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Rickettsia conorii Subspecies *israelensis* in Captive Baboons

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Hamadryas baboons (*Papio hamadryas*) may transmit zoonotic vector-borne pathogens to visitors and workers frequenting zoological parks. We molecularly screened 33 baboons for vector-borne pathogens. Three (9.1%) of 33 animals tested positive for *Rickettsia conorii* subspecies *israelensis*. Clinicians should be aware of potential health risks from spatial overlapping between baboons and humans.

Papio hamadryas baboons (order Primates, family Cercopithecidae) are frequently hosted in zoological gardens worldwide. The natural susceptibility of baboons to many zoonotic agents (*1*) may present a potential risk for transmission of emerging infectious diseases to humans. Nevertheless, few data are available on vector-borne pathogens of human concern that are hosted by baboons (e.g., *Rickettsia africae*, *Babesia microti*–like parasites, and *Anaplasma phagocytophilum*) (*1*). Data are likewise scarce on the role of *P. hamadryas* baboons in circulating arthropod vectors in zoological gardens and the resulting risk for transmitting vector-borne pathogens to persons frequenting such areas. We aimed to determine the occurrence of zoonotic vector-borne pathogens in a zoopark in the Apulia region of southern Italy and assess baboons' potential roles as reservoirs of emerging pathogens. Our study was approved by the University of Bari Aldo Moro ethics committee (Prot. Uniba 176/19).

During February–December 2020, we anesthetized baboons in the zoopark and housed them in cages for blood sampling. For each baboon, we recorded age, sex, weight, and body condition score (1–5); we obtained peripheral blood samples by cephalic vein puncture. To determine complete blood count and for molecular analysis, we collected 2 mL blood samples in Vacutainer K3-EDTA tubes. For biochemical analysis, we collected an additional 5 mL blood in Vacutainer clot activator serum tubes and centrifuged (15 min at 1,500 × *g* at room temperature), then delivered it to the University of Bari Department of Veterinary Medicine (Bari, Italy). We extracted DNA using QIAGEN QIAamp DNA Blood and Tissue kits (https://www.qiagen.com) and molecularly tested for vector-borne pathogens (Table) (*2*–*4*). We purified and sequenced amplicons in both directions using a Big Dye Terminator v3.1 Cycle Sequencing Kit in an Applied Biosystems 3130 Genetic Analyzer (ThermoFisher, https://www.thermofisher.com), then edited and analyzed them using Geneious version 9.0 (https://www.geneious.com). We then compared resulting sequences with those in GenBank. We performed complete blood counts using CELL-DYN 3700 Hematology Analyzer (Abbott, https://www.abbott.com), biochemical profile using a KPM Analytics SAT 450 random access analyzer (https://www.kpmanalytics.com), and protein electrophoresis analyses using Sebia Hydrasys 2 Scan Focusing (https://www.sebia.com). We calculated 95% CIs for proportions and χ^2 and odds ratios (OR) to assess differences in prevalence and infection risk stratified by age and sex. We used *t*-tests to compare mean laboratory values between baboons positive and negative for vector-borne pathogens. We considered p values <0.05 statistically significant.

We included 33 baboons: 21 male, 12 female; 13 juvenile, 16 adult, and 4 elderly. Blood samples from 3/33 (9.1%, 95% CI 3.1%–23.4%; 1 adult male, 1 adult female, 1 juvenile male) were positive for *R. conorii* subsp. *israelensis* by the *glt*A gene; all samples were negative by *omp*A and *omp*B genes. The only sequence type we identified showed 99%–100% nucleotide identity with *R. conorii* subsp. *israelensis* from GenBank; we deposited our sequence in GenBank (accession no. OQ360110). All baboons tested negative for other vector-borne pathogens.

Although we found adult and male baboons at higher risk for infection (OR 2.6), we found no significant difference by age or sex (p = 0.439). No baboon showed ectoparasitic infestation or clinical signs of vector-borne diseases, and all displayed good physical status (mean complete blood count 3, average bodyweight 17.5 kg). Hematologic and serum chemistry values were within normal ranges (Appendix Tables 1, 2, https://wwwnc.cdc.gov/EID/article/29/4/22-1176-App1.pdf) for both *R. conorii*–negative and –positive baboons (p >0.05).

Our study revealed a nonnegligible prevalence (9.1%, 3/33) of *R. conorii* subsp. *israelensis* in *P. hamadryas* baboons, representing a pathogen–host association previously demonstrated only among asymptomatic dogs and cats from Portugal (*5*) and in severe cases among symptomatic humans from Italy (*6*). This survey confirms circulation of rickettsiae among baboons, also reported in 1 study of *R. africae* in *P. cynocephalus* yellow baboons from Zambia (*1*).

Despite routine treatment of baboons (orally administering 0.4 mg/kg ivermectin every 15 days by ground bait), presence of ticks in the zoopark was supported by a previous finding of tickborne pathogens (*A. phagocytophilum*, *Coxiella burnetii*, and *Rickettsia* spp.) in a lion (*7*). Given the baboon grooming behavior of removing ectoparasites from their bodies, lack of *Rhipicephalus sanguineus* sensu lato ticks, a vector of rickettsiae (*8*), was not surprising (*9*). However, association between zoopark-dwelling baboons and *Rhipicephalus* spp. ticks, including *R. sanguineus* s.l., is well known (*9*). Because this tick species is prevalent in the study area in all developmental stages, exposure very likely occurs (*10*).

Taken together, the high density of *P. hamadryas* baboons, their close proximity to the zoopark, and the anthropophilic behavior of *R. sanguineus* s.l. ticks (*10*) highlight the threat to park visitors and workers from *R. conorii* subsp. *israelensis* infection. Absence of clinical signs in positive baboons and lack of

Table. PCR protocols used in study of vector-borne pathogens among baboons, Italy, 2020

Pathogen	Target gene	Primer	Sequence, 5′→3′	Fragment length, bp	Reference
Babesia/*Theileria* spp.	18S rRNA	RLB-F	GAGGTAGTGACAAGAAATAACAATA	460–520	(*2*)
		RLB-R	TCTTCGATCCCCTAACTTTC		
Ehrlichia/*Anaplasma* spp.	16S rRNA	EHR-16SD	GGTACCYACAGAAGAAGTCC	345	(*2*)
		HER-16SR	TAGCACTCATCGTTTACAGC		
Rickettsia spp.	*glt*A	CS-78F	GCAAGTATCGGTGAGGATGTAAT	401	(*2*)
		CS-323R	GCTTCCTTAAAATTCAATAAATCAGGAT		
Spotted fever group Rickettsiae	*omp*A	Rr190.70F	ATGGCGAATATTTCTCCAAAA	632	(*2*)
		Rr190.701R	GTTCCGTTAATGGCAGCATCT		
	*omp*B	120–2788	AAACAATAATCAAGGTACTGT	600	(*3*)
		120–3599	TACTTCCGGTTACAGCAAAGT		
Leishmania infantum	kDNA minicircle	Leish-1	AACTTTTCTGGTCCTCCG GGTAG	120	(*4*)
		Leish-2	ACCCCCAGTTTCCCGCC		

differences in hematological and biochemical parameters between negative and positive animals indicate the asymptomatic features of infection and make clarifying the baboons' role as a potential reservoir more urgent. Measures to control tick circulation should be established to reduce risk for transmission of *R. conorii* subsp. *israelensis* to zoopark visitors and workers.

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Prevention of *Thelazia callipaeda* Reinfection among Humans

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Thelazia callipaeda is a zoonotic vector-borne nematode that infects and causes eye disease among a wide range of domestic and wild mammals, including humans. We describe an unusual case of reinfection by this nematode in Serbia and call for a focus on preventive measures in endemic areas.

The genus *Thelazia* (order Spirurida, family Thelaziidae) comprises several species of nematode that cause ocular infections in different host mammals, including humans (*1*). Over the past 20 years, the *T. callipaeda* eyeworm has gained interest among

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Appendix

Appendix Table 1. Complete blood count and biochemical analyses results from baboons testing negative (n = 30) and positive (n = 3) *to Rickettsia conorii* subspecies *israelensis* DNA, during 2020, in Italy*

Parameter	Units	Mean BabR– †	Mean BabR+ ‡	Standard deviation	Standard error	*t*-test value	*p*-value	Range
Complete blood count	g/dL	12.7	12.2	1.7	1.0	0.5	0.632	12.6 ± 0.9
Hgb	K/µL	442.8	438.7	67.9	41.1	0.1	0.921	316 ± 83
Plt	Femtoliters	7.3	7.9	0.7	0.4	1.4	0.167	8.3 ± 1.0
MPV	K/µL	11.7	9.4	3.3	2.0	1.1	0.314	9.6 ± 2.9
WBC	M/µL	5.2	4.8	0.46	0.3	1.5	0.133	4.95 ± 0.32
RBC	%	40.7	36.6	3.6	2.2	1.8	0.069	38.2 ± 2.5
Hct	Femtoliters	77.6	78.6	2.6	1.6	0.6	0.530	77 ± 2.9
MCV	pg/dL	21.7	22.3	0.9	0.5	1.1	0.279	25.3 ± 0.9
MCH	g/dL	32.5	33.5	1.8	1.1	0.9	0.366	32.9 ± 0.7
MCHC	g/dL	31.7	32.2	1.9	1.1	0.5	0.650	NA
CHCM	pg/dL	3.7	3.4	0.4	0.2	1.2	0.224	NA
CHDW	%	14.0	14.5	0.7	0.4	1.2	0.247	NA
RDW	%	2.1	1.9	0.2	0.1	1.6	0.108	NA
HDW	K/µL	9.1	7.8	3.2	1.9	0.7	0.501	3.3 ± 1.9
Neu	K/µL	1.8	1.2	0.6	0.4	1.6	0.108	2.1 ± 1.3
Lym	K/µL	0.6	0.4	0.2	0.1	1.6	0.108	2.0 ± 2.0
Mon	K/µL	0.03	0.02	0.1	0.06	0.2	0.858	1.0 ± 1.0
Eos	K/µL	0.025	0.03	0.02	0.01	0.2	0.869	0.05 ± 0.05
Bas	%	0.3	0.3	0.07	0.04	0	1.000	NA
‡Biochemical analyses	%	41.2	40.8	3.2	1.9	0.2	0.838	NA
Pct	g/dL	23.1	20.9	3.3	1.9	1.1	0.279	NA
PDW	IU/L	564.0	477.0	622.6	377.0	0.2	0.819	NA
MPC	IU/L	49.6	31.7	16.3	9.8	1.8	0.079	NA
CPK	IU/L	33.4	18.7	32.6	19.7	0.7	0.462	NA
AST	IU/L	642.2	886.0	489.4	296.3	0.8	0.417	NA
ALT	IU/L	22.7	32.7	12.6	7.6	1.3	0.200	NA
ALP	IU/L	5,225.5	5,456.8	1,829.4	1,107.7	0.2	0.836	NA
GGT	mg/dL	0.4	0.3	0.1	0.06	1.6	0.111	NA
Cholinesterase	mEq/L	145.5	142.8	3.5	2.1	1.3	0.212	NA
Total bilirubin	mEq/L	4.2	4.5	0.7	0.4	0.7	0.485	NA
Natrium	mEq/L	22.0	18.7	5.8	3.5	0.9	0.355	NA
Potassium	mEq/L	111.0	107.4	3.5	2.1	1.7	0.099	NA
Natrium/potassium ratio	mmol/L	10.6	13.2	3.7	2.2	1.1	0.255	NA
Chlorine	mg/dL	120.3	105.6	30.5	18.5	0.8	0.432	NA
Anion gap	mg/dL	1.0	0.9	0.3	0.1	0.5	0.587	NA
Glucose	mg/dL	28.1	32.1	10.5	6.3	0.6	0.534	NA
Creatinine	mg/dL	9.0	9.5	0.7	0.4	1.2	0.247	NA
Urea	mg/dL	4.3	3.5	1.5	0.9	0.8	0.385	NA
Calcium	g/dL	6.6	6.8	0.5	0.3	0.7	0.514	NA
Phosphorus	mg/dL	0.18	0.2	0.05	0.03	0.7	0.510	NA
Total proteins	mg/dL	91.7	100.7	18.9	11.4	0.8	0.438	NA
Albumin	mg/dL	50.9	53.4	21.4	12.9	0.1	0.848	NA
Cholesterol	µg/dL	34.7	31.8	5.8	3.5	0.8	0.415	NA
Triglycerides	mmol/L	27.2	25.3	3.2	1.9	0.9	0.334	NA

*BabR–*R. conorii* subsp. *israelensis* DNA–negative baboons, BabR+ *R. conorii* subsp. *israelensis* DNA–positive baboons, HGB (hemoglobin), PLT (platelet), MPV (mean platelet volume), WBC (white blood cell), RBC (red blood cell), Hct (hematocrit), MCV (mean corpuscular volume), MCH (mean corpuscular hemoglobin), MCHC (mean corpuscular hemoglobin concentration), CHCM (cellular hemoglobin concentration mean), CHDW (cell hemoglobin distribution width), RDW (red cell distribution width), HDW (hemoglobin distribution width), Neu (neutrophils), Lym (lymphocytes), Mon (monocytes) Eos (eosinophils), Bas (basophils), Pct (procalcitonin), PDW (platelet distribution width), MPC (mean platelet component), CPK (creatine phosphokinase), AST (aspartate aminotransferase), ALT (alanine aminotransferase), ALP (alkaline phosphatase), GGT (gamma glutamyl transpeptidase).
†BabR–, Baboons tested negative to *Rickettsia conorii* subspecies *israelensis* DNA.
‡BabR+, Baboons tested positive to *Rickettsia conorii* subspecies *israelensis* DNA.

Appendix Table 2. Serum protein electrophoresis in baboons tested negative (n = 33) and positive to *Rickettsia conorii* subspecies *israelensis* DNA, during 2020, in Italy. The protein concentration values are expressed in g/dl

Parameter	Mean value *BabR⁻	Mean value †BabR⁺	Standard deviation	Standard error	*t*-test value	*p*-value
Albumin	3.9	4.2	0.6	0.4	0.8	0.415
α-1 globulins	0.2	0.14	0.06	0.04	1.7	0.106
α-2 globulins	0.7	0.5	0.2	0.1	1.6	0.108
β-1 globulins	0.5	0.4	0.1	0.06	1.6	0.111
β-2 globulins	0.5	0.48	0.1	0.06	0.3	0.745
γ-globulins	0.9	0.8	0.3	0.06	1.6	0.111
Total proteins	6.6	6.5	0.5	0.3	0.3	0.744
Albumin/globulins ratio	1.5	1.9	0.5	0.3	1.3	0.196

*BabR– *R. conorii* subsp. *israelensis* DNA–negative baboons, BabR+ *R. conorii* subsp. *israelensis* DNA–positive baboons