

## Supplementary material

**Table S1**

Details of target genetic markers used for testing microorganisms in ticks DNA

Target microorganism (Genus/Phylum)	Target microorganism (species)	Genetic marker (s)
<i>Anaplasma</i>	<i>Anaplasma</i> (A.) spp.	16S
	<i>A. marginale</i>	<i>msp1</i>
	<i>A. platys</i>	<i>groEL</i>
	<i>A. phagocytophilum</i>	<i>msp2</i>
	<i>A. ovis</i>	<i>msp4</i>
	<i>A. centrale</i> , <i>A. bovis</i>	<i>groEL</i>
Apicomplexa	Apicomplexa spp.	18S
<i>Aegyptianella</i>	<i>Aegyptianella pullorum</i>	<i>groEL</i>
<i>Babesia</i>	<i>Babesia</i> (B.) <i>microti</i> , <i>B. bovis</i>	CCTeta
	<i>B. ovis</i> , <i>B. bigemina</i> , <i>Babesia</i> sp. EU1, <i>B. venatorum</i>	18S
	<i>B. divergens</i> , <i>B. canis</i>	<i>hsp70</i>
	<i>B. caballi</i> , <i>B. gibsoni</i>	<i>rap1</i>
	<i>B. duncani</i>	ITS2
<i>Bartonella</i>	<i>Bartonella</i> (Ba.) spp.	<i>ssrA</i>
	<i>Ba. henselae</i>	<i>pap31</i>
	<i>Ba. barcilliformis</i>	<i>rpoB</i>
	<i>Ba. vinsonii</i> subsp. <i>berkhoffii</i>	ITS
<i>Borrelia</i>	<i>Borrelia</i> (Bo.) spp.	23S
	<i>Bo. miyamotoi</i> , <i>Bo. lonestari</i> , <i>Bo. theileri</i>	<i>glpQ</i>
	<i>Bo. anserina</i> , <i>Bo. afzelii</i> , <i>Bo. spielmanii</i>	<i>fla</i>
	<i>Bo. burgdorferi</i> s.s., <i>Bo. garinii</i> , <i>Bo. lusitaniae</i> , <i>Bo. bissettii</i>	<i>rpoB</i>
	<i>Bo. parkeri</i>	<i>gyrB</i>
	<i>Bo. turicatae</i>	ITS
	<i>Bo. valaisiana</i>	<i>ospA</i>
<i>Mycoplasma</i>	<i>Mycoplasma</i> (M) spp., <i>Candidatus Mycoplasma</i> (C.M.) <i>haemominutum</i> , <i>C.M. haematoparvum</i>	16S
	<i>M. wenyonii</i> , <i>M. haemocanis</i>	23S
	<i>M. haemofelis</i>	<i>dnaK</i>
	<i>M. ovis</i> , <i>M. suis</i> , <i>C.M. turicensis</i>	<i>rnpB</i>
<i>Coxiella</i>	<i>Coxiella</i> (C.) <i>burnetii</i>	IS1111, <i>icd</i>
<i>Cytauxzoon</i>	<i>Cytauxzoon felis</i>	ITS2

<i>Ehrlichia</i>	<i>Ehrlichia</i> (E.) spp.	16S
	<i>E. ruminatum</i> , Panola Mountain <i>Ehrlichia</i> , <i>E. canis</i>	<i>gltA</i>
	<i>E. chaffeensis</i> , <i>E. ewingii</i>	<i>dsb</i>
<i>Hepatozoon</i>	<i>Hepatozoon</i> (H.) spp, <i>H. americanum</i> , <i>H. canis</i>	18S
<i>Francisella</i>	<i>Francisella</i> spp.	<i>tul4</i> , <i>fopA</i>
<i>Leishmania</i>	<i>Leishmania</i> (L.) spp.	<i>hsp70</i>
	<i>L. infantum</i>	ITS
<i>Rangelia</i>	<i>Rangelia vitalli</i>	18S
<i>Rickettsia</i>	<i>Rickettsia</i> (R.) spp., <i>R. prowazekii</i>	<i>gltA</i>
	<i>R. parkeri</i>	<i>ompA</i>
	<i>R. felis</i>	<i>orfB</i>
	<i>R. akari</i> , <i>R. typhi</i> , <i>R. andeanae</i>	<i>ompB</i>
	<i>R. rickettsii</i> , <i>R. conorii</i> , <i>R. slovaca</i> , <i>R. massiliae</i> , <i>R. helvetica</i> , <i>R. aeschlimannii</i>	ITS
	<i>R. africae</i>	<i>sca1</i>
	<i>Neorickettsia</i> sp.	<i>groEL</i>
<i>Theileria</i>	<i>Theileria</i> (Th.) spp., <i>Th. velifera</i> , <i>Th. parva</i>	18S
	<i>Th. mutans</i>	ITS
	<i>Th. equi</i>	<i>ema1</i>
<i>Wolbachia</i>	<i>Wolbachia</i> spp.	<i>wsp</i>

**Table S2**

Host and location details of ticks tested and the real-time PCR cycle threshold (ct) values for microorganisms detected in this study.

Ticks	Host	Location	Lab ID	<i>Ehrlichia</i> sp.	<i>Rickettsia</i> sp.	<i>Bartonella</i> sp.	<i>Coxiella</i> -like sp.	Apicomplexa	<i>Theileria</i> sp.	<i>Hepatozoon</i> sp.
<i>Aponomma auruginans</i>	Bare-nosed wombat	Wilsons Promontory	11	-	-	-	+12.38	+19.42	-	-
			12	-	+26.42	-	+22.12	+21.54	-	-
<i>Ixodes antechini</i>	Agile antechinus	Boho South	23	-	+9.32	-	-	+17.44	+24.05	-
			24	-	+9.53	+19.69	-	+5.28	+7.66	-
			27	+18.72	+22.94	+17.4	-	+5.03	+17.6	-
			28	-	+24.48	+21.53	-	+15.2	+19.75	-
<i>Ixodes kohlsi</i>	Little penguin	Phillip Island	1	-	-	-	-	+25.4	-	-
			2	-	-	-	-	-	-	-
			3	-	-	-	-	+24.1	-	-
			4	-	-	-	-	+23.78	-	-
			5	-	-	-	-	-	-	-
			6	-	-	-	-	+22.77	-	-
<i>Ixodes tasmani</i>	Koala	Portland	22	-	+10.81	-	-	+11.04	+15.02	-
			7	-	+23.97	-	-	+23.67	-	-
			8	-	-	-	-	+11.6	+15.58	-
			9	-	-	-	-	+22.93	-	-
<i>Ixodes tasmani</i>	Mountain brushtail possum	Boho South	10	-	-	-	-	-	-	-
			14	-	-	-	-	+16.01	-	-
	Agile antechinus		25	-	-	-	-	+19.68	-	-
	Southern brown bandicoot	Koo Wee Rup	29	-	+8.5	-	-	+16.18	+19.84	-
			30	-	+12.07	-	-	+23.02	-	+24.24
			31	-	+10.66	-	-	+17.59	+23.07	-
			32	-	+9.47	+24	-	+15.95	-	-

			34	-	+/11.08	-	-	+/27.45	-	-
			36	-	+/10.5	-	-	+/24.01	-	-
			37	-	+/9.43	+/17.54	-	+/17.15	-	-
			38	-	+/9.51	-	-	+/24.66	-	-
<i>Ixodes trichosuri</i>	Mountain brushtail possum	Boho South	13	-	+/24.16	-	-	+/4.42	-	-
			15	-	+/8.5	+/22.68	+/25.86	+/17.09	+/22.97	-
			16	-	+/11.62	-	-	+/23.28	-	-
			17	-	-	-	-	-	-	-
			18	-	-	-	-	-	-	-
			19	-	-	-	-	+/24.49	+/23.66	-
			20	-	+/11	-	-	-	-	-
			21	-	+/24.2	+/26.15	-	+/34.97	-	-
			26	-	-	+/27.13	-	+/22.35	-	-
			33	-	+/26.07	-	-	+/22.6	-	-
	Southern brown bandicoot	Koo Wee Rup	35	-	+/24.85	-	-	+/22.84	-	-
			39	-	-	-	-	+/15.29	-	-
		Cranbourne	40	-	-	-	-	+/22.29	-	-
			41	-	+/25.74	-	-	+/24.84	-	-
			42	-	-	-	-	+/22.84	-	-
			43	-	+/24.8	-	-	+/20.77	-	-
			44	-	-	-	-	+/33.06	-	-
			45	-	+/27.59	-	-	-	-	-

**Table S3**

Pairwise percentage nucleotide similarities of 16S ribosomal RNA sequences of *Ixodes tasmani* determined in this study

Sequence ID	7-2020	10-2020	AD-2020	L-2020
7-2020		99.7	85.9	86.5
10-2020			85.7	86.2
AD-2020				98.2
L-2020				

**Table S4**

Pairwise percentage nucleotide similarities of *cox1* sequences of *Ixodes tasmani* determined in this study

Sequence ID	7-2020	AD-2020	L-2020
7-2020		88.7	89.2
AD-2020			98
L-2020			