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### Case Report

## Abortion Related to *Toxoplasma gondii* Infection in a Bactrian Camel (*Camelus bactrianus*) in Greece: A Case Report

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Received 12 Jan 2021 Accepted 19 Mar 2021	<b>Abstract</b> A female, 5 yr old Bactrian camel was presented to the Exotic and Wildlife Medicine Unit, School of Veterinary Medicine, Aristotle University of Thessaloniki, Thessaloniki, Greece, with severe dehydration, depression, anorexia, mild dyspnea and diarrhea. Supportive treatment immediately initiated with fluids, electrolytes and broad-spectrum antibiotics. The general condition of the animal was stable for the next 3 days, but at 4 <sup>th</sup> day became worse, since the camel remained in sternal recumbency, denied to drink water and abortion of a mummified fetus was noticed. The aborted fetus and fetal membranes were submitted for laboratory examinations (bacterial cultures, MZN, cytology, PCR) that revealed <i>Toxoplasma gondii</i> infection. Treatment with sulfadimidine improved the situation of the animal that returned to its farm 1 week later. This seems to be the first reported case in the literature of confirmed toxoplasmic abortion in camels.
<b>Keywords:</b> Camel; Toxoplasmosis; Abortion; Sulfadimidine; Greece	
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### Introduction

**T**oxoplasmosis is one of the most widespread zoonotic diseases caused by the obligatory intracellular protozoan para-

site *Toxoplasma gondii*. The domestic cat and wild Felidae serve as a natural reservoir and the definitive host of the parasite. However, a



wide range of mammals including humans, birds and reptiles could be affected and thus represent the intermediate host of the parasite (1-4).

Toxoplasmic abortion has been described extensively in sheep and goats (3). According to the best of the authors' knowledge, documented abortions related to *T. gondii* infection in camels are rare, as well as in other camelids (llama, alpaca) (4-6). In this article, a case report of an abortion accompanied by systemic signs in a pet camel, as well as the successful therapeutic management of the disease is presented.

### Case Report

A female, 5 yr old Bactrian camel (*Camelus bactrianus*) was presented to the Exotic and Wildlife Medicine Unit, School of Veterinary Medicine, Aristotle University of Thessaloniki, Greece. The examination and all the clinical process was made under the Greek legislation with the written consent of the animal owner. The animal had been purchased from the Netherlands about 1 year ago and was reared as a single one in a farm together with other animal species, as llamas, deer, small ruminants, equids, hens, geese and pheasants. The camel's diet consisted of alfalfa hay, straw, carrots, barley, wheat and corn. Therefore, 3 days before its admission to the clinic, the camel had been already treated unsuccessfully for indigestion problems by a local veterinarian, since it showed signs of abdominal pain and discomfort along with anorexia.

### Clinico-pathological examinations and findings

The clinical signs on presentation included severe dehydration, depression, anorexia mild dyspnea and diarrhea, while the rectal temperature was 37.2 °C. Haematological examinations revealed increased values over the reference range (7, 8) for hematocrit (PCV), hemoglobin, white blood cells count (WBC), while platelet number (PLT) was within the normal limits (Table 1). From the blood serum bio-

chemistry, increased values of blood urea nitrogen (BUN), creatinine, glucose, lactate dehydrogenase (LDH) were noticed.

Supportive treatment immediately initiated with fluids, electrolytes and broad-spectrum antibiotics. Penicillin-streptomycin (Vetricillin®-CEVA containing 20 x 10<sup>6</sup> procaine penicillin per 100 ml and 20,000 mg dihydrostreptomycin per 100 ml) at a rate 1 ml/20 kg was the antibiotic of choice. In addition, clanobutin (Bykahepar®-Schering-Plough) and vitamin B<sub>12</sub> with Butaphosphan (Catosal®-Bayer) were administered for the improvement of appetite. The general condition of the animal was stable for the next 3 days, and then antibiotic treatment with enrofloxacin (Baytril®-Bayer) initiated instead of penicillin-streptomycin. Abortion of a mummified fetus about 27 kg was noticed in the morning of the 4<sup>th</sup> day (Fig. 1), whereas the condition of the camel became worse, since it remained in sternal recumbency and denied to drink water.

The aborted fetus and fetal membranes were submitted for laboratory examinations (bacteriological and parasitological).

Stomach content and spleen from all the aborted fetuses were submitted for microbiological examination according to standard culture techniques (9). Stomach contents were spread directly to proper culture media while 0.5 cm<sup>3</sup> of each spleen tissue was first homogenized with 200 µl PBS using a tissue grinder (MINILYS Bertin France). Specifically, Farell's medium was used for the recovery of *Brucella* spp., a two-stage enrichment procedure using half and full strength Fraser liquid medium and Aloa agar was used for the recovery of *Listeria* spp., XLD medium was used for *Salmonella* spp. recovery and PPLO broth and PPLO agar was used for the recovery of *Mycoplasma* spp. The procedure followed for the *Campylobacter* spp. and *C. jejuni* was according the OIE Manual 2012 (10).

Finally, direct smears from cotyledons as well as from the fetus, lung and liver were stained by Modified Ziehl-Neelsen stain, whereas the presence of *Chlamydia* spp. was

investigated using dark field microscope. All manipulations were carried under appropriate

biosafety conditions according OIE 2013 (11), in a BSL 3 chamber.

**Table 1:** Haematological and biochemical values of the camel at the first day (day 1) in the clinic and 3 days later (day 4)

<i>Day</i>	<i>1</i>	<i>4</i>
Haematocrit-HCT (%)	48.8	50.2
Haemoglobin-Hgb (g/dl)	16.2	16.6
White Blood Count-WBC (/μl)	31,020	31,240
Platelets-PLT (/μl)	581,000	599,000
Total solids-TS (g/dl)	7.2	8.2
Neutrophils-NEUT (/μl)	23,340	22,640
Lymphocytes-LYMPH (μl)	6,090	7,340
Monocytes-MON (/μl)	1,450	1,090
Eosinophils-EOS (/μl)	100	80
Total Proteins-TP (g/dl)	7.00	6.80
Albumins –ALB (g/dl)	4.50	4.1
Blood Urea Nitrogen-BUN (g/dl)	233	245
Creatinine-CREA (g/dl)	15.1	10.5
Glucose-GLU (g/dl)	278	115
Total Bilirubin-TB (g/dl)	0.3	0.4
Direct Bilirubin-DB (g/dl)	0.01	<0.01
Alkaline Phosphatase-ALP (U/l)	204	357
Alanine Aminotransferase-ALT(U/l)	30	22
γ-Glutamyl Transferase-γ-GT (U/l)	91	47
Creatinine Phosphokinase-CK (U/l)	89	190
Lactate Dehydrogenase-LDH (U/l)	1,268	1,319
Phosphate-P (mg/dl)	10.5	6.5
Calcium-Ca (mg/dl)	11	11.1
Potassium-K (mg/dl)	4.5	5.7
Sodium-Na (mg/dl)	145	154



**Fig. 1:** The aborted camel fetus

A blood sample from the aborting camel was collected by jugular venepuncture, the serum was separated on the same day and it was stored at -20 °C. To detect *T. gondii* specific antibodies, ELISA was performed using soluble *T. gondii* antigen (from in vivo cultured parasites) in a final concentration of 3 µg/mL. Unfortunately, as not any positive control serum sample from a *Toxoplasma*-infected camel was available, dilutions of the sera were performed (1/100, 1/200, 1/400, 1/800, 1/1600 and 1/3200) in order to demonstrate the possible comparative positive reaction and the title.

Impression smears of the brain were examined for *T. gondii* tissue cysts (12). A portion of cerebrum (enough to fill an area under a 22 mm coverslip) was crushed between a glass slide and coverslip and examined unstained for tissue cysts in 3-4 brain smears at 100x magnification. One-half of the brain was ground in a manual glass homogenizer using saline. A small portion of brain homogenate was spread on 2-3 slides, which allowed in air to dry. Then samples stained with Giemsa stain and examined for *T. gondii* tissue cysts at 100x magnification.

Genetic material (DNA) was extracted using DNA purification kit (Promega, USA) from a pooled sample of different tissues (placenta, spleen, liver, lung and brain) collected from the aborted foetus. The PCR assay targeted the 529 bp fragment, which is highly sensitive and a preferred marker for the detection of *T. gondii* in human and animal tissues (13). The PCR product was examined by electrophoresis in a 2% agarose gel stained with ethidium bromide and visualized under UV light.

Using ELISA, we confirmed that the consecutive dilutions of the sera sample remained positive up to 1/1600 confirming the strong positivity of the mother's blood sample to *T. gondii* antigen (title 1/1600).

*T. gondii* tissue cysts were found in direct smears of the aborted fetus brain (Fig. 2, 3) and of the fetal membranes, while the moth-

er's antibody titer in blood serum was found strongly positive to toxoplasmosis. Polymerase chain reaction (PCR) was found positive and *T. gondii* DNA was detected in the pooled sample from the tissues of the aborted foetus. Fetal membranes and stomach content originated from the aborted fetus were examined for the presence of *Brucella* spp., *Chlamydia* spp., *Coxiella burnetii*, *Salmonella* spp., *Listeria* spp., *Mycoplasma* spp. and *Campylobacter* spp. All the above examinations were found negative.

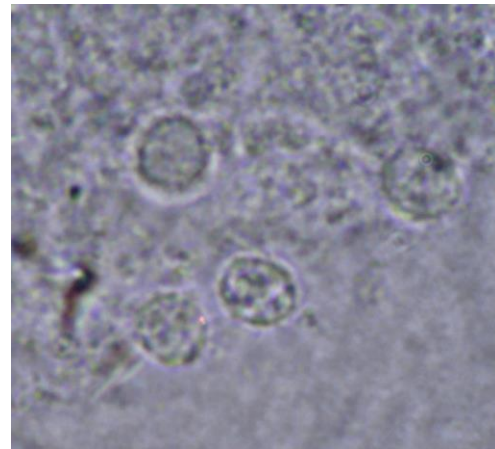


Fig. 2: *Toxoplasma gondii* tissue cysts in fresh brain smear (x100)

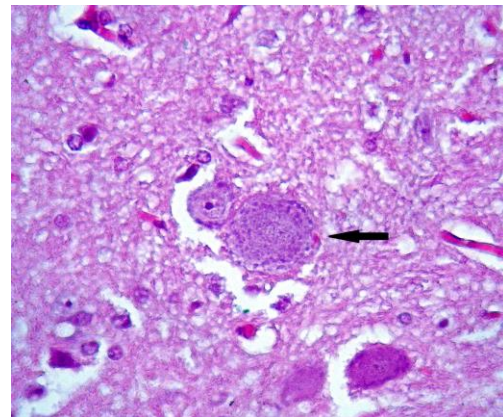


Fig. 3: *Toxoplasma gondii* tissue cyst (brain histology, haematoxylin-eosin x40)

Abortion of the camel was related to toxoplasmosis infection, based on the positive fetus direct smear results, PCR and the mother's antibody titer, therefore—toxoplasmosis treat-

ment, already applied in small ruminants, with sulphadimidine (Sulfadimidine<sup>®</sup>-CEVA) at a dose of 33 mg/kg (14, 15), immediately initiated. The camel responded quickly to treatment since 24 h after the first treatment the animal started to eat. During the next days the animal's general condition improved gradually, as well as its hematological and biochemical profile, so was discharged after 11 days of hospitalization and returned to the farm. The camel's general condition was re-assessed 1 month later after a visit to the farm, but no blood samples were taken since the camel was not easy to be restrained. The camel remained healthy 4 years later, according to the owner.

## Discussion

Camels represent one of the most common animals used for meat, milk, transportation and sport in many African and Asian countries, as well as in Australia, since they are important for their economies. During the last years, camels have gained popularity as pets or show animals in Europe and USA.

Toxoplasmosis is a common parasitic zoonosis caused by the protozoan parasite *T. gondii*, related to morbidity and mortality in both human and other warm-blooded animals. Camels are usually asymptomatic according to several serological surveys (5, 8, 16, 17). On the contrary, congenital toxoplasmosis results in weak, unable to suckle calves or sudden deaths, immediately after birth (18). The only clinical case of documented toxoplasmosis in a pet camel has been reported by Hagemoser et al in 1990 (5). The 6 yr old female dromedary camel, presented with a history of anorexia and dyspnea. The animal had aborted 4 weeks before admission, but the cause of the abortion had not been investigated. Cytology of smears from the pleural fluid revealed the presence of tachyzoites of *T. gondii*. In addition, results of indirect hemagglutination antibody test for *T. gondii* were strongly positive and the camel was diagnosed with acute toxoplasmosis. That animal died a few days later

despite the treatment with trimethoprim-sulfadiazine for 7 days. Presumably, its abortion could be attributed to toxoplasmosis. The case presented thereby seems to be the first, but not investigated and confirmed, case of toxoplasmic abortion in camels. In contrast, in the present manuscript toxoplasmic abortion was confirmed by laboratory examinations.

In described spontaneous and experimental cases of toxoplasmosis in small ruminants (14, 19, 20) the affected animals usually have fever and inappetence, some hours before the abortion, but these signs resolve without medical treatment after abortion. In the present case, the camel was probably bought pregnant about 12 months ago, since there were no other camels in the farm and gestation period in camels lasts 15 months, and the owner was unaware of the animal's pregnancy.

The vast majority of natural *T. gondii* infections in domestic animals are subclinical. However, *T. gondii* infection is an important cause of abortion and perinatal mortality in sheep and goats (2, 20). In our case severe general signs as dehydration, depression, anorexia mild dyspnea and diarrhea, were observed in the camel 7 days before the abortion. They resolved 48 h later after the supportive and anti-toxoplasmic treatment with sulfadimidine that started 24 h after abortion. Sulfadimidine, a drug that is used to control toxoplasmic abortions in sheep and goats (12, 13, 20) seems to had good results for the treatment of toxoplasmosis in the aforementioned camel. In the case of Hagemoser et al. (5), the camel did not respond to anti-toxoplasmic treatment with sulphonamides and died, possibly because there were severe complications in the pleural cavity. The response of the treatment is probably related to the type and severity of the clinical signs as well as the quick diagnosis and treatment initiation.

## Conclusion

Toxoplasmosis in camels is important because of its zoonotic importance, since camels are major sources of meat in many countries. The prevalence of *T. gondii* infection even though is common in pastoral camels in many countries, and of economic and public health significance, it seems to be not rare in pet camels as well; especially when these animals are kept in private collections as pets and in contact with other species, without taking any specific health prevention measures. In fact, it is important to be included in the differential diagnosis of camel abortions.

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## Conflict of interest

The authors declare that there is no conflict of interest.

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