

A Comparative Field Survey of Iraqi Camels with Trypanosomiasis between Karbala Governorate and Najaf Governorate

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Abstract

This field survey was conducted to find out the prevalence of trypanosomiasis in Iraqi camels in the governorates of Karbala and AL- Najaf and to find out which of the two governorates has the highest incidence of infection. Collected (300) blood samples randomly for the period from (1/7/2018 to 31 /7/2019). The samples were collected from camels that graze naturally in the pastures of the Western Badia in the city of AL-Najaf , the Sea of AL-Najaf , the natural pastures in Karbala ,from the camels brought for treatment in the veterinary clinic in AL-Najaf and Karbala and from the camels brought for slaughter in the slaughterhouses of AL- Najaf and Karbala. The camels from which blood samples were collected, some of them were in good health others suffered from respiratory and digestive diseases, had skin diseases and various wounds. Some of them have been observed to have accumulation of edema in different areas of the animal's body. The ages of the camels ranged between (1 year - 18 years). After collecting the blood samples, wet smears were made, thick and light dry blood smears were stained with Leishman stain to view the parasite, and a mercury chloride test was done. By examining the smears of all the samples, it was found that there were (191) healthy animals by (63.7%), and (109) infected animals by (36.3%), where the number of infected females was higher than the number of infected males in number (78) by (26) While the number of infected males was (31) by (10.3%). Thus, the results of the statistical analysis show the existence of a significant statistical difference at the level of ($p \leq 0.01$) between males and females. The highest infection rate was in camels whose ages ranged between (13 years - 15 years) with a number (26) by (8.6%), then came in the second degree for camels whose ages were (10 years - 12 years) with (24) by (8%), while the lowest rate was recorded in aged (12-15 years) with a number (9) by (3%). Thus, the results of the statistical analysis show the existence of a significant statistical difference at the level of ($p \leq 0.01$) between the camel groups according to the age group. As for the results of trypanosomiasis in the governorates of Karbala and AL-Najaf, the number of infection in the governorate of AL-Najaf was higher than the number of infection in the governorate of Karbala, the number of infected camels in the governorate of AL-Najaf was (66) infected animals by (22%) of the total (150) blood samples that were examined and the number of infected camels in Karbala governorate is (43) infected animals by (14.3%) of the total (150) blood samples. Thus, the results of the statistical analysis show the existence of a significant statistical difference at the level of ($p \leq 0.01$) between the infection rate in the two governorates .

Key words: Iraqi camels, field survey, trypanosomiasis, comparison, Karbala, AL-Najaf, Governorate.

Introduction

Camels have been known since ancient times as a ruminant animal that spreads in different parts of the world and is associated with its presence in desert regions from Morocco to China ⁽¹⁾ Arabian camels are herbivorous animals that feed mainly on thorny plants, dry herbs, and cysts of salt; However, they are not selective and

feed on every type of plant that grows in the desert ⁽²⁾. The Arabian camel is a herding animal in general. Herbs and weeds make up about (70%) of its diet. It spends (8 to 12) hours per day grazing, corresponding to an equal number of hours it spends ruminating ⁽³⁾. When feeding, camels spread across a vast area of land, picking only a few leaves from each plant, and this form of scavenging

reduces pressure on plants and reduces competition with other types of municipal herbivores⁽⁴⁾. For camels, this scavenging form reduces the risk of poisoning from any fluids that the plant secretes to protect itself^[3,4]. These animals need to consume salt that is six to eight times more than the amount needed by other animals, in order to be able to preserve and store water^[3,4]. As a result, a third of their food must be halophytes. Arab camels can graze trees up to a height of (3.5) meters, by breaking the branches and stripping them in one movement^[4]. The camels use their lips to grasp their food, and then chew each bite for between (40 and 50) times. They keep their mouth open when they chew the spiny plant^[3]. Trypanosomiasis is one of the most important diseases that camels are exposed to in many parts of the world because of this disease has important pathological effects on camels represented by recurrent miscarriage and immunosuppression and its effect on the productive and reproductive efficiency of animals^[5,6] The disease is widespread among camels in the Middle East region. If the infection is neglected without treatment, it is fatal^[7,8]. It is called by several names according to the regions and the different dialects of the Badia. Among these names are: Surra, Zorji, or Dabab. The cause of the disease is a single-celled parasite belonging to the Trypanosoma family called Trypanosoma Evansi, which is a protozoan^[9]. This disease was first recorded in India in (1880) and was called Sora disease. The vector of disease is flies of the genus Tabanus, or the bloodsucking bat in South America by automatically transferring the parasite from the infected animal to the susceptible animals^[9,10]. In a study proved that horned flies (Lyprosia) transmit the parasite to different animals^[11]. One of the signs of the acute phase of the disease is camels' fear of light, which is called in the desert (camels deviate from the sun, according to the breeder's expression)^[12]. The predominant form of the disease is the chronic phase^[13,14] that may last for three years, during which the animal will notice anemia, general sluggishness, lack of appetite, and skin ulceration with the appearance of skin crusts and the creation of a suitable environment

for infection with external parasites such as mites^[5]. Among the important pathological signs of the disease is edema in the lower areas of the body (the extremities), the front of the chest, the genitals of males and females, the eyelids, and the occurrence of recurrent miscarriage in pregnant females due to high temperature or the presence of antigens that prevent blood from reaching the fetus in the chronic phase. Haematuria is also noted^[15]. As for the subacute phase, the clinical signs in it are characterized by gradual wasting as a result of loss of appetite and intermittent fever associated with parasitic waves in the blood, and paleness of the mucous membranes as a result of anemia of varying degrees that occurs due to the decomposition of red blood cells as a result of infection with the parasite and also may notice hematuria on the animal^[16,17].

Materials and Methods

1. Research design:-

The research was designed on the basis of collecting blood samples from Iraqi camels, (300) blood samples were collected from camels randomly for the period of time from the beginning of (1/7/2018 to 31/7/2019). The samples were collected from camels that graze naturally in the pastures of the Western Badia in the city of Najaf, the Najaf Sea and Karbala also from camels brought for treatment in the veterinary clinic in Najaf and Karbala, from camels that brought for slaughter in the slaughterhouses of Najaf and Karbala. The camels from which blood samples were collected, some of them were in good health, others were ill, as they were afflicted with various respiratory and digestive diseases, various wounds and skin diseases, in addition to the presence of external and internal parasites, some of them were observed areas of edema in neck, in front of chest, genitals and limbs. Collecting (300) blood samples from camels of different ages ranging between (1 year -18 years) by (150) models for Najaf Governorate and (150) models for Karbala Governorate, then they were divided into six groups according to the age group, as shown in Table No. (1).

Table (1) shows the numbers of total blood samples tested for camels and the ages of both healthy camels and infected camels with trypanosomiasis in Karbala and AL-Najaf governorates

Age / year	Numbers of studied cases	Numbers of positive cases	Numbers of negative cases
1≤ year- 3 years	37	9	28
4years –6years	48	17	31
7years – 9 years	42	19	23
10years – 12 years	78	24	54
13years – 15 years	50	26	24
16years – 18 years	45	14	31
summation	300	109	191

2. Clinical examination and clinical signs:-

The clinical examination was performed after the camels were seated by tying one of their front limbs to allow them to sit, then tying the animal's head well so (the head and neck were bent towards the far side from the examiner), often linked to the back leg of the animal itself when it was in the desert, then the animal's mucous membranes, especially (conjunctiva) are examined, temperature, pulse and breathing speed are measured. Clinical signs are observed for each animal from which a blood sample is collected. We saw some of cases were afraid of light (photophobia), also some cases of recurrent miscarriage, enlarged lymph nodes and edema were recorded in the lower parts of the body such as the lower jaw front of the chest ,genitals and back legs.

3. Blood samples collection:-

Collected (300) blood samples for both sexes and for both governorates, the number of males was (123) and the number of females was (177). The blood was drawn by a syringe measuring (10) ml and needle (G22) to withdraw (5 ml) of blood volume from the jugular vein after sterilizing the site of collection by wiping it with ethyl alcohol. Put about (0.5-1) ml in a plastic tube containing an anticoagulant Ethylene diamine tetra acetic acid (EDTA) at a concentration of (1-2) mg / ml of blood, it combines with the calcium ions that enter the coagulation process, directly closure the tube then moved it carefully and quietly in the form of a number (8) several times to dissolve the anticoagulant and mix

well with the blood [18]. The other part of the blood is placed in special plastic tubes without anticoagulant, which are closed well and set aside in order to allow the blood to clot [18]. After collecting the samples, they are transferred as quickly as possible to the blood test laboratory in Al-Sadr Teaching Hospital in Najaf Governorate to examine the samples collected from Najaf Governorate or in the veterinary clinic laboratory in Karbala Governorate for samples collected from Karbala Governorate. Centrifugation (10,000) revolutions / minute for a period of (3-5) to separate the serum. Then it is withdrawn quietly by special sterile plastic droppers (single use), then placed in other sterilized and clean tubes [18].

4. Making blood smears:-

A- Wet blood smear.

A drop of blood containing an anticoagulant is placed on the slide and covered with the cover of the slide to observe the parasite's movement under the microscope with a force of magnification (40x), where the clear movement of the parasite is seen [19, 20].

B- Dry blood smear.

A drop of blood containing an anticoagulant is placed on a slide and drawn with a second slide at an angle of (45) to make a light blood smear and a thick blood smear, then fixed with absolute methyl alcohol at a concentration of (95%) and stained with Leishman stain in the laboratory of Al-Sadr Teaching Hospital in

Najaf Governorate or the laboratory of the veterinary dispensary in Karbala governorate, and then examined under the oily lens to see the parasite in the infected cases [21].

5.Preparation of the mercury chloride solution:-

The mercury chloride solution was prepared at a concentration of (1: 3000) by dissolving (0.03) mg of mercury chloride salt in one liter of distilled water. The serums of all samples of the collected animal blood were examined by placing (1) cubic millimeters of mercury chloride solution and then adding a drop of the animal’s blood serum into the solution and stirred for several seconds to minute, we see a white precipitate immediately in the positive cases, while the negative cases, no change occurs and the solution remains clear [22,9].

6.Statistical analysis:-

Use the ready-made SPSS statistical program on the calculator to analyze the statistical results.

Results and discussion :-

The results we obtained after examining (300) animals from camels for both sexes and for both governorates by (150) animals for each governorate, so it was found that there are (191) healthy animals by (63.7%) and (109) infected animals at a rate of (36.3%). The highest number and rate of infected was recorded in the governorate of AL-Najaf with (66) by (22%) while in Karbala governorate with (43) by (14.3%) as shown in Table no. (2) , this percentage is high compared to the study conducted by [23] in the central and southern regions in the country where the percentage was (11%) , also higher than the study [24] that was conducted for AL-Najaf governorate by (8.4%) and higher than the study [25] that conducted in the slaughterhouses in AL-Najaf governorate and Kufa, where it scored (12.8%), but this percentage was lower from a study [26] about the disease in the Qadisiyah governorate, the disease rate was recorded (45.1%).

Table (2) shows the numbers and percentage of total samples of camels and the numbers of positive and negative samples examined for both sexes in the governorates of Karbala and AL- Najaf

Governorate	Numbers of studied cases	Ratio %	Numbers of positive cases	Ratio %	Numbers of negative cases	Ratio %
AL-Najaf	150	50	66	22	84	28
Karbala	150	50	43	14.3	107	35.7
summation	300	100	109	36.3	191	63.7

Table No. (2) shows the numbers and percentage of total tested blood samples of camels, the number and percentage of both positive and negative samples for both sexes in the governorates of Karbala and AL-Najaf also shows that the highest number and percentage of infected was in AL-Najaf Governorate than it is in Karbala Governorate in number (66) cases were recorded by (22%).

The clinical signs of infected camels were characterized by an increase in pulse and respiration rate, high temperature, loss of appetite, weakness,

lethargy, intermittent fever, moving away from the herd, anemia,(edema in front of the chest, abdomen, genitals, limbs) and fear of light this corresponds to [16,17,27] . The important clinical signs of infected animals saw the frequent miscarriage of pregnant females, this is confirmed by the aducators and veterinarians in the veterinary dispensary of AL-Najaf Governorate and Karbala Governorate due to the high temperature and presence of the parasite in the blood, this is confirmed [29,28]. Table No. (3) shows the number and percentages of the total examined camels in both governorates in

terms of gender. (177) females were examined at a percentage (59%) of the total number of examined camels in different ages while the number of males examined are (123) by (41%) also of different ages, for both governorates.

Table (3) shows the number and percentage of examined males and females camels in the AL-Najaf and Karbala governorates .

sex	number	Ratio %
She camel	177	59
camel	123	41
summation	300	100

Table No. (3) shows the numbers of total blood samples of camels examined in relation to the gender of the animal, where the number of examined females was (177) and the number of males was (123) for the governorates of Karbala and AL-Najaf for all ages, from (less than one year to 18 years).

Table No. (4) shows the total number of examined samples by gender (males and females) in both governorates, the number of samples infected with trypanosomiasis, represented by the positive samples, and the number of samples without infected that were represented in the table with negative samples, so the number of samples of females with trypanosomiasis was (78) by (26%) of the total number of females examined, while the number of infected males is (31) by (10.3%)

of the total number of males examined Thus, the number of infected females is higher than the number of infected males, and a significant difference at a level ($p \leq 0.01$) appeared between the sexes, as shown in table (4). It has been shown from our study that breeders sell males to slaughter and keep females for the purposes of reproduction and milk production. Also, our study confirms the sensitivity of females to disease as a result of stress factors that they are exposed to during the months of pregnancy, childbirth, lactation in addition to the role of female hormones in attracting the carrier of the disease, where the transmission medium prefers to feed on the blood of females over males, this is consistent with [30, 31], also the study agrees with [24] but it does not agree with [25] which says the infection in males is higher than it is for females.

Table (4) shows the numbers , percentage of the total tested camels and the number , percentage of healthy and infected camels with trypanosomiasis in Karbala and AL-Najaf governorates by gender

sex	Numbers of studied cases	Ratio %	Numbers of positive cases	Ratio %	Numbers of negative cases	Ratio %	p-value
She camel	177	59	78	26	99	33	p≤ 0.01
camel	123	41	31	10.3	92	30.7	
summation	300	100	109	36.3	191	63.7	

Table No. (4) shows the total number of examined samples according to the gender (males and females) in both governorates. It shows that there is a significant difference at a level ($p \leq 0.01$) between the sexes, where the highest percentage of infection for females was recorded than it is for males with the number (78) by (26%) of the total number of females examined, while the number of infected males is (31) by (10.3%) of the total number of males examined.

As for the relationship of the disease and the age group, it is shown in table no. (5). It shows that the

highest number of infected animals was in the age group (13 years - 15 years) with the number (26) by (8.6%), followed by the age group (10 years - 12 years) with the number (24) by (8) , while the lowest affected age group was (1≤ year - 3 years) with a number (9) and a percentage (3%). This does not agree with [32], while it is consistent with [33] in terms of the lack of infection in young ages because they have acquired immunity from the mother. Across the placenta and through breastfeeding from her mothers, the results record a statistically significant difference ($p \leq 0.01$) between age groups.

Table (5) shows the percentage and total numbers of the camels samples tested ,positive and negative samples for both sexes in Karbala and Najaf governorates according to the ages of the groups.

Age / year	Numbers of studied cases	Ratio %	Numbers of positive cases	Ratio %	Numbers of negative cases	Ratio %	p-value
1≤ year - 3 years	37	12.3	9	3	28	9.3	$p \leq 0.01$
4years – 6 years	48	16	17	5.7	31	10.3	
7years – 9 years	42	14	19	6.3	23	7.8	
10years –12 years	78	26	24	8	54	18	
13years –15 years	50	16.7	26	8.6	24	8	
16years –18 years	45	15	14	4.7	31	10.3	
summation	300	100	109	36.3	191	63.7	

Table No. (5) shows the relationship of trypanosomiasis infection and groups of the age. The results of the table indicated a significant statistical difference at the level of ($p \leq 0.01$) between groups of the age, also shows the highest number of the infection was in the group of the age (13 years -15 years) and the lowest number of infection was in the group of the age (1≤ year - 3 years).

The results of confirming parasite infection with laboratory tests are shown in two tables no. (6,7). Table no. (6) shows the relationship of laboratory examination and sex, while table no. (7) shows the relationship of laboratory examination and the age group, thus the results of the two tables show that the examination of wet blood smear recorded less positive number and

percentage in a number (3) by (1%) of the total number (300) blood samples examined for both governorates, this is consistent with [5,25], which explains that the disease in the chronic phase is less because the less of the number of the parasite in bloodstream. The dry thin and thick smear recorded higher than the wet smear, with the number (7) by (2.3%).

As for the results of the mercury chloride test, it is recorded the highest percentage of positive results than that of the wet and dry smears tests. The results of the examination were recorded as (68) by (22.7%) of the total blood samples examined ,thus, a statistically significant difference at a level ($p \leq 0.01$) was recorded between the types of tests approved for diagnosis The results of the mercury chloride test in our study were confirmed by

[34] as it indicated the adoption of the test on the basis of protein precipitation in the serum of infected animals, so a white precipitate appears in the cases that are positive

for the test and does not appear in the negative cases of the test, and our results are consistent with the study [26].

Table (6) shows the types of laboratory tests approved for the diagnosis of trypanosomiasis, the number and percentages of examinations for each test by sex.

sex	Numbers of positive cases	Wet Smear	Ratio %	Dry Smear	Ratio %	Mercury chloride test	Ratio %	summation	p-value
She camel	78	3	1	7	2.3	68	22.7	26	p≤ 0.01
camel	31	1	0.3	2	0.7	28	9.3	10.3	
summation	109	4	1.3	9	3	96	32	36.3	

Table No. (6) shows the types of laboratory tests used to confirm the diagnosis of trypanosomiasis and its relationship to the sex of the animal. The results of the table are shown in the presence of a statistically significant difference at a level ($p \leq 0.01$) between the types of tests approved for the diagnosis, also the table shows that the examination of wet smears for blood samples recorded the lowest number and percentage of the positive total blood samples examined for both governorates, the dry thin and thick smears were higher than the wet smears. The highest recorded test to confirm infection was the mercury chloride test, as shown in the table.

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Conflict of Interest: None to declare.

Ethical Clearance: All experimental protocols were approved under the Technical Institute of Karbala and all experiments were carried out in accordance with approved guidelines.

References

- 1- Robert I. Camel (natural and cultural history). Publishing house: Abu Dhabi Authority for Culture and Heritage (Word Project). 1st.Edition. 2012; 263
- 2- Oakland Z. Dromedary Camel. Available online: 1997.
- 3- Schmidt-Nielsen K. Desert Animals, Physiological Problems of Heat and Water. New York: Dover Publications Inc. 1979.
- 4- Busch G. Animal Bytes: Dromedary Camel. Available online, 1996;2020-5-23.
- 5- Shebar S J. Some pathological changes and the role of auto-antibodies in rabbits infected with Trypanosom evansi. Master's thesis - College of Science - University of Baghdad. 1976.
- 6- Losos GJ, Ikede BO. Review of pathology of disease in domesticated and labrotory animals caused by T.congolense ,T.vivax ,T.gambiense. Vet. Pathol. 1972;9:1-71 .
- 7- Oxidation. Camel Research and Development Network. The Arab Center for the Studies of Arid Zones and Arid Lands. 2002;100:70.
- 8- Diseases that afflict camels and their treatment methods, Animal Production Forum, April, 2011;21.
- 9- Soulsby EJJ. Helminthes, Arthropods & Protozoa of domesticated animals 7th . ed.Balliere& Tindall, London, 1982;1 – 35.
- 10- Gill BS, Singh H, Gill JS, Kwattra MS. Trypanosoma evansi infection in pig in India .Vet. Rec. 1987;120(4)72.
- 11- Wilde J K H. Tick borne disease & their vectors.

- Edinburgh University press. 1978;177.
- 12- Kazem, Fawzia S. A study of the role of the stable fly in the transmission of *Trypanosoma evansi* parasite in laboratory mice and the changes occurring in the parasite in the insect's digestive tract. Master Thesis - College of Veterinary Medicine - University of Baghdad. 1980.
 - 13- Losos GJ. Infectious Tropical Diseases of Domestic Animals. Chapter 3, Trypanosomiasis, 1995;182-318 .
 - 14- Higgins AJ. Protozoal diagnosis of camels . Brit. 1985;141:87 .
 - 15- Loher KF, Pholpark S, Siriwan P, Leeirihul N, Strikijakarn L, Staak C. T.evansi infection in Buffaloes in northeast Thailand.11.Abortion.Trop. Anim.Hith.Frod. 1986;18:103–108 .
 - 16- Luekins AG. Method for diagnosis of Trypanosomiasis in live stock. Men. Inst. Dswaldo cruz , Riode Janero . 2002;96 (5):409 – 415 .
 17. Radostitis OM, Gay CC, Blood DC, Hinchcliff K W. Veterinary Medicine . 10th ed., W.B.Saunders. 2002;55 – 60:390 – 405:705:1032 – 1050:1562-1714 .
 18. Schalm OW, Jain NC, Carroll EJ. Veterinary haematology. 4th. ed. Philadelphia . 1986;16- 48: 66 – 68:164 – 188:321:409 – 411.
 19. Richardson U F, Kendall S B. Veterinary protozoology, 2nd edition. Oliver and Boyd Ltd (Edinburgh), 1957;85-88.
 20. Adam K M, Paul J, Zaman V. Medical and Veterinary Protozoology . Churchill Livingstone Edinburgh and London 1971;170.
 21. Rutter T E G. Disease in camel. Vet. Bull. 1967 ;(37):611.
 22. My L N, Wicher H, Tam P T. Comparative study of diagnosis methods for trypanosomiasis in buffaloes infected by *Trypanosoma evansi* .Sci. Tech. Vet. Vietnam 2000;7-12:6-14.
 23. Awkati VJ, Al-Katib GM. Trypanosomiasis in domestic animals of Iraq. J. Egypt. Vet. Med. Ass, 1972;32:203-206.
 24. Zahra M, Al-Hakak. Over wounded Iraqi camel disease (Asera) Trypanosomiasis. Proceeding of The 2nd International Scientific Conference of Genetic and Environment. Baghdad, Iraq. 2012.
 25. Al-Haboubi, Asaad H M. Diagnostic study of naturally infested camels with *Trypanosoma evansi*, Master's thesis - College of Veterinary Medicine - University of Baghdad. 1989.
 26. Al-Amishi, Ghada B A. Field survey on *Trypanosoma evansi* in camels in Al-Qadisiyah Governorate. Master thesis - College of Veterinary Medicine - University of Baghdad. 2004.
 27. Stephen L E. Trypanosomiasis A veterinary Perspective .Oxford : Pergamon on Press. 1986.
 28. Anosa V O. Hematological & Biochemical changes in humane and animal trypanosomiasis .Revu. Elev. Med Vet. Pays. Trop. 1988;41(6): 65-78.
 29. Hunter A G Luckins A G. Trypanosomiasis. In: Hand book on Animal Diseases in the Tropics .4th eds. M.M.H.Sewell & D.W. Brookesby, Londone : Baill icere Tindall. 1990; 204-226.
 30. Mackenzie PK, Gruickshantes JG. Phagocytosis of Erythrocytes & Leukocytes in sheep infected with *Trypanosoma congolense*. Res. Vet. Science , 1975;15(2):256- 262.
 31. Mahmoud M M. *T. congolense* :Latex – Fixation test for diagnosis of rabbit infections. Exp. Parasitol. 1974;31:109 -119.
 32. Ross JPJ. The detection of circulating trypanosomal antibodies by capillary tube agglutination test , ANN, Trop. Med. & Parasitol. 1971;65(3):327-333.
 33. North MJ. Comparative biochemistry of the proteinases of eukaryotic micro- organism. Microb. Rev. 1983;49:308 – 340.
 34. Murry M, Murry PK. An improved parasitological technique for diagnosis of African Trypanosomiasis .Trans. Roy. Sco .Trop .Med . Hyg. 1976;71:325 -326.
 35. Bradford PS. Large animal internal medicine , 3rded. Mosbey . 2002.
 36. Benjamin MM. Outlin of veterinary clinical pathology ,3rd.ed. 1986.