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Molecular Detection of Zoonotic Pathogens in the Blood and Tissues of Camels (*Camelus dromedarius*) in Central Desert of Iran

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Dromedary camels (*Camelus dromedarius*) play a major economic role in many countries in Africa and Asia. Although they are resistant to harsh environmental conditions, they are susceptible to a wide range of zoonotic agents. This study aimed to provide an overview on the prevalence of selected zoonotic pathogens in blood and tissues of camels in central Iran. Blood, liver, portal lymph node, and brain were collected from 100 apparently healthy camels at a slaughterhouse in Qom city to assess the presence of DNA of *Brucella* spp., *Trypanosoma* spp., *Coxiella burnetii*, and *Bartonella* spp. PCR products were sequenced bidirectionally and phylogenetic analyses were performed. Eleven percent of camels tested positive for *Brucella abortus* (3%) and *Trypanosoma evansi* (8%). *Coxiella burnetii* and *Bartonella* spp. DNA was not detected. Our data demonstrate that camels from Iran contribute to the epidemiology of some zoonotic pathogens. Performing proper control strategies, such as vaccination of camels and humans in contact with them, test-and-slaughter policy, and education of the general population is necessary for minimizing the risk of zoonotic infection.

INTRODUCTION

The *Camelidae* family is comprised of three species of Old World Camels (OWC; *Camelus dromedarius*, *C. bactrianus*, *C. ferus*) and four species of New World Camels (syn. South American camelids, NWC; *Lama*

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Abbreviations: DNA, deoxyribonucleic acid; PCR, polymerase chain reaction; OWC, old world camelids; NWC, new world camelids; Q fever, *Coxiella burnetii* infection; min, minute; sec, second; *gltA*, citrate synthase; *rpoB*, beta subunit of RNA polymerase; *ITS*, internal transcribed spacer; APOL1, apolipoprotein L1.

Keywords: *Bartonella*, *Brucella abortus*, camel, *Camelus dromedarius*, *Coxiella burnetii*, Iran, One Health, PCR, *Trypanosoma evansi*, zoonosis

Author Contributions: AB: Conceptualization; Supervision; Project administration; Funding acquisition; Review and Editing. AG: Experiments; Data Analysis; Writing; Review and Editing. SA: Experiments; Data Analysis; Review and Editing. SK: Experiments; Data Analysis; Review and Editing. SE: Experiments; Data Analysis; Review and Editing. BBC: Review and Editing. AS: Conceptualization; Supervision; Project administration; Funding acquisition; Data Analysis; Writing; Review and Editing.

glama, *Lama guanicoe*, *Vicugna pacos*, *Vicugna vicugna*). The world population of one-humped camels, also known as dromedary (*Camelus dromedarius*), is on the rise [1]. The dromedary camel is specifically adapted to life in hot, arid areas of the world, notably Western Asia, Africa, and India, with considerable (> 1 million individuals) feral population in Australia, where they were introduced in the 19th century [2]. Dromedaries are an important source of meat and milk in 47 countries, playing a crucial role in their economy. Their importance as "food security livestock" is increasing with the ongoing earth's desertification. Evidence of this is that between the years 2008 and 2018, the world camel population increased by 21% compared to only 5%, 9%, 15%, and 4% for cattle, sheep, goats, and pigs, respectively [1]. However, despite being extremely resistant to harsh environmental conditions, dromedaries can get infected with several zoonotic pathogens, thus posing a public health risk [3,4]. An overview of the most important zoonotic viral bacterial, fungal, and parasitic diseases of camelids was performed by Wernery and Rodriguez Caveney [5].

Brucellosis, a contagious disease caused by a zoonotic bacterium of the genus *Brucella*, is of greatest concern, not only because it is zoonotic but also causes severe economic losses for farmers and ranchers across the world in lost milk, reduced fertility, stillbirths, and abortions [6,7]. Camels can be infected with different biovars of either *B. abortus* and *B. melitensis*, both of which are the main causative agents of human brucellosis [8]. Camel brucellosis is endemic in all camel-rearing countries with exception of Australia [9]. Human infection due to *Brucella* from camels is known to occur mostly through consumption of raw milk, but also through close contact with camels [10,11]. Brucellosis has been diagnosed in some camel herds and slaughterhouse workers in Iran [12-15].

Query (Q) fever (or Coxiellosis) is another zoonotic infection with a global distribution with the exception of New Zealand [16]. The causal agent, *Coxiella burnetii*, is an intracellular and pleomorphic Gram-negative coccobacillus that affects mostly livestock and humans [16]. Camels like ruminants acquire the infection most frequently through inhalation of aerosolized bacteria that are spread in the environment by infected animals after delivery or abortion. However, *C. burnetii* is also present in urine, feces, and milk of infected animals [17,18]. There is also growing evidence on the importance of ticks in the epidemiology of Q fever [19,20]. Serological studies reported prevalence of up to 90% in dromedaries [21]. In studies that tested different animal hosts, antibodies against *C. burnetii* were more prevalent in dromedaries than cattle, sheep, and goats, suggestive of genetic susceptibility of camels to *C. burnetii*, or host preference of tick vectors to camel [18,21-23]. Interestingly, a serosurvey on pastoralists and their livestock revealed that camel breeders had a nine times higher risk of being *C. burnetii* seropositive compared to the general public [22]. An overview of Q fever in camelids has been compiled by Wernery et al. [24].

Bartonellosis is a globally emerging zoonotic bacterial disease. The genus *Bartonella* is comprised of Gram-negative intracellular bacteria that infect the endothelial cells and erythrocytes of mammals. These vector-borne bacteria are mainly transmitted through contaminated fecal materials of hematophagous arthropod vectors [25]. Currently, more than 40 *Bartonella* named and Candidatus species are recognized, several of them being zoonotic [26]. There is limited information on camel bartonellosis. However, DNA of zoonotic *B. henselae* and *B. rochalimae* in addition to *B. bovis* and *B. dromedarii* has been detected in the blood of camels and ticks collected from them in West Asia and North Africa [27-29].

Camels are affected by several zoonotic *Trypanosoma* species, such as *T. evansi*, *T. brucei*, *T. congolense*, and *T. vivax* [3]. *Trypanosoma evansi*, the agent of "surra", is the most pathogenic and economically important protozoan parasite of camels in the world [30]. It affects a wide range of domestic and wild mammals in Africa, Asia and South America. Since this species can be mechanically transmitted by virtually all biting flies, its geographical distribution is potentially unlimited [31]. This parasite is now considered as an emerging zoonotic parasite. So far human cases of *T. evansi* infection have been reported from India, Sri Lanka, Egypt, and Thailand [3,32,33].

An estimated 195,500 dromedaries are raised in 21 of the 31 Iranian provinces mainly for meat but also for milk and wool production [1,34]. According to the latest official reports 27,000 camels were slaughtered for production of > 5,400 tons of meat in 2019 [35]. Given previous reports on the presence of brucellosis, Q fever, bartonellosis, and trypanosomosis in camels of Iran [13,36-38], this study aimed to examine blood, brain, liver, and lymph nodes of camels by molecular techniques for the detection of these zoonotic pathogens.

MATERIALS AND METHODS

Study Area

This study was performed from January to June 2018 on 100 apparently healthy camels in a local slaughterhouse in Qom (34.6416° N, 50.8746° E), a region in the center of Iran characterized with a hot desert climate (Köppen–Geiger BWh) (Figure 1). Two breeds of dromedaries (Kalkooyi and Baloochi) are raised in four villages of Qom, ie, Seyd-Abad, Kaj-Abad, Qom-Rood, Kooh-Sefid. Camels were from both sexes (90 males and 10 females) and age ranged between 12–30 months (average=21 months).

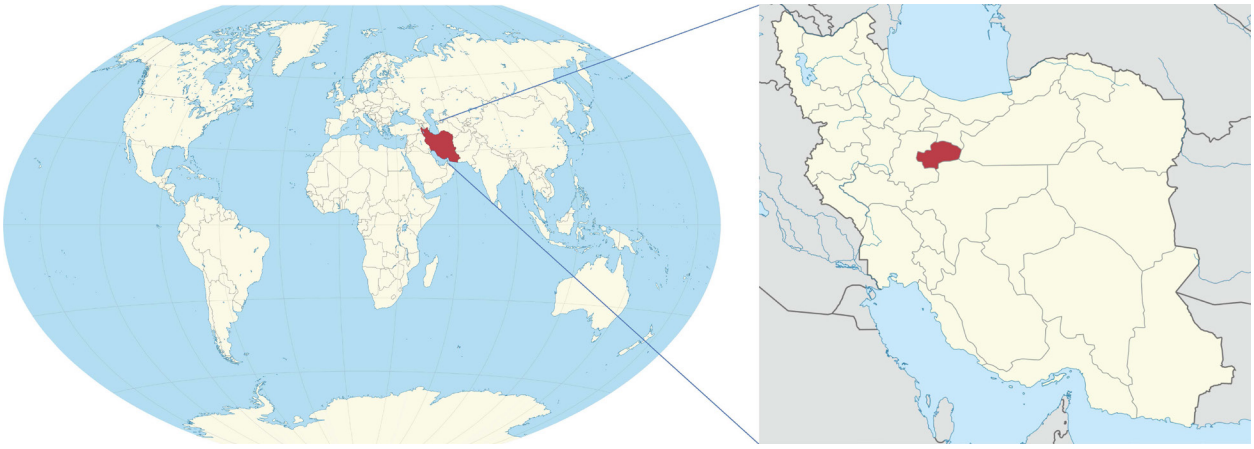

Figure 1. Samples were collected from Qom province in Iran.

Collection of Samples

Immediately after slaughter, jugular vein blood samples (10 ml) were collected in K_3EDTA coated tubes for DNA extraction. Also, from each camel tissue specimens from brain, liver and portal lymph node were collected. Tissue samples (approximately 1 × 1 cm) were stored at −80 °C freezer until DNA extraction.

Ethical Approval

Not applicable.

DNA Extraction

Genomic DNA was extracted from 300 µl blood and 10–20 mg of tissues ie, brain, liver and portal lymph node of the camels using WizPrep™ (Wizbiosolutions, Seongnam, South Korea) ie, WizPrep™ DNA Extraction Kit for blood and WizPrep™ gDNA Mini Kit for tissues according to the manufacturer's instructions. Quantitative and qualitative assessments were performed using NanoDrop 2000™ (Thermo Scientific, Waltham, MA, USA) and 0.8% agarose gels. All samples were used tested for the presence of *Brucella* spp., *Bartonella* spp., *Coxiella* spp., and *Trypanosoma* spp. DNA by using conventional PCR (cPCR).

PCR Assay

For the detection of *Brucella* spp. after an initial screening with B4/B5 genus-specific primers [39] a combinatorial PCR which detects *B. abortus*, *B. melitensis*, *B. canis*, and *B. suis* simultaneously was used [40]. Positive controls were *B. abortus* strain RB51 and *B. melitensis* strain Rev1 purchased from Razi vaccine and Serum Research Institute, Karaj, Iran.

For the detection of *Bartonella* spp. three PCR protocols were used: i) primers BhCS.781p and BhCS.1137n amplifying citrate synthase gene (*gltA*) [41], ii) primers 1400F and 2300R amplifying beta subunit of RNA polymerase (*rpoB*) [42], iii) primers 325s and 1100as amplifying 16S-23S ribosomal DNA internal transcribed spacer (*ITS*) [43]. Positive control was *B. henselae* Houston 1 strain.

Detection of *Coxiella* spp. DNA was performed using the primers Trans1 and Trans2 targeting the transposon-like repetitive region of *Coxiella* [44]. Positive control was phenol-killed, purified, and lyophilized cells of the *C. burnetii* Nine Mile, phase I, strain (RSA 493) (Slovakia).

For the detection of *Trypanosoma* spp. primers Kin1 and Kin2 amplifying the *ITS1* (a conserved regions situated between 18S and 5.8S rRNA genes of kinetoplastid species) were used [45]. This PCR protocol can detect and differentiate African livestock trypanosomoses eg, *T. vivax*, *T. simiae*, *T. theileri*, *T. evansi*, *T. equiperdum*, *T. brucei*, and *T. congolense* in a single PCR [46]. Positive control DNA was *T. evansi* isolated from diseased camels in Yazd, Iran by the last author (GenBank® Accession numbers MW272927 and MW272928). All primers and cycling conditions used for the detection of zoonotic pathogens are summarized in Table 1.

All PCRs were performed with the Taq DNA Polymerase Master Mix RED® (Ampliqon, Odense, Denmark) in a SimpliAmp® thermal cycler (Applied Biosystems, Waltham, MA, USA). The amplified products were detected by electrophoresis on 2% agarose gels stained with ethidium bromide (SinaClon, Tehran, Iran).

Sequencing and Phylogenetic Analysis

Purification and sequencing of PCR products were performed with Applied Biosystems 3500 Genetic An-

Table 1. Primers and target genes used in this study.

Pathogen	Primers	Target gene	Amplicon size (bp)	Cycling conditions	Ref
Brucella spp.	B4: TGGCTCGGTTGCCAATATCAA B5: CGCGCTTGCCTTTCAGGTCTG	*BCSP31*	224	95 °C–5 min; (× 40) 95 °C–1 min, 60 °C–1 min, 72 °C–1 min; 72 °C–7 min	[39]
	JPF: GCGCTCAGGCTGCCGACGCAA	*omp2*		95 °C–5 min; (× 35) 95 °C–1 min, 65 °C–1 min, 72 °C–1 min; 72 °C–7 min	[67]
	JPR-ab: CCTTTACGATCCGAGCCGGT		186		[40]
	JPR-ca: CCTTTACGATCCGAGCCGGTA		187		[40]
	1S: GTTCGCTCGACGTAACAGCTG 1AS: GACCGCCGGTACCATAAACCA	*omp31*	249	95 °C–5 min; (× 35) 95 °C–1 min, 65 °C–1 min, 72 °C–1 min; 72 °C–7 min	[40]
Bartonella spp.	BhCS.781p: GGGGACCAGCTCATGGTGG BhCS.1137n: AATGCAAAAAGAACAGTAAACA	*gltA*	379	94 °C–5 min; (× 35) 95 °C–20 sec, 51 °C–30 sec, 72 °C–2 min; 72 °C–7 min	[41]
	1400F: CGCATTGGCTTACTTCGTATG 2300R: GTAGACTGATTAGAACGCTG	*rpoB*	825	94 °C–5 min; (× 35) 94 °C–30 sec, 53 °C–30 sec, 72 °C–1 min; 72 °C–7 min	[42]
	325s: CTTCAGATGATGATCCCAAGCCTTYTGGCG 1100as: GAACCGACGACCCCCTGCTTGCAAAGCA	*ITS*	604	95 °C–2 min; (× 55) 94 °C–15 sec, 66 °C–15 sec, 72 °C–15 sec; 72 °C–7 min	[43]
Coxiella spp.	Trans1: TATGTATCCACCGTAGCCAGT Trans2: CCCAACAACACCTCCTTATTC	transposon-like repetitive region	687	94 °C–10 min; (× 35) 94 °C–30 sec, 63 °C–1 min, 72 °C–1 min; 72 °C–7 min	[44]
Trypanosoma spp.	Kin1: GCGTTCAAAGATTGGGCAAT Kin2: CGCCCGAAAGTTCACC	*ITS1*	*T. vivax*: 305 *T. simiae*/*T. theileri*: 455 *T. evansi*/*T. equiperdum*/*T. brucei*: 540 *T. congolense*: 680 or 750 or 780	94 °C–3 min; (× 4) 94 °C–1 min, 58 °C–1 min, 72 °C–1 min; (× 8) 94 °C–1 min, 56 °C–1 min, 72 °C–1 min; (× 23) 94 °C–1 min, 54 °C–1 min, 72 °C–1 min; 72 °C–7 min	[46]

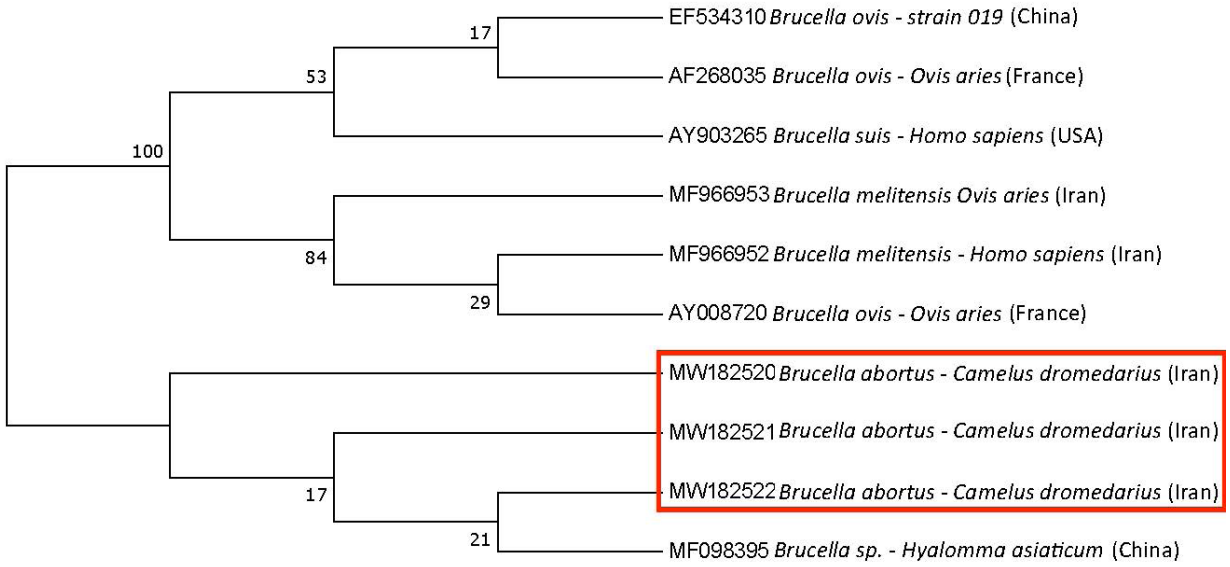

Figure 2. Phylogenetic relationship of *Brucella abortus* in this study (in boxes) to other *Brucella* spp. based on a partial sequence of the *omp2* gene. The analyses were performed using a Neighbor-Joining method [47] and evolutionary analyses were conducted on 1000 bootstrap replications [48] in the MEGA7 software [49].

alyzer (Thermo Fisher Scientific, Waltham, MA, USA) by Pishgam Biotech Company (Tehran, Iran). Sequence reads were curated manually by removing all primer sequences and compared with those available in the GenBank® database using Basic Local Alignment Search Tool (http://blast.ncbi.nlm.nih.gov/Blast.cgi).

In order to investigate the phylogenetic relationships between the *omp2* gene sequences of *Brucella* spp. and *ITS1* gene sequences of *Trypanosoma* spp. obtained in the present study and sequences from GenBank database, we used Neighbor-Joining method [47] and evolutionary analyses were conducted on 1000 bootstrap replications [48] in the MEGA7 software [49]. Trees were drawn to scale, with branch lengths in the same units as those of the evolutionary distances used to infer the phylogenetic tree. The evolutionary distances were computed using the Maximum Composite Likelihood method [50] and were in the units of the number of base substitutions per site. Codon positions included were 1st+2nd+3rd+Noncoding. All positions containing gaps and missing data were eliminated.

RESULTS

Using B4/B5 primers, *Brucella* spp. DNA was detected in three camels (3%) ie, blood of two camels (camels Nos. 44, 73), liver and lymph node of one camel (camel No. 12) (Table 2). All of the four specimens were positive for *B. abortus*-specific primers targeting the *BCSP31* and *omp2* genes. Nucleotide sequences of *omp2* gene were identical for all three camels, so only one was deposited in GenBank® (accession number: MW182522). The ML tree that involved 12 nucleotide sequences supported isolated *B. abortus* in a distinct separation of species-specific branch (Figure 2).

Trypanosoma spp. DNA was detected in specimens from eight camels (8%) including six positive blood samples (camels Nos. 20, 32, 33, 43, 52, 94) and two positive liver samples (camels Nos. 46, 47), using Kin1/Kin2 primers (Table 2). Amplicons sized approximately 540 bp, suggestive for *T. brucei*-group parasites ie, *T. evansi*, *T. brucei*, and *T. equiperdum*. The sequences (GenBank® accession numbers: MW282046–MW282047, three accession numbers under NCBI processing) were > 99.7% identical to *T. evansi* isolates from camels in Iran (KX898420– KX898421), camels in Egypt (AB551919–AB551922), camels in Iraq (MH595480, MH571705), buffalo in China (FJ712715), deer (AY912279), cattle (DQ472705), and buffalo in Thailand (MN121259, DQ472705, AY912270). The ML tree that involved 19 nucleotide sequences supported isolated *T. evansi* in a distinct separation of species-specific branch (Figure 3).

DNA of *Bartonella* spp. and *Coxiella burnetii* were not detected in the examined specimens.

DISCUSSION

Camels are more resistant to a wide range of pathogens, but several studies have confirmed that they are susceptible to various pathogens including zoonotic viruses, bacteria and parasites posing a risk to public health [3,4,51-53]. In this study we detected the zoonotic bacte-

Table 2. Demographic data of infected camels in this study.

Camel ID	Age (months)	Sex	Area of origin	Breed	Detected pathogen	Infected organ	GenBank Accession number
12	24	female	Kaj-Abad	Kalkooyi	*Brucella abortus*	Liver, lymph node	MW182522
20	26	male	Kaj-Abad	Kalkooyi	*Trypanosoma evansi*	Blood	MW282046
32	15	male	Kaj-Abad	Kalkooyi	*Trypanosoma evansi*	Blood	–
33	24	male	Kaj-Abad	Kalkooyi	*Trypanosoma evansi*	Blood	MW282047
43	15	male	Qom-Rood	Kalkooyi	*Trypanosoma evansi*	Blood	–
44	24	male	Kaj-Abad	Kalkooyi	*Brucella abortus*	Blood	MW182521
46	12	male	Kaj-Abad	Kalkooyi	*Trypanosoma evansi*	Liver	–
47	27	male	Kaj-Abad	Kalkooyi	*Trypanosoma evansi*	Liver	MW519523
52	18	male	Seyd-Abad	Kalkooyi	*Trypanosoma evansi*	Blood	MW519524
73	15	male	Kaj-Abad	Kalkooyi	*Brucella abortus*	Blood	MW182520
94	21	female	Sey-Abad	Baloochi	*Trypanosoma evansi*	Blood	MW519525

rium *B. abortus* and zoonotic protozoan parasite *T. evansi* in the blood and tissue of camels in the central desert of Iran.

Three percent of the camels were infected with *B. abortus*. In the phylogenetic analysis, our isolates were clearly distinct from *B. melitensis*, *B. ovis*, and *B. suis*. Although, camel brucellosis is endemic in all camel-rearing countries with exception of Australia [9], there is no official policy for the control of camel brucellosis in any of the major camel-raising countries [54]. In Iran, several reports have been published since 1986 on the incidence of *Brucella* infection in camels in various regions using serological and molecular methods [36]. So far, *B. melitensis* biovar 1 and 3, and *Brucella abortus* biovar 5 have been isolated from milk, lymph nodes, and aborted fetuses of dromedaries [12-14]; surveys reported seroprevalence of up to 43.5% [55]. In a recent study, DNA of *B. melitensis* was detected in lymph nodes of two seronegative camels highlighting the complementary role of PCR on different organs for a better screening of *Brucella* infection in camels [56]. In general, the highest incidence of camel brucellosis is found when camels are kept together with infected ruminants [7]. Hence, higher prevalence of *B. abortus* or *B. melitensis* in camel herds in each region will depend on the mixed farming of camels with large ruminants (cattle, buffalo) or small ruminants (sheep, goat). Human brucellosis is a very important health issue in Iran, with an average incidence of 100 cases per 100,000 individuals, reaching up to 276/100,000 in some regions [57]. However, the role of camels in the spread of infection in the population has not been evaluated in the country. In Western Asian societies, it is generally believed that the boiling of milk spoils the taste and removes the goodness of milk [58]. In such social context, it is necessary that health policy makers implement prevention strategies such as livestock vaccination and to educate the public about the risks of drinking unpasteurized milk.

We found 8% of camels to be infected with *T. evansi*. Of note, livers of two camels tested positive while their blood tested negative, suggestive of the value of multiple organ testing in epidemiological studies. In the phylogenetic analysis our isolates grouped with other *T. evansi* camel isolates from Iran and Algeria, buffalo from India, Thailand, and China, and white-tailed deer from Thailand. In Iran, *Trypanosoma* prevalence between 0 and 20% depending on the region and season of sampling has been reported in dromedary camels [37]. For a decade, it was hypothesized that human susceptibility to *T. evansi* was linked to insufficient or missing levels of human trypanocide apolipoprotein L1 (APOL1), a trypanocidal component of normal human serum [59]. However, a report of infection in a Vietnamese patient with fever, headache, and arthralgia in 2016 with no previous immu-

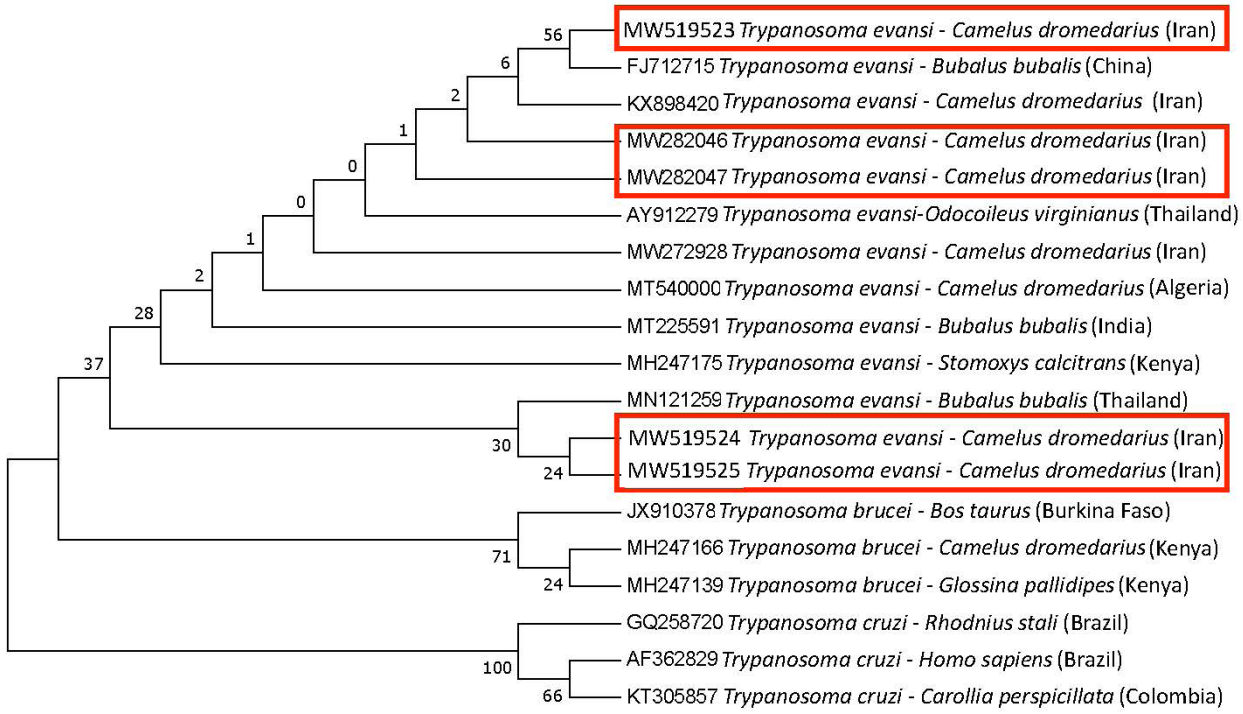

Figure 3. Phylogenetic relationship of *Brucella abortus* in this study (in boxes) to other *Trypanosoma* spp. based on a partial sequence of the *ITS1* gene. The analyses were performed using a Neighbor-Joining method [47] and evolutionary analyses were conducted on 1000 bootstrap replications [48] in the MEGA7 software [49].

nological risk, 2 wild-type APOL1 alleles and a normal serum APOL1 concentration did not support that hypothesis [32]. The patient possibly contracted the infection via a wound while butchering raw meat. This finding suggests that *T. evansi* is a true zoonosis with a risk of infection for the general population. Although "surra" is considered a debilitating disease, apparently healthy camels have also been diagnosed infected with *T. evansi* [30,60]. Hence, it is possible that farmers, slaughterhouse workers, veterinarians, and stay-at-home parents can get infected from asymptomatic camels and camel meat. While occupational risk might be inevitable, further research on the longevity of trypomastigotes in meat and other edible tissues of camels is suggested.

In the present study, DNA of *Bartonella* spp. was not detected in 100 camels from Qom province in the central desert of Iran. Similarly, in another study on 200 camels of two Iranian southern and south-eastern provinces (Kerman and Sistan-va-Baloochestan) no case of bartonellosis was detected [52]. However, 18 of 106 (17%) dromedaries in the Iranian province of Fars were infected with *B. dromedarii* [38]. Further research is warranted to better understand the epidemiology of *Bartonella* infection in camels in Iran and which vectors may be involved. In Israel, nine of 51 (17.6%) dromedaries were bacteremic with *Bartonella* spp. The bacterium was diagnosed as a novel species and was named *B. dromedarii* [27]. Following that report, DNA from *B. bovis* and *B. rochalimae* was detected in *Hyalomma dromedarii* ticks collected from a single camel in Palestine [28]. However, none of the 19 blood samples from camels were positive for *Bartonella* spp. [28]. Recently, *B. henselae* was identified in the blood of 15 (3.6%) of 412 dromedaries in Tunisia highlighting the importance of camels in the circulation of zoonotic bartonellosis [29]. Further research is needed to identify the vectors of *B. dromedarii* and if there is a zoonotic risk for humans raising or caring for these animals.

Similarly, *Coxiella* DNA was not detected in our study, which could be due to limited bacteremia after infection. Serological studies have reported high prevalence of 70, 80, and 90% in dromedaries in Egypt, Chad, and Ethiopia, respectively [21-23]. Comparative studies on different livestock species showed that antibodies against *C. burnetii* were more prevalent in dromedaries than in cattle, sheep, and goats [18]. However, so far no clinical signs have been attributed to *C. burnetii* in camels, possibly because it has not been systematically sought [24,61]. In a study by Wernery et al. [62], no *C. burnetii* organisms were found in fresh, unheated camel milk by PCR although all tested dromedaries from which the milk originated were serologically positive. However, high antibody titers were more frequent in female camels that had a history of abortion in a Tunisian study [63].

Absence of reports regarding clinical disease and abortion in camels in Iran associated with coxiellosis is partly because camels mainly live in areas with limited access to diagnosis and treatment services. In the first serologic study of different animal species in Iran in 1951, antibodies against *C. burnetii* were detected in the serum of 2 of 12 (16.7%) tested dromedaries [64]. In another study, *C. burnetii* DNA was detected in the blood of 2.4% of tested dromedaries while 28.7% of them were seropositive [65,66]. In general, urine, feces, milk, and mainly birth products of camels (*C. burnetii* has affinity to the udder, placenta, and other female reproductive organs) represent the most important source of human infection [17,18]. However, to our knowledge *C. burnetii*–induced abortion in dromedary camels has not been evaluated so far. In Chad, a serosurvey of pastoralists and their livestock found that camel breeders had a nine times higher risk of being *C. burnetii* seropositive compared to the general public [22]. Further research should be assessed to evaluate the role of camels in *C. burnetii* transmission to humans keeping in mind that infection with *C. burnetii* in humans is most often asymptomatic. Subtyping camel *C. burnetii* strains might elucidate their role in the infection risk to farmers, veterinarians, slaughterhouse workers and other people in contact with camels. Testing the efficacy of available livestock *C. burnetii* vaccines in camels to reduce infection and environmental contamination is highly recommended.

In conclusion, the herein reported data provide more knowledge of zoonotic diseases in camels of Iran. Of the detected pathogens, brucellosis is of great concern for its pathogenicity to humans. Effective control strategies such as vaccination of both camels and people in their close contact, test-and-slaughter policy, and education of the public is necessary for minimizing the risk of zoonotic infection. Finally, although *Brucella* spp. and *Trypanosoma* spp. produce diseases in camels, the epidemiological role of camels in human societies need further investigation. Also, it is completely unclear if and how *C. burnetii* is excreted and if people are really at risk.

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