

Attachment 1. Thermocycler programs, primers, and probes for polymerase chain reaction (PCR) protocols.

Pathogen	Protocol	Temperature (Celsius) and time	Cycles	Primer/probe	Primers and probes (5'-3')
<i>Borrelia burgdorferi</i>	qPCR	50 for 2 minutes		Primer Primer	FWD: CGAGTCTTAAAAGGGCGATTTAGT ³ REV: GCTTCAGCCTGGCCATAAATAG ³
		95 for 10 minutes		Probe	FAM-AGATGTGGT/ZEN/AGACCCGAAGCCGAGTG ³
<i>Anaplasma marginale</i>	qPCR	95 for 15 seconds	40 cycles	Primer	FWD: TTGGCAAGGCAGCAGCTT ¹
		60 for 1 minutes		Primer Probe	REV: TTCCGCGAGCATGTGCAT ¹ 56-FAM/TCGGTCTAACATCTCCAGGCTTTCAT ¹
<i>Theileria cervi</i> and <i>Babesia</i> spp.	Nested PCR external primer	95 for 15 seconds		Primer Primer	FWD: CCTGGTTGATCCTGCCAGTAGT ⁴ REV: CTCCTTCCTTTAAGTGATAAG ⁴
		95 for 1 minute 48 for 1 minute 72 for 1 minute 72 for 1 minute	30 cycles		
<i>T. cervi</i> and <i>Babesia</i> spp.	Nested PCR internal primer	94 for 15 seconds		Primer Primer	FWD: GAGGTAGTGACAAGAAATAACAATA ^{2,4} REV: TCTTCGATCCCCTAACTTTC ⁴
		94 for 1 minute 50 for 1 minute 72 for 1.5 minutes 72 for 10 minutes	40 cycles		

¹Carelli et al. 2007

²Gubbels et al. 1999

³Xu et al. 2016

⁴Yabsley et al. 2005a