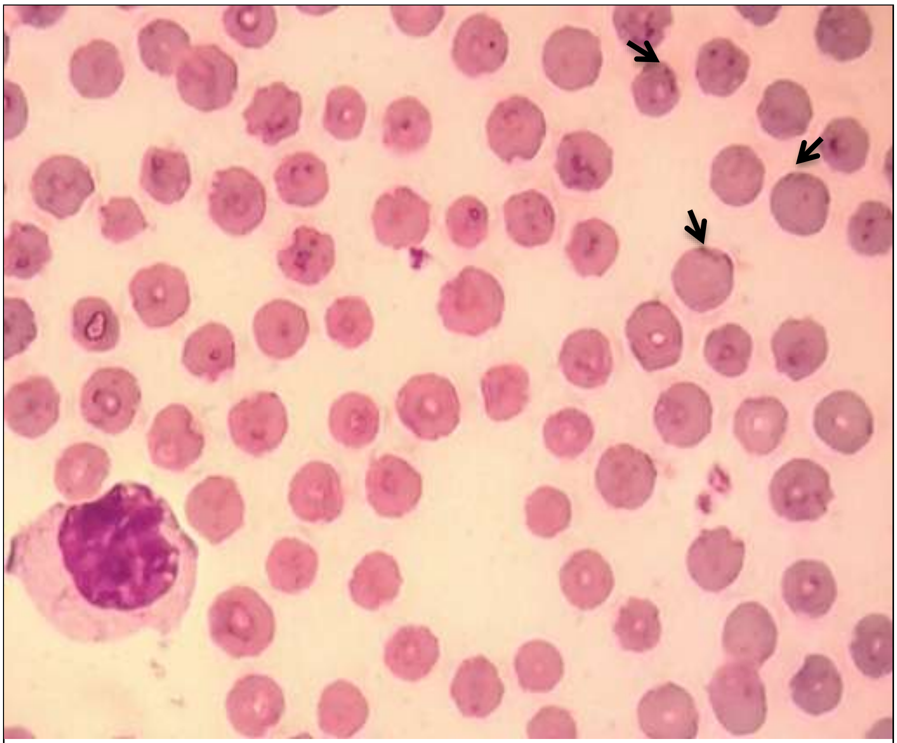
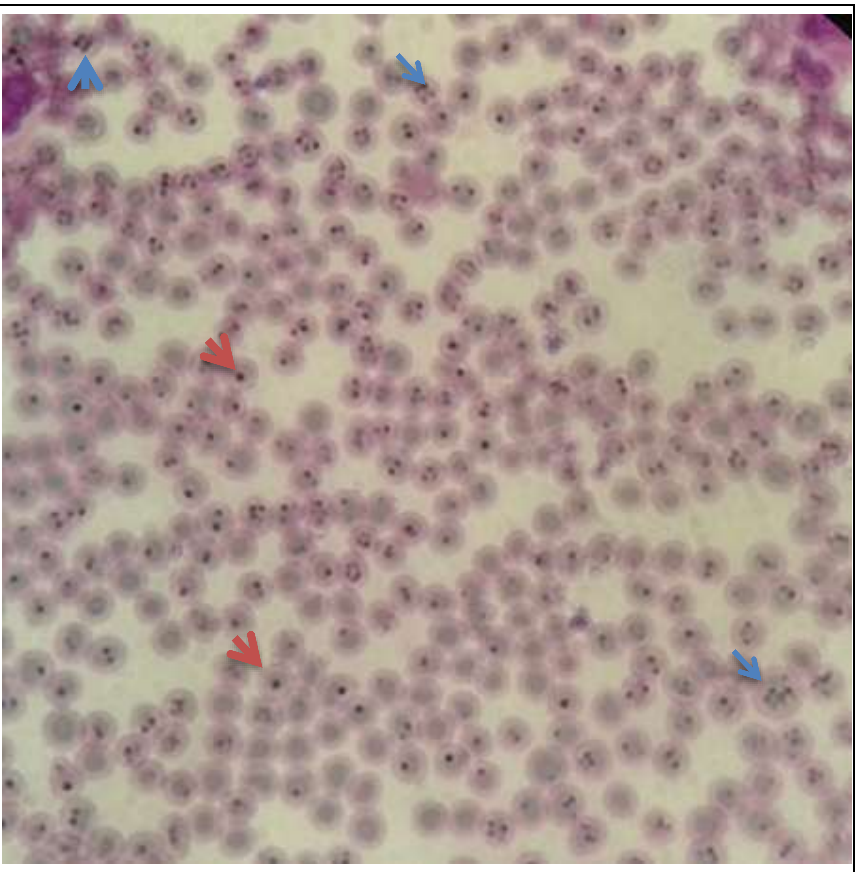


Figure 4-1: Mixed infection with: E. wenyoni and b: H.bovis in blood smears of cattle (100x).

Babesia spp. parasites appeared in two to four of as shapes intra- ethrocytes. In acute diseased animals about 75-85% of blood red cells infected (Fig. 4-2), theleiria spp. observed as irregular shape from 4 to 8 particles in red blood cells (Fig. 4-3), the genus Eperythrozoon appeared in blood smear as cocci, spheres clustered on the surface of the red cells (extracellular) and are found in the intercellular space. In heavy infections, chains of the parasites surrounding the margin of the red cell (Fig 4-1).



Figur4-2 Show the the mixed infection of babesia and anaplasosis in havey infcted blood semear.

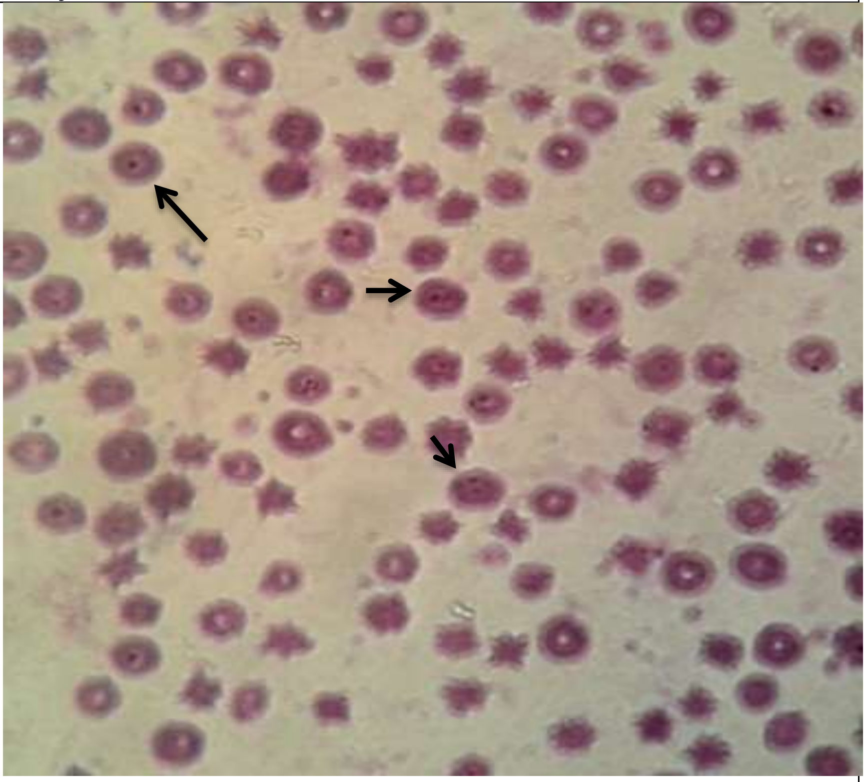


Figure 4-3: Anaplasma Centrale and Anaplasma Marginale in blood smear of cattle (100x),

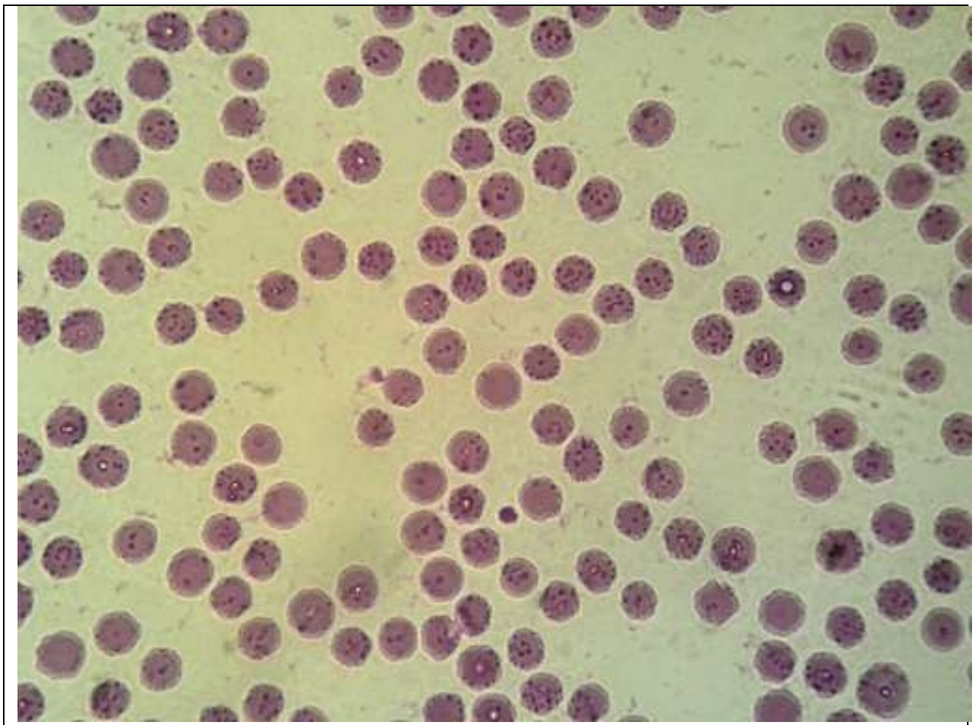


Figure 4-4: Haemobartonella in blood smears of cattle – shaped form is located around the periphery of the red cells (100x).

Hemoplasma spp. in blood smear of cattle appeared as minute bluish coloured cocci, rings, spheres clustered on the surface of RBCs and found free in plasma (Fig. 4-4). Haemobartonella bovis appeared chains of rods are usually around the periphery of RBCs (Fig. 4-5). Many authors made similar reports on these parasites

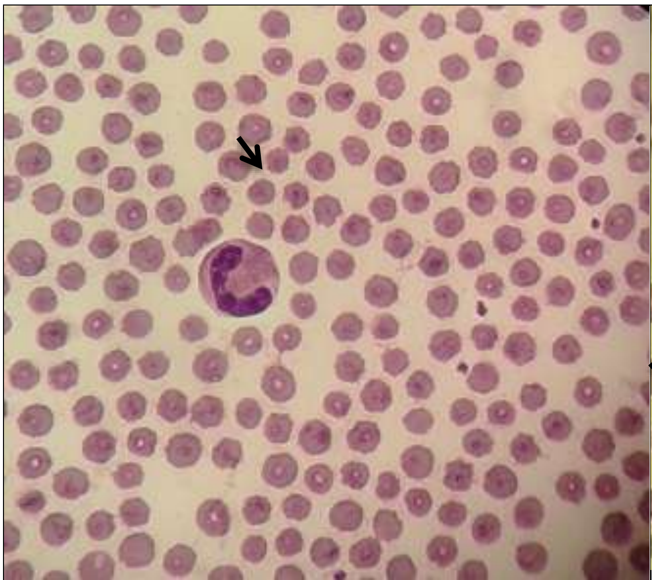


Figure (4-5) show Hemoplasma spp. as extraerthrocytic cocci adherted with RBCs

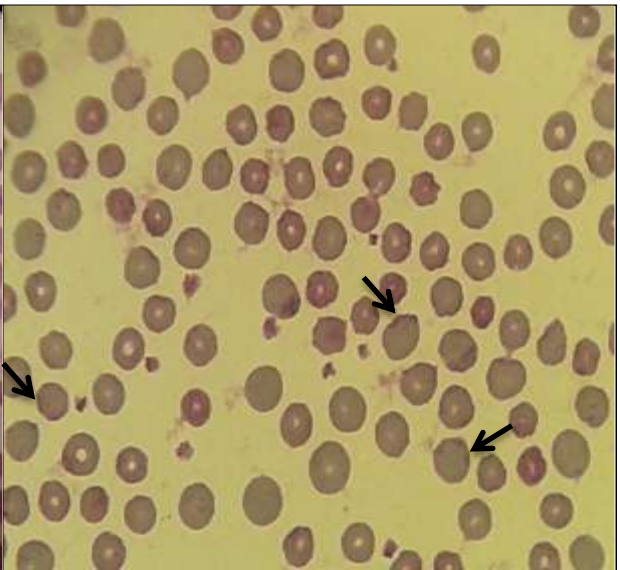


Figure (4-6) show the Haemobartonella bovis a short chain with margins of red blood cells

4-2- Breed, Age and Sex relationship:

In this survey, depending on the microscopic examination, some epidemiological elements as age, sex and breeds were investigated.

To analyze the risk related with cattle breed associated with the hemoparasites infection, two breeds (cross breed and local breed) of Iraqi cattle were infected. The highest percentage of infection were among cross breed with percentage of 39 (78%), and the local breed had lower percentage 18 (36%) (*P*= 0.01). Statically the results revealed that there were high significant differences between cross and local breeds of cows for their susceptibility to BLV infection (Table 4-3).

Table 4-3: Identification of inter-erythrocytic parasites infection regarding to breed, age and sex investigated in this study.

Cattle	Cattle Breed		
	Cross Breed	Local Breed	Total
Infected	39(78%)	18(36%)	57(57%)

The ages of cows affected by hemoparasites is summarized in Table (4-4). The results of this study revealed that there were no significant differences between ages of cattle for their susceptibility to blood parasites.

Table 4-4: the distribution of intra-erthrocytes parasitic infection regarding age of cattle in this study.

	Age		
Cattle	< 2 yr.	≥ 2 yr.	Total
Infected	39(60%)	18(51.4%)	57(57%)

The results also showed that there were no significant differences between both sex of animals for their susceptibility to hemoparasites and Table (4-5) exhibits that parasites detected in 4 (4.34%) and 53 (7.34%) samples from male and female respectively.

Table 4-5: The distribution of intra-erthrocytes parasitic infection regarding sex investigated in this study.

Cattle	Sex		
	Male	Female	Total
Infected	6(39.9%)	51(60%)	57(57%)

5- Discussion:

This study revealed an overall haemoparasite infection rate of 57% in cattle in Al-Qadisiyah Farmers in Iraq, due to their subclinical nature of presentation or due to their chronic nature on the affected animals, leading to reduction in production and eventual death of the affected animals. It is noteworthy that most of the cattle studied were located in southern part of the country.

In this study the diagnosis of theilerea spp. and babesia spp. in blood smears of cattle was made in Al-qadisiyah city, the rate of occurrence for bovine hemoparasitic diseases were studied in Iraqi cattle during 6 months. Theileriosis showed a higher peak of occurrence during the period of samples collection was . while the mean occurrence of babesiosis was 8.5, and this result could have a twofold because the high predominance of B. bigemina in the region, since this hemoparasite shows greater infectivity for bovidae.

Therefore this is the first report on the parasites in Al-qadisiyah, Iraq. Previous study by (Hasan, 2012) in Mosul in Iraq, and (Cem, 2007) reported E. wenyoni in Turkey in cattle. And E.wenyoni was also reported in cattle of Saudi Arabia (Al-khalifa, *at al.,* 2008 and Al-Khalifa, *et al.,* 2009). The results of this study showed that the clinical sings observed in infected animals were in agreement with the results of other studies carried out in other countries (Cem, *et al.,* 2009 Love, et al., 2003 and Adejinmi, *et al.,* 2004). These parasites cause anaemia, fever, anorexia, weight loss and decreased milk production and infertility (Smith, *et al.,* 1990 – Montes, *et al.,* 1994). Most of the tested animals were subclinical infected (Montes, *et al.,* 1994). In this study the percentages of Eperythrozoon in cattle and sheep were 28.33%, 40.0% respectively. Whereas Al-khalifa *et al*. (2009) recorded 1-4% of infection with E.wenyoni in cattle and 2-9% of infection with E.ovis in Saudi Arabia. The mixed infection with Eperythtozoon and Haemobartonella were diagnosed in blood smears of cattle, which were also described by (Love and Hutchinson, 2003, Messick, 2004) In Australia Eperythrozoon infection was reported in 10% of weaner and 51% of adult sheep (Nicholls, 1986). Eperythrozoon and Haemobartonella 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 tick. (Zintl, *et al.,* 2003 and Kahn, 2010) referred that the Eperythrozoon and Haemobortonella are transmitted mechanically by arthropods such as Fleas and Mosquitos transmission the Eperythrozoon in sheep and goats, Ticks (Rhipicephalus) are vectors of Eperythrozoon for cattle, also transmission may occur via surgical procedures through blood contaimination of instruments (needle contamination during vaccination).

E.ovis appeared as minute bluish coloured cocci, rings, spheres clustered on the surface of RBCs and found free in plasma. Haemobartonella bovis appeared chains of rods are usually around the periphery of RBCs. Many authors made similar reports on these parasites (Adam, *et al.,* and Coles, *et al.,* 1983). The percentages of parasitaemia of E.bovis and E.ovis were 1.81%, 2.29% respectively. Cem Ecmel SAKi (2009) mentioned that the parasites numbers of E.wenyoni ranging from 1-50, in highlydense parasitaemia, While (Coles, *et al.,* 1983) attributed to the anaemia. Parasitaemia develops during the pyretic stage at which time parasites can be found in great numbers on the red cells and in the plasma (Jain, *et al.*, 1983). In case of Eperythrozoon infection, the mechanism of disease appears similar to that occurs of other blood parasites, where in inmmunemediated hemolysis within macrophages follows attachment of the organisms to erythrocytes (Mc Gavin, 2001) In this study, during the examination of 60 blood smears of cattle, Trypanosoma was diagnosed in 2 animals with percentage of 3.33%. These parasite appear in blood smear as a small size about 8-10µ in length, with blunt posterior end, and no free flagellum therefore determined the species as T.congolense dependened on (Soulsby, 1982 and Adam, 1971). Probably introduced the affected animals with T.congolense from different regions into the Mosul city and because the transmission of T.congolense by Glossina and as well as they can also be transmitted mechanically by other biting flies such as Tabanas and Stomoxys. Authors (Rodistitis, *et al.,* 2007) mentioned that after trypanosomes have been introduce into a herd transmission is possible even in the absence of Glossina-Biting flies such as Tabanidas, Stomoxyinae and Hipoboscidae are capable of mechanically transmitting trypanosomes. Mechanical transmission can through the needle during inoculations and in carnivores infected carcasses. Samdi etal., (2011) recorded that 50% in cattle in kaduna central abattoir, North central Nigeria were infected with T. congolense. whereas ohaeri (Ohaeri, 2010) recorded the percentage of infectin in cattle with T.congolense of 42% in Nigeria. However these parasites are more predominant in small ruminants. The variation in results in different studies may be because the variation in strain of parasite, the vector, breed and management of animals as well as the season of the year during the study. Some of the factors that affect the prevalence of trypanosomiasis in Nigeria include: animal breed, type of management season, of the year and the type of vegetation (Maclaennnan, 1983).

45% incidence of blood parasites in cattle and buffaloes, respectively. The higher rate of prevalence of Anaplasma marginale as compared to other blood parasites recorded in the present study is also in agreement with the findings of Burriro et al. (1994). It could be concluded from the present study that our local animals harbour a sub-clinical infection of blood parasites. Further studies are needed on a broader scale so as to determine the ill effects of such sub-clinical infections, if any.

The mere appearance of a band of expected size does not always correlate with the presence of the virus genome. Hence confirmation of the PCR positivity needs to be performed by one of the several methods available such as nested PCR, southern blot, hybridization and sequencing. Further re amplification with the second set of internal primers served to verify the specificity of the first round product. With nested PCR, the transfer of reaction products from the first reaction effectively (Parthiban, et al. 2010).

Seasonal fluctuations of both tick species in the three localities were very similar but their burdens in cattle in Qwa-Qwa were significantly highest compared to the other two localities. A first possible reason for higher tick infestations in Qwa-Qwa was that the body condition of cattle in this locality was always poorer compared to that in the other two localities. This can result in a lowered tick resistance resulting in higher tick burdens (De Castro & Newson 1993). A possible reason for the poor body conditions could have been the severe overgrazing of the veld in this specific locality. Farmers in this area appeared unmotivated and unwilling to herd their cattle to better grazing further away. A second possible reason for the higher tick burdens could have been the higher stocking densities, resulting in more contact with hosts (Hlatshwayo 2000). Another possible reason for higher tick burdens in Qwa-Qwa is that farmers in this area do not control ticks or tick-borne diseases. A possible reason these farmers not to control ticks is that most of them are resource-poor farmers, and they do not have the necessary resources and knowledge to control ticks (Hlatshwayo 2000).

The high prevalence of haemoparasites observed in this study is similar to the work of Oduye & Dipeolu (1976) that reported a prevalence of about 50% infections in 800 dogs sampled in Ibadan, Nigeria. It also agreed with the result of Dipeolu *et al.,* (1982) who observed 81% prevalence of haemoparasitic infections in local pigs at Ibadan, Nigeria. Other similar findings include 50.2% prevalence of blood protozoan (Talabi *et al.,* 2009) in the Trans boundary areas of Ogun State, Nigeria and 54%prevalence in sheep at Abeokuta, Ogun State (Takeet *et al.,* 2009). The high incidence of haemoparasites recorded could be as a result of favorable environmental condition that helps in the survival and proliferation of the arthropod vectors responsible for the transmission of these parasites (Adejinmi *et al.,* 2004)

All the animals were sero-negative for B. bovis and this is probably because the tick vector (Boophilus microplus) which transmits the disease is not present in the Free State Province. Two tick species belonging to the family Ixodidae were found on cattle, namely Boophilus decoloratus and Rhipicephalus evertsi evertsi. In the present study significant differences in seasonal burdens of B. decoloratus occurred, with the highest infestations recorded from February to June. The presence of R. evertsi evertsi throughout the year without any or with small fluctuations in winter months was observed, with a peak from February to May

6-1- Conclusions:

1. There are high pervalences of intra-erthrocytic parasites in southern region of Iraq.\
2. Perioplasmosis, Recktiosis, hemoplamoisis and Haemobartenlosis are present in the RBCs of cattle.
3. Theileriosis was higher percentage of infection.
4. There are no predilection of Breed, sex, and age for infections.

6-2- Recommendations:

1- There is a need for further investigations using modern serological and molecular techniques for the identification of the carriers of the tick-borne pathogens.

2- Investigation of relationships of infections with poor condition scours and loses of productivity.

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