

Molecular detection and phylogenetic analysis of *Babesia caballi* in domestic donkeys (*Equus asinus*) in Baghdad Zoo, Iraq

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Abstract

In Iraq, equine piroplasmiasis is considered a severe haem-protozoan infection of Equidae caused by the tick-borne protozoa *Babesia caballi*. The present study collected blood samples (n = 70) of domestic donkeys from the Baghdad Zoo in the Al Zawra'a Gardens area, Baghdad, Iraq, from April to September 2018. The molecular method to detect *B. caballi* was based on partial amplification and sequencing of the 18S rRNA gene. Additionally, the haematologic parameters of all blood samples were also studied. The results of PCR products (540 bp) of the 18S rRNA gene showed that 65 blood samples (92.9%) were positive. The phylogenetic analysis of five sequences of samples (NCBI accession numbers: MN723600.1, MN723592.1, MN723851.1, MN723855.1, and MN723849.1) compared with global and neighbouring countries isolates revealed five separate clades with homology 99% in four samples. The results of haematologic parameters showed no statistically significant changes in blood values in infected and uninfected donkeys. The study concludes that donkeys are carriers of piroplasmiasis and can be a reservoir and source of infection for other equids. This study is the first molecular report to confirm piroplasmiasis in domestic donkeys in Iraq.

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Introduction

Tick-borne disease was thought to be one of the most common blood-protozoan intra-erythrocytic parasites that affected a lot of different types of equids (horses, donkeys, mules, and zebras), caused by ticks transmitting two protozoa (*Theileria equi* and *Babesia caballi*) inside the erythrocytes (1,2). Ticks transmit both pathogens belonging to *Hyalomma*, *Rhipicephalus*, and *Dermacentor* species (3-5). The presence of transmitter vectors in tropical and subtropical areas makes piroplasmiasis endemic. The disease spreads worldwide, causing economic loss and affecting the movement of horses across borders (2,6,7). Equine piroplasmiasis has been studied in Iraq, and previous articles studied piroplasmiasis using molecular and serological methods in horses and serological detection in donkeys (8-

11). Molecular methods have been used as sensitive techniques for identifying and characterizing *B. caballi* in horses and donkeys (12-15). For example, Sunday Idoko *et al.* (12) in Nigeria used conventional PCR for detecting *rap-1* of *B. caballi* in horses and donkeys, and this study suggested that donkeys act as reservoirs of these parasites. Bashiruddin *et al.* developed a PCR technique that amplified targets of 659 bp regions of 16S rRNA genes to detect *B. caballi* (13). Abedi *et al.* (14) modified the multiplex PCR of 18S rRNA to detect *T. equi* and another *B. caballi* designed by Alhassan *et al.* (15). The donkeys with piroplasmiasis were asymptomatic. Still, they could act as carriers (14). Salinas-Estrella *et al.* used a duplex qPCR method and nested PCR test for amplification of the *rap1* gene of *B. caballi*; this study suggested the duplex qPCR was more sensitive than nested PCR (16). Mshelia *et al.* (17) used

nested PCR to screen the 18S rRNA of *B. caballi* and *T. equi*, and they used nested PCR to identify specific species of the *B. caballi rap1* gene. The comparison between nested and duplex real-time PCR (nPCR) and duplex PCR showed that nPCR can detect *B. caballi* with more sensitivity and specificity than duplex real-time PCR. In contrast, duplex real-time PCR can readily diagnose *B. caballi* infection in equines, but this test has low sensitivity (18).

The present study used the conventional polymerase chain reaction (PCR) method to detect *B. caballi* in the blood of 70 donkeys domesticated in the Baghdad Zoo of the Al Zawra'a Gardens area, Baghdad, Iraq, from April to September 2018. In addition, the 18S rRNA gene of 5 isolates was sequenced, and phylogenetic variations of *B. caballi* sequences were compared with other countries.

Materials and methods

Ethical approve

All animals and procedures in this research were approved by the Committee of University of Baghdad, committee number 16 in the 6 June/2022.

Blood collection and hematological analysis

Blood samples (n = 70; 59 females and 11 males) were collected from domestic donkeys from April to September 2018 in the Baghdad Zoo of the Al Zawra'a Gardens area, Baghdad, Iraq. These donkeys were prepared to be food for carnivores in the zoo. A total of 2.5 mL of blood samples were collected from the jugular vein using sterile tubes containing Ethylene Diamine Tetra Acetic Acid (EDTA) anticoagulant. The haematologic parameters of all blood samples were investigated, and from each sample, 25 µl blood was analyzed using blood haemolyze (HumaCount-30TS-Veterinary Mode, Germany). The blood samples were kept at -20°C for further molecular analysis in the Department of Internal and Preventive Veterinary Medicine at the College of Veterinary Medicine at the University of Baghdad. Blood samples.

Extraction of genomic DNA and PCR

The DNAs were extracted from all blood samples using the Blood gDNA Miniprep kit (Promega, USA) according to the manufacturer's instructions for amplification of the 18S rRNA gene of *B. caballi*, the forward primer UFP (5-TCG AAG ACG ATC AGA TAC CGT CG-3) and the reverse primer CabR (5-CTC GTT CAT GAT TTA GAA TTG C-3) (15) (Microgen Bioproducts, UK) were used. The PCR reaction was 25 µl consisting of 12.5 µl Green Master Mix (Promega, USA), 1 µl of each primer, 8.5 µl nucleus free water, and 2 µl genomic DNA.

The PCR reaction used Thermal Cycler (TC9610, Inc. USA) to amplify the 18S rRNA gene according to the following conditions: initial denaturation for 10 min. At 96 °C, followed by 36 cycles, including a first step of

denaturation at 96 °C for 60 s, an annealing step at 55.9 °C for 60 s, and an extension step at 72 °C for 60 s, while the final extension was done at 72 °C for 10 min. For each PCR, a negative control was added using deionized pathogen-free distilled water to the mixture instead of the DNA template. Afterward, the PCR products were loaded at 5 µl volume on a 1.2% agarose gel prepared in 1X TBE buffer, stained with Ethidium Bromide, and electrophoresed for 45 minutes at 90 volts, 110 Ampere. A ladder of 100 bp was used as a size marker. The DNA bands on the agarose gel were visualized by a UV transilluminator and photographed by a digital camera. The annealing temperature 55.9°C was optimized using a clinically positive sample with severe clinical signs of piroplasmosis (bloody urine, icterus, and intraerythrocytic parasites).

Sequence analysis

The PCR products of five samples were analysed and sequenced for the 18S rRNA gene according to the manufacturer's instructions (Macrogen, Korea). The nucleotide sequences were aligned with the NCBI data. The software Molecular Evolutionary Genetics Analysis version (MEGA 6.0) generated phylogenetic trees and the sequence analysis.

Statistical analysis

SPSS was used for the statistical analysis (version 20.0, USA). To calculate the risk factors of sex with *B. caballi*, the odd ratio was used. To compare means at the level of statistical probability at P<0.05, analysis of variance (ANOVA) was used in this study.

Results

The infection rates and hematological features

In the present study, the *B. caballi*-positive cases were 65 out of 70 donkeys, representing an infection rate of 92.8%. All 65 DNA samples were positive for *B. caballi* and showed a band of about 540 bp (Figure 1).

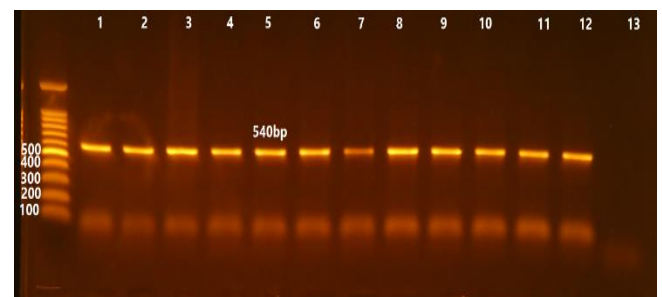


Figure 1: Partial fragments 540 bp of the 18S rRNA gene of *B. caballi* in donkeys. Lanes 1-12 represent positive cases, and lane 13 is the negative control.

The results revealed a non-significant increase in the risk factor (1.10 odds ratio) for males compared to females, where the infection rate between females was 91.5% compared to males at 100% (Table 1).

Table 1: The infection rates of *B. caballi* in donkeys

Infected/Total (%)	Genders [Infected/Total (%)]	
	Females (59)	Males (11)
65/70 (92.8%)	54/59(91.5%)	11/11 (100%) *

*Refer to non-significant difference at (P>0.05) (1.10 odds ratio).

The changes in blood values in animals infected with *B. caballi*, compared with uninfected animals, showed non-significant differences in mean±SE (Table 2) despite an apparent decrease in hemogram values, but it is insignificant. Infected donkeys showed severe emaciation, signs of a lack of nutrition, and pale mucous membranes because of poor management. *Babesia caballi* infection did not have a significant effect on blood values.

Table 2: The hematological values of donkeys infected with *B. caballi*

Tests	Non infected donkeys (10) mean±Std	Total infections (65) mean±Std
RBC (×10 ⁶ / μ L)	5.02±0.72 A	5.03±0.27 A
Hb (g/dl)	10.53±0.45 A	10.48±0.36 A
HCT (%)	30.77±1.36 A	30.15±1.08 A
MCV (fl)	58.68±1.26 A	59.69±0.63 A
MCH (pg)	21.36±0.48 A	20.84±0.20 A
MCHC (g/dl)	34.01±0.57 A	34.66±0.24 A
WBC (×10 ³ / μ L)	9.34±1.85 A	8.89±0.59 A
PLT (×10 ³ / μ L)	237±17.52 A	200±8.19 A

Differences in letters horizontally refer to significant differences at P<0.05.

Phylogenetic analysis of *B. caballi*

A phylogenetic tree of *B. caballi* 18S rRNA sequences of five DNA samples was analyzed and submitted in the NCBI (accession numbers: MN723600.1, MN723592.1, MN723851.1, MN723855.1, and MN723849.1) (Figure 2). The sequences were grouped into five individual clades: the first clade involved MN723600 and has 81% resemblance with global isolates from Sereba, Bosnia and Herzegovina, Tobago, Brazil, China, and other countries with high heterogeneity of this clade, while the second clade included MN723592 with a matching percentage of 99% with isolates from one of the neighboring countries of Iraq (Turkey, MN481269, unpublished but data of sequence available in NCBI website). The other three clades of *B. caballi* infecting donkeys in this study (MN723851.1, MN723855.1, and MN723849.1) were similar to each other at 98-99% and also

to the Indian isolate (India, MF384422.1, unpublished but data of sequence available in NCBI website).

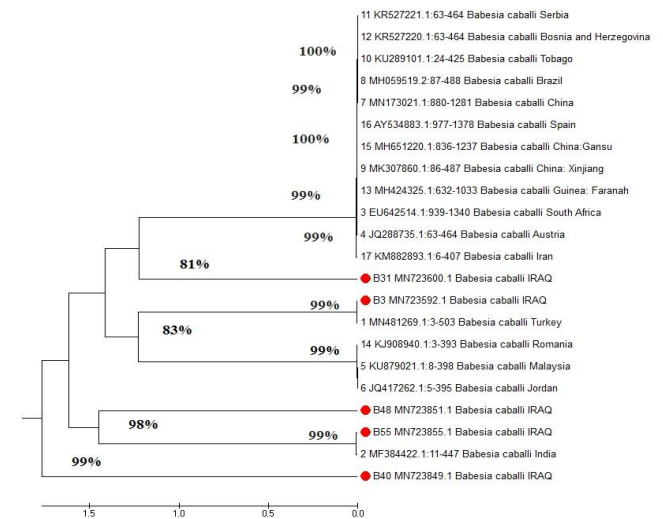


Figure 2: Molecular phylogenetic tree of partial 18S rRNA gene sequences of *Babesia caballi* infecting domestic donkeys in Baghdad Zoo and other sequences obtained from NCBI database. Phylogenetic analysis was carried out using the MEGA6 program illustrated by five Clades.

Discussion

In donkeys, a few studies on *B. caballi* are available. Most epidemiological studies show that *B. caballi* is less widespread than *Thaleria equi* (19). However, a high rate of *B. caballi* infection was seen in Egypt, and the infection rate in donkeys was 15.7% (20). In another recent study, an infection rate of *B. caballi* of 8.8% in Nigeria was recorded in donkeys, and no case was diagnosed in horses (12). Most of the DNA samples amplified by PCR were positive for *B. caballi* with bands between about 540 bp; the band size is similar to that obtained by the researchers who designed the primers (15), who mentioned that the expected positive result was about 540 bp where they used the multiplex method. Therefore, conventional optimization of the annealing temperature for diagnosing *B. caballi* is recommended due to its high sensitivity.

These donkeys live in crowded environments and are neglected in the Baghdad Zoo, where the donkeys gather from most areas of Iraq. This increases the possibility of infection among them. The donkeys were suffering from severe emaciation, signs of nutritional deficiency, and pallor of the mucous membranes due to poor management conditions and neglect. These donkeys were prepared to be food for carnivores in the zoo. For these reasons, the infection rate in the present study was higher than that in other research papers performed by other researchers in Iran (14,15). The Iranian researchers (14) noticed, when using the

same type of forward and reverse primers by multiplex PCR, that no infection with *B. caballi* was recorded in donkeys. In contrast, a high infection rate with *Theileria equi* was observed 50.9%. Alhassan *et al.* (15) were the first to use primers (UFP and CabR) to diagnose *B. caballi* with a 17.9% infection rate.

This study suggests that donkeys are more infected than other Equidae species due to the long hair of donkeys and the lack of grooming hair. In our opinion, the skin of the donkeys became good hideouts for the ticks that transmit *Babesia*. Since the donkeys received no anti-babesia treatments, these animals became reservoirs for *Babesia*. Using PCR assay, the most recent studies in Israel found that 9.7% of horses had been infected with *B. caballi*, and the prevalence of infection depended on breed, tick infestation, and geographical area (21). According to case history, the donkeys in Baghdad Zoo do not receive adequate veterinary care, unlike horses.

According to Abedi (14), both infected and uninfected donkeys had low hemoglobin levels; this means that donkeys do not get enough food and are neglected, making it hard to find control donkeys living in the same conditions for comparison. The blood tests for the group infected with *B. caballi* showed that their hemoglobin was 10.48 g/dl, and their HCT was 30.15%. These results are similar to donkeys living in similar conditions in Egypt (20), where the hemoglobin levels were 10.37 g/dl and the HCT was 29%. They recorded a significant decrease in these values compared to the control group. This reinforces the result that the animals in Iraq generally suffer from anemia. In Iraq, one study estimated the normal hematological values of donkeys in Iraq, where these values ranged from PCV 32.4-48.7%, Hb 9.6-14.3 g/dl and RBC $4.69-7.6 \times 10^6/\mu\text{L}$ (22). This study indicated that the hematological values in donkeys were low under normal conditions.

Davitkov *et al.* (23) found a very low incidence of *B. caballi* infection 2.1%. While the annealing temperature of the Sant *et al.* (24) isolate Tobago, KU289101.1 was reduced to 51.3, this study also revealed a low infection rate of *B. caballi* in horses of 3.6%. The common infection rates in these studies, when compared to the infection rate reported in this study, might be because of a difference in the geographical distribution among countries, animal species, or differences in the annealing temperature of PCR.

The present study first investigated the prevalence of *B. caballi* in donkeys in Baghdad/Iraq by molecular approach. Al-Mola *et al.* (25) were clinical and pathological studies of equine babesiosis in Mosul/ Iraq in draught horses. Alsaad *et al.* (10) was seroprevalence reported of *B. caballi* in Mosul, Iraq, in both horses and donkeys. Alsaad *et al.* (26) detected equine babesiosis in horses in Basrah/ Iraq by hematological and clinical assay. Another study was designed to detect equine piroplasmiasis by molecular technique in the tick activities season (27). The importance of molecular investigation of protozoa ticks gives additional information

about the life cycle of the protozoa (28). The hematological studies and investigation of protozoa were conducted on donkeys in Iraq, but this species required further investigations on internal and blood parasites (22,29,30).

From what was presented in the discussion, it is necessary to determine the optimal conditions for PCR to use the primers designed by Al-Hassan (15) and delete the variable nucleotide base (T) that is at the end of the reverse primer (cabR) to detect *B. caballi* and conventional PCR should be used instead of multiplex PCR. All of these procedures give more accurate results and make the PCR more sensitive as well.

In the phylogenetic tree of *B. caballi*, the heterogeneity of the sequence of one isolate was 19% compared to other countries, which may be due to inadequate sequencing techniques. In contrast, the other four isolates had a high identity of 99%, especially with neighbouring countries. For this reason, we sent five isolates to be sequenced. Some *B. caballi* isolates had a high matching rate of 98-99% with other isolates from nearby countries, such as the Turkish isolate in the horse MN481269.1 and the Indian isolate in the horse MF384422.1, both of which have not been published but have information available in NCBI. The sequences of isolates in the results matched each other at a high rate, except for two isolates (MN723600.1 and MN723592.1). There may have been more Turkish horses coming into and out of Iraq, or there may have been more free movement of animals between Iraq and Turkey.

Conclusion

The study concluded that donkeys are carriers of *B. caballi* and act as reservoirs. These donkeys may be a major source of infection for other equids. This study is the first to use molecular diagnosis to confirm the presence of *B. caballi* in donkeys in Iraq.

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Conflict of interest

The authors declare that there is no conflict of interest.

References

1. Qablan MA, Obornik M, Petrželková KJ, Sloboda M, Shudiefat MF, Hořín P, Modrý D. Infections by *Babesia caballi* and *Theileria equi* in Jordanian equids: Epidemiology and genetic diversity. *Parasitol.* 2013;140(9):1096-1103. DOI: [10.1017/S0031182013000486](https://doi.org/10.1017/S0031182013000486)
2. Lei R, Wang X, Zhang D, Liu Y, Chen Q, Jiang N. Rapid isothermal duplex real-time recombinase polymerase amplification (RPA) assay

- for the diagnosis of equine piroplasmosis. Sci Rep. 2020;10(1):1-11. DOI: [10.1038/s41598-020-60997-1](https://doi.org/10.1038/s41598-020-60997-1)
3. Camino E, Dorrego A, Carvajal KA, Buendia-Andres A, de Juan L, Dominguez L, Cruz-Lopez F. Serological, molecular and hematological diagnosis in horses with clinical suspicion of equine piroplasmosis: Pooling strengths. Vet Parasitol. 2019;275:108928. DOI: [10.1016/j.vetpar.2019.108928](https://doi.org/10.1016/j.vetpar.2019.108928)
 4. Scoles GA, Ueti MW. Vector ecology of equine piroplasmosis. Annu Rev Entomol. 2015;60:561-580. DOI: [10.1146/annurev-ento-010814-021110](https://doi.org/10.1146/annurev-ento-010814-021110)
 5. Sumbria D, Moudgil AD, Singla LD. Equine Piroplasmosis: Current status. Vet. 2014;1(1):9-14. [\[available at\]](#)
 6. Hansford KM, Gillingham EL, Cull B, McGinley L, Medlock JM, Phipps LP, Peaty M. Disease and ticks on horses. Vet Rec. 2019;184(19):592. DOI: [10.1136/vr.12056](https://doi.org/10.1136/vr.12056)
 7. Salim B, Bakheit MA, Kamau J, Sugimoto C. Current status of equine piroplasmosis in the Sudan. Infect Genet Evol. 2013;16:191-199. DOI: [10.1016/j.meegid.2013.02.008](https://doi.org/10.1016/j.meegid.2013.02.008)
 8. Faraj AA, Hade BF, Al-Amery AM. Conventional and molecular study of *Babesia spp.* of natural infection in dragging horses at some areas of Baghdad city, Iraq. Iraqi J Agric Sci. 2019;50(3):909-915. DOI: [10.36103/ijas.v50i3.707](https://doi.org/10.36103/ijas.v50i3.707)
 9. Khalid JA, Al-Barwary LO. Epidemiological study of equine piroplasmosis (*Theileria equi* and *Babesia caballi*) by microscopic examination and competitive-ELISA in Erbil province north-Iraq. Iran J Parasitol. 2019;14(3):404-412. [\[available at\]](#)
 10. Alsaad KM, Hassan SD, Al-Obaidi QT. Detection of *Babesia equi* and *Babesia caballi* antibodies in horses and donkeys in Mosul, Iraq. Res Opin Anim Vet Sci. 2012;2(4):291-294. [\[available at\]](#)
 11. Alssad KM. Acute babesiosis in foals. J Anim Vet Adv. 2009;8(12):2585-2589. [\[available at\]](#)
 12. Sunday II, Tirosh-Levy S, Leszkowicz MM, Mohammed AB, Sikiti GB, Wesley ND, Steinman A. Genetic characterization of piroplasms in donkeys and horses from Nigeria. Anim. 2020;10(2):324. DOI: [10.3390/ani10020324](https://doi.org/10.3390/ani10020324)
 13. Bashiruddin JB, Camma C, Rebelo E. Molecular detection of *Babesia equi* and *Babesia caballi* in horse blood by PCR amplification of part of the 16S rRNA gene. Vet Parasitol. 1999;84(1-2):75-83. DOI: [10.1016/S0304-4017\(99\)00049-7](https://doi.org/10.1016/S0304-4017(99)00049-7)
 14. Abedi V, Razmi G, Seifi H, Naghibi A. Molecular detection of equine piroplasms in donkeys (*Equus asinus*) in north Khorasan province, Iran. Iran J Vet Res. 2015;16(2):202-204. [\[available at\]](#)
 15. Alhassan A, Pumidonming W, Okamura M, Hirata H, Battsetseg B, Fujisaki K, Igarashi I. Development of a single-round and multiplex PCR method for the simultaneous detection of *Babesia caballi* and *Babesia equi* in horse blood. Vet Parasitol. 2005;129(1-2):43-49. DOI: [10.1016/j.vetpar.2004.12.018](https://doi.org/10.1016/j.vetpar.2004.12.018)
 16. Salinas-Estrella E, Ueti MW, Lobanov VA, Castillo-Payró E, Lizcano-Mata A, Badilla C, Martínez-Ibáñez F, Mosqueda J. Serological and molecular detection of *Babesia caballi* and *Theileria equi* in Mexico: A prospective study. PLoS One. 2022;17(3):e0264998. DOI: [10.1371/journal.pone.0264998](https://doi.org/10.1371/journal.pone.0264998)
 17. Mshelia PW, Kappmeyer L, Johnson WC, Kudi CA, Oluyinka OO, Balogun EO, Richard EE, Onoja E, Sears KP, Ueti MW. Molecular detection of *Theileria* species and *Babesia caballi* from horses in Nigeria. Parasitol Res. 2020;119(9):2955-63. DOI: [10.1007/s00436-020-06797-y](https://doi.org/10.1007/s00436-020-06797-y)
 18. Lv K, Zhang Y, Yang Y, Liu Z, Deng L. Development of nested PCR and duplex real-time fluorescence quantitative PCR assay for the simultaneous detection of *Theileria equi* and *Babesia caballi*. Front Vet Sci. 2022;9: 873190. DOI: [10.3389/fvets.2022.873190](https://doi.org/10.3389/fvets.2022.873190)
 19. Cortés MM, Fernández-García JL, Martínez-Estélez MH. Seroprevalence of *Theileria equi* and *Babesia caballi* in horses in Spain. Parasit. 2017;24:14. DOI: [10.1051/parasite/2017015](https://doi.org/10.1051/parasite/2017015)
 20. Mahmoud MS, El-Ezz NA, Abdel-Shafy S, Nassar SA, El Namaky AH, Khalil WB, Suarez CE. Assessment of *Theileria equi* and *Babesia caballi* infections in equine populations in Egypt by molecular, serological and hematological approaches. Parasit Vectors. 2016;9(1):1-10. DOI: [10.1186/s13071-016-1539-9](https://doi.org/10.1186/s13071-016-1539-9)
 21. Tirosh-Levy S, Mazuz ML, Savitsky I, Pinkas D, Gottlieb Y, Steinman A. Serological and molecular prevalence of *Babesia caballi* in apparently healthy horses in Israel. Pathogens. 2021;10(4):445. DOI: [10.3390/pathogens10040445](https://doi.org/10.3390/pathogens10040445)
 22. Al-Hadithy HA, Al-Salmayy AM, Badawi NM. Evaluation study of the normal hematological parameters of donkeys in Baghdad city/Iraq. Al-Anbar J Vet Sci. 2016;9(2):19-23. [\[available at\]](#)
 23. Davitkov D, Vucicevic M, Stevanovic J, Krstic V, Slijepcevic D, Glavinic U, Stanimirovic Z. Molecular detection and prevalence of *Theileria equi* and *Babesia caballi* in horses of central Balkan. Acta Parasitol. 2016;61(2):337-342. DOI: [10.1515/ap-2016-0044](https://doi.org/10.1515/ap-2016-0044)
 24. Sant C, Allicock OM, d'Abadie R, Charles RA, Georges K. Phylogenetic analysis of *Theileria equi* and *Babesia caballi* sequences from thoroughbred mares and foals in Trinidad. Parasitol Res. 2019;118(4):1171-1177. DOI: [10.1007/s00436-019-06240-x](https://doi.org/10.1007/s00436-019-06240-x)
 25. AL-Mola G, Al-Saad MK. Clinical and pathological study of equine babesiosis in draught horses in Mosul. Iraqi J Vet Sci. 2006;20(1):89-101. DOI: [10.33899/ijvs.2006.45787](https://doi.org/10.33899/ijvs.2006.45787)
 26. Alsaad KM, Alsaad EA, Al-Derawie HA. Clinical and diagnostic study of equine babesiosis in draught horses in some areas of Basrah province. Res J Anim Sci. 2010;4(1):16-22. DOI: [10.3923/rjnasci.2010.16.22](https://doi.org/10.3923/rjnasci.2010.16.22)
 27. Al-Rammahi HM, Hatem AA, Al-Atabi AC. Molecular detection and occurrence of equine theileriosis in Arabian horses in Al-Najaf province/Iraq. Braz J Vet Res Anim Sci. 2020;57(3):e166996-e166996. DOI: [10.11606/issn.1678-4456.bjvras.2020.166996](https://doi.org/10.11606/issn.1678-4456.bjvras.2020.166996)
 28. Hajeel MS, Abd Alfatlawi MA. Sequencing-based phylogenetic-study of *Babesia spp* detected in tick tissues in Al-Diwaniyah province, Iraq. Iraqi J Vet Sci. 2019;33(1):9-12. DOI: [10.33899/ijvs.2019.125512.1034](https://doi.org/10.33899/ijvs.2019.125512.1034)
 29. Esmael SA. Clinical and hematological study of the internal parasites in native donkeys in Mosul city. Iraqi J Vet Sci. 2010;24(2):129-135. DOI: [10.33899/ijvs.2010.5591](https://doi.org/10.33899/ijvs.2010.5591)
 30. Hussain J. Prevalence of *Toxoplasma gondii* in native donkeys in Mosul. Iraqi J Vet Sci. 2011;25(2):111-115. DOI: [10.33899/ijvs.2011.5654](https://doi.org/10.33899/ijvs.2011.5654)

الكشف الجزيئي وتحليل تطور السلالات للبابيزيا كابالي في الحمير في حديقة الحيوانات في بغداد، العراق

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³ كلية التربية، جامعة تيشك الدولية، أربيل، العراق

الخلاصة

يعد داء الكثرات في العراق واحد من الأمراض شديدة العدوى وخصوصا في الفصيلة الخيلية والتي تسببها نوع من الأولي الدموية التي تسمى بابيزيا كابالي في هذه الدراسة تم جمع ٧٠ عينة من الحمير في حديقة حيوانات الزوراء في بغداد للفترة من نيسان إلى أيلول من العام ٢٠١٨. أجريت الفحوصات الجزيئية على هذه العينات في مختبر الفحوصات الجزيئية في كلية الطب البيطري جامعة بغداد وذلك لتضخيم جزء من الجين الخاص بالمسبب المسمى ثماني عشر أس للحمض النووي الرايبوزي وكذلك تم عمل تعاقب القواعد النيوكليوتيدية لخمس عينات من ناتج تفاعل البلمرة المتسلسل. فضلا عن الفحوصات الجزيئية تم عمل بعض المقاييس الدمية. أظهرت نتائج التحليل الكهربائي

في شجرة تطور السلالات مع باقي العزلات في العالم. عند دراسة القيم الدمية لم تظهر القيم الدمية أي فروقات معنوية بين الحيوانات المصابة والحيوانات غير المصابة. خلصت الدراسة أن هذه الحمير كانت حاملة للمرض ويمكن أن تكون كخازن للكثيريات ومصدر للإصابة لباقي الفصيلة الخيلية وكانت هذه الدراسة هي أول دراسة جزيئية عن داء الكثيريات في الحمير في العراق.

للعينات الموجبة في تفاعل البلمرة المتسلسل حجم قطعة تقدر بحوالي ٥٤٠ قاعدة على الهلام لجين ثماني عشر أس للحمض النووي الرايبوزي الخاص بالمسبب باييزيا كابلي وكانت عدد العينات الموجبة ٦٥ عينة بنسبة إصابة ٩٢,٩%. أظهر التحليل لتطور السلالات لخمسة عينات موجبة والتي تم تسجيل التعاقب لها في المركز الوطني لمعلومات التقانة الحيوية وتم تصنيف هذه العزلات إلى خمس تفرعات و مقارنتها