بسم الله الرحمن الرحيم

Sudan University of Science and Technology

College of Graduate Studies

Prevalence and Risk factors of Major ticks Borne Disease in Bovine in Khartoum State – Sudan

A thesis Submitted to the Collage of Graduate Studies, Sudan University of Science and Technology in partial fulfillment for the requirements for the Degree of Master of Preventive Veterinary Medicine (MPVM)

Candidate

SHaza alyasmin Adam Ibrahim Nemiry

BVSC. (2013), Faculty of Veterinary Medicine, Sudan University of Science & Technology

Supervisor

Professor: Galal Eldeen ELazhri Mohammed

Department of Medicine and,Sudan University of Science & Technology, Faculty of Veterinary Medicine

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DEDICATION

I dedicate this work to my soul late father, Mr. Adam Ibrahim Nemiry for his insistence to see me attain higher education and achieve better things. My mother

for urging me to be self dependent and be a family asset. My elder brother, Mr. Mohommed ELmoatz Adam Ibrahim Nemiry , for the encouragements and helps ,my sister ,and brothers they were giving me to push on to higher heights and be a better person and and my children Heba and Mozaa and my future sons

. Finally God almighty for good health.

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I am sincerely thankful to my supervisors, prof .Galal ELdin ELzahri Mohammed for their generosity and willingness in sparing time for supervision. I also acknowledge him for the corrections they made to the dissertation so that it could reach acceptable standards. I am greatly humbled for Dr.Ehab Mohuomed Nuor for their helps and guidance during the dissertation write up and Iam grateful to Dr Mohamed Tag ELdeen for their help me critical and analytical approach during the data analysis . I am most grateful to Dr. Tag ELseer Elnour for his helps in laboratory diagnosis in soba center veterinary lab I wish to thank the prof Abd ELwahab elketaibi ,prof .Mohoumed abd elslam for their efforts. The farmers are also acknowledged for allowing me access to their animals Last but not the least, my sincere thanks to Prof. Abd ELhamed ELfadel who have been or acted as Heads of preventive medicine and public health department during this period He was very supportive and encouraged me to soldier on and for helping me through the proposal formulation.

Iwish to express my special sincere thanks to Mr. Nag ELdeen ELgid for providing assistance which enabled me complete the whole research work with fewer problems last but not the lest I would like to convey my special thanks to my family

Abstract

A cross-sectional study was conducted in Khartoum State to estimate the prevalence of the major Ticks Borne Diseases and identified associated risk factors ,case history(age ,sex ,body condition ,dehydration, facial condition),body temperature ,packed cell value . This study was done between August to December 2015 in cattle farms in 3 districts of Khartoum State ,(Khartoum ,Omdurman and Bahri) blood were collected from cattle randomly selected from sampling farms and at the same time data such temperature and case history .

The collected blood were screened for TBDs using microscopic examination after blood smear stained with giemsa stain preparation .The risk factor identification was done using a pre-tested questionnaire which was administered simultaneously with the blood sampling. The factors assessed included types of cattle breeds ,sex ,age ,body condition ,dehydration ,faces ,vector problems ,vector control and lymph node enlargement related to Theileriosis. Prevalence estimates, including the 95% confidence interval, were determined for each localities as a whole. The Fisher's exact test was used to test for associations between diseases positivity on the farm and the hypothesized risk factor and student sample t-test was done to determined the effect of Temperature ,PCV and Hb among sex, ROC graf draw to estimate sensitivity of PCV to detected anemia .

A total of 100 blood samples from Khartoum (n=40) Bahri (n=30) and Omdurman(n=30) were screened. The estimated prevalence in Khartoum State was 34, 38% and 20% (95% CI) for Theileriosis ,Anaplasmoisis and Bebesiosis respectively ,*Anaplasma* spp considered highest prevalence in the Khartoum state followed it *Theileria* spp and *Babesia* spp, The prevalence's according to localities, Khartoum district had high prevalence in Theileria and Anaplasma compared to other localities

A univariate analysis of risk factors showed that the method of acquiring a farm was the most important risk factor. However, other factor such as significant at 75% confidence level. The study therefore highlights the need for effective control measures to be put in place to reduce the observed TBDS prevalence's

الخلاصة

أجريت دراسة استطلاعية بولاية الخرطوم لحساب نسبة الإصابة بالأمراض المنقولة بواسطة القراد والتعرف على عوامل الخطر المتعلقة بحدوث المرض (العمر ، الجنس ، حالة الجسم ، الجفاف ، درجة حرارة جسم الحيوان ، حجم الدم الكلي) .

أجريت الدراسة في الفترة من أغسطس إلى ديسمبر 2015 م بمزارع تربية الماشية بمدن ولاية الخرطوم الثلاث (أم درمان ، الخرطوم ، بحري) .

جمعت عينات دم بطريقة عشوائية من الأبقار حيث تم مسحها لمعرفة الأمراض لمعرفة الأمراض المنقولة بواسطة القراد باستخدام الفحص ألمجهري للعينات المصبوغة بصبغة " حمسا " .

عوامل الخطر تم التقصي عنها بواسطة استبيان أجرى بالتزامن مع أخذ العينات تقييم عوامل الخطر وهي تتضمن الآتي : (نوع سلالة الأبقار ، الجنس ، العمر ، حالة الجسم ، التجفاف ، الروث ، مشاكل الناقل والتحكم في الناقل وتورم الغدد الليمفاوية المتعلقة بمرض التاليريا .

تم حساب نسبة الإصابة بمرض التاليريا بعامل 95% كحد ثقة لكل مدينة على حدا ، كما تم استخدام اختبار " فيشر " لمعرفة مدى العلاقة التي تربط بين ايجابية الإصابة بالمرض لحيوانات المزرعة وعوامل الخطر الافتراضية .

اختبار " T " أجري لتحديد تأثير درجة الحرارة و الحجم الكلي للدم وقياس الهيموقلوبين على جنس الحيوان . كما رسم منحنى "ROC " لحساب حساسية الحجم الكلي للدم لكشف فقر الدم .

أجمالي عينات الدم التي جمعت من ولاية الخرطوم عددها 100 عينة كانت كالآتي : 40 و 30 30 جمعت في مدينة الخرطوم ، الخرطوم بحري و أم درمان على التوالي .

معدل الأصابة تم حسابه بواقع 34% ، 38% ، 20% ، لكل من داء التاليريا والأنابلازما والبابيزيا على التوالي . سجلت أعلى إصابة وكانت بداء الانابلازما وتليها الإصابة بمرض التاليريا والبابيزيا . مدينة الخرطوم سجلت أعلى نسبة إصابة بدائي التاليريا والأنابلازما مقارنة بباقي المدن .

أوضحت عوامل التحليل الأحادي لعوامل الخطر أو العامل المكتسب هو الأكثر أهمية لحدوث المرض من باقي العوامل .

كذلك الدراسة أظهرت مدى الحوجة لتطبيق أنظمة التحكم الفعال لتقليل نسبة الإصابة بالأمراض المنقولة بواسطة القراد .

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CHAPTER ONE

Introduction

Back ground:-

Tick-borne diseases (TBDs) are widespread in the Sudan (FAO 1983b). Most important TBDs of cattle in Northern Sudan are tropical Theileriosis (*Theileria annulata* infection of cattle), Babesiosis (*Babesia bigemina* and *Babesia bovis* infections), and Anaplasmosis (*Anaplasma marginale* and *Anaplasma centrale* infections), beside the nonpathogenic species *Theileria mutans (*El hussein *et al*, 2004).

Tick-borne diseases (TBDs) are widely distributed throughout the world especially in tropical and subtropical regions including sadun (Jongejan , Uilenberg, 2004; Khan *et al*, 2004). It has been estimated that TBDs cause US $ 13.9 to 18.7 billion loss per annum in world 80% cattle population are at risk of ticks and TBDs (De Castro, 1997).

Theileriasis is a protozoan infection of wild and domestic Bovidae which occurs Throughout much of the world. The Theileria spps. are transmitted by Ixodid ticks and have complex life cycles in both vertebrate host and vector, with sexual reproduction occurring in the tick , Theileriosis is an economically important protozoan disease of cattle in tropical and subtropical regions (Robinson, 1982). *T. annulata* is transmitted by ticks of the genus Hyalomma after cyclical development in these ticks (Uilenberg, 1981).

The main effects of these parasites are reflected in reduction of production, loss of weight and death of substantial proportion of the affected animals. One of the ways in which livestock production can be increased is by reduction of losses due to tick-borne diseases(Hoogstraal , 1956),(Karrar *at el*, 1963), (Osmane *at el* ,1982), ticks infesting livestock in the Sudan are

mainly *Hyalomma anatolicum anatolicum*, *Hyalomma dromedarii*, *Hyalomma* marginatum rufipes, *Hyalomma impressum*,. Although different species of ticks and the diseases they transmit occur in different ecological regions of the world, their impact on animal production is similar in nature and importance (FAO, 1984). Despite considerable progress made in combating ticks and tick-borne diseases, the latter seem to be increase in both in prevalence and severity. Tick-borne diseases are responsible for hundreds of millions US dollar losses per year in tropical and temperate areas where they pose a problem. In the Sudan no accurate economic evaluations have been made. However, economic losses in Khartoum State alone due to tropical Theileriosis have been estimated to be between 4 and 6 million US$ (Latif ,1994). *Theileria annulata* is a protozoan parasite, which causes tropical Theileriosis, a disease transmitted by several tick species of the genus Hyalomma (Dolan, 1989). It is endemic in the area around the Mediterranean, the Middle East and reaches the southern parts of Asia (Brown, 1990).

The Theileria are protozoan parasites infecting wild and domestic animals throughout much of the world. They have a schizogonous reproductive cycle, usually in lymphocytes of the vertebrate host, and a piroplasm stage in erythrocytes Transmission, as far as is known, occurs via ticks of the family Ixodidae and there is increasing evidence of a sexual cycle in the arthropod They are classified in the phylum Apicomplexa, class Sporozoea, subclass Piroplasmea in the order Piroplasmida along with another important genus infecting domestic animals, Babesia and theileria The speciation of Theileria is a maze of synonyms, homonyms and subspecies which will be resolved only when immunological, biochemical and molecular biological techniques are developed to complement morphological, host range and vector differences in discriminating between species. The most recent attempt to unravel the Theileria was made by (Uilenberg, 2005).

Babesiosis is caused by intraerythrocytic protozoan parasites of the genus *Babesia*. Transmitted by ticks, babesiosis affects a wide range of domestic and wild animals and occasionally people. Although the major economic impact of babesiosis is on the cattle industry, infections in other domestic animals, including horses, sheep, goats, pigs, and dogs.Two

important species in cattle—*B. bigemina* and *B. bovis*—are widespread in tropical and subtropical areas.(The merck veterinary manual).

Anaplasmosis, formerly known as gall sickness, traditionally refers to a disease of ruminants caused by obligate intraerythrocytic bacteria of the order Rickettsiales, family Anaplasma, genus *Anaplasma*. Cattle, sheep, goats, buffalo, and some wild ruminants can be infected with the erythrocytic *Anaplasma*. Anaplasmosis occurs in tropical and subtropical regions worldwide(The Merck Veterinary Manual).

the most common vectors of *A. marginale* has not been identified. *B. microplus* ticks are known to be efficient vectors of *A. marginale* in many parts of the world (Khan *et al*, 1994).

After feeding on an infected animal, intrastadial or trans-stadial transmission may occur. Transovarial transmission may also occur, although this is rare, even in the single-host *Rhipicephalus spp*. A replicative cycle occurs in the infected tick. Mechanical transmission via biting dipterans occurs in some regions. Transplacental transmission has been reported and is usually associated with acute infection of the dam in the second or third trimester of gestation (The merck veterinary manual).

blood parameters is helpful in assessing the health status of animals. Common diseases in the tropics may lead to anaemia, include tick-burden and tick-borne infections such as Theileriosis ,Babesiosis and Anaplasmosis. Measurement of anemia is said to give a reliable indication of the disease status and production infected animals (Nwoha & Anene, 2011). Laboratory diagnosis of anemia is based on the hemoglobin (Hb) concentration, the number of red blood cells and the haematocrit or packed cell volume (PCV) values (Aiello, 1998). Anemia is most simply and reliably estimated by measuring PCV percent using the haematocrit method, while determining the Hb concentration gives accurate information on the type of anaemia (Murray *et al*, 1983), (Quinto *et al*, 2006) .

General Objectives:-

To estimate the prevalence of bovine TBDs in Khartoum state ,Sudan

Specific objective:-

1\ To estimate the prevalence of bovine TBDs (Theileriosis ,Babesiaosis and Anaplasmosis) in Khartoum State.

2\ To Investigate the risk factors association with the disease.

3\ To know the relation ship between TBDs and Anemia in different localities.

CHAPTER TOW

Literature views

2-1-Etioiogy:-

Theileriases are a group of tick borne diseases caused by Theileria spp. A large number of Theileria spp are found in domestic and wild animals in tick-infested areas of the Old World. The most important species affecting cattle are *T. parva* and *T. annulata*, which cause widespread death in tropical and subtropical areas of the Old World. *T .lestoquardi, T. lowenshuni*, and *T. uilenbergi* are important causes of mortality in sheep(The Merck Veterinary Manual2015).

2-2- Classification of the causative agent:-

Genus Theileria, Family Theileriidae, Order Piroplasmida, Subclass Piroplasmia, Phylum Apicomplexa.(Oie, 2009).

2-3 Taxonomy of Theileria (Levine, 1988):-

Domain	Eukaryota
kingdom	Chromalveolata
Sub kingdom	Protozoa;single cell eukaryotes
Phylum	Apicomplexa;apical complex present at least in some stages;reproduce sexually by syngamy
Class	Sporozoea;sporogonic stage producing sporozoites
Sub class	Piroplasma;piroform,rodshaped or amdoebiod;parasite inerythrocytes and some other cells
Order	Piroplasmida;asexual and sexual reproduction;ticks are vectors.
Family	Theileriidae;schizonts stages in lymphocytes.
Genus	Theileria;piroplasm stage in erythrocytes lacks pigments

2-4-Mammals species aftected by Theileria species:-

2-4-1 Tropical Theileriosis :-

Theileria annulata, the causal agent of tropical theileriosis, is widely distributed in north Africa, the Mediterranean coastal area, the Middle East, India, the former USSR, and Asia. It is transmitted by several species of ticks of the genus Hyalomma. *T. annulata* can cause mortality of up to 90%, but strains vary in their pathogenicity, anemia is often a feature of the disease. Characteristic signs include fever and swollen superficial lymph nodes. If the disease progresses, cattle rapidly lose condition. The schizonts and piroplasms are morphologically similar to those of *T. parva*. Animals that recover from infection are immune to subsequent challenge. (The Merck Veterinary Manual).

2-5-Epidemiolody of Bovine Theileriosis:-

2-5-1-Hosts:-

T. annulata infects cattle and yak (*Bos grunniens*), with milder infections usually seen in water buffalo; the water buffalo is considered to be the natural host in which the parasite evolved. The taurine breeds of cattle, introduced into endemic areas, have a much more severe form of the disease than do indigenous zebu cattle (OIE 2009).

2-5-2-Transmission:-

T. annulata are spread by ticks, its transmitted by ticks of the genus Hyalomma ,Ticks can remain infected on the pasture for up to 2 years depending on the climatic conditions, Disease is not maintained in the absence of these field vectors, Theileria sporozoites are transmitted to susceptible animals in the saliva of the feeding tick

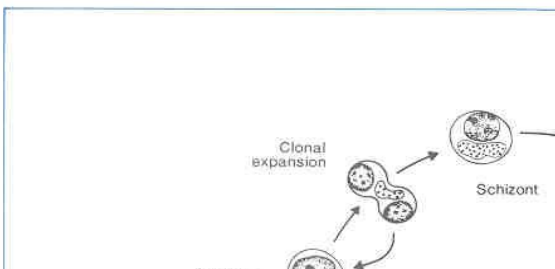
Ordinarily, *T. Annulata* only mature and enter the saliva after the tick attaches to a host; usually, a tick must be attached for 48–72 hours before it becomes infective; however, if environmental temperatures are high, infective sporozoites can develop in ticks on the ground and may enter the host within hours of attachment ,Transovarial transmission does not occur with

in *T. Annulata*. Inside the host, Theileria sporozoites undergo a complex life cycle involving the replication of schizonts in leukocytes and piroplasms in erythrocytes ,Cattle that recover from Theileria infections usually become carriers(Oie 2009).

2-6- Theileriosis life cycle stages:-

The complex life cycle of Theileria is illustrated schematically in Fig. 1. The two main stages in the tick - gametogony and sporogony ,we shall be primarily concerned with the stages in the bovine host. The bovine host is infected by the inoculation of sporozoites by infected ticks during feeding and the sporozoites invade leukocytes, probably within 5-60 minutes. After entry into the host cell, the sporozoite develops into the trophozoite which after nuclear division develops into the macroschizont, inducing the host cells to become large lymphoblastoid cells which divide in synchrony with the macroschizont. As a result of the parasite-induced lymphoproliferation, a large population of parasitised cells develops in the infected animal. These further develop into microschizonts and ultimately merozoites which are released from the lymphocyte. The merozoites invade erythrocytes and develop into piroplasms, this stage completing the life cycle within the bovine host. Although the lymphoproliferation has pathogenic consequences for the host, the anaemia induced by the destruction of the high levels of infected erythrocytes is probably the major pathogenic change in the infected animal .

A priori, a protective immune response could arise to either the extracellular stages (sporozoite or merozoite) or to antigens exposed on the surface of infected host cells, (macroschizont or piroplasm stages). Protective immunity clearly occurs as cattle, after recovery from primary infection, are immune to homologous challenge and often to challenge with heterogonous stocks of the parasite. Such immunity can be induced naturally as a result of sporozoite-induced infections, by artificial inoculation using ground-up infected tick stabilities or by infection with attenuated culture-derived macroschizont-infected leukocytes. The role of the immune response to sporozoites in protection has not been specifically evaluated; the ability to protect using macroschizont-infected cell lines implies that immune responses to the sporozoite in natural immunity may be limited, to improved immunity if sporozoite immunization was



undertaken in addition to schizont-infected cell line immunizatic

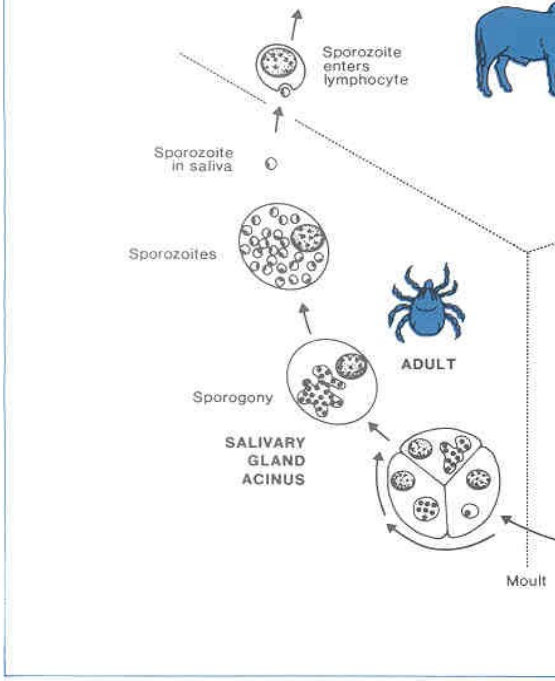


Figure 1: The life cycle of *Theileria annulata*. Sporozoite forms of the parasite are transmitted from the tick to the bovine host in tick saliva as an infected tick feeds on an animal. In the animal host organism complete its complex life cycle and development in to adult piroplasm in side RBCs .(OIE, 2008).

2-7- Economic losses due to Theileriosis:-

Theileriosis causes economic losses to individual farmers and governments by lowering cattle production and through costs incurred for controlling the disease and providing research, training and extension services pertaining to the disease .such economic losses vary widely within and among countries, due to differences in livestock production systems, cattle type, level of disease risk, disease control policies and programmers, and cost, and price structures. there is little information available on the effect of theileriosis caused by *T.annulata* on live stock production and its control.(The epidemiology of theileriosis in Africa,1987).

2-7-1-Production losses:-

2-7-1-1-Direct production losses:-

Direct production losses are those that are directly attributable to the presence of the disease in the cattle population through morbidity and mortality. Cattle which become severely infected usually die unless they are treated. morbidity and mortality in certain breeds of susceptible cattle may approach 100% (Cunningham, 1977;hooke,1981).mortality is particularly high in dourine and zebu or sanga cattle developed to improve the productivity of the (Morzararia, *et al*. 1988).indigenous breeds are also at risk in situations where they are subjected to intensive ticks control ,or when they are move from disease free to endemic areas ,case fertility rate can very from zero to 50% under endemically stable conditions(Staak,1981;Mollelal,1984,1986;Ngulo,1985; Ngulube *et al*,1985, Berkvens *et al*.,1989;Otim,1989)and to as high as 80%to100%under endemically unstable condition or

epidemic condition(Juiia,1985)although mortality in indigenous cattle in endemic areas can below, calf growths often severely impaired (Moll *et al*,1984), animal recover from Theileriosis may suffer from weight loss, produce low milk yields, provide less draught power and may experience reduced fertility and delays in reaching maturity (Lawrence,1981;Brown1985).

2-7-2-Indirect Production losses:-

Rresult from disease actS as constraint on live stock production and improvement. In the affected areas, farmers face a substantial risk if they try to keep dourine or crossbreed cattle due to their high susceptible to the disease .many farmers are there fore on strained or prohibited from improving livestock productivity and efficiency(Callow,1983).

2-8- Pathogenesis of bovine theileriosis:-

T. annulata sporozoites are injected into cattle by infected vector ticks. An occult phase of 5–10 days follows before infected lymphocytes can be detected in Giemsa-stained smears of cells aspirated from the local draining lymph node. Subsequently, the number of parasitized cells increases rapidly throughout the lymphoid system, and from about day 14 onward, cells undergoing merogony are observed. This is associated with widespread lymphocytolysis, marked lymphoid depletion, and leukopenia. Piroplasms in RBCs infected by the resultant merozoites assume various forms, but typically they are small and rod-shaped or oval.(The Merck Veterinary Manual2015).

2-9- Geographical Distribution of Theileriasis:-

According to Purnell (1978) the disease is prevalent throughout tropical and subtropical zones, North Africa, South wards down to Sudan and Eriterria and east wards up to South East Europe, the Near East and across the Indian subcontinent to China and the far east Fig2 *T.annulata* has not been reported in East Africa and the southern limit of its distribution is the Sudan and this limit does not overlap with that of *T.parva* and *T. lawrenci* (Neitz, 1957). Distribution of *T.annulata* has been shown to coincide with that of it is vector

Hyalomma.d.detritum in the Mediterranean basin and *H.a.anatolicum* in Central Western Asia, Egypt and Northern Sudan (Fischer and Say, 1989- Yassir Ali, 1998).

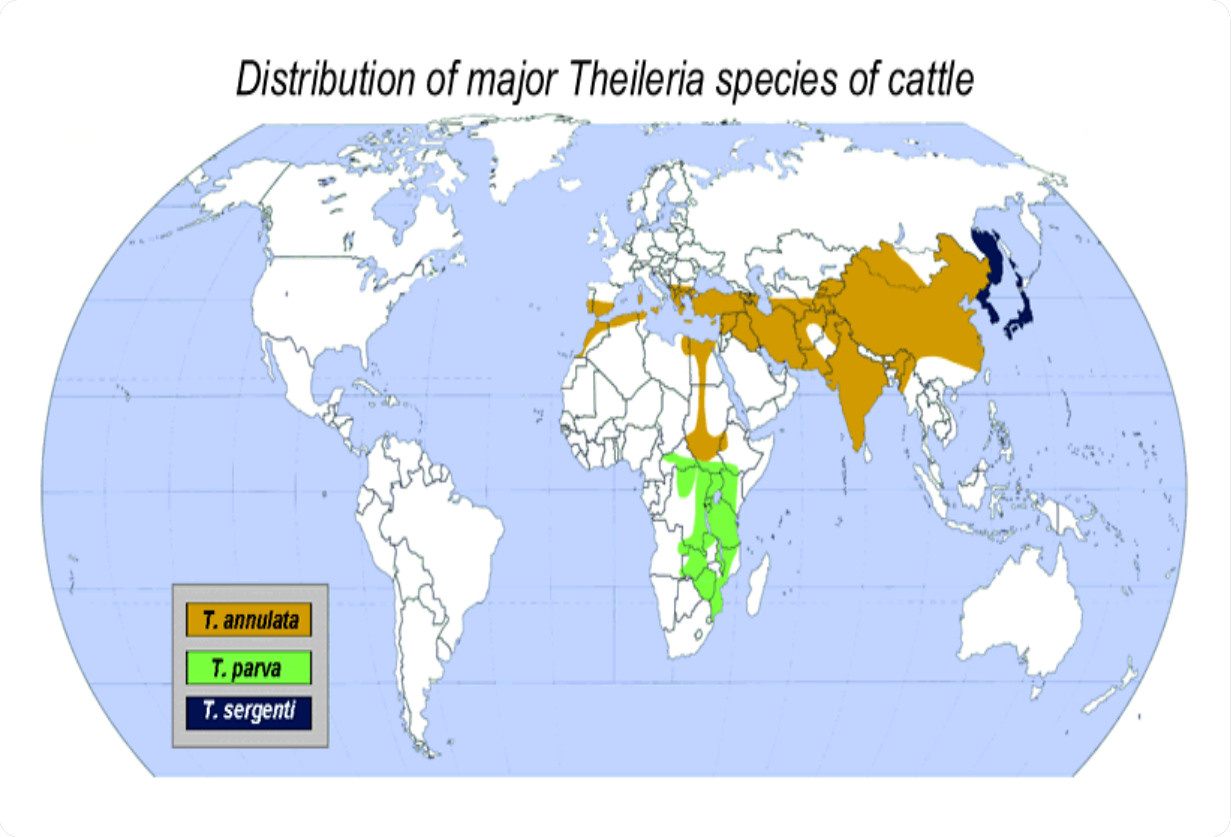


Figure 2 : Distribution of Theileria spp through the World (The Center for Food Security and Public Health.2008)**.**

2-10-Diagnosis of Theileriosis:-

2-10-1- General clinical examination:-

The clinical examination was conducted following standard method of Blood *et al*, (1985). Physical condition, posture, gait, abnormal behavior were examine as well. Other important parameters such as percussion of the lung, respiration rate, pulse rate, body temperature were measured.

2-10-2- Clinical findings of bovine Theileriosis:-

Clinical signs vary according to the level of challenge, and they range from inapparent or mild to severe and fatal. Typically, fever occurs 7–10 days after parasites are introduced by feeding ticks, continues throughout the course of infection, and may be >106°F (41°C). Lymph node swelling becomes pronounced and generalized. Lymphoblasts in Giemsa-stained smears of needle aspirates from lymph nodes contain multinuclear schizonts. Anorexia develops, and the animal rapidly loses condition; lacrimation and nasal discharge may occur. Terminally, dyspnea is common. Just before death, a sharp decrease in body temperature is usual, and pulmonary exudate pours from the nostrils. Death usually occurs 18–24 days after infection. The most striking postmortem lesions are lymph node enlargement and massive pulmonary edema and hyperemia. Hemorrhages are common on the serusal and mucosal surfaces of many organs, sometimes together with obvious areas of necrosis in the lymph nodes and thymus. Anemia is not a major diagnostic sign (as it is in babesiosis) because there is minimal division of the parasites in RBCs, and thus no massive destruction of them. (The Merck Veterinary Manual).

Animals that recover are immune to subsequent challenge with the same strains but may be susceptible to some heterologous strains. Most recovered or immunized animals remain carriers of the infection(The Merck Veterinary Manual2015).

2-10-3-Labrotary diagnosis:-

The simplest diagnostic test available is the Giemsa- stained blood or tissue smears in order to detect either macroschizonts or piroplasms within an infected animal. As sick animals usually have relatively high parasitaemias, such tests are effective and have the added advantage of detecting active infections. In addition, indirect immunofluorescence antibody tests have been used, based on those

developed for *T. parva* using either piroplasms or cultured schizonts as antigen. In this context, antigens derived from schizont cell cultures are suitable, as schizonts are relatively abundant in infections and thus infected animals produce high titre antibodies to this stage. The use of a serological test, however, suffers from the disadvantage that it is not possible to distinguish between animals with an active infection and those which are recovering or immune whin the availability of monoclonal antibodies to both the schizont and the piroplasm stage ,it would be possible to develop ELISA assays aimed at detecting circulating antigens or immune complexes and thus to detect active infections. Similarly, with the availability of cloned parasite genes ,it would be possible to develop sensitive methods based on DNA hybridisation to detect parasites within samples of blood or tissues. At the present time, the need to develop such sensitive tests for active infection has not been apparent. However, if low parasitaemias needed to be detected, such methods could have the sensitivity that would make them superior tothemethods currently used. In this context the extra-chromosomal element described (Hall *et al* ,1988). might be very useful as it is present in all life-cycle stages, all strains of *T. annulata* examined and in multiple copies per genome. This latter property would increase the sensitivity of any test using DNA hybridisation with a cloned fragment of the element.(The epidemiology theileriosis in Africa,1992)**.**

2-10-4- Histophathology for Theileria annulata in calf:-

Histopathological lesions The nodules described in the various tissues were found to be formed by round cells, similar to lymphoid neoplastic cells with moderate pleomorphism .In the lymph nodes and spleen, moderate infiltration by these cells was accompanied by hypoplasia of the lymphoid tissue. Some lymph nodes also showed heavy diffuse lymphocytic infiltration with no clear cortico-medullary demarcation(figure 5). In the skin, the infiltration rarely reached the superficial dermis and did not infiltrate the epidermis, being mostly limited to the middle and deep dermis. The neoplastic-like cells had a cloudy cell membrane, a scarce and slightly basophilic cytoplasm, a high nucleus/ cytoplasm ratio, occasionally indented nuclei, chromatin orming granules near the nuclear membrane, and signs of cariorrexis ,The cells also displayed moderate anisocaryosis and had a high mitotic index, with 2 to 3 mitoses per field (×400). Occasionally, macroschizonts of *T. annulata* detected by Giemsa stain were identified in the cytoplasm of these cells (Sandra Branco, *et al .1987*).

Petechial and ecchymotic hemorrhages are often found on the serosal surfaces of internal organs(figure 7) and the body cavities may contain serous fluid. In acutely infected animals, the lymph nodes are usually

enlarged and may be edematous and hemorrhagic(figure6). In chronic cases, they may be shrunken. The spleen is typically enlarged. The liver may also be larger than normal, and white foci of lymphoid infiltration (pseudoinfarcts) may be present in the liver and kidney(figure6),**(The Center for Food Security and Public Health.2008).**

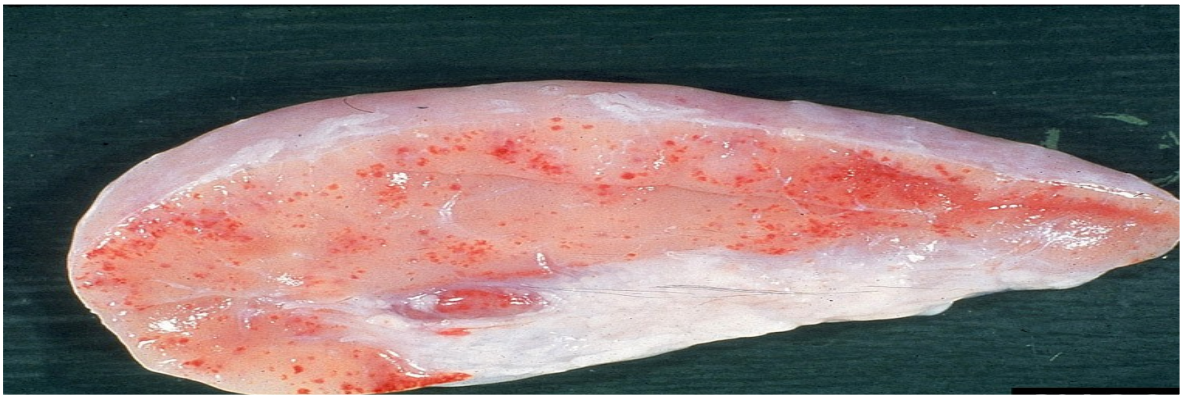


Figure 3 Bovine popliteal lymph node. The node is enlarged and diffusely pale, and contains numerous petechiae The Center for Food Security and Public Health.2008).

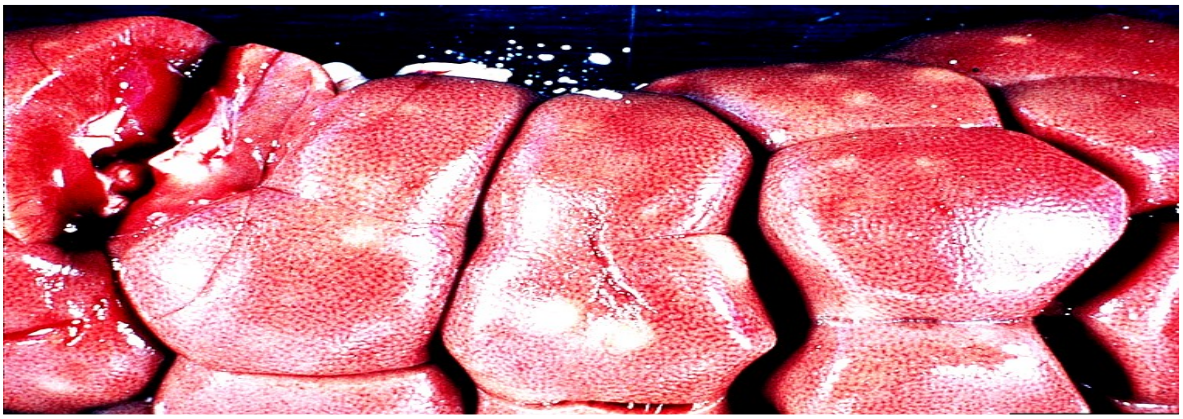


Figure 4 Bovine, kidney. The multiple pale foci on the cortical surface are lymphoid infiltrates. The Center for Food Security and Public Health.2008).

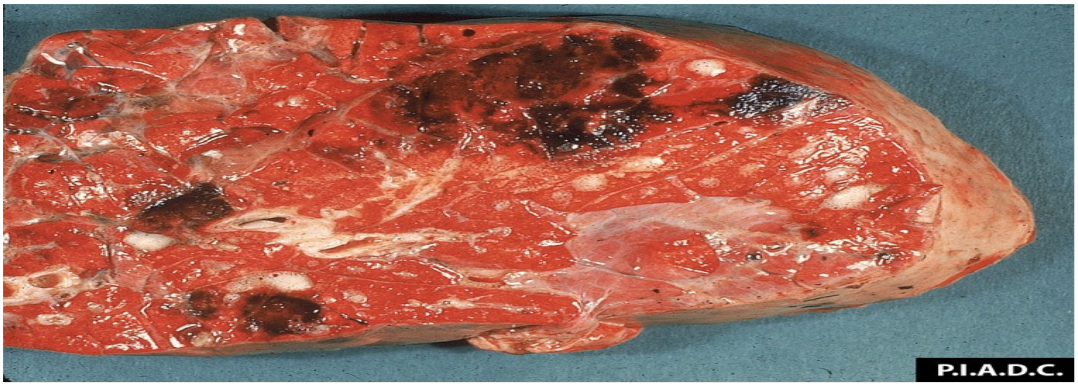


Figure 5 : Bovine, lung. Lung tissue is noncollapsed, contains multiple foci of hemorrhage, and there is fluid/foam within interlobular septa and bronchi. The Center for Food Security and Public Health.2008).

2-11-current control measure:-

The main control measures available for tropical theileriosis are acaricides (applied by dipping), chemotherapy with or without low levels of sporozoite-induced infection and vaccination by attenuated cell line vaccines. The latter control measure is by far the most widely used against T. annulata; successful cell line vaccines have been developed in Israel (Pipano. 1989.), Iran (Hashemi-fesharki r.1988), India (Singh d,k,1990) and the (Ussr –Stepanova ,*et al* 1989). Although acaricides have been applied successfully against *T. parva* they have not been used extensively for the control of tropical theileriosis. There are a number of logistical and theoretical problems associated with their use:

A.a highly organised dipping programme needs to be set up to ensure that all cattle are treated

B. long-term application can lead to acaricide resistance in ticks

C. If tick density is significantly reduced and the cattle become free of *T. annulata*, then over time these animals will become highly susceptible to the disease as a result of waning immunity due to the reduction in challenge. Thus, if infected ticks migrate into a treated area after cessation of acaricide treatment, many animals will become infected with a high level of mortality. The major advantage of acaricide-based control methods is that simultaneously all tick-borne diseases are controlled, thus reducing the need for different measures for each disease. Chemotherapy too has been used only sparingly for the treatment of *T. annulata* infections although the efficacy of drugs such as parvaquone and halofuginone -.(Dolan *et al* 1981) , (Schein .1981). is established and these drugs have been used successfully in the treatment of *T. parva* infections. However, (Hashemi-Fesharki r,1988) reports that in Iran, treated cases can still die from acute disease and this has been reported by others (Uilenberg , 1986). Despite reports (Uilenberg g,1986 and Singh d,1986). of the effectiveness of chemotherapy, for many developing countries the cost of chemotherapy is considered to be too high in relation to animal health budgets and the system of livestock rearing. The most widespread control measure used against *T. annulata* infection has been the use of attenuated macroschizont cell lines as vaccines. It has been clearly established that the vaccination of animals with such cell lines leads to the establishment of infection and the transfer of the parasite from the donor cells to those of the recipient (Brown ,1981 and Brown C,*et al* 1987). No reports are available to show that such vaccines 'break down' against either heterogonous challenge (Gill *et al*,1980) or from a decreased immunity with time. The vaccine lines can be cryopreserved (Pipano ,1981).(Singh ,1990) for storage and transport, although once thawed the vaccine has a limited shelf-life unless further culture is initiated. The ability to cryreserve the vaccine circumvents many of the problems of transport, although this is a potential limitation in some parts of the world. The limitations and potential problems of such cell line vaccines are primarily theoretical. However, they should be borne in mind as the use of the vaccine becomes widespread. The testing and long culture period needed to produce a vaccine is a limitation, as well as the possibility that indefinite culture may lead to the loss of the ability of the macroschizonts to transfer to the recipient cells, thus reducing the ability to immunise. Markers for attenuation and the definition of attenuation at a molecular level could speed production of cell line vaccines. A further potential problem is that vaccination on a large scale

may lead to the infection of cattle which therefore would allow transmission to continue. This is probably not a problem where fully attenuated cell lines are used as these do not produce the infected erythrocyte stage (Pipano ,1989) and so transmission cannot occur. However, if a virulent strains of the parasite are used for immunization (Ouhelli *et al*,1898), the infected erythrocyte stage occurs after immunization and vector transmission can occur. Furthermore, although cattle are protected by the attenuated cell line vaccine, it does not prevent the appearance of the erythrocyte stages when animals are challenged. If the parasite is maintained in this way, the potential for reversion to virulence or for antigenic variation to occur is present and, if either or both occurred, the consequences would be severe. In addition, one must also consider the possibility of transferring other diseases by the use of such vaccines, although this could be eliminated by appropriate screening if available). Clearly the issues raised here need to be investigated in order to obtain further information and the improvement of the cell line vaccines However, none of these considerations should detract from their undoubted value.

2-12-Transmission and Epidemiology of bovine Babesiosis :-

The main vectors of *B bigemina* and *B bovis* are 1-host *Rhipicephalus (Boophilus)* spp ticks, in which transmission occurs transovarially. Although the parasites can be readily transmitted experimentally by blood inoculation, mechanical transmission by insects or during surgical procedures has no practical significance. Intrauterine infection has also been reported but is rare.

In *Rhipicephalus* spp ticks, the blood stages of the parasite are ingested during engorgement and undergo sexual and asexual multiplication in the replete female, infecting eggs and subsequent parasitic stages. Transmission to the host occurs when larvae (in the case of *B bovis*) or nymphs and adults (in the case of *B bigemina*) feed. The percentage of larvae infected can vary from 0–50% or higher, depending mainly on the level of parasitemia of the host at the time the female ticks engorge. Under field conditions, the rate of tick transmission is generally higher for *B bigemina* than for *B bovis*.

In endemic areas, three features are important in determining the risk of clinical disease: 1) calves have a degree of immunity (related both to colostral-derived antibodies and to age-specific factors) that persists for ~6 mo, 2) animals that recover from *Babesia* infections are generally immune for their commercial life (4 yr), and 3) the susceptibility of cattle breeds to ticks and *Babesia* infections varies; eg, *Bos indicus* cattle tend to be more resistant to ticks and the effects of *B bovis* and *B bigemina* infection than *Bos taurus*–derived breeds. At high levels of tick transmission, virtually all calves become infected with *Babesia* by 6 mo of age, show few if any clinical signs, and subsequently become immune. This situation can be upset by either a natural (eg, climatic) or artificial (eg, acaricide treatment or changing breed composition of herd) reduction in tick numbers to levels such that tick transmission of *Babesia* to calves is insufficient to ensure all are infected during this critical early period. Other circumstances that can lead to clinical outbreaks include the introduction of susceptible cattle to endemic areas and the incursion of *Babesia*-infected ticks into previously tick-free areas. Strain variation in immunity has been demonstrated but is probably not of practical significance in the field.(The Merck Veterinary Manual, 2015) .

2-12-1-Clinical Findings and Pathogenesis :-

B bovis is a much more virulent organism than *B bigemina*. With most strains of *B bigemina*, the pathogenic effects relate more directly to erythrocyte destruction. With virulent strains of *B bovis*, a hypotensive shock syndrome, combined with generalized nonspecific inflammation, coagulation disturbances, and erythrocytic stasis in capillaries, contribute to the pathogenesis.

The acute disease generally runs a course of ~1 wk. The first sign is fever (frequently ≥106°F [41°C]), which persists throughout, and is accompanied later by inappetence, increased respiratory rate, muscle tremors, anemia, jaundice, and weight loss; hemoglobinemia and hemoglobinuria occur in the final stages. CNS involvement due to adhesion of parasitized erythrocytes in brain capillaries can occur with *B bovis* infections.

Either constipation or diarrhea may be present. Late-term pregnant cows may abort, and temporary infertility due to transient fever may be seen in bulls.

Animals that recover from the acute disease remain infected for a number of years with *B bovis* and for a few months in the case of *B bigemina*. No clinical signs are apparent during this carrier state. .(The Merck Veterinary Manual, 2015) .

2-12-2- Lesions:-

Lesions (particularly with *B bovis*) include an enlarged and friable spleen; a swollen liver with an enlarged gallbladder containing thick granular bile; congested, dark-colored kidneys; and generalized anemia and jaundice. Most clinical cases of *B bigemina* have hemoglobinuria, but this is not invariably the case with *B bovis*. Other organs, including the brain and heart, may show congestion or petechiae. .(The Merck Veterinary Manual, 2015) .

2-12-3-Diagnosis :-

Clinically, babesiosis can be confused with other conditions that cause fever, anemia, hemolysis, jaundice, or red urine. Therefore, confirmation of diagnosis by microscopic examination of Giemsa-stained blood or organ smears is essential. From the live animal, thick and thin blood smears should be prepared, preferably from capillaries in the ear or tail tip.

Smears of heart muscle, kidney, liver, lung, brain, and from a blood vessel in an extremity (eg, lower leg) should be taken at necropsy.

Microscopically, the species of *Babesia* involved can be determined morphologically, but expertise is required, especially in *B bovis* infections in which few organisms are present. *B bovis* is small, with the parasites in paired form at an obtuse angle to each other and measuring ~1–1.5 × 0.5–1 µm. *B bigemina* is larger (3–3.5 × 1–1.5 µm), with paired parasites at an acute angle to each other. Single forms of both parasites are also commonly

seen. .(The Merck Veterinary Manual, 2015) .

2-13- Anaplasmosis :-

Anaplasmosis, formerly known as gall sickness, traditionally refers to a disease of ruminants caused by obligate intraerythrocytic bacteria of the order Rickettsiales, family Anaplasmataceae, genus *Anaplasma* Cattle, sheep, goats, buffalo, and some wild ruminants can be infected with the erythrocytic *Anaplasma*. Anaplasmosis occurs in tropical and subtropical regions worldwide (~40°N to 32°S), including South and Central America, the USA, southern Europe, Africa, Asia, and Australia. .(The Merck Veterinary Manual, 2015) .

2-13-1-Lesion :-

Lesions are typical of those found in animals with anemia due to erythrophagocytosis. The carcasses of cattle that die from anaplasmosis are generally markedly anemic and jaundiced. Blood is thin and watery. The spleen is characteristically enlarged and soft, with prominent follicles. The liver may be mottled and yellow-orange. The gallbladder is often distended and contains thick brown or green bile. Hepatic and mediastinal lymph nodes appear brown. There are serous effusions in body cavities, pulmonary edema, petechial hemorrhages in the epi- and endocardium, and often evidence of severe GI stasis. Widespread phagocytosis of erythrocytes is evident on microscopic examination of the reticuloendothelial organs. A significant proportion of erythrocytes are usually found to be parasitized after death due to acute infection. .(The Merck Veterinary Manual, 2015) .

2-13-2-Diagnosis :-

Microscopic examination of Giemsa-stained thin and thick blood films is critical to distinguish anaplasmosis from babesiosis and other conditions that result in anemia and

jaundice. Blood in anticoagulant should also be obtained for hematologic testing. In Giemsa-stained thin blood films, *Anaplasma* spp appear as dense, homogeneously staining blue-purple inclusions 0.3–1.0 μm in diameter. *A marginale* inclusions are usually located toward the margin of the infected erythrocyte, whereas *A centrale* inclusion bodies are located more centrally. (The Merck Veterinary Manual, 2015) .

2-13-3-Treatment, Control, and Prevention :-

Tetracycline antibiotics and imidocarb are currently used for treatment. Cattle may be sterilized by treatment with these drugs and remain immune to severe anaplasmosis subsequently for at least 8 mo.

Prompt administration of tetracycline drugs (tetracycline, chlortetracycline, oxytetracycline, rolitetracycline, doxycycline, minocycline) in the early stages of acute disease (eg, PCV >15%) usually ensures survival. A commonly used treatment consists of a single IM injection of long-acting oxytetracycline at a dosage of 20 mg/kg. Blood transfusion to partially restore the PCV greatly improves the survival rate of more severely affected cattle. The carrier state may be eliminated by administration of a long-acting oxytetracycline preparation (20 mg/kg, IM, at least two injections with a 1-wk interval). Withholding periods for tetracyclines apply in most countries. Injection into the neck muscle rather than the rump is preferred.

Imidocarb is also highly efficacious against *A marginale* as a single injection (as the dihydrochloride salt at 1.5 mg/kg, SC, or as imidocarb dipropionate at 3 mg/kg). Elimination of the carrier state requires the use of higher repeated doses of imidocarb (eg, 5 mg/kg, IM or SC, two injections of the dihydrochloride salt 2 wk apart). Imidocarb is a suspected carcinogen with long withholding periods and is not approved for use in the USA or Europe.

In some areas, sustained stringent control or elimination of the arthropod vectors may be a

viable control strategy; however, in other areas immunization is recommended. .(The Merck Veterinary Manual, 2015).

2-14-Prevalance of Theilerisos from Prevoious Studies:-

In Omdurman.(Safieldin *et al*,2011) ,a study was conducted in Al-rodwan project in Omdurman to investigate the prevalence blood parasite in dairy cattle during different season .Atonal of 290 animals were examined during three season: dry cool (100),dry hot(95),and wet hot(95).this result showed that the prevalence of blood parasites during different seasons was8,5.25 and 6.32%for dry cool, dry hot and wet hot season, respectively. The prevalence of Theileria species infection was found to be 7, 5.25 and 6.32% for dry cool, dry hot and wet hot season, respectively. While the prevalence of Babesia species infection was only recorded in the dry cool season as (1%). There was no effect (χ 2= 0.6, p> 0.05) of season on the occurrence of blood parasites. Strong association (t-test= –43.6, p< 0.05) was found between presence of blood parasites and milk yield.

In Irag.(Afkar *et al*,2012), the application of thick blood smear technique based on Giemsa-stain confirmed the presence of high rate of ticks infection 58.3% is revealed to endemic area of Theileriosis and Babesiosis,in Iraq. Abdomen area (gut and ovary) of hard ticks*: Hyalomma a. anatolicum* revealed high rate of infection with Theileria sp.43% and Babesia sp. 15.2% . Babesia was recorded for the first time in Iraq from ticks. Females appeared positive smears more than males that total rate of infection (39) 54.1%. Distribution of infection rate in ticks was discussed with two periods of collection from May to October 2009. We concluded from this study that *Hyalomma a. anatolicum* tick at least one of Theileria and Babesia sp. Infective for cattle in Iraq; and this technique to be useful in identifying the species of protozoa in potential tick vectors .

In Kayseri - Turkey Anıl İÇA *et al*, (2006) a study was carried out to detect and compare the prevalence of bovine Theileria and Babesia species in the vicinity of Kayseri by microscopic examinations (ME) and reverse line blotting (RLB). A total of 337 cattle usually grazed on pasture in 13 different regions of Kayseri were sampled randomly. Blood samples were collected into tubes containing EDTA from jugular veins. Thin blood smears were prepared from ear capillaries. On microscopic examination of smears, 51 (15.1%) were positive for piroplasms. In the RLB assay, 61 (18.1%) were positive for *T. annulata* and 3 (0.9%) for *T. buffeli* /orientalis. Two (0.6%) of the animals were infected with *B. bigemina* and also had a concurrent infection of *T. annulata*. No animals were positive for *B. bovis or B. divergens*. The differences between me and RLB results were statistically significant ($P < 0.05$)**.**

CHAPTER THREE

Materials and methods

3-1-study area:-

The study was conducted in Khartoum State. The State lies between longitude 31.5 -34o and latitude 15-16o in an area about 28.165 square kilometres. It is bordered to the north and the east side by the River Nile State, to North Western by the Northern State, and to the east and south-eastern and south by Kassala, Gedarif and Gezira state respectively and to west by North Kordofan . Most of the Khartoum State lies in the climatic semi-desert region, while northern areas lie in desert zones. The climate of the state is ranging from hot to very hot. The weather is rainy in summers, cold and dry in winters. Average rainfall reaches 100-200 mm in the north-eastern areas and 200-300 mm in the North Western areas. Temperature ranges in summer between 25-40 degrees in the months from April to June, and 20-35 in the months from July to October. In winter, however, temperatures continue to decline between November to March from 25-15oC. Geographically, Khartoum State is divided into three regions:

A / First region: This is a Khartoum region which starts from the Mugran, i.e. the confluence of the two rivers (the blue and white Niles). Being confined between them, this block extends southwards to the boundaries of the Gezira state. Administratively, it is divided into two localities, Khartoum and Gabal Owlia localities.

B/ Second region: This is Bahri region which lies east to the Blue Nile and the River Nile. It includes the localities of Khartoum Bahri and east Nile

C / Third region : This is Um durman region which is the one located west of the White Nile and the River Nile and includes three localities, which are: umm durman, Um Badda and Karary localities.2005).

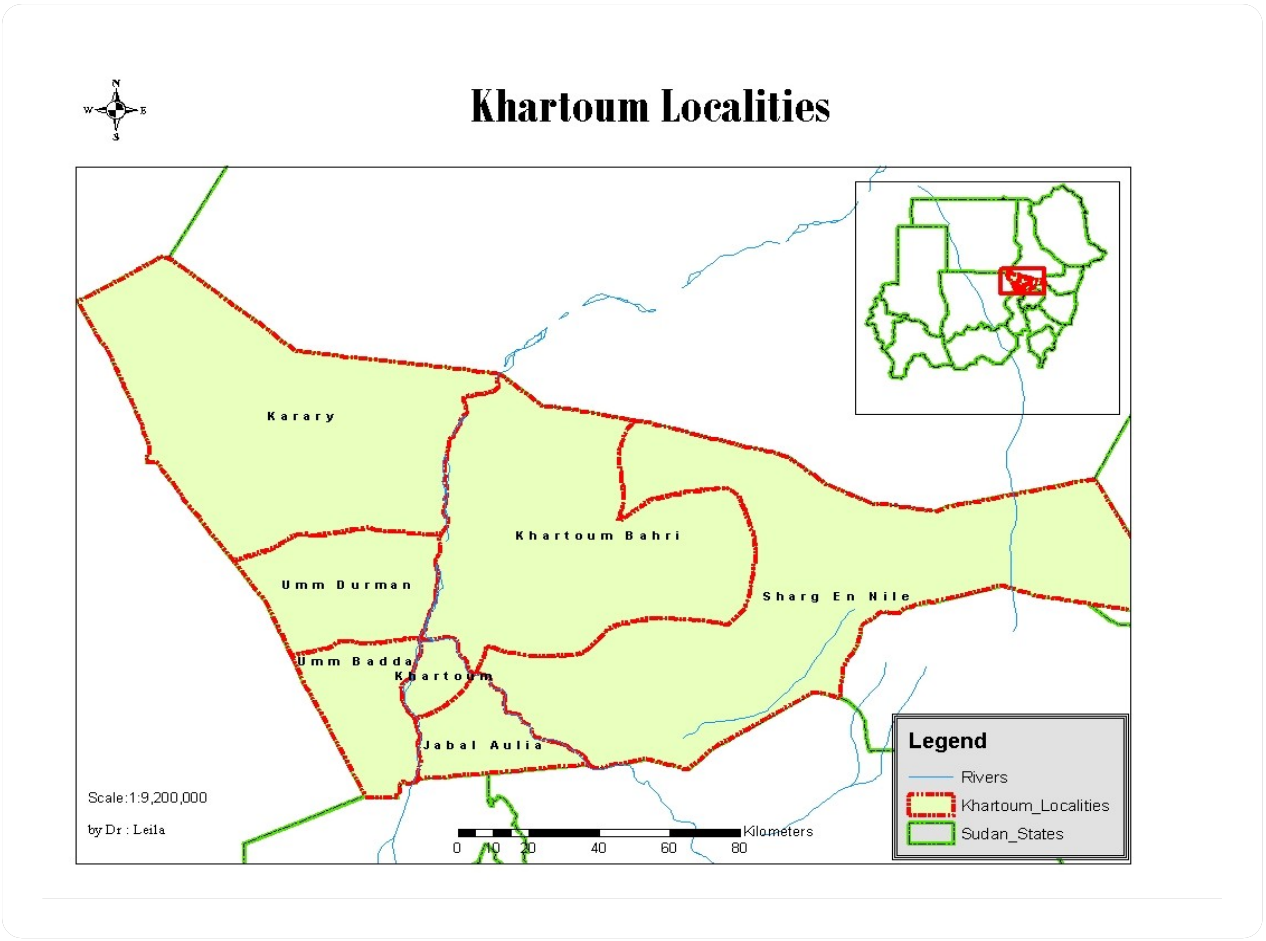


Figure 6 Khartoum localities site for sample collections.

3.2.Type of study:-

The study design was a cross-sectional study to estimate the prevalence of the TBDs , and investigate the risk factors associated with TBDs , also provides some information on occurrence of a disease .

3.3.Sampling methods:-

Non Probability sampling methods (multistage sampling method) were used to select the animals from three localities of the state. From each locality selected two unit after divided in to two administration units and samples were taken from subunit farm . Finally, animals had been selected by using simple random sampling from animals in each farm.

$$Prevalence\,rate = \frac{No.\,of\,diseased\,animals\,with\,theilaria\,x\,100}{Total\,no.\,of\,cattle\,at\,a\,particular\,point \in time}$$

3.4.Sample size determination:-

The sample size was calculated by the formula:-

$$N = \frac{4\,x\,P^{x}\,Q^{¿}}{L2}$$

N = sample size

P^ = expected prevalence

L2 = desired absolute precision

Q^ = (1-P^). (Martin , *et al*. 1987.)

x =multiply mark

From the previous studies the samples size was , from ALradwan project (Safieldin *et al*, 2011)according to then study on prevalence of bovine theileriosis in dry cool ,dry hot and wet hot season was 7, 5.25 ,6.32% ,7respectively. The prevalence was estimated about 7% then the sample size was :-

$$N = \frac{4\,x(0.07)\,x(0.93) = 104.16 = 105\,animal\,samples}{(0.0025)}$$

Samples size equal105 animals.

3.5.blood sample:-

Field samples of blood from cow (n = 105) randomly were collected from different farms in Khartoum localities (Khartoum ,Bahri and Omdurman)

Blood was collected from the jugular vein of individual cow by disposable syringe professionally after cleaning the place. Then put about 5to3ml in vacationers tube with ethylene diamine tetra-acetic acid (EDTA) anticoagulant ,and the same label for each of syringe and tube. The samples were transported under refrigeration using ice packs to the Sudan University of science and technology laboratory, in cal veterinary not exceeding 2–3 hours of collection during a day (OIE ,2009). Thin blood smears on clean glass slide were prepared from each animal by spreading and placing another slide 45 angle and pull. Thin blood films are air-dried, fixed in absolute methanol for 10–60 seconds described by (OIE ,2010) Each slide was placed in the designated inside a wooden box until use .Immediately after arrived to laboratory were done Hematological test.

3.6.Hematological parameters: -

3.6.1.Packed cell volume “PCV”:

A sample of heparinzed blood was drawn in the capillary tube, sealed with crystaseal centrifuged for 5-minutes. Then read at the haematocrit reader.

3.6.2 Hemoglobin concentration:-

Sahli’s acid hematin method:-

Procedure:-

Hemoglobin was converted to acid hematin by the action of HCL. The acid hematin solution is further diluted until
its colour matched exactly with that of the permanent standard of the comparator block. The hemoglobin concentration was read directly from the calibration tube.

Cleaned the hemoglobinometer tube and pipette and ensure that they are dryFill the hemoglobinometer tube with N / 10 HCl up to its lowest mark (10 per cent or 2 g%),Allow a large drop of blood to form on the finger tip, and then dip and tip of the hemoglobinometer pipette into the blood-drop and such blood up to 20 cu mm mark of the pipette, air bubbles then bring down the blood column to the mark by tapping the pipette against the finger tip the tip of the pipette. Immediately transfer the 0.02ml of blood from the pipette into the hemoglobinometer tube containing N / 10 HCl by immersing tip of the pipette in the acid solution and blowing out blood from the pipette. Rinse the pipette two to three times by drawing up and blowing out the acid solution. Withdraw the pipette from the tube, Leave the solution in the tube in the hemoglobinometer, for about ten minutes (for maximum conversion of haemoglobin to acid haematin, which occurs in the first ten minutes),After ten minutes, dilute the acid haematin by adding distilled water drop by drop. mixed it with the stirrer. Matched the colour of the solution in the tube with the standards of the comparator, After addition of every drop of distilled water, the solution should be mixed and the colour of the solution should be compared with the standard. While matching, take care to hold the stirrer above the level of the solution If the colour of the test solution is darker, then continued dilution till it matched with that of the standard . The read of the lower meniscus of the solution should be noted as the result. One more drop of distilled water should be added and the colour should be observed to check the result. The colour will be lighter than the standard if the previous read
was accurate.

3.6.3.First:microscope examination:-

Central VeterinaryResearchLaboratory (Soba):-

A total of 105 cows and calves were examined in August 2015 to find out the prevalence of TBDs in Khartoum State. The Previous preparation the thin blood smears slides from sudan

university for science and technology stained with working dilution of Giemsa stain (1:9) for 30 minutes (10% Giemsa in phosphate buffered saline (PBS) at pH 7.4)(Oie ,2010). The smears were then washed with tap water to remove extra stain and air dried .The stained blood smears were examined under oil immersion lens of microscope (100X) (as a minimum) a ×8 eye piece and a ×60 objective lens(Oie ,2010). At least 50 fields were searched per slide for the presence of Theileria and other blood parasites (Babesia and Anaplasma),all organisms were identified by their morphological characteristics as described in standard texts. ((The Center for Food Security and Public Health.2008).

3.7. Questionnaire:-

Data regarding the characteristics of cow (age, gender, breed, body condition, presence number of ticks and other individual risk factor) and complete case history (lymph node palpation, mm, temperature of animals, breathing status, other management factors).All the animals from each farm examined and fill out the questionnaire by asking the owner ,and other notices. Then divided these risk factors to categories described Thrusfield (1995).Questionnaire were collected through questionnaires completed by investigators on sampling sites in order to calculate the risk factors involved in the spread of Bovine TBDS

3.8.Statistical analyses:-

All the data collected about the risk factors and the results of the study were analyzed using statistical package of social science (SPSS)program, version 16.Descriptive statistical analysis was displayed in frequency distribution and cross tabulation tables . Univariate analysis using the chi-square for qualitative data .describe the variable, number of tested animals and degree of freedoms , chi-square p-value ($p<0,25$)was considered as significant association and the risk factor was then selected to enter the multivariate analysis.

Multivariable logistic regression model was used to describe the risk factor , number of positive cases , odds ratio, confidence intervals (95%) and p-value ($p<0.05$)indicated significant association between the disease and the risk factors.

Anova and T -test were performed. Statistical significance was set at a p-Value of =0.05.

ROC graf done to determined sensitivity of PCV measured to estimated anemia**.**

CHAPTER FOUR

Results

4.1.Microscopic examination:-

A total of 100 cows were examined to detect the prevalence of Theileriosis ,Anaplasmiosis ,Babesiosis. the parasite within RBCS accoding to there morpholical characteristics are show in figures(1 .2 .3 .4).

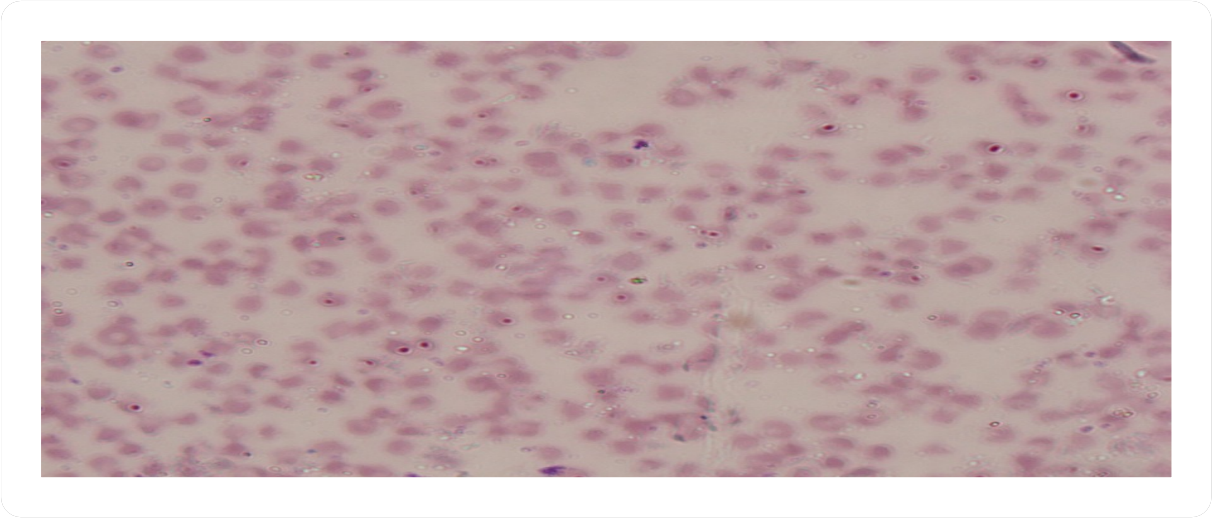
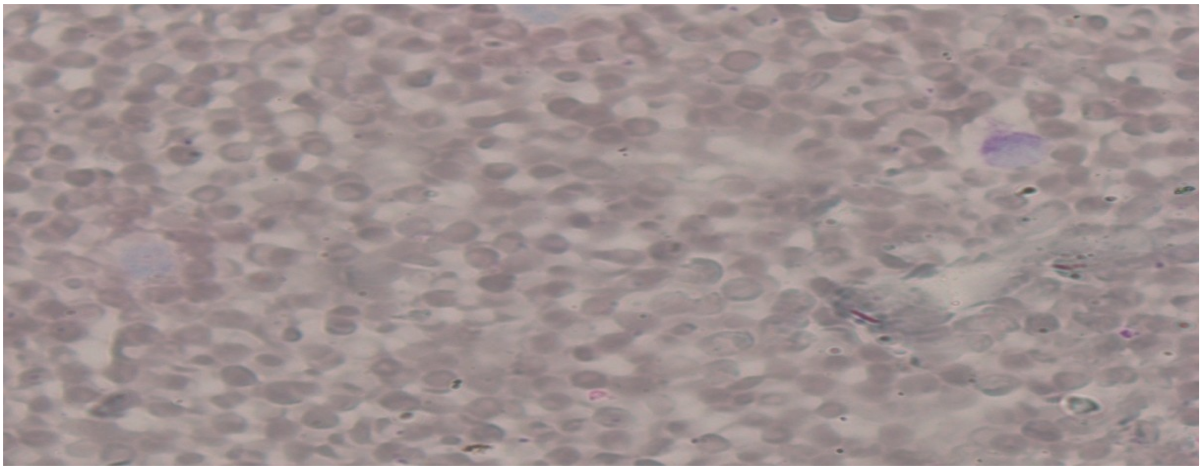


Figure (1) Blood smear from a bovine with *Babesia ssp* Arrow heads denote Babesia in an infected erythrocytes .



Figure(2). Blood smear from a bovine with *Babesia ssp* Arrowheads denote Babesia in erythrocytes with clear apex ,nucleus and cytoplasm

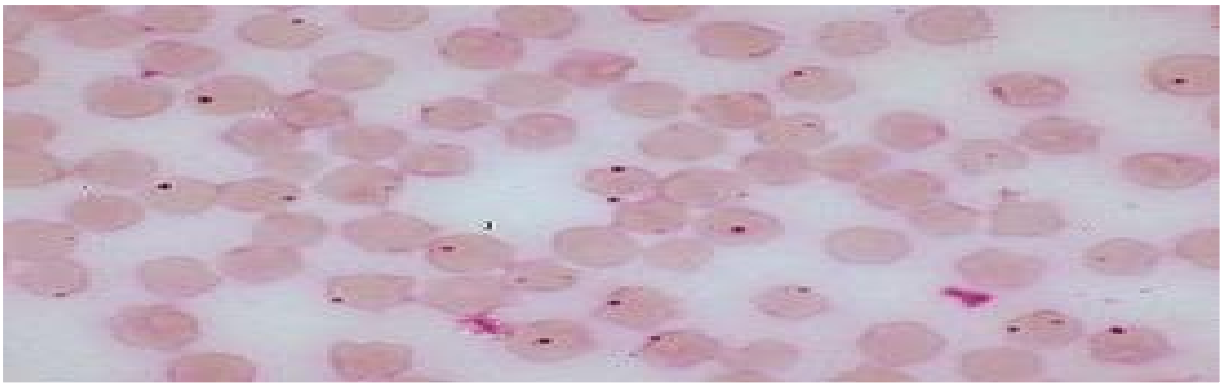


Figure 3 . Blood smear from a bovine with *Anaplasma central* A rrowheads denote Anaplasma in erythrocytes.

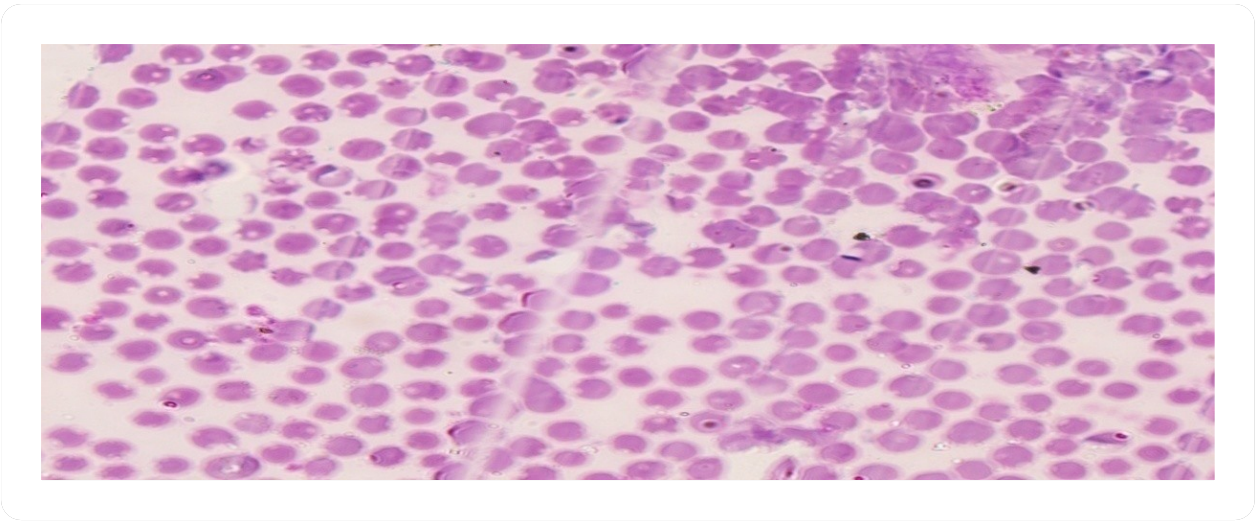


Figure4. **Blood smear from a bovine with *Theileria Annulata* Arrowheads denote piroplasms in erythrocytes**

4.2. Distribution of total results:-

1. A total of 100 cattle and calve from Khartoum State were examined for bovine theileriosis, 34 (34%) animals were positive for bovine theileriosis and 66 (66%) animals were negative in the region (Table 4.1) .

2. A total of 100 cattle and calve from Khartoum State were examined for bovine anaplasmoisis, 38 (38%) animals were positive for bovine anaplasmoisis and 62 (62%) animals were negative in the region (Table 4.2) .

3. A total of 100 cattle and calve from Khartoum State were examined for bovine babesiosis, 20 (20%) animals were positive for bovine babesiosis and 66 (62,9%) animals were negative in the region (Table 4.3)

.2. Descriptive statistical analysis frequency tables, cross tabulation and association tables between the disease and risk factors:-

4.2.1. Age:-

Different ages were examined in this study . The distribution of cattle ages showed in frequency table (4.7) ,5 of cattle were less than 1 years , 12 cattle from 1-2 years and 73 were more than 3 years .The rate of bovine theileriosis was(20 ,46.7 ,26.7) respectively ,anaplasmiosis was (66.7,16.7,8.3)respectively and babesiosis was (31.538.4,20.5)respectively.show in table(4.4.1 , 4.5.1 ,4.6.1,) respectively.

4.2.2.Sex :-

cattle examined for bovine, theileriosis ,anaplasmiosis and babesiosis were as follaw the number of female examined was 81 animals, while the total number of male examined were 19 animals (table 4.2.1). the rate of infection among male was (36.8) %for theileriosis ,(31.6)%for anaplasmiosis and (21.1)% for babesiosis , Rate of infection within females was (33.3)%for theileriosis (39.5)%for anaplasmiosis and(19.8)% details in table (4.4.2 ,4.5.2 ,4.6.2) respectively.

4.2.3. Breed:-

From a total of 100 cattle tow breed were examined, cross breed and local breed . the total of local breed examined was 18 and 82 was cross breed . the rate of infection within cross breed was (32.9)% for theileriosis , (37.8)% anaplasmiosis and (20.7%) for babesiosis while the rate of infection within local breed was (38.9)% theileriosis ,(38.9)% anaplasmiosis and babesiosis (table 4.42) .

4.2.4. Body condition:-

By visual examination the body condition of animals had been investigated, and its association with bovine Theileriosis, Anaplasoisis and Babesiosis, cattle were 67 found to be in good condition and the rate of infection was(37.3)% Theileriosis,(34.3)% Anaplasmoisis(17.9)% while 33 cattle were found to be in poor condition and the Rate of infection (27.3)% for Theileriosis ,(45.4)% Anaplasoisis and (24.3)% table(4.4.4 , 4.5.4 ,4.6.4) respectively.

4.2.5. Vector problem:-

The results of this study showed distribution of 100 cattle examined for bovine Theileriosis, Anaplasmosis and Babesiosis ,[

by visual examination identified ticks in the animals body, by Yes or No. Total number of yes tick present was 43 animals while the total number of no ticks present was 57 (table 4.13.1). Among the animals yes ticks present, the Rate of infection was (32.5)%for theileriosis ,(39.5)% anaplasmiosis and (20.9)% for babesiosis while among these animals no ticks present on the animals body the rate of infaction was (35.1) for Theileriosis ,(36.8)% Anaplasmoisis and (19.3)% Babesiosis . (Table 4.4.6 ,4.5.5 , 4.6.5).respectively .

4.6. Vector Control:-

The results of this study showed distribution of 100 cattle examined for bovine theileriosis, anaplasmosis and babesiosis by vector control, Information identified by answering Yes or No. Total number of answering yes vector control examined was 32 animals, while the total number of answering no vector control examined was 68 (table 4.13.1) ,the rate of infection within vector control was (18.7)% for theileriosis, (31.2)% Anaplasmiosis and (34.4)% while The rate of infection without vector control was (41.1)% for Theileriosis ,(41.2)%Anaplasmiosis and(13.2%) Babesiosis (Table 4.4.6 ,4.5.6 ,4.6.7).respectively.

4.2.7.dehydration :-

By visual examination the animals dehydration has been investigated, for bovine Theileriosis, Anaplasmosis and Babesiosis ,45 cattle were found to be dehydration and the rate of infection was(28.9)% for Theileriosis ,(42.2)% Anaplasmiosis ,(26.7)% while 55 cattle were found to be in no dehydration and the rate of infection (38.2)for Theileriosis ,(34.5)% Anaplasmiosis and (4.5)% Babesiosis table (4.4.7 ,4.5.7 , 4.6.8).respectively

4.8. Localities:-

The result of this study showed distribution of 100 cattle examined for bovine Theileriosis, Anaplasmosis and Babesiosis, by deferent locality in the state,Omdurman, Bahri and Khartoum the distribution of cattle in the Localities was (30 ,30 ,40) respectively .Rate of ir Theileriosis was (10)%in Omdurman ,(30)% Bahri and (.30)%in Khartoum state and the rate of

Anaplasmioisis was (40)% in Omdurman ,(36.7)% Bahri and(13.3)% in Khartoum and the rate of Babesiosis was (47.5)% in Omdurman (45)%Bahri and(17.5)% in Khartoum showed in Table (4.4.9,4.5.8,4.6.10).respectively

4.2.9.Faces :-

The results of this study showed distribution of 100 cattle examined for bovine Theileriosis, Anaplasmosis and Babesiosis by showing fecal nature normal or abnormal Total number of animal had normal feces was (27,32,17) respectictively and the rate of theilerisis was (31.8)% ,(37.6)% Anaplasmiosis and (20%)% Babesiosis ,and animal was found abnormal faces (7 ,6 ,3)respectively and the rate of Theileriosis was (46.7)% ,(40)% Anaplasmosis and (20)% Babesiosis) . show in table (4.4.8 ,4.5.8 ,4.6.9).respectively.

4.2.10.Anemia:-

The results of this study showed distribution of 100 cattle examined for bovine Theileriosis, Anaplasmosis and Babesiosis by hemoglobin measurement ,animals divided in to tow groups(anemic and normal) in the abnormal group the rate of Theileriosis was (42.9)% , (28.6)% for Anaplasmosis and (25) % in Babesiosis and while .the Rate of infection in normal group was(57)% for Theileriosis ,(71.4)% Anaplasmosis and (75)% Babesiosis . table (4.4.11 ,4.5.10 , 4.6.10) respectively.

4.2.1l.ymoh node enlargement:-

By hand palpation for sub scapular lymph node on the animals boby A100 cattle was examined for bovine Theileriosis 54 animal were enlarged and the rate of infection 15(32.6%),while 46 animals were not enlarged lymph node and the rate of infected animals was 19(35.2%) show in table (4.4.10).

4.1:Frequency table for distribution of bovine Theileriosis.

Results	Frequency	Relative Freguency	Cumulative Freguency

+ve	**34**	**34.0**	**34.0**
-ve	**66**	**66.0**	**100.0**
Total	**100**	**100.0**	

Table 4.2:Frequency table for distribution of bovine Anaplasmosis.

Results	**Frequency**	**Relative Frequency**	**Cumulative Percent**
+ve	**38**	**38.0**	**38.0**
-ve	**62**	**62.0**	**100.0**
Total	**100**	**100.0**	

Table4.3:Frequency table for distribution for bovine Babesiosis.

Results	**Frequency**	**Relative frequency**	**Cumulative frequency**
+ve	**20**	**20.0**	**20.0**
-ve	**80**	**80.0**	**100.0**
Total	**100**	**100.0**	

Table 4.4.1:deserbution and prevalence of bovine theileriosis in 100 cattle examined by age :

Results	**Age**			**Total**
	<1	1-3	>3	
+ ve **% of total**	3 20%	8 66.7%	23 31.5%	34 34 %
- ve	12 80%	4 33.3	50 68.5%	66 66%
% of total	15(%)	12(%)	73(%)	100(%)

Table 4.4.2:Distribution and prevalence of bovine theileriosis by sex from 100 cattle examined:

Results	**Sex**		**Total**
	Female	**Male**	
+ ve%	27	7	34
of total%	33.3%	36.8%	34 %
- ve	54	12	66
%	66.6%	63.2%	66%
% of total			
% of total	81(%)	19(%)	100(%)

Table 4.4.3:Distribution and prevalence of bovine Theileriosis in 100cattle examined by breed.

Results	**Breed**		**Total**
	Local	**Cross**	
+ ve	7	27	34
% of total	38.9%	32.9%	34%
- ve	11	55	66
% of total	61.1%	67.1%	66%

Table 4.4.3:Distribution and prevalence of bovine Theileriosis in 100cattle examined by breed.

% of total	18(%)	82(%)	100%

Table 4.4.4: Distribution and Prevalence of Bovine Theileriosis within Body condition from 100 cattle examined in Khartoum State:

Results	**body condition**		**Total**
	Poor	**Good**	
+ ve	9	25	34
% of total	27.3%	37.3%	34%
- ve	24	42	66
	72.7%	62.7%	66%
% of total	33(%)	67(%)	100%

Table 4.4.5:Distribution and Prevalence of Bovine Theileriosis by vector problem on the animals from 100 cattle examined Khartoum state.

Results	**Vector problem**		**Total**
	Yes	**No**	
+ ve	14	20	34
% of total	32.5%	35.1%	34%

- ve	29 67.4%	37 64.9%	66 66%
% of total	43(%)	57(%)	100(%)

Table 4.4.6: Distribution and prevalence of bovine Theileriosis by vector control: from 100 cattle examined in Khartoum state.

Results

Vector control

Total

No

Yes

+ ve

% of total

28

41.1%

6

18.7%

34

34%

- ve

% of total

40

58.8%

26

81.3%

66

66%

% of total

68(%)

32(%)

100(%)

Table 4.4.7: Distribution and prevalence of bovine Theileriosis in 100cattle examined by dehydration .

Results

	Dehydration	
Total		
Yes		
No		
+ ve		
% of total		
13		
%28.9		
21		
38.2%		
34		
34 %		
- ve		
% of total		

32

%71.1

34

%61.8

66

%66

Total %

45(%)

55(%)

100(%)

Table 4.4.8 : Distribution and prevalence of bovine Theileriosis in 100cattle examined by faces :

Results

Faces

Total

Abnormal

Normal

+ ve

% of total

7

46.7%

27

31.8%

34

%

- ve

% of total

8

53.3%

58

68.2%

66

66%

Total %

15(%)

85(%)

100(%)

Table 4.4.9: Prevalence of bovine Theileriosis within localities from 100 cattle examined in Khartoum State:

Results	Localities			Total
	Omdurman	Bahri	Khartoum	
+ ve % of total	3 10%	12 40%	19 47.5%	34 34%
- ve % of total	27 90%	18 60%	21 42.5%	66 66%
% of total	30(%)	30(%)	40(%)	100(%)

Table 4.4.10 : Distribution and prevalence of bovine Theileriosis in 100cattle examined by Lymph node enlargement:

Results	Lymph node enlargement		Total
	Yes	No	
+ ve % of total	15 32.6%	19 35.2%	34 34%
- ve % of total	31 67.4%	35 64.8%	66 66%
Total %	46(%)	54(%)	100(%)

Anemia :-

Table 4.4.11 : Distribution and prevalence of bovine Theileriosis in 100cattle examined by anemia measurement:-

Results	Anemia		Total
	Normal	Anemic animal	
+ ve	22	12	34%
% of total	30.6%	42.9%	
- ve	50	16	66%
% of total	69.4%	57.1%	
Total %	72%	28%	100%

Table 4.5.1 : Distribution and prevalence of bovine Anaplasmiosis in 100 cattle examined by age :-

Results	Age			Total
	<1	1-3	>3	
+ ve%	8	2	28	38
of total%	53.3%	16.7%	38.4%	38%
- ve	7	10	45	62
	46.7	83.3	61.6	62%
% of total	15(%)	12(%)	73(%)	100(%)

Table 4.5.2 : Distribution and prevalence of bovine Anaplasmoisis within sex from 100 cattle examined:

Results	Sex		Total
	Female	Male	
+ ve % of total	32 39.5%	6 31.6%	38 38%
- ve % of total	49 60.5% 81(%)	13 68.4% 19(%)	62 62% 100(%)

Table 4.5.3 : Distribution and prevalence of bovine Anaplasoisis in 100cattle examined by breed .

Results	Breed		Total
	Local	Cross	
+ ve % of total	7 38.9%	31 37.8%	38 38 %
- ve % of total	11 61.1%	51 62.2%	62 %
100%	18(%)	82(%)	100(%)

Table 4.5.4: Distribution and prevalence of bovine Anaplasmosis within body condition from 100 cattle examined in Khartoum State:

Results	Body condition		Total
	Poor	Good	

+ ve	**15**	**23**	**38**
% of total	**45.4%**	**34.3%**	**38 %**
- ve	**18**	**44**	**62**
	54.5%	**65.7**	**62%**
% of total	**33(%)**	**67(%)**	**100(%)**

Table 4.5.5: Distribution and prevalence of bovine anaplasmosis by vector prpblem on the animals from 100 cattle examined Khartoum state.

Results	**Vector problem**		**Total**
	Yes	**No**	
+ ve	**17**	**21**	**38**
% of total	**39.5%**	**36.8%**	**38%**
- ve	**26**	**36**	**62**
% of total	**60.5%**	**63.1%**	**62%**
% of total	**43(%)**	**57(%)**	**100(%)**

Table4.5.6:Distribution and prevalence of bovine Anaplasmosis within vector control: from 100 cattle examined in Khartoum state:

Results	**Vector control**		**Total**
	No	**Yes**	

Results	Vector control		Total
+ ve	28	10	38
% of total	41.2%	%31.2	% 38
- ve	40	22	62
% of total	58.8%	68.7%	62%
% of total	68(%)	32(%)	100(%)

Table 4.5.7:Distribution and prevalence of bovine Anaplasmoisis in 100cattle examined by dehydration :

Results	Dehydration		Total
	Yes	**No**	
+ ve	19	19	38
% of total	42.2%	34.5%	38 %
- ve	26	36	62
	57.8%	65.4%	62%
Total%	45(%)	55(%)	100(%)

Table 4.5.8: prevalence of bovine Anaplasmiosis within localities from 100 cattle examined in Khartoum State:

Results	Localities			Total
	Omdurman	**Bahri**	**Khartoum**	
+ ve	9	11	18	38
% of total	30%	36.7%	45%	38%

Results	Localities			Total
- ve	21	19	22	62
% of total	70%	63.3%	55%	62%
% of total	30(%)	30(%)	40(%)	100(%)

Table 4.5.9 : Distribution and prevalence of bovine Anaplasmiosis in 100cattle examined by faces :

Results	Faces		Total
	Abnormal	**Normal**	
+ ve	**6**	**32**	**38**
% of total	**40%**	**37.6%**	**38%**
- ve	**9**	**53**	**62**
% of total	**60%**	**62.4%**	**62%**
Total %	**15(%)**	**85(%)**	**100(%)**

Table 4.5.10 : Distribution and prevalence of bovine Anaplasmoisis in 100cattle examined by anemia measurement:-

Results	Anemia		Total
	Normal	**Anemic animal**	
+ ve	**30**	**8**	**38%**
% of total	**41.7%**	**28.6%**	
- ve	**42**	**20**	**62%**

% of total	**58.3%**	**71.4%**	
Total %	**72%**	**28%**	**100%**

Table 4.6.1 : Distribution and prevalence of bovine Babesiosis in 100 cattle examined by age

Results	**Age**			**Total**
	<1	**1-3**	**>3**	
+ ve	**4**	**1**	**15**	**20**
% of total	**26.7%**	**8.3%**	**20.5%**	**20%**
- ve%	**11**	**11**	**58**	**80**
% of total	**73.3%**	**91.7%**	**79.5%**	**80%**
% of total	**15(%)**	**12(%)**	**73(%)**	**100(%)**

Table 4.6.2 : Distribution and prevalence of bovine Babesiosis within sex from 100 cattle examined:-

Results	**Sex**		**Total**
	Female	**Male**	
+ ve	**16**	**4**	**20**
% of total	**19.8%**	**21.1%**	**20%**
- ve	**65**	**15**	**80**
% of total	**80.2%**	**78.9**	**80%**
% of total	**81(%)**	**19(%)**	**100(%)**

Results	Sex		Total
	Female	Male	

Table 4.6.3:Distribution and prevalence of bovine Babesiosis 100 cattle examined by breed.

Results	Breed		Total
	Local	Cross	
+ ve % of total	3 16.7%	17 20.7%	20 20%
- ve % of total	15 83.3%	65 79.3%	80 80%
% of total	18(%)	82(%)	% of total

Table 4.6.4: Distribution and prevalence of bovine Babesiosis within body condition from 100 cattle examined in Khartoum State:

Results	body condition		Total
	Poor	Good	
+ ve% of total	8 24.3%	12 17,9%	20 %
- ve % of total	25 75.7% 33(%)	55 82.1 67(%)	80 80% 100(%)

Table 4.6.5: Distribution and prevalence of bovine Babesiosis within vector problem on the animals from 100 cattle examined Khartoum state.

Results	Vector problem		Total
	Yes	No	
+ ve	9	11	20
% of total	20.9%	19.3%	20 %
- ve	34	46	80
	79.1%	80.7%	80%
% of total	43(%)	57(%)	100(%)

Table 4.6.7: Distribution and prevalence of bovine Babesiosis within vector control: from 100 cattle examined in Khartoum state:

Results	Vector control		Total
	No	Yes	
+ ve	9	11	20
% of total	13.2%	34.4	20%
- ve	59	21	80
% of total	86.8%	65.6%	80%
% of total	68(%)	32(%)	100(%)

Table 4.6.8 : Distribution andprevalence of bovine Babesiosis in 100cattle examined by dehydration .

Results	Dehydration	Total

Results	Vector control		Total
	No	Yes	
+ ve % of total	9 13.2%	11 34.4	20 20%
	Yes	No	
+ ve % of total	12 26.7%	8 14.5%	20 20%
- ve % of total	33 73.3%	47 85.4%	80 80%
Total %	45(%)	55(%)	100(%)

Table 4.6.9 : Distribution and prevalence of bovine Babesiosis in 100cattle examined by faces :

Results	Faces		Total
	Abnormal	Normal	
+ ve % of total	3 20%	17 20%	20 20%
- ve % of total	12 80%	68 80%	80 80%
Total %	15(%)	85(%)	100(%)

Table 4.5.10: prevalence of bovine Babesiosis within localities from 100 cattle examined in Khartoum State:

Results	Localities			Total
	Omdurman	Bahri	Khartoum	
+ ve % of total	9 30%	4 13.3%	7 17.5	20 20%
- ve % of total % of total	21 70% 30(%)	26 86.7% 30(%)	33 82.5% 40(%)	80 80% 100(%)

Table 4.6.10 : Distribution and prevalence of bovine Babesiosis in 100cattle examined by anemia measurement:-

Results	Anemia		Total
	Normal	Anemic animal	
+ ve % of total	13 18%	7 25%	38%
- ve % of total	59 82%	21 75%	66%
Total %	72%	28%	100%

Table 4.7:Frequency Table which determined distribution risk factor of animals on the farms.

	Frequency	RalativeFrequency	Cumulative Frequency

Age <1 1-2 > 3	 15 12 73	 15.0 12.0 73.0	 15.0 27.0 100
Sex female	81	81.0 19	81.0 100

Continue table 4.7

Breed Local Cross	18 82	18.0 82	18.0 100
Bodycondition Good Poor	67 33	67.0 33.0	67.0 100
Vector Yes No	43 57	43.0 57.0	43.0 100
Localities Omdarman Bahri Khartoum	 30 30 40	 30.0 30.0 40.0	 30.0 60.0 100
Vector control Yes No	 30 70	 30.0 70.0	 30.0 100
Dehydration Yes No	 45 55	 45.0 55.0	 45.0 100.0
Feaces Normal Abnormal	 81 19	 81.0 19.0	 81.0 100
Anemia Anemic Normal	 28 72	 28 72	 28 100

Table 4.8.1: Summary of univariate analysis for potential risk factors of bovine Theileriosis in 100 cattle examined in Khartoum state using the Chi- square test:P-value ≤ 0.25

Risk factors	Number of animals examined	Number of positive (%)	df	X^2-value	p-value
Age: <1 1-2 >3	 15 12 73	 3 (20%) 8(66.7%) 23(31.5%)	2	7.219	0.027
Sex Mal Female	 19 81	 7(36.8%) 27(33.3%)	1	0.084	0.771
Breed local cross	 18 82	 7 (38.9%) 27 (32.9%)	1	0.234	0.629
Body condition Good Poor	 67 33	 25 (37.3%) 9 (27.2%)	1	0.993	0.319
Vector No ticks Yes ticks	 20 14	 57(35.1%) 43(32.5%)	1	0.070	0.791

Continue table 4.8.1

dehydration Yes No	 45 55	 13(28.9%) 21(38.2%)	1	0.952	0.329
feaces normal abnormal	 85 15	 27(31.8%) 7(46.7%)	1	0.687	0.407

Vector control yes No	 32 68	 6(18.7%) 28(41.1%)	1	4.877	0.027
Localities: Omdurman Bahri Khartoum	 30 30 40	 3(10%) 12(40.0%) 19(47.5%)	2	11.430	0.003
Lymph node enlarged no yes	 46 54	 15(32.6%) 19(35.2%)	1	1.973	0.016

Table 4.8.2: Summary of univariate analysis for potential risk factors of bovine Anaplasoisis in 100 cattle examined in Khartoum state using the Chi- square test:P-value ≤ 0.25

Risk factors	No.of animals examined	Number of positive (%)	df	X^2 –value	p-value
Age: <1 1-3 >3	 15 12 73	 8 (53.3%) 2(16.7%) 28(38.4%)	2	3.819	0.148
Sex Mal Female	 19 81	 6(31.6%) 32(39.5%)	1	0.410	0.522
Breed local cross	 18 82	 7 (38.9%) 31 (37.8%)	1	0.007	0.932
Body condition Good Poor	 67 33	 23(34.3%) 15(45.4%)	1	1.162	0.281
Vector No ticks Yes ticks	57 43	21(36.8%) 17(39.5%)	1	0.075	0.784
Dehydration Yes No	 45 55	 19(42.2%) 19(34.5%)	1	0.619	0.431
Feaces normal abnormal	 81 19	 32(42.2%) 6(40%)	1	0.030	0.863
Vector control yes No	 30 70	 10(31.2%) 28 (41.2%)	1	0.910	0.340
Localities: Omdurman Bahri Khartoum	 30 30 40	 9(22.5%) 11(36.6%) 18(45%)	2	1.669	0.434

Continue table 4.8.2

Table 4.8.3: Summary of univariate analysis for potential risk factors of bovine Babesiosis in 100 cattle examined in Khartoum state using the Chi- square test:P-value ≤ 0.25

Risk factors	No.of animals examined	Number of positive (%)	df	X^2 –value	p-value
Age: <1 1-3 >3	 15 12 73	 4 (26.7%) 1(8.3%) 15(20.5%)	2	1.451	0.484
Sex Female Mal	 81 19	 16(19.8%) 4(21.1%)	1	.016	0.899
Breed local cross	 18 82	 3 (16.7%) 17 (20.7%)	1	0.152	0.696
Body condition Good Poor	 67 33	 12 (24.3%) 8 (17.9%)	1	0.554	0.457
Vector No ticks Yes ticks	 57 43	 11(19.3%) 9(20.9%)	1	0.041	0.840
Dehydration Yes No	 45 55	 12 (26.7%) 8 (4.5%)	1	2.273	0.132
Feaces normal abnormal	 81 19	 17(20%) 3(20%)	1	0. 00	1.000
Vector control yes No	 30 70	 11 (34.4%) 9 (13.2%)	1	6.078	0.014
Localities: Omdurman bahri Khartoum	 30 30 40	 9(30%) 4(13.3%) 7(17.5%)	2	2.865	0.239

Vector					
yes ticks	14(32.5%	Ref	-	-	
			0.180	1.763	0.324
Vector control					
Yes	6(18.7)	ref	-	-	0.154
No	28(41.1)	2.467	0.712	8.555	
Faeces					
Normal	27(31.8)	Ref	-	-	0.762
Abnormal	7(46.7)		0.284	5.568	
Locality					
Omdurman	3(10)	Ref	-	-	0.003
Bahri	12(40)	0.172	0.028	1.045	
Khartoum	19(47.5)	0.055	0.010	0.308	
Lymph node-enlargement					
No	15(32.6)	ref	-	-	0.912
Yes	19(35.2)	0.936	0.291	3.006	

Continue table 4.9.1

Table 4.9.2: Multivariate analysis of bovine Anaplasmoisis and potential risk factors in 100 cattle examined in Khartoum State:

Risk factors	**No.of positive (%)**	**Exp(B)**	**95% Confidence interval. For EXP(B)**		**p-value**
			Lower	**Uppe**	
Age					
1-3	2(16.7%)	Ref	-	-	0.6000
<1	8(53.3%)	2.405	0.920	35.476	
	%)	1.198	0.600	5.622	
Sex					
Male	6(31.6%)	Ref	-	-	0.523
Female	32(39.5%)	0.707	0.244	2.050	

Continue table 4.9.2

Risk factors	No.of positive (%)	Exp(B)	95% Confidence interval. For EXP(B)		p-value
Breed					
cross	31 (37.8%)	ref	-	-	0.919
local	7 (38.9%)	0.931	0.237	3.65	
Body Condition					
good	23 (34.3%)	ref	-	-	0.400
poor	15(45.4%)	1.229	0.483	6.199	
Dehydration					
no	19(34.5%)	ref	-	-	0.957
Yes	19(42.2 %)	0.967	0.286	3.270	
Vector					
no ticks	21(31.2%)	Ref	-	-	0.996
yes ticks	17(39.5%)	0.997	0.340	2.928	
Vector control					
Yes	10(31.2)	ref	-	-	0.247
No	28(41.2)	1.901	0.641	5.633	
Faeces					
normal	32(37.6)	Ref	-	-	0.170
abnormal	6(40)	3.065	0.619	15.182	
Locality					
Omdurman	9(30)	Ref	-	-	0.939
Bahri	11(36.6)	0.815	0.191	3.471	
Khartoum	18(45)	1.006	0.269	3.756	
HB					
normal	30(41.7)	-	-	-	1.786
anemic	8(28.6)	1.786	0.695	4.591	

Table 4.9.3: Univariate analysis of bovine Babesiosis and potential risk factors in 100 cattle examined in Khartoum State:

Risk factors	No.of positive (%)	Exp(B)	95% Confidence interval. For EXP(B)		p-value
			Lower	Upper	
Age 1-3 <1 >3	 1 (8.3%) 4(16.7%) 15(20.5%)	 Ref 3.028 1.565	 - 0.126 0.081	 - 72.805 30.311	0.775
Sex Female Male	 16(19.8%) 4(21.1%)	 Ref 1.320	 - 0.084	 - 20.813	0.843
Breed **Continue table 4.9.3**	%) 7%)	ref 1.154	- 0.218	- 6.106	0.866
Body condition good poor	 12 (17.9%) 8(24.3%)	 ref 0.679	 - 0.163	 - 2.817	0.593
Dehydration No yes	 8(4.6 %) 12(26.7%)	 ref 2.105	 - 0.522	 - 8.484	0.295
Vector no ticks yes ticks	 11(19.3%) 9(20.9%)	 Ref 0.673	 - 0.158	 - 2.862	0.591
Vector control no yes	 9(13.2) 11(34.4)	 ref 0.363	 - 0.112	 - 1.173	0.09
Faeces Normal Abnormal	 17(20) 3(20)	 Ref 0.821	 - 0.146	 - 4.625	0.823

Locality Bahri Khartoum Omdurman	 4(13.3) 7(17.5) 9(30)	 Ref 0.815 1.006	 - 0.191 0.269	 - 3.471 3.756	0.544
HB Normal Anemic	 13(18.1%) 7(25%)	 ref 0.661	 - 0.232	 - 1.880	0.438

Table 4.10: The effect of Temperature , HB and PCV among sex.

	Temperatures	**HB**	**PCV**
Sex		9.66 ±1.43	6.99±6.99
Age >1 1-3 >3	**Temperture** 38.82±0.85 39.41±0.80 38.91±0.66	**HB** 10.46±1.38 9.208±2.34 8.68±2.14	**PCV** 37.55±5.99 30.25±6.38 33.84±6.50
p-value	**0.05**	**0.012**	**0.015**
Location Omdurman Bahri Khartoum	 39.25±0.57 39.16±0.66 38.59±0.72	 10.06±1.39 8.68±1.99 8.482.46	 36.20±5.18 31.84±5.93 33.88±5.93
P-value	**0.000**	**0.05**	**0.037**

livestock farms located at Attock district and Islamabad, Pakistan as compared to this study.

Zahid *et al*. (2005) reported the prevalence of Babesia spp. to be 5% on a government Livestock

Experiment Station, Kasur district. Similarly, Ahmad and Hashmi (2007) have reported the overall occurrence of *B. bigemina* to be 6.6% in cattle from Malakand Agency.

While Afridi *et al*. (2005), had reported a lower prevalence of 1.75% for *B. bigemina* from Peshawar and adjoining areas as compared to present study. AFkar *at el*(2012) reported the prevalence of theileriosis was 42%. And in Irag Safieldin ,*et al(2011*) reported The prevelance of theileria annulata was7%,5.2%,6.32% for dry cool ,dry hot, wet hot season respectively while the prevalence of babesiosis infection was 1% in the dry cool season ,The prevalence varies from region to region, host, management and environmental factors (agro-ecological and geo-climatic conditions) influence the prevalence of ticks and tick-borne diseases (Kivaria, 2006)

12,8,7 animals were positive to diseases (Theileriosis ,Anaplasmoisis and Babesiosis) respectively and showed the sign of anemia compared while Riond *et al*,(2007) reported that 34 animals out of 94 positive cattle showed signs of haemoglobinuria while all the positive animals showed the sings of anaemia ,

Previous reports lack distribution of TBDs on the basis of age, sex, breed, body condition,vector control,ticke problems and herd. Moreover, there was difference of area and cattle breeds under this study. A higher prevalence of Anaplasma ssp and Babesia ssp was apparent in animals of less than one years years of age while *T. annulata* was more prevalent in the 1 to 3 year- though these findings were statistically significant in the present study except in babesia there is no significant different between babesiaosis and age group disagrement with present study Kocan and associates also reviewed that cattle of more than one year of age are more frequently affected by Anaplasmosis (Kocan *et al*, 2010).disagreement with these study. A higher infection rate in males as compared to female cattle has been recorded in the present study except in Aanaplasa ssp. Though this difference was not statistically significant, these findings are in disagreement with the results of Durrani (2008) and Rajput *et al*. (2005) who reported higher prevalence of *T. annulata* in female animals and agreement with present study in higher prevalence in female infected with *A.naplasma* spp. The immunosupression in advanced pregnancy and or lactation in high producing study animals are the possible reasons for the higher

prevalence of A. marginale in female cattle (Kocan *et al*, 2010). non significant relationship of different breeds on the prevalence of TBDs was found. differently, Khan *et al*. (2004) mentioned

higher prevalence of tick-borne disease in local breed cattle (38.9,%) Theileria ssp then (38.9%) Anaplasma ssp and Babesia ssp (20.7%)but in cross breeds. The significant relationship of different localities in Khartoum State showed that the highest prevalence of Anaplasma ssp recorded in Khartoum (45)%then Bahri(36.7)%and Omdurman(30)%and Babesia ssp was recorded higher in Omdurman and higher prevalence of theileriosis occurred in Khartoum, highest prevalence of TBDs in the farm when the owners didn't use acaricides to control ticks in there animals and its farms was (42.2)% in Anaplasma ssp then (41.1)% in theilera ssp and there was significant association fount between TBDs and vector control except in. Anaplasma parasitic diseases constituted a real problem in the area. In addition, from the low consumption of the external parasites drugs (the lower one), it became obvious that, cattle keepers knowledge about the acaricides and prevention was inadequate. Therefore, ticks infestation was very high and represents a major problem needs more extensive control measures. animal with poor body condition were susceptible to TBDs more than other and the higher prevalence found to be in anaplasma spp (45.4)and (24.3) in babesia ssp and vice versa in theileria ssp .there was signifecent association between theileriosis and lymph node enlargement in univeriate analysis p-value-0.016. and no singniicent association between TDBs in present study and anemia ,the higher prevalence of anemia in animals which infected with theileriosis was (42.9%) anaplasmiosis (28.6)% babesiosis (25)% ,the prevalence of anemia in the healthy animal more than the prevalence of diseased animals that means there were other diseases and factor causing anemia such as internal parasite and nutritional insufficiency Safieldin,*et al*,(2011), Gill *et al* , (1977) and callow and pepper (1970) agreement with present study the anther reason may due to low number of parasite in the blood of diseased animals and enzootic stability.

In T.test analysis there was no significant different between the mean in level of Hemoglobin ,Temperature and Packed cell value between male and female.

Highly significant relationship between Temperature ,HB and PCV in different location Omdurman ,Khartuom and Bahri to occarance of TBDs (table 4.12).

PCV measurement was considered poor test to detect anemia because it had low sensitivity the result under the area equal 0.624. in ROC graf(statistics for veterinary animal science .2006).

Conclusion :-

 There was high prevalence's of TBDs in Khartoum State ,Sudan. There is a need for further investigations using modern serological and molecular techniques for the identification of the carriers of the tick- borne pathogens.

 Tick control by dipping or spraying can reduce the risk. Strategic tick control is good, as it is a level of control that prevents ticks becoming a nuisance, but allows enough ticks to remain for infection to occur at an early age so that the animals become protected against the diseases
 Try to keep only tropical breeds which are more resistant to ticks and tick-borne diseases .

Recommendations :-

 The prevention and control of tick-borne diseases are very complex and varies in different areas. Ask your animal health technician or veterinarian for advice about the best methods in your area .

 Animals should preferably be exposed to the parasites at a young age so that they can develop natural immunity in areas where the diseases occur.

 astrategic treatment and application of preventive measures. The of livestock owners use the drugs randomly without a clear plan .

  using prophylactic drugs to Control Ticks borne diseases .

 Indigenous animals should have sufficient population of vector (continual infection – reinfection process = premunition).

 Rigorous programmes of tick eradication lead to sterile animals (naive animals). It may lead to loses .

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Appendix1

Questionnaire:-

Investigation of Bovine TBDs in Khartoum State.

Conducted by: The Epidemiology Department of Sudan university of science & technology .

Locality _____ date _____

Farm Owner_____

Animal name _____

Address _____

The individual risk factors:-

1-Age :(years)

<1 ( ) 1-3 ( ) >3 ( )

2-sex:

Males ( ) Females( )

3-Breed:

Local ( ) Cross ( )

4-Body condition:

Good ( ) Poor ( )

5- ticks on the animal :

No ticks ( ) yes ticks ( )

6-Tick problem:

Yes ( ) No ( )

7-Vector control:

Yes ( ) No ( )

8-dehydration

Yes ( ) No ( )

9-Faces

nomal ( ) Abnomal ( )

10-lymph node enlargement(for theileria ssp)

yes ( ) no ( )

11-localities:

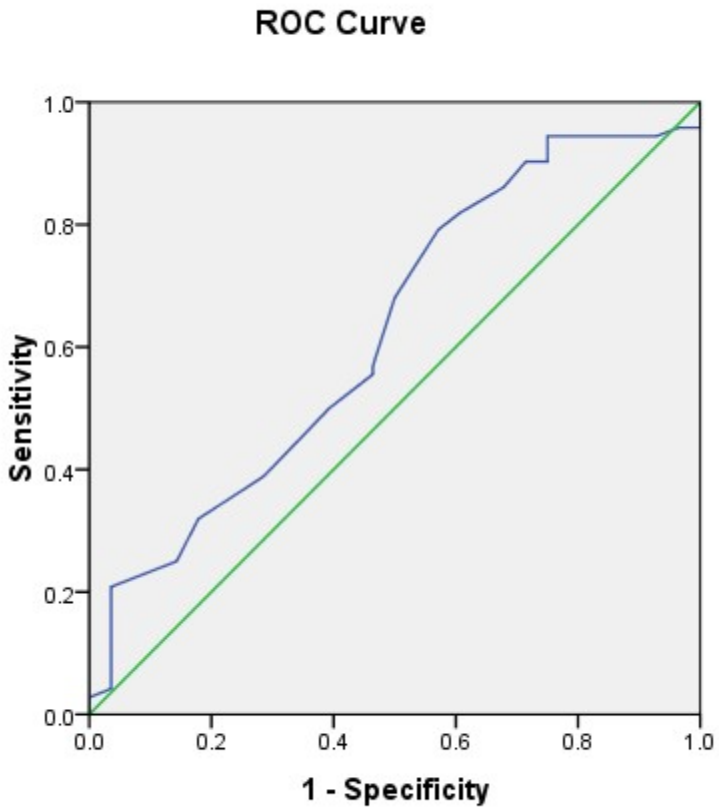
Omdurman ( ) Bahri ( ) Khartoum ( )

12-Animal temperture ( )C

13-other notice and comment

……
……

Appendix 2


ROC Curve
Sensitivity
1 - Specificity
0.0
0.2
0.4
0.6
0.8
1.0
Diagonal segments are produced by ties.