Zoonotic Protozoan Parasites Infecting Camels, Diagnosis and Control –

A Review

Nagwa I. Toaleb¹ and Raafat M. Shaapan²*

¹Department of Parasitology and Animal Diseases, Veterinary Research Institute, National Research Centre, 33 El-Bohouth St., Dokki, Giza, P.O. Box 12622, Egypt

²Department of Zoonotic Diseases, Veterinary Research Institute, National Research Centre, 33 El-Bohouth St., Dokki, Giza, P.O. Box 12622, Egypt

Camels are an important domesticated animal that provides milk and dietary meats, as well as they are working animals that represent vital means of transportation, especially in desert habitats. However, they are susceptible to many parasitic diseases, which lead to substantial economic losses related to reducing productivity and performance along with fatalities in severe cases. Camels also carry pathogens and may transmit them to humans and other animals. Parasitic infections in camels include protozoa, helminths, and arthropods. There are many protozoal diseases that affect and endanger camels such as blood protozoa (trypanosomiasis, babesiosis and theileriosis), gastrointestinal protozoa (coccidiosis and cryptosporidiosis), and tissue protozoa (sarcocystosis and toxoplasmosis). Microscopical diagnosis of parasitic protozoan diseases is usually dependent on the detection of the parasites in blood smear in case of blood parasites and fecal examination to detect the protozoal oocysts. Other methods such as micro-hematocrit centrifugation (MHCT) and quantitative buffy coat methods are used in the diagnosis of certain parasites such as the detection of motile and live trypanosomes. Histopathology can also be used, and different serological assays. Moreover, molecular diagnosis is used for the accurate identification of parasite species such Camel, Parasites disease, Zoonotic Protozoa, Diagnosis, Control as Polymerase chain reaction (PCR), sequencing, and phylogenetic analyses. This review highlights different diagnostic techniques and strategies for controlling parasitic protozoan diseases in camels. Also outlines of different control and management protocols to reduce the risk of this parasitic infection in camels by using chemical drugs, natural products (plant extracts and bee products), biological control, and by some vaccination trials.

Keywords: Camel, Parasites disease, Zoonotic Protozoa, Diagnosis, Control

Introduction

Camel is an important unique animal that has evolved to live and breed in the desert's extreme heat and thirst. Camel, often known as the "Ship of the Desert," is an important source of food and transportation for desert nomads in Asia and Africa. Camel's milk is one of the most helpful types of milk due to its higher vitamin C and iron content than cow's milk [1]. The genus Camelus includes three species: Camelus dromedaries, Camelus bactrianus, and Camelus bactrianus ferus. Dromedaries are mainly present in central Australia, the Middle East, parts of Africa, and south Asia [2]. The C. dromedaries is nearly found in 47 countries and represent about 95% of the total population of the Old-World camels [3].

The ability of camels to serve as a disease vector is deeply concerning due to rising human demand for meat, particularly in Egypt and other African countries, and the lack of biosecurity regulations and biosafety in many locations, as well as the growth of camel herds in wildlife with nondomestic species [4]. As a result, the camel breeding sector is becoming a major source of income for herders. The world's annual camel meat and milk production records were 532, 198,000 and 2, 696, 337 tonnes, respectively [5].

*Corresponding author: Raafat M. Shaapan, E-mail: rmshaapan2005@yahoo.com. Tel.:00201005280571
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Parasitic diseases are the major problem of livestock production especially on camels they are divided into Protozoan, Helminthic, and tick-borne diseases. Gastrointestinal and blood parasites are particularly had bad effects on the camel health which is causing anemia, wasting, and death in heavy infection [6]. Most camel infecting parasites are enteric and blood and tissue parasites they lead to nutritional and immune deficiency, stunted growth, delayed development, and production loss in terms of morbidity and mortality, results in significant economic losses [7]. They might be asymptomatic or subclinical, and they reduce camel performance potential as well as the quality of camel meat and milk [8].

Currently, in addition to clinical signs, numerous laboratory approaches are employed to detect and diagnose parasite infections. However, in recent years, research has concentrated on alternative methods to improve parasitic disease diagnosis. Immunoassays, molecular-based approaches, and proteomics using mass spectrometry platforms technologies are among these sophisticated techniques, and early identification using these advanced techniques is critical for treating and preventing parasite infections [9]. So, this review summarizes the progress in new approaches and provides an overview of the major parasitic protozoan disease present in dromedary camels, and the various diagnostic methods focusing on microscopic examination, serological, and molecular approaches that are being used in diagnosis of protozoan and discusses some of the merits and disadvantages of these tests. Also, it presents the more efficient and sustainable control methods to integrate the ongoing scientific advances.

**Parasitic protozoan of camel**

Protozoa are single-cell organisms that multiply by simple binary division in their hosts, increasing in their numbers to cause overwhelming infection. They divided into three groups according to their infections; blood parasites, gastrointestinal parasites, and intercellular parasites (Fig. 1), [10].

Fig. 1. Parasitic protozoan of camel. Shaapan [10].

**Blood protozoan parasitic diseases**

More than 48% of camels are infected with blood protozoan parasites (Haemoprotozoan parasites) such as *Trypanosoma* sp., *Babesia* sp and *Theileria* sp. [11].

**Trypanosomiasis**

Several *Trypanosoma* spp. infect camels, but *Trypanosoma evansi*, the causative agent of "Surra," is the most common and considered an emerging zoonotic parasite. The rate of trypanosomiasis infection in Asia is higher than in Africa. Due to trypanosomiasis, around 12 million camels are at risk around the world, costing an estimated 1.3 billion US dollars when compared to the cost of meat and milk [12]. In an acute case, trypanosomiasis produces an increase in body temperature, starvation, and mortality, whereas in a chronic condition, the disease causes anaemia, intermittent fever, emaciation, hair loss, oedema, and abortion. This condition is known as 'Tibersa' since it lasts three years or longer [13]. The trypanosomiasis in the camel (*T. evansi*) is transmitted in several ways; mechanically by a
number of species of haematophagous biting or sucking insects (*Tabanus, Haematobia, Hippobosca* and *Stomoxys*) (Fig. 2 a-d), per-oral when carnivores eat infected prey, rarely through the use of non-sterile surgical instruments during mass treatment and vaccination. Transmission in milk and during coitus has been also documented (3). In Egypt, *Trypanosoma* infection was found in 36.7% camels and 12.24% of female camels in Upper Egypt. By ELISA, Camels were found to be naturally infected with *Trypanosoma evansi* in 8.0% and 24.0% of females. The prevalence of *Trypanosoma* infection in Egypt's northern west coast ranged from 20.24% to 67.06%. *T. evansi* infection was found in 5.63 % of screening camels in Halayeb and Shalateen in Upper Egypt [14].

![Trypanosoma evansi; Tabanus Vector; (A), Haematobia (B), Hippobosca (C) and Stomoxys (D). Desquesnes et al. [23.]](image)

**Fig. 2.** *Trypanosoma evansi; Tabanus Vector; (A), Haematobia (B), Hippobosca (C) and Stomoxys (D). Desquesnes et al. [23.].**

### Diagnosis of trypanosomiasis

**Microscopical diagnosis**

Wet blood examination (WBE) was used, in which a drop of fresh whole blood was placed on a clean microscope slide and mix gently, covered with coverslip and examined under x40 objective of a microscope which is easily detect the active motile *T. evansi* parasites. Also, Giemsa-stained thin blood smears (GSBS) under light microscopy is used to diagnosis of *T. evansi* infection in the field and acute cases, but this method is not suitable for chronic cases or for the detection in carrier animals (Fig. 3 a, b) [15].

![Trypanosoma evansi in wet blood film (motile trypomastigote indicated by red arrow) (a) and Giemsa-stained blood smear (b). Hassan-Kadle et al. [15.]](image)

**Fig. 3.** *T. evansi* in wet blood film (motile trypomastigote indicated by red arrow) (a) and Giemsa-stained blood smear (b). Hassan-Kadle et al. [15.].

The GSBS have a low sensitivity equivalent to $10^5$ trypanosomes/ml so, micro hematocrit technique (MHCT) and quantitative buffy coat method (QBC) were detect the live parasite in the blood of the camel which are simple, easy, cheap, and more sensitive than microscopic examination. These techniques detect motile and live trypanosomes in blood of asymptomatic carriers as a rapid method of diagnosis in the field while incapable of identifying different trypanosome species [16].
Serological diagnosis

Immunodiffusion test

It is easy test requires few reagents and equipments, in which the antigen and antibodies are added in different wells into a semi-solid medium such as agar gel set onto a slide, the positive sample formed a white line. The disadvantage of this test, is a time-consuming for detection of antibodies in suspected animals and its sensitivity is moderat [17].

Card agglutination test

This test determine antibodies of T. evansi experimentally and naturally infected animals, while this test cannot determine T. vivax or T. congolense because of the difficulty of identifying suitable variable antigens of these species [18].

Enzyme-linked immunosorbent assays (ELISA)

While antibodies (Abs) detection by ELISA cannot differentiate between active infection and past infection as Abs persists after the treatment of the affected animals, but it is useful as an epidemiological tool. ELISA proved to be more sensitive and specific than stained blood films. The ELISA assay detected IgM of T. evansi provides sensitivity and specificity ranged from 90% to 95% [19].

Latex Agglutination

Surratex detects circulating trypanosome antigens The Surratex reagent is a suspension of latex particles which sensitized with a monoclonal antibody against T. evansi internal antigen [20].

Immunohistochemical

Inducible nitric oxide synthase (iNOS) and interleukin-17 (IL-17) were expressed immunohistochemically in brain, spleen and liver of infected animals increased significantly with progression of the T. evansi infection [21].

Molecular Diagnosis

Many molecular tests, such as conventional and real time PCR have been used to diagnose camel trypanosomiasis through the detection of trypanosomal DNA in blood samples in both the camel and the insect vectors. Molecular tests are more sensitive than other techniques, since it is 90% sensitive when compared with parasitological and serological techniques and have the advantage of being capable of classifying parasites at the subspecies level [22].

Control of trypanosomiasis

Control strategies of trypanosomiasis include four categories; (1) treatment using chemotherapy and chemoprophylaxis drugs to kill T. evansi may be either curative drug, such as melarsomine (Cymelarsan), (2) vector control by using traps and/or impregnated screens and also, using insecticides in form of vapors on livestock or in its environment (Fig. 4a, b), (3). avoid infected introduction of the animals (isolation) and stay for 4-weeks quarantine to maintain the status of non-infected camels, and (4) vaccination of Trypanosoma spp. which may failed due to the different antigens isolated from different types and strains of trypanosome species [23].

Babesiosis

Camel babesiosis is an acute to chronic infectious disease that is distributed all over the world that transmitted by tick such as Hyalomma, Rhipicephalus and Dermacentor. The infective stages of babesia are found intra and extra of the red blood corpuscles. The disease is responsible for deteriorative effects such as, fever, anemia,
hemoglobinuria, high morbidity, and substantial economic losses [14].

**Diagnosis of Babesiosis**

The firstly diagnosis of babesiosis is achieved by using by microscopic examination using GSBS which give lower prevalence of 43.6%, while by PCR method the infection rate was 74.5%. So, the molecular and phylogenetic diagnostics are more accurate in determining the species of parasites [24].

**Control of Babesiosis**

The control planes of Babesia spp infection were not commonly available, successful control and prevention is dependent on the suitable vector control using acaricides such as organophosphates, pyrethroids, and amidines. And treatment either by S/C or I/M injection of some drugs such as Imidocarb (1-3 mg/kg), or Diminazene aceturate (3-5 mg/kg) Different vaccine candidate of Babesia spp are available, it gives 80% protection and the immunity lasts for about 6 months [25].

**Theileriosis**

Theileriosis s one of the most common tick-borne diseases. It has a broad distribution extended from North Africa to China. *Theileria spp.* is an intracellular protozoan parasite infecting leukocytes and erythrocytes of animals, such as *T. cervi*, *T. capreoli*, *T. annulata*, *T. mutans*, and *T. ovis*. The theileriosis disease causes heavy economic losses low productivity of milk, and may the death of infected animals besides the high cost of the treatment [26].

**Diagnosis of Theileriosis**

The pathognomonic clinical signs include, increase in camel temperature, enaciation, watery ocular lacrimation, diarrhea, in addition to enlargement of the superficial lymph nodes. The haematological picture of diseased camels indicated progressive haemolytic anaemia with differentiation in clotting factor. Molecular diagnosis by PCR revealed positive general *Theileria* spp (70 %), while 60 % for *T. annulata and 10 % for other *Theileria* spp. Moreover, amplify protozoan 18S rRNA gene sequences with primers A/B was used to detect and differentiate *Theileria* spp [27].

**Control of Theileriosis**

The control of theileriosis can occur through treatment of infected animals by use acaricides and vaccination. The attenuated live vaccines established from local strains of *T. annulata* are available in Tunisia, Sudan and Egypt [28].

**Gastrointestinal protozoan parasites**

The most investigated gastrointestinal protozoan parasites of camels are including, *Cryptosporidium* spp., *Eimeria* spp. (Coccidiosis), *Giardia duodenalis*, *Balantidium coli* and *Enterocytozoon bieneusi* [29].

**Cryptosporidiosis**

The disease is caused by *Cryptosporidium* spp., which is a ubiquitous apicomplexan parasite that mainly infects the gastrointestinal epithelium of human and animal hosts. Three species of *cryptosporidium* spp.; *C. muris*, *C. parvum*, and *C. andersoni* found in camelids [30]. Infected camels suffer from severe watery diarrhea, dehydration which leads to death in newborn camel and in severe cases and camel plays a role in the transmission of zoonotic species of *Cryptosporidium* [8].

**Diagnosis of Cryptosporidiosis**

**Microscopical diagnosis**

Microscopical examination of feces and abomasal mucosa of camels and staining with different kinds of stains as Nomarski interference-contrast and modified Ziehl-Neelson were used for identification of oocysts; which is the most sensitive (83%) and specific (99%) and with a relatively low cost per test. The oocysts appear spherical organisms contain four sporozoites (Fig. 5 a, b, c) [31].

**Immunological Diagnosis**

Immunological assays are based on either antigen detection or antibody detection. They have been reported to yield good sensitivity and specificity in the range of 93%–100% (Fig. 5 d) [32].

**Histopathological Diagnosis**

Histopathological examination showed a large number of variably-sized (3–5 μm in diameter) circular basophilic oocysts arranged in rows adherent to the mucosal surface and occasionally free within the lumen (Fig. 5 e) [33].

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Fig. 5. *Cryptosporidium* sp. oocysts in camel feces; wet mount (a), Nomarski interference-contrast stained (b), modified Ziehl-Neelsen stained (c), labeled with immunofluorescent antibodies (d) and histopathological stained intestinal epithelia with H&E (e) embedded in the mucosal crypts (black arrows). Ghazy et al. [31].

Molecular Diagnosis

PCR technique is a sensitive, rapid, and more specific method for the direct detection of *C. parvum* in stool specimens and additionally, PCR-based heat shock protein gene of *C. parvum* in camel fecal samples. PCR with sequencing of about 800 base-pair fragments of the gene encoding 18S rRNA commonly used for identification *Cryptosporidium* species [34].

Control of cryptosporidiosis

Actually, the management of cryptosporidiosis is difficult because of the stability of its oocysts in the environment, low infective dose, and the huge number of sporulated oocysts shed from different hosts. Several drugs have reported to reduce *cryptosporidium* oocysts number in vitro as Nitazoxanide, Aminoglycoside, Dinitroanilineoryzalin and lasalocid [31]. Some preventive hygienic measures are recommended to manage the parasite isolation of ill animals, using alternative oocyst disinfectants such as ozone and UV radiation, using activated-sludge, sand filtration systems in treatment of wastewater and sewage and no effective vaccines are available to prevent cryptosporidiosis, and then control often depends on prevention, risk reduction, and rehydration therapy [35, 36].

Eimeriosis (Coccidiosis)

Coccidiosis is an intestinal protozoan infection caused by apicomplexan parasites of the genus *Eimeria*. Five *Eimeria* spp. can infect the camel intestine including; *E. cameli, E. rajasthani, E. bactriani, E. dromedarii, E. pellerdyi* and *E. leuckarti* (Fig. 6 a-d). Eimeriosis disease has a great economic importance because of losses due to enteritis, diarrhoea and decrease in weight of camels, also affects meat yield and quality [37].

Diagnosis of Eimeriosis

Camel faecal samples examined to diagnosis the prevalence of *Eimeria* species, fecal smears stained with modified Ziehl–Neelsen used to detect the oocysts of *Eimeria* spp. [38].

Control of Eimeriosis

There are no anti-coccidial drugs approved specifically for camels. The disease progression is so rapid that any therapeutic treatment may be too late, for this reason, continuous medication in food or water often used for prophylactic treatment. Sulfadimidine used as treatment for camels’ coccidiosis given as an aquatic suspension orally for 10 days in a dose 30 mg/ kg body weight [29].
Intracellular (tissue) protozoan parasitic diseases

They invade tissues and produce tissue cysts, such as *Sarcocystis* spp. and *Toxoplasma gondii* [39].

**Sarcocystosis (Sarcocystis cameli infection)**

There are two main species of *Sarcocystis* that have been reported from camels; *S. ippeni* (microscopic) and *S. cameli* (macroscopic & microscopic). Camels serve as intermediate hosts and can be affected after ingestion of sporulated oocysts that excreted in final host (carnivores). Sporozoites excyst in the camel gut then migrate to the muscles to produce the *Sarcocystis*. Sarcocystosis causes a significant economic loss due to the downgrade of camel meat quality, productivity and milk yield [40].

**Diagnosis of Sarcocystosis**

*Macropscopic and Microscopical examination*

Direct examination of *Sarcocystis spp.* infections in post mortem of slaughtered animals is the usual diagnostic method that are characterized by the presence of parasite-full sacs that ranged in size from few micrometers to several centimeters, in the heart, diaphragm, and oesophagus. The tissue digestion method is used for the detection of bradyzoites in different samples of the organs and to confirm the results obtained by direct examination (Fig. 7 a, b, c) [41].

Transmission Electron Microscopy (TEM)

In which the primary cyst wall s appeared as a thick electron-dense layer with spine-like protrusions [39].

**Histopathological examination**

Histopathological examination revealed the presence of morphologically distinct *Sarcocystis* embedded within the muscle fibers, cyst walls. A palisade-like thick wall or a smooth thin wall *Sarcocystis* of camel appear dark blue-colored due to the presence of many bradyzoites inside the cysts, no inflammatory reaction in the tissue surrounding the cysts, the size of bradyzoites were 14–15 × 3–4 µm, the total thickness of the sarcocysts wall was 2.3-3.0 µm [40].
Serological diagnosis

Several serological techniques such as ELISA and indirect fluorescent antibody test (IFAT) were used for the diagnosis of Sarcocystis infection. However, these methods have low sensitivity and specificity because of the cross-reactivity between the various Sarcocystis spp. [42].

Molecular Diagnosis

Two samples R10C (thick-walled cyst) and D7S (thin-walled cyst), isolated from Camels dromedarius in Riyadh and Dammam respectively, by using Power SYBR™ Green PCR Master Mix and a 7500 Real-Time PCR System, the tree shows that MK948444 (D7S) and MK948442 (R10C) are placed in a clade with S. levinei. MK948443 (D10S) was grouped with Sarcocystis miescheriana. MK948441 (R16C) was grouped with Sarcocystis fayeri [39].

Control of Sarcocystosis

There is no effective treatment against chronic intracellular Sarcocystis because of the prey-predator life cycle of its most species. So, the main strategies of control are preventing the ingestion of prey carcasses or infected tissues by carnivorous animals and to reduce grass and water contamination with their feces. Drugs can be used to reduce illness and protect other animals against sarcocystosis. Amprolium used at 100 mg/kg/day for 30 days and vaccines against sarcocystis are not available for camels [42].

Toxoplasmosis

Toxoplasmosis caused by Toxoplasma gondii (T. gondii). It is an apicomplexan, obligate, and intracellular protozoan parasite. It has three life cycle stages named sporozoites, tachyzoites, and bradyzoites [43]. T. gondii infects camels after the ingestion of oocysts-contaminated food or by inhalation of sporulated oocysts from air, which is shed in the feces of cats or wild felids in the environment [44]. Toxoplasmosis is considered a major cause of reproductive losses in animals worldwide. It causes a huge economic loss in the form of abortion, stillbirth, and neonatal losses (43). In addition, toxoplasmosis is a relevant zoonosis and infection in camel may play a major role in its transmission to humans after subjected to undercooked meat or polluted milk [45, 46].

Diagnosis of toxoplasmosis

According to the non-specificity of toxoplasmosis clinic signs and the difficulty of direct examination, diagnosis of T. gondii infection is mainly through isolation of the infective stages, serological and molecular diagnosis [47].

Biologic Diagnosis (Microscopy Findings)

Direct visualisation of the parasite infective stages; tachyzoites, tissue cysts with bradyzoites, and oocysts in biopsy (ant-mortem) or autopsy (post-mortem) investigated material specimens under the microscope (Fig. 8 a, b, c) [48].

Serological Diagnosis

Serodiagnosis of T. gondii is based on using native antigens to detect a specific antibody against T. gondii such as Modified agglutination test (MAT), Enzyme linked immunosorbant assay ELISA and Latesx agglutination test (LAT) [49, 50].
Molecular diagnosis
Various recombinant antigens were developed and used in PCR assay, B1 gene is considered as more effective diagnostic tool [51].

Control of toxoplasmosis
Hygienic control was applied to reduce animal feed contamination by T. gondii oocysts [52]. keep cats out of the animal barns, and also, pregnant female animals should avoid contact with cat feces and no vaccine is found for camels [53].

Conclusion
Camels are important animals raised in arid and semi-arid areas of the world. Even though it provides many advantages to people, camel has received very little attention as compared to other species of domesticated animals. Camels are affected by several diseases. The most dangerous and zoonotic diseases infecting camels are protozoal infections such as; trypanosomiasis, cryptosporidiosis, and toxoplasmosis. Early diagnosis of parasitic infections by various techniques such as microscopic, immunological (such as ELISA, DOT ELISA, the immunofluorescent antibody test (IFAT), card agglutination test (CAT), and latex agglutination), and also the molecular approaches which was the most sensitive and specific than both microscopic and serological diagnosis for the detection of parasitic diseases and identification of parasites in camels. Chemical drugs, biological control and natural products can be used to control and prevent the parasite infections. Besides the presence of some trials to produce specific vaccines.

Recommendations
- In my point of view, more attention is needed to the modern animal health programs by camel owners and veterinary services.
- Additional researches on camel diseases are recommended with special emphasis on diagnosis especially of zoonotic infections and control methods for certain diseases such as trypanosomiasis, cryptosporidiosis, and toxoplasmosis.
- In addition, camel pastoralists need to separate infested camels from healthy ones, clean fomites and utensils before and after use and treating herds when one or more camels show clinical signs.
- More attention to camel health and management of its diseases should be arisen by governmental and non-governmental organizations working on livestock health.
- Increasing the awareness of the potential of medicinal plants and their role in development of much needed new safe, low cost, and easily used anti-parasitic drugs.
- Further studies should be done using apitherapy in different branches of medicine especially in the control of parasitic infections due to its antimicrobial effect.
- Finally, integrated control system, which includes more than one approach can result in potent impact on parasites elimination than a single approach.

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References


