



Sudan University of Science and Technology
College of Graduate Studies



**Prevalence and Risk factors of *Trypanosoma evansi* Infections
and Classification of Its Vectors in Camel in Tamboul
Locality - Gezira State - Sudan**

نسبة الإصابة وعوامل الخطر لعدوى مثقبة ايفانساى وتصنيف نواقله في الإبل
في محلية تمبول - ولاية الجزيرة - السودان

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بِسْمِ اللَّهِ الرَّحْمَنِ الرَّحِيمِ

قال تعالى :-

(أَفَلَا يَنْظُرُونَ إِلَى الْإِبِلِ كَيْفَ خُلِقَتْ)

صدق الله العظيم

سورة الغاشية الآية (17)

DEDICATION

To my father & my mother

To my brothers & my sisters

For the deceased soul Aietizaz

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ABSTRACT

Surra, a vector borne disease caused by *Trypanosoma evansi*, is considered as a major enzootic disease mainly for the dromedary camel. Therefore, a cross-sectional study was conducted to determine the prevalence of camel trypanosomiasis and to assess the distribution and dynamics of the vectors responsible for transmission of the disease in camels in Tamboul market, Gezira State, Sudan from December 2018 to November 2019. A total of 213 blood samples were collected from camels, thin blood smears stained by Giemsa and the Buffy coat (Haematocrit Centrifugation technique (HCT)) were used to detect the protozoan. The both methods showed the same prevalence ratio was 11 (5.2)% Aquestionnaire which included age, sex, breed, body condition and season was designed. The study revealed that there was no relationship between the disease and all risk factors ($p \leq 0.05$). This study demonstrated that there was no difference between thin smear and buffy coat methods for diagnosis of trypanosoma when using Roc curve. Concerning density of Tabanus flies , NZI traps were used for collection of the flies through the year to identify the type of flies in this area. One hundred eighty one flies were collected. Seventy five *Tabanid agrestis*, 74 *Tabanus sufis* and 32 *Tabanus teaniola* . The largest number of *Tabanus sufis* (30%) was captured in April, followed by the *Tabanus agrestis* (22.6%) in October and *Tabanus taeniola* (21.8%) in November. Diagnosis of trypanosomiasis in camels by using microscopic examination and haematocrit centrifugation is easy tests and quick. Identification of vectors Important because the disease leads to health and productivity loses.

المستخلص

السرة ، وهو مرض ينقله الناقل الناتج عن التريبانوزوما إيفانساوي، يعتبر من أمراض الدم الرئيسية المتوطنة في الإبل (الجمال) . لذلك، تم إجراء دراسة مستعرضة لتحديد مدى انتشار داء مثقبيات الإبل وتقييم توزيع وديناميكية النواقل المسؤولة عن إنتقال المرض في الإبل في سوق تمبول، ولاية الجزيرة، السودان من ديسمبر 2018 إلى نوفمبر 2019. تم جمع عدد 213 عينة دم من الإبل. تم إستخدام لطخات الدم الرقيقة المصبوغة بجيمسا وطريقة البوفي كوت (تقنية الطرد المركزي الهيماتوكريت (HCT) للكشف عن الطفيل. تم تصميم إستبيان وتضمن العمر الجنس، السلالة، حالة جسم الحيوان والمواسم . وكان معدل الإيجابيات 11 (5.2)٪ في كلتا الطريقتين. أوضحت هذه الدراسة أنه لا يوجد فرق بين المسحة الرقيقة والبوفي كوت باستخدام إختبار منحني Roc Curve في تشخيص هذا الطفيل كشفت الدراسة عن عدم وجود علاقة بين المرض وجميع عوامل الخطر ($p \geq 0.05$). فيما يتعلق بكثافة ذباب تباويد ، تم إستخدام مصائد NZI لجمع الذباب على مدار العام لتحديد نوع الذباب في هذه المنطقة ، حيث تم جمع 181 ذبابة ، 75 Tabanid ، 74 ، agrestis ، Tabanus sufis و 32 Tabanus teaniola. تم إمساك أكبر عدد من Tabanus sufis (30٪) في أبريل ، يليه Tabanus agrestis (22.6٪) في أكتوبر و Tabanus taeniola (21.8٪) في نوفمبر. تشخيص الأمراض المنقبة للإبل عن طريق الفحص المجهرى والطرد المركزي بالهيماتوكريت هي اختبار سهل سريع للكشف عن الطفيل والتعرف على ناقلات المرض ذو اهمية لأن المرض يؤدي إلى خسارة أكبر بكثير بين الصحة والإنتاجية.

INTRODUCTION

Camels (*Camelus dromedarius*) are important animals. They play an important role in sustainable agricultural resources for millions of people in the arid and semi-arid zones. Camels also provide milk, meat, wool and are used for water traction and the bear of burden. Moreover, the exportation of camels contributes to foreign currency earnings (Abd-Elmajid, 2000). Sudan ranks second, after Somalia, in camels' population (FAO, 2008). The Camel population was estimated at three millions. They are distributed in Northern Kordofan and Darfur in the West and the Red Sea, Kassala and Butana in the East (Abd-Elmajid., 2000). The protozoan parasite *Trypanosoma evansi* (*T. evansi*) has a large diversity of mammalian hosts. It can be found in intra- and extra-vascular fluids of mammals and causes the surra disease in Africa, Asia, Europe and Latin America (Desquesnes *et al.*,2008). Surra constitutes one of the major veterinary problems worldwide (Omer *et al.*,2004). Infection with *T. evansi* leads to high mortality and morbidity rate in camel's population. Additionally, since 2008, it is stated that the presence of surra in any area should be reported to the OIE (OIE,2008). Furthermore, the first human trypanosomiasis caused by *Trypanosoma evansi* in India was reported by Joshi *et al.* (2005) which make it a potential human pathogen. Tsetse fly is considered as the main vector of animal trypanosomiasis, however, *T. evansi* is transmitted mechanically, non-cyclically, by Haematophagus flies such as: *Tabanus*, *Stomoxys*, *Lyperosia*, and *Chrysops*. Because the trypanosomes remain infective for only a short period, such transmission occurs when the biting flies feed on more than one host and transmit the organism through their mouth parts (Desquesnes *et al.*, 2013).

T. evansi affects the health, working capacity and productivity of dromedary camels however, clinical signs are not pathognomonic

therefore, diagnosis must be confirmed by laboratory methods. Additionally, assessment of the prevalence of *T. evansi* would be better performed by means of parasitological (Giemsa stained thin smear), serological and molecular tests (Singh *et al.*, 2004). The control of trypanosomiasis can be performed either by using chemotherapy or by vector control (Abd-Elmajid., 2000) which is necessary for improved camel health and productivity. This cannot be attained unless several investigations on the epidemiology of the disease are made.

Objectives:

The objectives of this study are:

1. To determine the current situation of *Trypanosoma evansi* in sudanese camels in Tamboul market at Gezira state, Sudan.
2. To investigate the potential risk factors such as (age, sex, breed, body condition and season), which may be associated with *T. evansi* infection in the study area.
3. To determine the density of tabanids vectors in Tamboul area.

CHAPTER ONE

1. LITERATURE REVIEWS

1.1. Etiology of trypanosomiasis

Trypanosoma evansi belongs to the subgenus Trypanozoon and it is the causative agent of camel trypanosomiasis. It is hypothesized that *T. evansi* originated from *Trypanosoma brucei* by adaptation to a non-cyclical transmission and loss the ability to develop and differentiate in the tsetse fly vector (Luckins, 1998). Camels become infected following contact with biting insect and when these camels moved to non-tsetse areas. Other species of *trypanosoma*, such as *Trypanosoma congolense*, *Trypanosoma brucei* and *Trypanosoma vivax* have been also isolated from camels in Sudan, (Mahmoud and Gray, 1980; Elamin *et al.*, 1999, Mossaad, *et al.*, 2017), but their effects in camel needs to be elucidated.

1.2. Classification of trypanosomes

Trypanosomes are classified according to (Taylor *et al.*, 2016) as follows:

Kingdom: Protozoa

Phylum: Euglenozoa

Class: Kinetoplasta

Order: Trypanosomatida

Family: Trypanosomatidae

Genus : *Trypanosoma*

Species: *evansi*

The principal pathogenic trypanosomes causing animal Trypanosomiasis in the Sudan are *T. congolense*, *T. vivax* and *T. brucei* which affected cattle, sheep, goats, horses and donkeys, while *T. evansi* affects mainly camels and may affects horses (Karib, 1961).

1.3. Morphology and biology of Trypanosoma sp

T. evansi is morphologically identical with subgenus Trypanozoon, and indistinguishable from the other members of this subgenus. *T. evansi* can be distinguished from *T. brucei* by isoenzyme electrophoresis (Gibson *et al.*, 1983). *T. evansi* enters the tissue or other body fluids and it may cross the blood brain barrier (Leese, 1927). It can also enter the joint fluids, thus being less accessible to the chemotherapy (Jennings *et al.*, 1977).

1.4. Distribution of Trypanosomiasis

Trypanosomiasis is widespread in different parts of the world and considered as a major constraint for camel production (Elamin *et al.*, 1999). Generally, the geographical distribution of Trypanosomiasis is related to the movements of infected animals (Gutierrez *et al.*, 2010). In Africa, the trypanosomiasis is present in all countries, where camels are present, from the northern part of Africa through the Middle East to the South-East Asia. The disease exists in Mauritania, Morocco, Algeria, Tunisia, Libya, Egypt, Sudan, Eritrea, and Ethiopia, also in the northern parts of Mali, Burkina Faso, Niger, Nigeria, Chad, Somalia, and Kenya (Hoare, 1972).

Trypanosomiasis is continuously present eastwards, in Saudi Arabia, Oman, the United Arab Emirates, Jordan, Israel, Lebanon, Syria, Iraq, Turkey, Iran, Kasakhstan, Afghanistan, Pakistan and Bulgaria (Srivastava *et al.*, 1984; Desquesnes *et al.*, 2003, 2009; Hasan *et al.*, 2006).

Trypanosomiasis also exists in India, China, Mongolia, Russia, Nepal, Myanmar, Laos, Vietnam, Cambodia, Thailand, Malaysia, Philippines, and Indonesia (Luckins, 1988; Reid, 2002). The extension of Trypanosomiasis toward the West is more recent (Hoare, 1965).

1.4.1. The Camel Trypanosomiasis in Sudan

The most important protozoan disease of camels in the Sudan is trypanosomiasis, especially infection with *T.evansi*. According to Karib (1961) camel trypanosomiasis was first recorded in 1904. By 1908 the disease had been diagnosed in Kordofan province north of the 12° parallel, the whole of the White Nile province and the area between Suakin and Kassala along the border with Eritrea (Mahmoud and Gray, 1980). At present, camel trypanosomiasis is wide spread in all of the camel breeding areas, occurring in acute and chronic forms. In the acute form the disease is almost always fatal. In the chronic form there is usually loss of production and bodily condition, and anemia (Mahmoud and Gray, 1980).The disease is common in Kordofan and Darfur states in the west, Kassala, Gadaref and Red sea states in the east and to a lesser extent in central Sudan, in the Gezira, Sennar, Blue Nile and Khartoum states (karib,1961). Then successful studies to evaluate a simple PCR based technique for field diagnosis of *T. evansi* infection in camels from Eastern and Western regions of the Sudan were conducted by Nahla *et al.* (2011). During a camel trypanosomiasis survey in different areas of Sudan, Ibrahim *et al.* (2011) concluded that the decrease in the packed red cell volume (PCV%) can be used as indicator for the infection of camel trypanosomiasis in the Sudan. Later camel trypanosomiasis (Guffar) caused by *T.evansi* was reported in Omdurman west plain, western Sudan, using direct smear, MicroHematocrit Centrifugation Technique (MHCT) and Card Agglutination Test for *T.evansi* (CATT/*T. evansi*) by Babeker and Hassab Elrasoul.(2014).

1.4.2. Tabanus Distribution in Sudan

Transmission of *T. evansi* is reported to be exclusively mechanical, carried out by a number of species of haematophagous biting flies, including

Tabanus and *Stomoxys* (Mahmoud and Gray, 1980) found a definite correlation between the seasonal outbreak of *T. evansi* infections and the increase in number of tabanids during the rainy season in the Sudan (June-October). The prevalence of tabanid species throughout the year ensures permanent possibility of infections with *T. evansi*, with outbreaks occurring during the rainy season (Malik and Mahmoud, 1978) found that cattle, sheep, goats and donkeys undergo a protracted infection which may result in a carrier state, and these species may act as reservoir hosts. Previously it is reported that camels do not frequently come into close contact with cattle in the Sudan, however, they do come into contact with sheep, goats and equines. This situation has now changed as a result of the recent drought, so that wherever cattle are kept in northern Sudan, camels also exist, thereby increasing the possibility of contracting the infection with *T. evansi* and other trypanosomes. Yagi and Abdel Razig ,(1972) reported that *Tabanus taeniola* was prevalent throughout the year, while *Atylotus agrestis* have a predisposition to appear at the end of the rainy season. *Atylotus fuscipes* has the same ecological preferences as *A. agrestis* and usually appears at the same season. It was also mentioned that *Tabanus biguttatus* was also common throughout the year, while *Ancalalatipes* occurs during the rainy season together with *Philoliches magrettii*, but its flight season was very short. Suliman (1992) reported that seven species were collected from Sinnar area including: *Tabanus Taeniola*, *T. gratus*, *T. biguttatus*, *T. sufis*, *Atylotus agrestis*, *A. fuscipes* and *Philoliches magrettii*. He stated that the first three were found in all of the surveyed areas (Central State) while *T. biguttatus* was also found.

1.5. Hosts of Trypanosoma Evasi

Although *T. evansi* also has a huge range of domestic and wild hosts worldwide (Lun and Desser, 1995). It is highly pathogenic in Camelids and Equids. *T. evansi* can infect camels, horses, donkeys, dogs, cattle, water

buffaloes and elephants. Equines and dogs are very susceptible and usually die after an acute course of the disease (Leese, 1927). Cattle, sheep, goats and antelopes often carry the parasite sub-clinically and acting as asymptomatic reservoirs.

In Africa and the Middle East *T. evansi* is mainly a parasite of camels (*Camelus dromedarius*) which act as the main host, and also it is pathogenic in other Camelidae, such as the *Bactrian camel* (*Camelus bactrianus*). Moreover, *T. evansi* is highly pathogenic in Equidae, especially in horses, asses, donkeys and also in their crossbreeds (mules). Additionally, *T. evansi* can infect cattle (Dia and Desquesnes, 2007), pigs, sheep, goats (Reduth *et al.*, 1994), domestic cats (Tarello, 2005) and dogs, which may act as sentinel animals as observed in the surroundings of tamboul market).

In Asia; Trypanosomiasis due to *T. evansi* is considered as an economically important disease, which affects not only horses and buffaloes, but also cattle, pigs, and goats (Dargantes *et al.*, 2009). Cattle in Asia are more receptive for the infection than in Africa or Latin America, and they can exhibit strong clinical signs (Tuntasuvan and Luckins, 1998). *T. evansi* has been also isolated from elephants in India (Stephen, 1986), and Thailand (Hin *et al.*, 2004). Generally, horse, donkeys, mules, camel, dogs, cats and Asian elephant are more susceptible than sheep, goat, bovines and pigs. Rats and mice are highly susceptible as experimental hosts (Reid *et al.*, 2001).

1.6. Life cycle of Trypanosoma Evasi

The trypanosoma is replicated by longitudinal binary fission in both host and vector, in which the flagellum and kinetoplast were dividing together (Liu and liv, 2005). Adding to that, the cyclical transmission was not observed at any stage of *T. evansi* in the mechanical vectors. Consequently, a procyclic or insect stages (epimastigotes) do not exist in *T. evansi*, which

is attributed to lack of maxi circles in the kinetoplast DNA (Ellie *et al.*, 1999).

1.7. Transmission of Trypanosoma

T. evansi is transmitted in several ways, via biting insects, sucking insects, and vampire bats. The transmission can also be vertical, horizontal and peroral route, with various epidemiological significances, depending on the season, the location, and host species (Desquesnes *et al.*, 2019).

Mechanical transmission occurs in cases of interrupted feeding, where a fly began feeding on infected animal and completed it in another animal. So it passes infection from an infected animal to non- infected susceptible animals through contamination. The most important mechanical vectors are flies of genus *Tabanus*, *Haematopota*, *Lyperosia*, *Stomoxys* and *chrysops* (Mohiuddin, 2007). This mode of transmission is sufficiently effective in maintaining *T. vivax* and *T. evansi* outside tsetse infected area. Wells (1972) reviewed the importance of mechanical transmission in nagana and related it to the presence of *T. vivax* in countries outside Africa where tsetse flies are present. Raymond (1990) was able to prove the role of *Tabanus importunes* in *T. vivax* transmission in French Guyana Mihok *et al.* (1995) studied the ability of African *Stomoxyniae* to transmit trypanosomes; they concluded that five species of the family were capable of transmitting the disease mechanically. Transmission by other means include infection of carnivores with *T. evansi* and *T. brucei* by ingesting meat or organs of infected animals and transmission of *T. evansi* in South America by the bites of vampire bats, but these ways were considered to be of less importance (Ulenberg, 1998).

1.7.1. Other types of Transmission

Besides vector transmissions and the contamination of wounds, iatrogenic transmission caused by the use of non-sterile surgical instruments or needles especially during vaccination and mass treatments (Davila and

Silva, 2000). Sexual transmission or transmission from dam to calf/ foal could occur in particular cases, especially when mucosa are altered or in cases of very close contact (licking) with secretions (mucus, lacrimation, etc.). However, the real impact of horizontal transmission has not been estimated (Sina *et al.*, 1979; Gardiner and Mahmoud, 1990).

Transplacental infections have been described in *T. equiperdium* and *T. brucei* (Sina *et al.*, 1979; Gardiner and Mahmoud, 1990). Adding to that vertical transmission of *T. evansi* has also been demonstrated as shown in a review on transplacental transmission of trypanosomes (Ogwu and Nuru, 1981). *Trypanozoon*, especially *T. evansi*, may be transmitted by pre-oral contamination. This mechanism could occur quite easily when the oral mucosae is damaged, especially when the carnivores eating fresh meat, blood, offal, or bones from animals infected with *Trypanosoma* (Bhaskararao *et al.*, 1995). Transmission by the vampire bat is a new biological system that has been established in Latin America (Hoare, 1972).

1.8. Epidemiology of Trypanosomiasis

Trypanosomiasis is often referred as African trypanosomiasis. However, there are certain types of trypanosomes cause infection outside this continent. *T. evansi*, the causative agent of Surra, occurs not only in Africa, but also in Central and South America, Middle East, and in Asia. Although Surra has a wide host spectrum, the main host species varies with the geographical region. In East Africa, camels are the most important host, whilst in Central and South America the horse is principally affected (Dia *et al.*, 1997). In Asia, a wider range of hosts is involved, including camel, cattle, buffalo, horses and pigs (Pacholek *et al.*, 2001). In contrast, there is a little evidence to suggest that domesticated livestock other than camels and horses in Africa and South America respectively, are infected with *T. evansi* (El-Sawalhy and Seed, 1999). The ability of *T. evansi* to be

transmitted by blood sucking insects allows *T.evansi* to extend its geographical range to the north of Sahara desert, Asia, Pakistan, India, the USSR, China, Sumatra, Java, the Philippines, Mauritius, Madagascar, and South and Central America. Moreover, through camels' exportation the disease was introduced into Australia, North America and South-West Africa. Introduction of the parasite into new areas is generally characterized by a high incidence rate where the mortality rate ranging between 30 to 100% (Elamin *et al.*, 1999).

1.9. Course of infection

Not all animals get the infection with trypanosomes showing clinical signs; some may recover without any signs. In camel, the period between initial infection with *Trypanosoma* and the onset of the clinical signs is extremely variable, but generally ranges between 5 and 60 days. However, longer periods such as 3 months have been reported (Aha, 2005). Furthermore, the interval between the infection and detection of parasites in the blood stream is usually less than 14 days (Aha, 2005).

A number of factors that can increase the severity of the disease such as initial infective dose, stress, pregnancy, lactation, infection with another parasite, especially helminthes, malnutrition and travelling for long distances (Getahun and Demeke, 1998). Surra affects camels of all ages with a higher incidence rate in young camels shortly after weaning (Evans *et al.*, 1995). Additionally, the severity of the disease depends also on the number of vectors that an animal experiences in a given time. In camels the disease is manifested by elevation of body temperature, which is directly associated with parasitaemia. Infected animals show progressive anaemia, marked depression, dullness, loss of condition, and often rapid death. Anaemia was observed to be a major clinical finding in camel trypanosomiasis (Rami *et al.*, 2003). Some camels develop oedema, urticaria plaques and petechial haemorrhages in serous membranes. Death

finally follows if animals are not treated (Tuntasuvan *et al.*, 1997). Other signs are also reported such as abortion (Lohr *et al.*, 1986), reduced draught power (Luckins, 1998) and nervous signs like circling movement and trembling, unusual aggressiveness, aimless running and sudden collapse in severely stressed and overworked animals (Manuel, 1998). At post mortem, necrotic foci in the liver and spleen as well as generalized lymphoid tissue hyperplasia are common in camels suffering from surra (Rottcher *et al.*, 1987).

1.10. Immunity against *T. evansi*

An increase in gamma-globulin (IgM) during both acute and chronic *T. evansi* infections in camels has been reported (Boid *et al.*, 1981). However, these antibodies are not protective, as the majority of these antibodies are auto antibodies (Anosa, 1988). Increasing in leucocytosis, neutrophilia and eosinophilia have been reported in infection with *T. evansi* in camels (Anosa, 1988). Generally, the eosinophilia is a feature of parasitic infection and is associated with immediate-type hypersensitivity reactions. In the acute phase of the disease, lymph nodes and spleen are remarkably reactive. This may account for the generalized lymphoid tissue hyperplasia, which is a characteristic sign for *T. evansi* infections, while in the late stage of infection the immune system become depleted of lymphoid cells (Losos, 1980).

Circulating and tissue-mediated immune complexes have been demonstrated in laboratory animals infected with *Trypanosoma*. These immune complexes are likely to have wide varying pathological effects, including anemia, tissue damage, vascular dilatation and increased permeability (FAO, 1979).

The host immune response to a variety of antigens has been found depressed in animals infected with *Trypanosoma* under experimental conditions (Baltz *et al.*, 1981; Anene *et al.*, 1989; Enwezor and Ekejindu,

1998). Several hypotheses have been put forward to explain trypanosome-induced immune suppression, and the most favored appeared to be the action of Trypanosome enzymes, such as phospholipases (Tizard *et al.*, 1978), neuraminidases (Esievo, 1983) and proteases (Lonsdale-Eccles and Grab, 1986) which all have been implicated in membrane fluidity and cellular damage.

1.11. Diagnosis of trypanosomiasis

Many diagnostic techniques including parasitological and serological ones were devised. The techniques differ in their reproducibility, specificity and sensitivity and each can be applied according to the prevailing situation (Abdel Rahman *et al.*, 2001). However, characteristic clinical symptoms of emaciation and anemia are still used for the provisional diagnosis of the disease (Luckins *et al.*, 1979).

1.11.1 . Parasitological diagnosis

Parasitological methods include microscopic examination of blood, parasite concentration techniques and animal inoculation. The easiest and most frequently used of the three techniques is a direct microscopic examination of blood. In the wet blood smear, the trypanosomes are seen between blood cells (FAO, 2000). Haematocrit centrifugation is one of the concentration techniques that have been used to detect trypanosomiasis (Kihurani, 1995).

1.11.1.1. Wet blood films

A small drop of blood is placed onto a clean glass slide and covered with a cover-slip to spread the blood as a monolayer of cells. The slide is examined using light microscopy (x200) to detect any motile trypanosomes. Although, this technique is simple, inexpensive and gives immediate results, it is not enough to identify the species of the trypanosome properly. Final confirmation of the species is made by the examination of the stained smears. Although, the sensitivity of this method

is generally low, the examines experience and the level of parasitemia can improve the sensitivity (OIE, 2013).

1.11.1.2. Stained Thick smears

Smears are made by placing a drop of blood on a clean microscope slide. By using the corner of another slide, the blood was spreading on an area of approximately 2 cm. The film is dried by rapidly waving in the air, and without fixation the smear is stained for 30 minutes using 4% diluted Giemsa (diluted in phosphate buffered saline, pH 7.2). The stained smear will be washed with buffered water and examined at $\times 1000$ magnification. This method is simple and relatively inexpensive, but the result is delayed because of the staining process. Trypanosomes are easy to recognize depend on the general morphology (OIE, 2013).

1.11.1.3. Stained Thin smears

A small drop of blood is placed on the clean microscopic slide around 20 mm from the edge and spread an angle of 30 using another microscopic slide to obtain a thin smear. The film is air-dried briefly and then fixed using methyl alcohol for 2 minutes and then allowed to dry. The smears are stained using 4% diluted Giemsa for 30 minutes. The stain will be washed off using distilled water. The stained thin film will be viewed under a light microscope using X100 oil immersion objective lens. This technique permits morphological detail and identification of the trypanosome species. Usually, both a thin and thick smear is made from the same blood sample. Thick smears contain more blood than thin smears, thus it has a higher diagnostic sensitivity. On the other side, thin smears allow the identification of the species of *Trypanosome* (OIE, 2013).

1.11.1.4. Haematocrit centrifugation technique (HCT)

The procedure was described by Woo (1970). Briefly, each capillary tube will be filled with approximately 70 μl of EDTA fresher blood and centrifuged at 12,000 g for 5 min. The capillary tube is then gently snapped

at the junction between the lower Buffy-coat and RBC layer using a diamond pen. Plasma and Buffy-coat layer will be evacuated onto a clean slide, covered by 18x18 mm cover slip and examined under microscope using x100 or x400 objective lens. This technique can detect around 50–200 trypanosomes/ml of blood (Desquesnes and Tresse, 1996).

1.11.1.5. Animal inoculation

Laboratory animals may be used to reveal sub-clinical infections in domesticated animals and also when highly sensitive detection is required. This method is able to detect as few as 1.25 *T. evansi*/ml blood (Reid *et al.*, 2001). *T. evansi* has a broad spectrum of infectivity for small rodents, rats and mice. Rodent inoculation is not 100% sensitive, but further improvement in its efficacy can be obtained by the use of Buffy coat technique (Monzon *et al.*, 1990).

1.11.2. Serological diagnosis

Generally, serological tests are useful to apply for prevalence or incidence studies, seasonal or inter-annual variations and for vector control (Desquesnes *et al.*, 2011). Different methods have been used to detect either specific humeral antibodies or trypanosomal antigens, such as direct or indirect agglutination tests, complement fixation test (CFT), IFAT (Desquesnes, 1997; Uilenberg, 1998), ELISA and CATT (Desquesnes *et al.*, 2011).

1.11.2.1. Indirect fluorescent antibody test (IFAT)

Although the technique is not usually applied to large-scale surveys, it is still useful to screen a small number of samples. The IFAT- *T. evansi* sero-conversion can be used after 60–90 days from the infection (Jacquiet *et al.*, 1993). Compared with the CATT, IFAT is more sensitive, probably because it can detect infections with low parasitaemia in animals (Dia *et al.*, 1997).

1.11.2.2. Enzyme-linked immune sorbent assay (ELISA)

The principle of this technique is that specific antibodies against trypanosomes can be detected by using enzyme-linked anti-immunoglobulin and solid-phase polystyrene plates coated with soluble antigen (Warburg and Christian, 1942).

Soluble antigens from whole lysate of *T. evansi* are able to detect the immunoglobulin's present in blood of the host infected with *T. evansi*. This method can also detect the infections with *T. vivax*, *T. congolense* and *T. cruzi*. The technique is fast and allowing between 500–1000 samples to be tested daily (Laha and Sasmal, 2008).

1.11.2.3 . Card agglutination tests (CATT)

The CATT-*T.evansi* is a quick and easy test which can be performed under field conditions for serological diagnosis of Surra in camels (Luckins, 1988). Demonstration of trypanosomal antigens in the blood of the infected animal would be completed with a parasitological diagnosis (Voller and Desavigny, 1981).

1.11.2.4. Complement fixation test (CFT)

The complement fixation test (CFT) is one of the first techniques used in the diagnosis of *T. evansi* in camels. The test is also used successfully for the detection of *T. equiperdum* in horses. However, cross reaction with sera of horses infected with other trypanosomes may be occurring. Comparing with ELISA, CFT is less sensitive in the diagnosis of the Dourine disease in equines (Wassal, 1998; Gillbert *et al.*, 1998).

1.11.3. Molecular diagnosis

Molecular techniques are suitable for detecting parasites in the mammalian host and in the insect vector and currently are the main research tools. The Polymerase Chain Reaction (PCR) is based on the use of the enzyme DNA polymerase that will amplify the sequences of DNA bases, until sufficient DNA is produced to be detectable (Desquesnes *et al.*, 2001). A PCR

technique proved to be suitable for the diagnosis of *T. evansi* infection in camels. This technique has 90 % sensitivity as compared with other parasitological and serological tests which suggests that PCR can be a useful diagnostic tool for detecting *T. evansi* infected in the very early stages where the microscopic examination is confusing (Muhammad *et al.*, 2010).

1.12. Control of trypanosomiasis

1.12.1. Chemical Control

Various chemical compounds are being used for the treatment of trypanosomiasis. Among different compounds, the most widely used trypanocide compound is diminazene aceturate. Besides diminazene aceturate, other chemical compounds like isometamidium chloride, suramin, quinapyramine sulphate (curative), quinapyramine chloride (prophylactic) and cymelarsan (only for camel) are also available. The prophylactic drugs which are used as chemoprophylaxis. They not only kill parasites, but also prevent any new infection, due to the remanence of the sustainable in the blood of animals (Dia and Desquesnes, 2004).

1.12.1.1. Therapeutic and Chemoprophylactic Drugs

Therapeutic drugs aim to eliminate parasites from a sick animal. Although, Diminazene Aceturate (DA) is the most widely used therapeutic drug against Surra. Other drugs can be used also such as Isometamidium Chloride (IMC), Melarsomine dihydrochloride (cymelarsan), Suramin, and Quinapyramine (Dia and Desquesnes, 2004).

1.12.1.2 . Strategies for the Use of Trypanocides

The recommended drug for trypanosomiasis in buffalo, cattle, sheep and goats is diminazene aceturate with dose rate of 7 mg/ kg injected deep intramuscularly. Resistance against diminazene aceturate in trypanosomes has been reported in different parts of the world (Desquesnes, 2004; Peregrine and Mamman, 1993). If animal doesn't respond then,

isometamidium chloride or melarsomine hydrochloride may be used at dose rate of 0.5 mg kg⁻¹ b.wt. deep intramuscularly (Desquesnes, 2004). Alternate use of diminazene aceturate and isometamidium chloride was recommended, as these make a sanative pair, means that once resistance develops to one of the drugs, the other drug will be used to control the infection. Horses, dogs, and cats can be treated with diminazene aceturate or isometamidium chloride but adequate water should be provided to animal to avoid a toxic effect on the kidneys. Melarsomine dihydrochloride is the best drug for camel at the dose rate of 0.25-0.5 mg kg⁻¹ b.wt. Although other trypanocide drugs could also be used (Desquesnes *et al.*, 2013b). Melarsomine dihydrochloride is not recommended in buffaloes if treated with 0.75 mg kg⁻¹ body weight because chances of development of nervous signs.

1.12.2. Prevention and Control of trypanosomiasis

Not a single promising experimental result to develop a vaccine could be obtained till date because of the capacity of parasite to modulate its own antigen termed as antigenic variation that means ability of parasite to regularly switch its surface coat glycoprotein. Another reason is that the parasite undermining the host's capacity to mount an efficient immune response and to maintain its immunological memory termed as immunodeficiency to the host (Pays *et al.*, 2004). Although a solitary report, wherein formalin inactivated *T. evansi* (2×10^6 count) were administered in mice and found protective against homologous challenge (Tewari *et al.*, 2009). Immunization with *T. evansi* recombinant beta-tubulin, induced some protection against *T. evansi*, *T. equiperdum* and *T. brucei* infection in mice (Li *et al.*, 2007). Control of *T. evansi* includes use of anti trypanosomal drugs for therapeutic and prophylaxis uses, vector control and use of trypanotolerant breed (applicable for cattle) (Tewari *et*

al., 2012). Various chemical compounds may be used for treatment and some have prophylactic property (e.g. quinapyramine chloride).

1.12.2.1. Vector Control

Control of vector also reduces the prevalence of trypanosomiasis.

Glossina spp. can be easily controlled by using insecticide impregnated screens and insects sterilization techniques in livestock breeding areas (Rodtian *et al.*, 2012). Control of tabanid flies is difficult because of its high mobility and prolificacy and the larval stages of fly are generally spread over a wide area (Foil and Hogsette, 1994). Although control of tabanid flies is found efficient by insecticide sprays in small closed deforest areas. *Stomoxys* is another fly responsible for transmission of trypanosomiasis. This fly develops within the livestock area or the farm and is closely related to the farming systems and it can be controlled by trap systems and insecticides prays on animals (Foil *et al.*, 1991; Leprince *et al.*, 1991) or use of fly proof system with mosquito net.

1.12.2.2. Other Methods for Prevention

In situations where it is difficult to control the biting insect populations, it may be easier to control transmission. *Tabanids* are naturally persistent feeders and they do not leave the animal to bite another if the latter one is more than 50 meters away (Foil *et al.*, 1985). Therefore, 200m is considered to be a safe distance to control the mechanical transmission by biting insects (Foil, 1983; Foil *et al.*, 1985; Barros and Foil, 2007).

However, separating bovine from equine is highly recommended to prevent the transmission of *T. evansi* from a reservoir such as buffalo or cattle to the highly sensitive host such as camel and horses. Thus, it is advisable to housing the cattle and camel/ horses in completely different areas and the separated at least by several kilometers (Da Silva *et al.*, 2007; Vergne *et al.*, 2011).

As mentioned before carnivores may be infected when they eat the bones, flesh, or blood of an infected animal that has only just died. Rodents, which are omnivorous, may become infected like carnivores and get the infection via oral as demonstrated in a trial (Da Silva *et al.*, 2007; Vergne *et al.*, 2011). To control this type of transmission, the dead animal carcasses should be eliminated as soon as possible and the stray dogs, around the slaughterhouses, as well as around the farms should be controlled (Gutierrez *et al.*, 2010).

CHAPTER TWO

2. MATERIAL AND METHODS

2.1. Study Area

This study was conducted at Tamboul livestock market, which is located 3 km from the Faculty of Veterinary Medicine, University of Albutana, Sudan from December (2018) to November (2019) . Tamboul is located in the east part of Gezira state around 150 km south East Khartoum (Fig1). The district is located at latitude 14° 52' N and longitude 33° 31' E, (Fig 1).

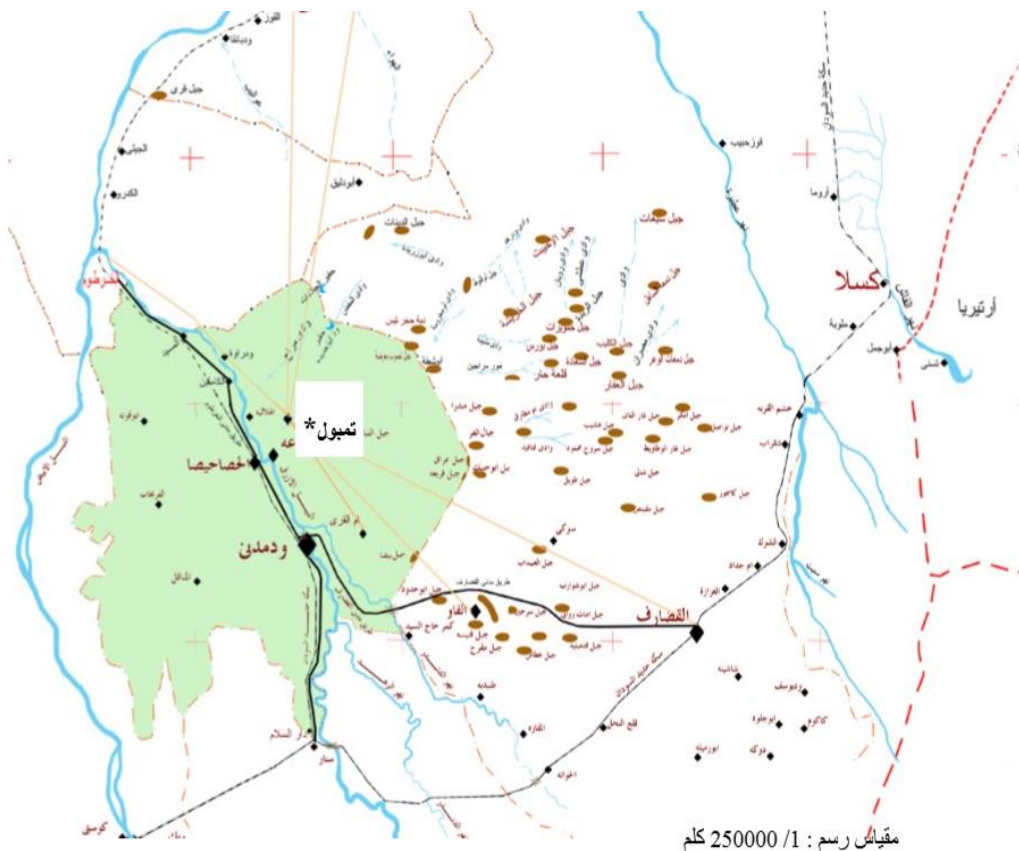


Fig (1): Map of the study area (Butana area –Taboul area)

Source: Government of Gezira State.

2.2. Study type

The study was a cross-sectional study to estimate the prevalence of camel trypanosomiasis , and the risk factors associated with the disease.

2.3. Sampling Method and Sample Size Determination

By using systematic random sampling methods the samples were selected. The size of the sample was determined by using the formula of Thru field (2007) and 95% confidence interval as following:

$$n=1.96^2 \cdot p_{exp} (1-p_{exp})/d^2$$

n = sample size

P_{exp}= expected prevalence

1.96 = the value of 95% confidence interval

d = desired accuracy level at 95% confidence interval

In a previous study performed in Tamboul livestock market, (Alsrug, 2017) found that, the prevalence of camel Trypanosomiasis was 4.8%. Referring to that, the size of the sample was calculated as following:-

$$n = 1.96^2 * (0.048) * (0.952)/(0.0025) = 71 \text{ animal samples.}$$

To increase the precision of the study, the sample size was multiplied by three. So the size of the sample became 213 samples.

2.4. Collection of blood samples

A total of 213 blood samples were collected from camels of examined for the presence of trypanosomes. three ml of blood were collected from the jugular veins of animals into a heparinized vacutainers tube (5ml). The tubes were labeled with a numbers, kept in ice box and were transported immediately to the laboratory of, Al Butana University Faculty of Veterinary Medicine.

2.5 . Diagnosis of Trypanosomiasis

2.5.1 . Thin blood smears

The Thin blood smears were performed as described by Murray *et al.* (1977). A small drop of blood was placed onto a clean microscope slide around 20 mm from the edge and then spread with a glass spread an angle of 30 to obtain a thin smear. The film was air-dried briefly and then fixed using absolute methyl alcohol for 2-3 minutes and then allowed to dry. The smears were stained using 4% diluted Giemsa for 30 minutes. The stain was washed off using distilled water. After air-drying the slides were examined under oil immersion objective lens (100x) to detect and identify the *Trypanosoma* species based on their morphological characters.

2.5.2 . Microhaematocrit centrifugation technique (MHCT)

heparinized capillary tube (75 × 1.5 mm) were filled with blood until two thirds its size and were then sealed from one side using crestaseal. The tubes were centrifuged for 4 minutes at 12,000 rpm/ for 5 minutes using microhaematocrit centrifuge. After centrifugation, the capillary tubes were placed on a microscopic slide and the interphase between the buffy coat layer and the plasma was examined under a microscope using 10X objective lens (Wernery *et al.*, 2001). After examination the capillary tubes was cut at 1mm below the buffy coat to include the upper layers of red blood cells. The contents of the capillary tube were gently spread onto microscopic slide, and then covered with a cover slips. The slides were examined under a microscope for the presence of the motile trypanosomes using 10x or 40x objective lenses (Murray *et al.*, 1977).

2.6. Collection of flies

Nzi trap was made of blue cotton, local white nylon mosquito netting and local matt black cotton cloth. The blue cloth was imported from Kenya (United Textile Industries Nairobi, Kenya) following ICIPE scientists

(Mihok,2002).The Flies were collected used Nzi trap two days a month from the researches of Tamboul, and University of Al-Butana, Faculty of Veterinary Medicine killed by isolation in plastic bottles and transferred to the Tsetse and Trypanosomiasis Control Department Veterinary Research Institute, Khartoum, where they were identified and counted (Fig 2).



(Fig2). A model of Nzi traps for capturing the vectors of trypanosomiasis.

2.7. Questionnaire

A questionnaire was designed to provide information about potential risk factors hypothesized to be associated with trypanosome infection. Data regarding the characteristics of individual camels, including age, sex, breed, body condition, and sources of animals were obtained by asking the owner during the collection of blood sample.

2.8. Data analysis

The overall prevalence was calculated by dividing the number of positive results by the total number of animals examined in this study. The whole data (laboratory results, risk factors such as age, breed.... *etc.*) were entered into Microsoft excel sheets (Microsoft office excel 2007) and then transferred to SPSS version (16.0) for statistical analysis. Data were analyzed by simple descriptive statistic using relative frequencies, cumulative frequencies and cross tabulation. A univariate analysis was then used to estimate the strength and statistical significance of the association between risk factors and the disease. Chi -square (X^2) test was used to determine the significance (p-value < 0.05) of each factors.

CHAPTER THREE

3. RESULTS

3.1. Overall prevalence of trypanosomiasis

Out of 213 examined animals 11 were found positive for Trypanosome evansi. The overall prevalence of camel trypanosomiasis in Tamboul market, Sudan was 5.2% (Table 1).

Table (1): The overall prevalence of camel trypanosomiasis (n=213) in Tamboul market Gezira state

Result	Frequency	Cumulative frequency%
+ve	11	5.2
-ve	202	94.8
Total	213	100.0

3.2. Distribution of the camels examined for Trypanosomiasis according to the potential risk factors in Tamboul market

Regarding animals age, 3(%) out of 58 young animals, 3(%) out of 78 adult animals and 5(%) out of 77 old camels were found to be infected with *T. evansi* (Table 2).

As for sex, the number of positive samples was 10(%) females out of 165 examined and for the 48 males only 1(%) sample was positive (Table 2).

Considering the breed, blood samples were taken from 51 Butana breed, 85 Darfur breed, 66 Kassala breed, 11 Kenana breed of camels; Results showed that 4(%) camels of Butana breed, 2(%) of Darfur breed, 4(%) of Kassala breed and 1(%) of Kenana breed were infected with Trypanosomiasis (Table 2).

According to the body condition, the number of positive samples was one out of 38 in the group of fat body condition, 8(%) positive samples out of 127 in the group of normal body condition, and 2(%) positive samples out of 48 in the group of poor body condition (Table 2).

Regarding seasonality the number of samples in dry hot , wet cold and cold dry were 71 and the parasites were found in 5(%), 3(%) and 3(%) samples respectively (Table 2).

Table (2): Distribution of camels (n= 213) examined for Trypanosomiasis according to the potential risk factor inTamboul market, Gezira state

Risk Factor	No. Tested	No. +ve
Age		
Young <5	58	3
Adult 5-10	78	3
Old >10	77	5
Total	213	11
Sex		
Female	165	10
Male	48	1
Total	213	11
Breed		
Butana	51	4
Darfur	85	2
Kassala	66	4
Kenana	11	1
Total	213	11
Body condition		
Fat	38	1
Normal	127	8
Poor	48	2
Total	213	11
Season		
Dry hot	71	5
Wet cold	71	3
Cold dry	71	3
Total	213	11

3.3. Examination of blood samples

Infection with *T. evansi* was diagnosed by examination of 213 camels using two different techniques, Giemsa's stain (**Fig 3**) and MHCT (Buffy coat techniques) (**Fig 4**). The numbers of positive samples detected in this study were 11 by using both Giemsa's stain and Buffy coat techniques.

Table (3): Prevalence of infected camels (n=213) with *T.evansi* using Giemsa stain and buffy coat in Tamboul market Gezira state

Result	N.using Gernsa Stain	N.using Buffy Coat	Prevalence %
+ve	11	11	5.2
-ve	212	212	94.8
Total	213	213	100

3.4. Risk factors analysis

Table (4): Summary of univariate analysis of the risk factors associated with trypanosomiasis in Tamboul market Gezira state

Risk factors	No. tested	No. +ve %	D.f	Chi	P.value
Age					
Young <5	58	3 (5.2%)	2	0.554	0.758
Adult 5-10	78	3 (3.8%)			
Old <10	77	5 (6.5%)			
Sex					
Female	165	10 (6.1%)			
Male	48	1 (2.1%)	1	1.201	0.273
Breed					
Butana	51	4 (7.8%)	3	2.574	0.462
Darfur	85	2 (2.4%)			
Kassala	66	4 (6.1%)			
Kenana	11	1 (9.1%)			
Bodycondition					
Fat	38	1(2.6%)	2	0.929	0.628
Normal	127	8 (6.3%)			
Poor	48	2 (4.2%)			
Season					
Dry hot	71	5 (7.0%)	2	0.767	0.682
Wet cold	71	3 (4.2%)			
Cold dry	71	3 (4.2%)			

According to age factor, the highest infection rate was detected in old camels (6.5%) followed by young (5.2%) and finally adult (3.6%). There was no significant association observed ($\chi^2=0.554$; $P = 0.758$) (table 4).

In relation to sex factor, female animals had higher prevalence rate (6.1%) than males (2.1%), no significant association was observed ($\chi^2= 1.201$; $P =0.273$) (Table 4).

As for breed the highest prevalence rate was detected in Kenana (9.1%) followed by Butana (7.8%) then Kassala (6.1%) and finally Darfur(2.4). The result showed that there was no association between *trypanosoma* infectin and breed observed ($\chi^2= 2.574$; $P =0.462$) (Table4).

Considering body condition of breed animals, fat animals had lower prevalence (2.6%) followed by poor and normal body condition animals (4.2% and 6.3%), respectively the result show no significant association was observed ($\chi^2= .929$; $P =.628$)(Table 4).

Regarding Season, in this study the higher prevalence was detected in the dry hot season (7.0%) followed by the wet cold and the cold dry seasons,(4.2%) There was no significant association observed ($\chi^2= 0.767$; $P = 0.682$) (Table4).

3.5. Classification of flies

The most abundant species of biting flies during the whole collection period was *T.sufis* (31%). They were highly abundant during April (31%) and least during May and June. Other species were *T.agrestis* and *T.taeniola* and their highest proportion was (22.6%) and (21.8%) and were during October, and November respectively.

Table (5): Relative abundance of biting flies (n=181) collected at species level during different months around Tambuol market Gezira state

Month	<i>T.sufis</i>	<i>T.taeniola</i>	<i>T.agrestis</i>	Mean	S .d
January	4 (5.4%)	2 (6.25%)	5 (6.66%)	2.1	.94
February	7(9.4%)	5 (15.6%)	7 (9.33%)	2	.88
March	8 (10.8%)	2 (6.25%)	3 (4%)	1.6	.87
April	23(31%)	3 (9.37%)	14 (18.7%)	1.77	.95
May	0 (0%)	0 (0%)	1(1.3%)	3	0
June	0 (0%)	0 (0%)	4 (5.33%)	3	.000
July	2 (2.7%)	1(3.12%)	5 (6.66%)	2.37	.92
August	2 (2.7%)	0 (0%)	1(1.3%)	1.66	1.2
September	2 (2.7%)	4 (12.5%)	5 (6.66%)	2.27	.79
October	2 (2.7%)	2 (6.25%)	17 (22.6%)	2.7	.64
November	15 (20.27%)	7 (21.8%)	6 (8%)	1.67	.82
December	9 (12.16%)	6 (18.7%)	7 (9.33%)	1.9	.87
Total	74	32	75		

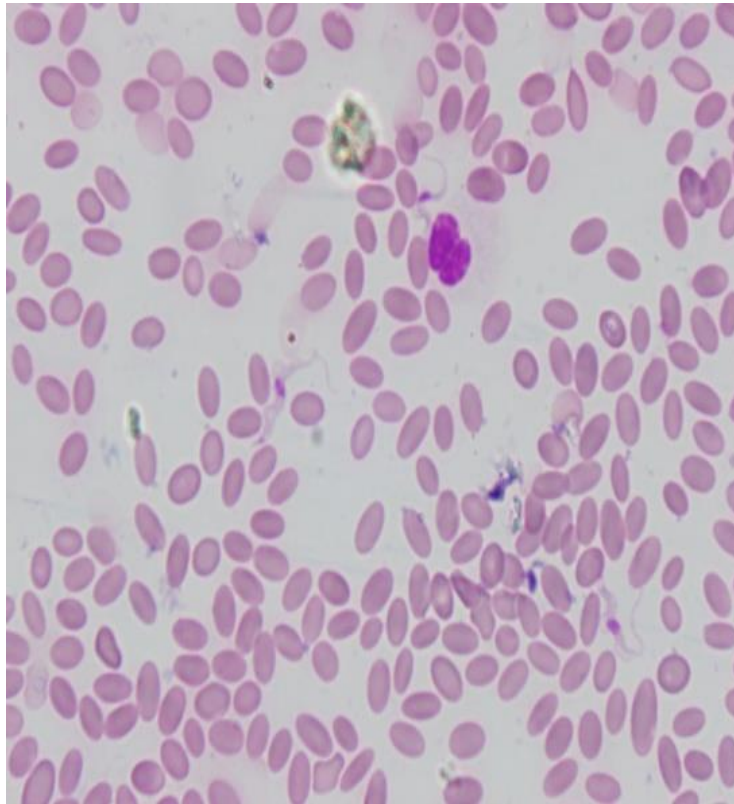


Figure (3): Photomicrograph of camel blood sample infected with *Trypanosoma evansi* and stained with Giemsa using light microscope (100×) in Tamboul market, Gezira state

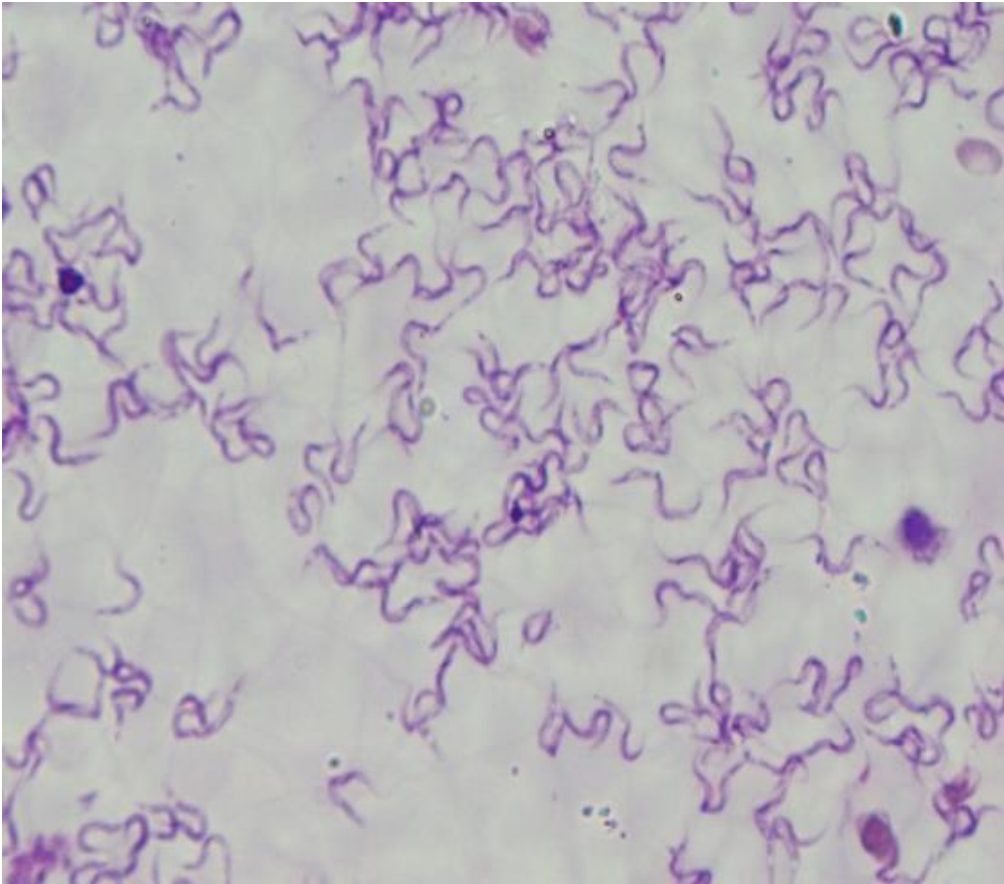


Figure (4): Photomicrograph of camel blood sample infected with *Trypanosoma evansi* using buffy coat, in Tamboul market, Gezira state



Figure (5): *Tabanus Sufis* was obtained from Tambuol region, Gezira state



Figure (6): *Tabanus atylotus agrestis* was obtained from Tambuol, Gezira state



Figure (7): *Tabanus taeniola* was obtained from tambuol, Gezira state

CHAPTER FOUR

4. DISCUSSION

Trypanosomiasis is a disease affecting the immune system of the host animal. *T. evansi* as purely extracellular parasites are permanently confronted with the multiple components of the host's immune system ranging from innate to adaptive immune defenses (Eyob and Matios, 2013.) In our study prevalence and risk factor of camel trypanosomiasis were investigated. The prevalence in our study was 5.2% and this indicate the importance of the disease in the area, this result is in agreement with the study which was carried out by Khalid (2015) who surveyed camel trypanosomiasis in Butana, Gedarif state, where the prevalence was 5.1%. On the other hand, our result was higher than that demonstrated by Alsrug (2017), where the prevalence was 4.8% and lower than that reported by Ibrahim *et al.* (2015) where the prevalence was 6.08%. This difference may be due to the difference in the number of camels investigated in each study, which may be reflected in the prevalence rate of the disease.

In the present study, the univariate analysis of the risk factors showed that there was no statistical significant ($p\text{-value} < 0.25$) association of the studied factors with trypanosomiasis infection. In this study a high infection rate was found in old camels >10 years (6.5%) and young <5 years (5.2%) than the adult 5-10 years (3.8%), this result agrees with that of Basaznew Bogale *et al.*, (2012). The higher prevalence of trypanosomiasis in old camels might be due to the heavy stress through their use for transportation and long-term exposure under poor management and exposure to the vector, young animals become susceptible to infection after weaning and thus losing the maternal immunity transferred to them through colostrum.

The present results showed that the prevalence rate was higher in females (6.1%) than males (2.1%). Similar results were reported by Najira *et al.*, (2003) and Eldaw (2009), who investigated camel trypanosomiasis in Kenya and Tamboul area in Sudan, respectively. This observation may be attributed to the stress induced by pregnancy and lactation (Tayib *et al.*, 2015). Moreover, the majority of animals slaughtered during this study period were females.

As for breed, the higher infection rate detected was 9.1% in Kenana and 7.8% in Butana, than Darfur (2.4%) and Kassala (6.1%) breeds. This result could be due to the fact that Kenana breed is used for transportation and working than other breeds, and kenana breed was the lower number of camels examined from this area.

Regarding body condition in this study. The infection rate was higher (6.3%) in animals with normal body condition, than in fat (2.6%) and poor (4.2%) body conditions. This could be due to the chronic nature of the disease.

Concerning seasonal distribution, our results showed that the prevalence of camels' infection was high in dry hot season (7.0%), compared with the wet cold (4.2 %) and dry cold season (4.2%). This could suggest that these animals may become infected by the end of wet cold season and during the dry cold when the highest fly count that acts as *T. evansi* vectors which is usually expected during the rainy season. The high prevalence in dry hot season could be explained by the fact that the owners in Butana usually take their animals in this season to the river Nile, Dindir national park or even to the Ethiopian border areas which are also favourable grounds for these flies.

In our study examination of blood samples of camels with both stained blood smear and buffy coat technique revealed an infection rate of (5.2%). This result indicates that the stained smear is specific as well as buffy coat

technique in diagnosing camel trypanosomiasis which is also at tribute to the good handling of the samples when preparing them at the laboratory.

In this study, a few number of *Tabanus* species were collected, which may probably be attributed to the low prevalence of *Trypanosoma evansi* in the study area.

The presence of *Tabanus* species all the year round ensures that the transmission of the parasite occurs wherever there is co-existence of reservoir hosts and susceptible hosts. Sporadic occurrence of the disease during the dry season and outbreaks during the rainy season has been reported to be associated with the abundance of *Tabanus* species (Njriu *et al.*, 2002). However, the efficiency of the different flies in transmitting *Trypanosoma evansi* reported is due to variation in geographic conditions and is also dependent on the interval between two successive feeds and the intensity of the flies' challenge (Lukins, 1998).

CONCLUSIONS AND RECOMMENDATIONS

CONCLUSION

The results showed that the overall prevalence rate of camels infected by *T.evansi* was 5.2 % by using Both Giemsa stained smear and buffy coat methods. There were no significant differences between the risk factors associated with the disease.

Diagnosis of the disease by microscopic examination and hematocrit centrifugation are the easiest and quick methods for detection , The vectors that transmitted the disease were identified in the area. whereas identification of vectors of importance because the disease leading to much greater loss amongst camels health and productivity.

RECOMMENDATIONS:

The following points are recommended:

- More sensitive diagnostic methods such as PCR could give a higher prevalence rates.
- Prevention and control measures should be designed against the parasite and their vectors to minimize the disease.
- Developing of control measures in affected herds to avoid spreading of the disease.
- Further epidemiological studies should be conducted in areas where surra or Camel trypanosomiasis is endemic in order to determine the impact of the disease on the productivity as well as its economic impact in the country.
- The vector that transmitted the disease were identified in the study area. Identification of Vector is of importance because trypanosomiasis leading so much greater loss amongst Camels health productivity.

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