

ORIGINAL ARTICLE

Changes of reproductive indices of the testis due to *Trypanosoma evansi* infection in dromedary bulls (*Camelus dromedarius*): Semen picture, hormonal profile, histopathology, oxidative parameters, and hematobiochemical profile

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ABSTRACT

Objectives: This study was designed for the investigation of the effect of infection by *Trypanosoma evansi* on the changes of reproductive indices of the testis, causing reproductive failure in dromedary bulls (*Camelus dromedarius*).

Material and methods: Seventy-five bulls were used for monitoring of the changes in the semen characteristics, reproductive hormones, hematobiochemical profiles, histopathological characters in the testis, and oxidative biomarkers. The animals were divided into two groups. Group A represented the uninfected or control group, while group B represented the infected group. Group B was again divided into two subgroups, such as acute and chronic infected animals.

Results: Results showed that the semen analysis of infected camels revealed the presence of alterations in the morphology of sperms, especially the heads and tails, as compared to control animals. The hormonal profile indicated a significant decrease in the luteinizing hormone, follicle-stimulating hormone, and testosterone levels, accompanied by the rise in the cortisol level in infected camels compared with the negative control. The histopathology and testicular degeneration were found to be associated with other disorders in infected camels. The oxidative profile and protein oxidation were promoted in infected testicles, indicating the occurrence of harmful effects in the cell.

Conclusion: It is concluded that *T. evansi* infection in dromedary bulls causes severe damage to the testicular tissue and decreases the reproductive hormone levels associated with severe morphological disorders in sperms due to oxidative stress resulting from the infection. All these findings indicate that *T. evansi* can cause reproductive failure and fertility damage.

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Introduction

Trypanosomiasis is a common and life-threatening disease. It causes massive economic damage in both milk and meat productions in various animal species, especially camels. Recent outbreaks of Trypanosomiasis have been reported in some countries such as Kuwait, Palestine, and Mauritius [1]. Trypanosomiasis is a blood protozoan disease caused by various trypanosome parasites, but the most dangerous

one is *Trypanosoma evansi*, which is the agent of “surra”. Surra is considered as a severe disease characterized by different pathological manifestations [2] in addition to its immunosuppressive effects.

Trypanosomiasis is well known as one of the leading causes of limiting livestock production in Africa, especially in the sub-Saharan region. From the economic point of view, it is one of the main factors which threatens the

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food security in Africa, especially a country like Nigeria [3]. Reproductive organs are recognized to be affected under *Trypanosoma* infection. Trypanosomosis has been reported to cause significant damage to reproductive aspects in animals. The reproductive harm resulted from its harmful effects on endocrine glands and gonads, which leads to hormonal perturbation either in secretion or in its concentration in the blood. Therefore, delayed puberty in young animals or disruption in semen production and its quality in adult animals were shown to occur from infection by *Trypanosoma* [4]. A previous study carried out in Yankasa rams infected with *T. evansi* reported that the infected rams suffered from the production of poor-quality semen, such as oligospermia and aspermia.

Furthermore, if a sperm is present, it is either dead or it suffers from poor motility if it is alive [5]. The harmful effects of *Trypanosoma* infection are also extend to females and even to humans. Sterility, menstrual disorder, and stillbirths are the drawbacks of Trypanosomiasis infection in humans. While in animals, the harmful effects include obstetrical disorders such as abortion, premature birth, and prenatal losses, whereas the gynecological effect includes the incidence of cystic ovaries [6].

Several studies on *Trypanosoma* infection have mainly investigated the harmful effects of that parasite on females at the expense of males. However, the defect in males leads to a more considerable drop in the total fertility of the herd due to the constricted number of the male to female ratio. Therefore, the effect of parasitic infestation on the part of reproduction needs more studies to have enough knowledge about the degree of involvement of the reproductive system with parasitic infection. Also, these studies should include the mechanism involved in the effects of infection on the host, with particular regard to reproductive hormones and their respective receptors, especially with Trypanosomiasis infection.

It is possible to find studies about the effect of *Trypanosoma* infection in the production of livestock, but reviews specifying its impact on the reproductive system are not available. Because of the significant harm of that parasite nowadays in different animals, especially in productive camels, we intend to present sufficient information on the factors associated with *Trypanosoma* infection belonging to symptomatology and physiopathology of reproductive aspects in bulls.

This study describes that *T. evansi* infection in dromedary bulls causes marked testicular tissue damage and a decrease in the levels of hormones of the reproductive system [such as luteinizing hormone (LH), follicle-stimulating hormone (FSH), and testosterone] associated with severe morphological disorders in sperms due to the increase in the levels of oxidative stress biomarkers and cortisol. All these findings indicate the incidence of complete

reproductive failure, which supports the idea that this parasite can contribute to infertility in male camels.

This study was designed for the investigation of problems related to reproduction, leading to the damage of the economy, due to the scarcity of the studies on *T. evansi* infection, which included the destructive influence of *T. evansi* in the reproductive system of bulls primarily related to histopathological changes of testicular tissue. Therefore, this study aimed to investigate the changes of reproductive indices of the testis and reproductive failure resulting from the *T. evansi* infection in dromedary bulls (*Camelus dromedarius*) through different diagnostic tools, such as evaluation of oxidative stress, testicular lesions, semen characteristics, hormone levels, and hematobiochemical parameters.

Materials and Methods

Ethical statement

The steps of the current study were carried out according to the ethical guidelines approved by the Ethics Committee of the Faculty of Veterinary Medicine, Aswan University, Egypt.

Animals and blood smears

Camel bulls (*Camelus dromedaries*) from the southeast region of Egypt in Aswan province were used in the current study. The animals reached Aswan province through the border in the area of Abo-simple and were kept in quarantine for 15 days for later slaughtering. Some of these bulls appeared in excellent and healthy conditions, while others were weak and had the symptoms of cachexia. This study was carried out in the breeding season (January and February) on 75 dromedaries aging between 10 and 14 years.

The animals were divided into two groups. Group A ($n = 25$) contained uninfected bulls and represented the control group, while group B ($n = 50$) contained animals infected with *T. evansi*. Group B was further divided into two subgroups comprising acute ($n = 25$) and chronic ($n = 25$) infected animals.

Several parameters were determined at the sampling time, such as general health condition, rectal temperature, and checking of the scrotal contents through palpation to detect testicular firmness and springiness. The body condition scores (BCS) ranged from 1 = thin to 5 = fat. Testicular firmness and springiness were recorded to detect the testicular tone depending on the scale, which ranged between 1 and 4. If the testicular tissue is firm and most of it is springy, the score is 1. If the testicular tissue has moderate firmness and springiness, the score is 2. On the contrary, if the testicular tissue is hard with no springiness, the score

is 3, and if the testicular tissue is soft with no springiness, the score is 4.

Collection of samples and diagnosis of trypanosome infection

Blood smears were observed daily for 15 days after staining with the Romanovsky method. Microscopic examination was done at 1,000× and focused on the number of the parasite per field (t/f). Blood was collected from all camels by jugular venipuncture. Whole blood was collected for hematology, while serum was prepared for biochemical analysis. Packed cell volume (PCV) and hemoglobin (Hb) concentration were estimated to detect the presence or absence of anemia.

Collection of the semen and its evaluation

The semen was collected from all the studied camels. Epidermal sperms are collected after the slaughtering of the animals. Testicles were collected postmortem in the abattoir. Both testicles were moved to the laboratory in a Styrofoam box under cool temperature. Testes and epididymis were separated from the scrotal sac. Caudae epididymides were separated from the testes and the surrounding connective tissue. The retrograde flushing technique was used for the collection of epididymal sperms [7]. Briefly, before flushing, cannulation was performed in the lumen of the ductus deferens using a blunted 22-gauge needle.

Cauda epididymidis and ductus deferens were isolated from the remaining part of the epididymidis by a scalpel, which was used to cut the junction of the corpus and the proximal cauda. A syringe containing a flushing solution composed of 2 ml of extender (37°C) was used for the flushing of sperm cells from the ductus deferens through the cauda epididymidis. The semen extender composed of the Tris-fructose extender, which was prepared according to that reported by Salamon [8]. A computerized cell motion analyzer was used for the evaluation of the extended semen [9].

Analysis of hematological parameters

An automatic cell counter using Exigo vet (Boule Medical AB, Spånga, Sweden) was used for the analysis of hematological parameters. The measured hematological parameters were PCV, Hb concentration, white blood cells (WBCs), lymphocytes, neutrophils, red blood cells (RBCs), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), and mean corpuscular hemoglobin concentration (MCHC) [10,11].

Biochemical analysis

Biochemical parameters, including urea, creatinine, Glutamate-Pyruvate Transaminase (GPT), Glutamic oxaloacetic transaminase (GOT), Gamma-glutamyl transferase

(GGT), and glucose, were measured using commercial kits (Spinreact S.A./S.A.U. Ctra. Santa Coloma, Spain). A spectrophotometer was used and the results were obtained after using standard formulae [12]. Serum calcium (Ca) and phosphorus (P) were also detected [13,14].

The determination of hormonal profile (LH, FSH, and testosterone) was done by the technique of Enzyme Immunoassay, in which (Cayman Chemical Company, Ann Arbor, MI) the manufacturer's instructions were followed. Cortisol was detected by commercially available enzyme-linked immunosorbent reagent kits (Diagnostic biochem Canada, Canada).

Histopathology

For histopathological evaluation, samples from testes were collected. Two slides were made for each sample, which was stained with the Hematoxylin and Eosin method. In each slide, germinal epithelium thickness and the degree of its degeneration were determined, in addition to checking the tubular lumen for the presence of sperms.

Biomarkers of cell injury

Checking the biomarkers of cell injury was used as a method for evaluating the degree of cell damage. Detection of the advanced oxidation protein product (AOPP) of the testis was done to evaluate protein oxidation [15]. Determination of the Thiobarbituric acid reactive substances (TBARS) was done to assess the lipid oxidation of the testis. The technique described by Tatsch et al. [16] was used to detect NO_x (nitrite/nitrate) quantity, which was used to determine the NO levels.

Statistical analysis

Statistical evaluation of the results was carried out using SPSS® (SPSS 16). A general linear model was used to evaluate significant differences.

Results

The results of this study showed a comparison between the infected group and the control group in some parameters, but there was no comparison between acute and chronic infections or between the acute group and control group due to the absence of significant differences between acute infection and control group in these parameters.

Progression of parasitemia

The results showed that 25 bulls were found free from Trypanosomiasis, which represent the control group, and 50 bulls were infected with Trypanosomiasis, which represent the infected group. Based on the clinical and hematological parameters (Table 1), it is revealed that 25 bulls had chronic infection with a significant decrease in the BCS, Hb

concentration, PCV percentage, and testicular tone scores (Tables 1 and 2). On the contrary, the acute infection in the other 25 bulls in the infected group was evidenced by checking the presence of significantly higher rectal temperature (Table 1).

Hematobiochemical parameters

Hematobiochemical parameters of the infected and non-infected bulls were examined and the results are shown in Tables 2 and 3. The levels of Hb, RBCs, PCV, MCV, MCH, and MCHC decreased significantly in chronically infected bulls compared to healthy animals. On the contrary, the WBCs, neutrophils, and lymphocytes exhibited an increase in infected animals compared to healthy control animals (Table 2). Also, there were changes in the biochemical

Table 1. Estimates of clinical and seminal parameters in infected and control group (means \pm standard errors).

Parameters	Control group	Infected group (B) (n = 50)	
	(A) (n = 25)	Acute infection (n = 25)	Chronic infection (n = 25)
BCS	4.2 \pm 0.1 ^a	4.0 \pm 0.3 ^a	2.1 \pm 0.3 ^b
Rectal temperature(°C)	36.8 \pm 3.0 ^a	39.1 \pm 4.0 ^b	36.2 \pm 3.5 ^a
Testicular tone scores	1.2 \pm 0.2 ^a	1.7 \pm 0.3 ^a	3.2 \pm 0.1 ^b
Sperm count(\times 10 ⁶ /ml)	121.0 \pm 12.0 ^a	114 \pm 15.0 ^a	30 \pm 11.0 ^b
Motility (%)	67.1 \pm 5.6 ^a	70.2 \pm 7.9 ^a	21.2 \pm 3.1 ^b
Live sperm (%)	72.9 \pm 7.4 ^a	75.8 \pm 8.5 ^a	25.5 \pm 2.9 ^b
Abnormalities (%)	3.2 \pm 0.85 ^a	2.8 \pm 0.7 ^a	29.8 \pm 2.9 ^b

Values in the same line have different superscripts differ significantly ($p < 0.05$).

Table 2. Hematological parameters of infected groups compared to the control group (means \pm standard errors).

Parameter	Control group	Infected group (B) (n = 50)	
	(A) (n = 25)	Acute infection (n = 25)	Chronic infection (n = 25)
Hb (gm/dl)	14.67 \pm 0.3 ^a	13.47 \pm 0.3 ^a	4.1 \pm 0.7 ^b
PCV (%)	31.5 \pm 1.3 ^a	29.2 \pm 3.3 ^a	10.2 \pm 1.6 ^b
WBC (10 ⁹ /l-1)	9.62 \pm 0.5 ^a	13.22 \pm 0.5 ^a	18.9 \pm 0.1 ^b
Neutrophil (10 ⁹ /l-1)	5.85 \pm 0.4 ^a	7.5 \pm 0.4 ^a	12.89 \pm 0.1 ^b
Lymphocytes(10 ⁹ /l-1)	3.09 \pm 0.7 ^a	3.09 \pm 0.7 ^a	4.94 \pm 0.2 ^b
RBC (10 ¹² /l-1)	9.82 \pm 0.8 ^a	9.52 \pm 0.8 ^a	7.79 \pm 0.1 ^b
MCV (fl)	33.17 \pm 0.4 ^a	32.17 \pm 0.4 ^a	32.76 \pm 0.5 ^b
MCH (pg)	15.33 \pm 0.5 ^a	13.73 \pm 0.5 ^a	10.73 \pm 0.4 ^b
MCHC (gm dl-1)	459 \pm 0.1 ^a	427 \pm 0.1 ^a	320.33 \pm 0.3 ^b

Values in the same line having different superscripts differ significantly ($p < 0.05$).

profile as creatinine, urea, GGT, GPT, and GOT concentrations were higher in the infected animals. On the contrary, glucose concentration decreased drastically in infected camels compared to the healthy group (Table 3). The mineral parameters of chronically infected animals showed a significant decrease in Ca and P compared to healthy or acute infected camels (Table 3).

Sperm morphology

Examination of the semen of the infected and non-infected bulls revealed that the sperm concentration was significantly decreased in the infected bulls in addition to the reduction in its motility percentage. On the contrary, the examination of sperm morphology revealed the presence of an increase in the percentage of abnormalities ($p < 0.01$) in the infected animals. Significant changes were observed in the head (tapered head), tail, and distal drop of the sperms in infected bulls compared to the uninfected bulls (Table 4).

Table 3. Biochemical parameters of infected groups compared to the control group (means \pm standard errors).

Parameter	Control group	Infected group (B) (n = 50)	
	(A) (n = 25)	Acute infection (n = 25)	Chronic infection (n = 25)
Kidney function profile			
Creatinine mg/dl	1.08 \pm 0.2 ^a	1.20 \pm 0.4 ^a	2.06 \pm 0.4 ^b
Urea liquid mg/dl	26.33 \pm 0.4 ^a	30.55 \pm 0.1 ^a	65.11 \pm 0.6 ^b
Liver function profile			
GPT U/l	8.6 \pm 0.2 ^a	12.5 \pm 0.2 ^a	57.96 \pm 0.1 ^b
GGT U/l	14.3 \pm 0.2 ^a	18.65 \pm 0.5 ^a	40.66 \pm 0.9 ^b
GOT U/l	37.7 \pm 0.5 ^a	45.23 \pm 0.6 ^a	195.0 \pm 0.2 ^b
Minerals profile			
Calcium mg/dl	11.27 \pm 0.8 ^a	11.25 \pm 0.2 ^a	10.326 \pm 0.4 ^b
Phosphorus mg/dl	6.66 \pm 0.3 ^a	6.02 \pm 0.7 ^a	4.00 \pm 0.7 ^b
Sugar profile			
Glucose mg/dl	131.7 \pm 0.2 ^a	124.25 \pm 0.8 ^a	68.00 \pm 0.4 ^b

Values in the same line having different superscripts differ significantly ($p < 0.05$).

Table 4. Morphological analysis of sperm in infected and control groups.

Sperm morphology (n = 200)	Control group (A) (n = 25)%	Infected group (B) (n = 50)%
Tapered head sperm	0.55 \pm 0.52	12.56 \pm 0.54 [*]
Tail folded sperm	1.36 \pm 0.70	15.26 \pm 0.75 [*]
Distal drop	0.21 \pm 0.16	4.25 \pm 0.56 [*]

Groups (results in percentage %). Values with superscripts in the same line differ statistically ($p > 0.05$).

Hormones

Examination of the hormonal profile (LH, FSH, and testosterone) of the infected camels revealed that these animals suffered from a significant decrease in the levels of these hormones ($p < 0.05$) when compared to the control group (Fig. 1).

Histopathology

Histological sections of testes of healthy camels showed normal seminiferous tubules lined with sertoli cells, which progressed to spermatocytes and ended to become sperm (Fig. 2a and b). On the contrary, those of infected camels showed the presence of degenerative changes of the seminiferous

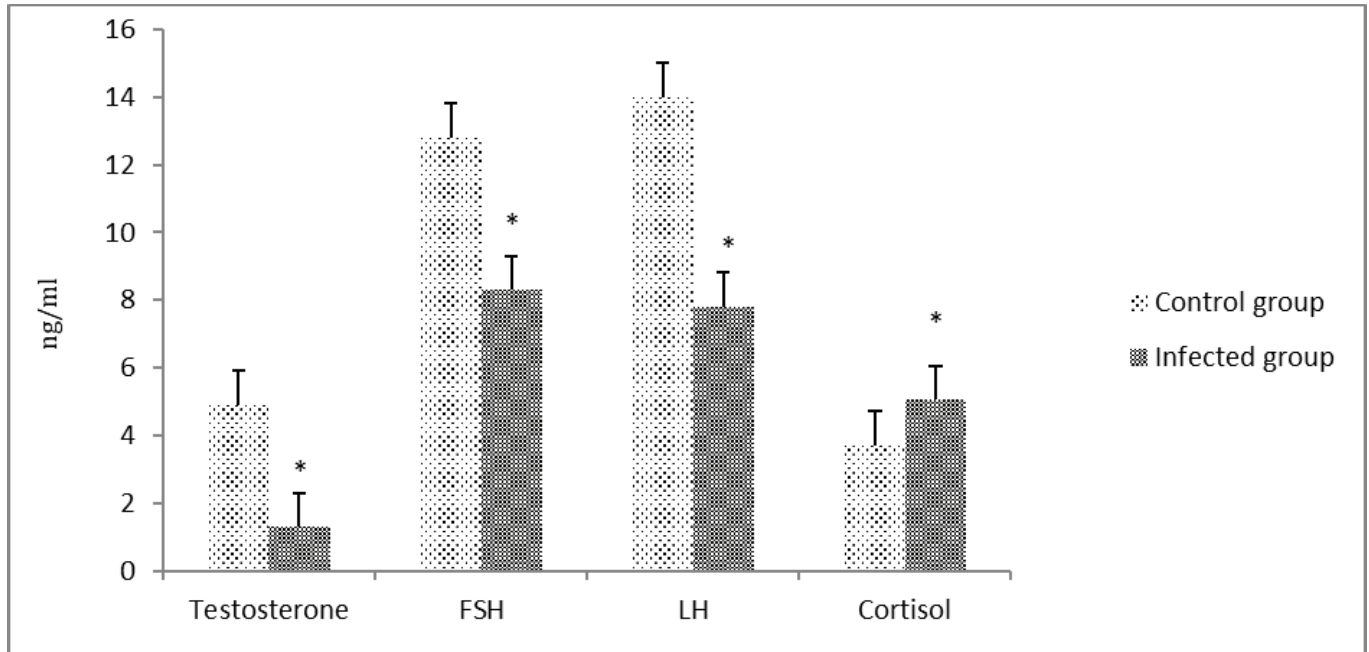


Figure 1. Hormonal parameters of infected groups compared to the control group. Columns with asterisks differ significantly ($p < 0.05$).

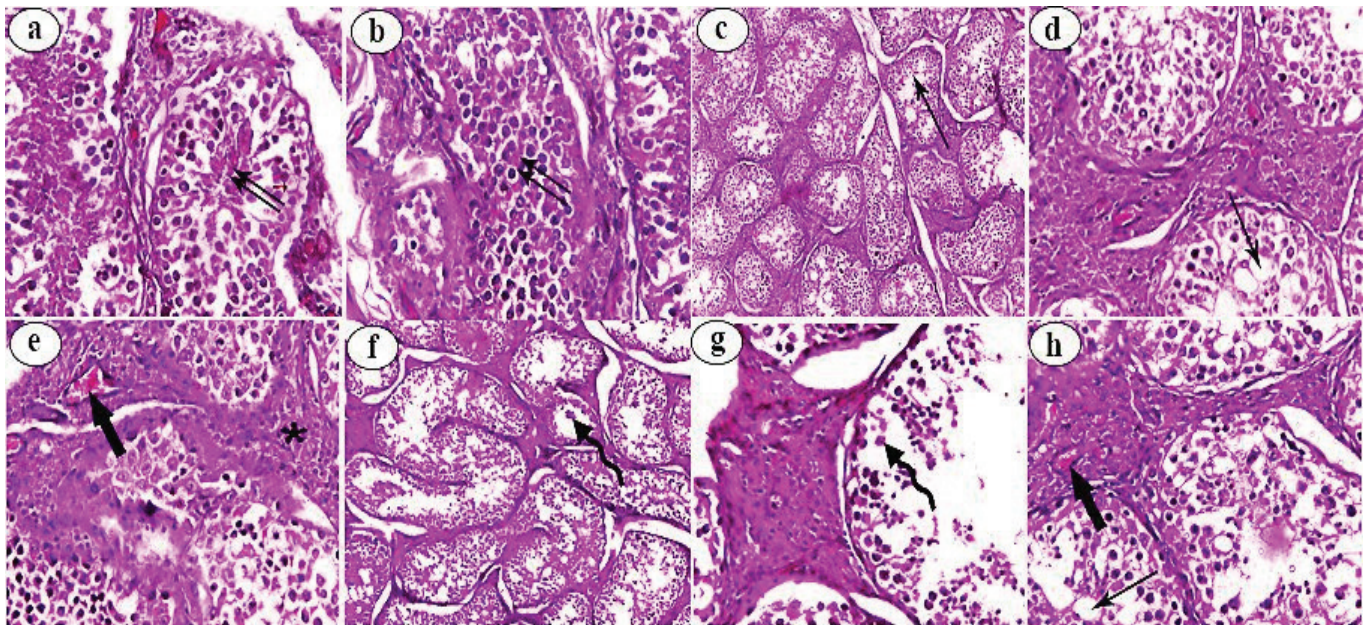


Figure 2. Histopathological examination of male camel testis stained with hematoxylin and eosin (HE). (a,b): control testes, (c-h): testes of an infected camel. Sperms (double arrows), vacuolation (thin arrow), thickening and swelling of the interstitial tissues (asterisk), congestion of the blood vessels (thick arrow), atrophied spermatocytes(^). [(a, b, d, e, g, h = 400 \times) and (c, f = 100 \times)].

tubules characterized by vacuolation (thin arrow) with scarce sperm cells and spermatids (Fig. 2d and h). Other sections showed atrophy and necrosis of seminiferous tubules comprising atrophied spermatocytes and sperm cells (Fig. 2f and g). Furthermore, Fig. 2e shows the thickening and swelling of the interstitial tissues (star) in addition to the congestion of the blood vessels (Fig. 2e and h) (thick arrow).

Cell injury biomarkers

Examination of the biomarkers of cell injury revealed that Nitrogen oxides (NO_x), TBARS, and AOPP increased significantly in infected camels compared with the control group ($p < 0.05$) (Table 5).

Discussion

In the present study, camels infected with acute Trypanosomiasis suffered from hyperthermia. These results are similar to other investigations that mentioned the presence of higher rectal temperatures in camels infected with Trypanosomiasis in acute forms. Furthermore, a study carried out in buffaloes revealed that infected animals suffered from not only high fever but also edema of face and legs, bulging eyes with lachrymation, and hyperemia of the eye mucosa [17].

The bad effects of parasitism on the health of mammals resulted from the usage of resources from the host, therefore affecting the energy balance of this host; however, a little knowledge is known about this effect [18], especially those related to energy consumption. The different life processes are controlled by the mitochondrial ATPase, which is responsible for the synthesis and hydrolysis of Adenosine Triphosphate (ATP). The *Trypanosoma*-induced hypothyroidism may lower the mitochondrial ATPase activity. Panicucci et al. [19] revealed that although the blood parasite *T. brucei* could produce ATP during its procyclic stage, it consumed ATP regularly during the mammalian bloodstream stage as it does not have the

respiratory chain, and this can be the explanation for the reason of chronically infected camels suffer from muscular wasting and emaciation [20].

Besides, anemia appeared in infected animals, which resulted from the products of parasites and which caused damage to blood cells in addition to extensive gastric bleeding. These results are in agreement with a study carried out in dromedary camels [21] and which showed that *T. evansi* could induce anemia, dullness, weakness, the rise of body temperature, and nervous manifestation. Furthermore, another study carried out in domestic cats infected experimentally by *T. evansi* showed that the same results were found in the infected cats [22].

Examination of hematological and biochemical parameters provides a picture of the condition of the living body, especially in cases of disease such as Trypanosomiasis. Trypanosomiasis causes a decrease in Hb and PCV in camels and different animal species [23–25]. Trypanosomiasis is associated with hemolysis, which may result in a reduction in the life span of RBCs and extensive erythrophagocytosis [26]. This reduction of the half-life of the RBCs resulted from the activation of the mononuclear phagocytic system, which leads to the erythrophagocytosis [27]. Similar to these findings were found in the current study, which revealed that Hb concentration, PCV, and RBCs decreased significantly in the animal that suffered from the infection compared to the control group. Besides, these findings were also confirmed by several studies [23,24,28–30]. Furthermore, infected animals were observed with higher means of WBCs, neutrophils, and lymphocytes when compared with the healthy control camels. These findings indicate that an initial enhanced immunological response occurred and it was followed by the immunosuppressive effect of trypanosome [23].

In the present study, Trypanosomiasis in camels showed higher urea and creatinine concentrations, especially in chronic conditions. These results are in agreement with a previous study in dromedary camels that were infected subclinically with *T. evansi* during its season of breeding [31]. These results revealed that Trypanosomiasis causes damage to host tissues, leading to hepatic and renal malfunction.

Trypanosomiasis-affected camels showed higher mean serum GPT, GGT, and GOT concentrations (Units/l) in infected camels compared to the healthy ones. These results are in accordance with the results reported by other studies [28,32]. These results revealed that Trypanosomiasis has its harmful effect, which may result in damage to the liver, heart, and brain.

The mean glucose concentration was found lower in infected dromedary bulls than healthy control animals. These results are similar to previous studies [33–35]. This phenomenon of hypoglycemia occurs because

Table 5. Biomarkers of cellular injury in testicles of Trypanosomes-infected camels compared to control healthy camels (means \pm standard errors).

Parameter	Control group (A) (n = 25)	Infected group (B) (n = 50)	
		Acute infection (n = 25)	Chronic infection (n = 25)
NO _x (μmol/l)	12.6 \pm 0.3 ^a	30.5 \pm 0.1 ^b	38.5 \pm 0.4 ^b
TBARS (μmol MDA/mg of protein)	15.3 \pm 0.8 ^a	25.6 \pm 0.2 ^b	27.9 \pm 0.7 ^b
AOPP (μmol/l)	35.8 \pm 0.5 ^a	50.9 \pm 0.3 ^b	52.7 \pm 0.6 ^b

Values in the same line having different superscripts differ significantly ($p < 0.05$).

trypanosomes are voracious consumers of host glucose, which is utilized for their metabolism, in addition to fever and hepatocellular damage associated with trypanosomes infection increase metabolic rate also [35].

In the current study, the examination of the semen showed that there were adverse changes in its characteristics, which include a reduction in sperm concentration and its motility. In contrast, the rate of dead sperm increased, accompanied by increased abnormalities in morphology. These results were the same as those reported in Yankas rams infected with *T. evansi* experimentally [5,36].

Our research group reported that checking the hormonal parameters in infected camels showed that a decrease in the levels of LH, FSH, and testosterone was significantly present. These results are similar to that of a study [9] carried out in male camels infected with *T. evansi* and showed that testosterone level decreased in infected animals and associated with a decrease of sperm counting, while the index of abnormal sperm increased. Faccio et al. [37] also confirmed that the experimentally infected male rat with *T. evansi* suffered from a reduction of testosterone, LH and FSH levels. Decrease in testosterone levels can be explained by a decrease in LH, which is associated with decreased stimulation of Gonadotropin-releasing hormone (GnRH), and subsequently, a drop in testosterone levels.

Cortisol is used as a marker of stress. Serum of infected camels contains high levels of cortisol, which may prohibit the secretion of GnRH; therefore, this explains the decreased concentrations of LH and FSH in the infected animals. Furthermore, seminiferous tubules degeneration may also cause a low level of testosterone in infected animals. Leydig cells that are responsible for the production of 95% of the circulating testosterone are located in the testis. Besides this, testosterone production is regulated by the secretion of LH, which was found to be decreased in the infected animals. LH also has a direct effect on the hypertrophy of Leydig cells [38].

The histopathology of the testis from infected camels in the current study showed degenerative changes of the seminiferous tubules characterized by vacuolation with scarce sperm cells and spermatids. The same results were detected in the experiment of rams infected with *T. brucei* and *T. evansi* [36]. Furthermore, another study was carried out in a deer infected with the same parasite, which revealed that the seminiferous tubule has degeneration accompanied by degeneration of sperm in the ducts of the epididymis [39]. Bezerra et al. [40] applied a study on sheep infected with *T. vivax* and revealed that the infected animals suffered from marked degeneration of the testis and concluded that these findings were leading to a decrease in LH and FSH hormones that are responsible for stimulating spermatogenesis and

steroidogenesis [41]. Thus, such deformities reduced the reproductive capacity of the infected male and ended with infertility with the chronicity of the disease.

The histopathology of other infected testes in the current study showed thickening and swelling of the interstitial tissues in addition to the congestion of the blood vessels. These results are almost in accordance with that found by Adamu et al. [42]. They noted that animals infected with *T. vivax* have hypoplastic seminiferous tubules, which results in a reduction of spermatogenic cells. In addition, the interstitial tissue was destructed and the sertoli cells disappeared with the involvement of epididymis parenchyma.

In the current study, camels infected with *T. evansi* suffered from oxidative stress due to increased levels of NOx, TBARS, and AOPP. These results are in agreement with those previously observed in the serum of rats infected with Trypanosomiasis [43]. Besides, other studies carried out in buffaloes naturally infected with *T. evansi* revealed that the oxidative indices in trypanosomiasis-affected buffaloes increased significantly compared to healthy control ones [17,24].

These findings reveal that the testicles of infected animals undergo oxidative stress. Therefore, cell injury was confirmed by the increase of these biomarkers and the histopathology of the organs.

The process of spermatogenesis occurring inside the testis is an extremely active replicative process which can produce a massive number of sperms per second. Cell divisions take place in excessive ratio inherent in this process, and this pushes the germinal epithelium to consume an excessive amount of oxygen. But the problem is the low oxygen tension in the testis which is the result of bad vascularization. This shows that spermatogenesis and steroidogenesis are exposed to oxidative stress [44], which is responsible for the production of harmful free radicals.

Unfortunately, trypanosome infection induces the production of free radicals, as some reports have revealed that the free radicals which are responsible for the induction of oxidative stress play a significant role in the pathogenesis of Trypanosomiasis [45,46]. This explains the purpose of *T. evansi* in causing damage to the testicular tissue through the induction of oxidative stress.

Consequently, it is evident that *T. evansi* causes critical damage to the fertility of infected adult males leading to severe damage to the economy, which becomes more apparent in excessive genetic animals and in the herds which use these males for the fertilization process. Therefore, there is a high need for the improvement of local research about how to control parasitic infection because the available strategies commonly utilized is no longer positive due to lack of knowledge of its biology.

Conclusion

It is concluded that *T. evansi* infection in dromedary bulls causes severe damage to the testicular tissue and a decrease in the reproductive hormone levels associated with severe morphological disorders in sperms due to oxidative stress resulted from the infection. All these findings indicate that *T. evansi* infection induced complete reproductive failure, leading to infertility in male camels, either directly or indirectly.

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Conflict of interest

All the authors have no conflict of interest to declare.

Authors' contribution

Yahia conceived and designed the study. Yahia, Enas, Samer, and Hassan executed the experiment and analyzed the sera and tissue samples. Yahia, Rana, and Hassan analyzed the data. All the authors interpreted the data, seriously revised the manuscript for necessary intellectual contents, and approved the final manuscript.

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