
SCIENTIFIC PUBLICATIONS

Bhoora, R., Franssen, L., Oosthuizen, M.C., Guthrie, A.J., Zwegarth, E., Penzhorn, B.L., Jongejan, F., Collins, N.E., 2009. Sequence heterogeneity in the 18S rRNA gene within *Theileria equi* and *Babesia caballi* from horses in South Africa. *Veterinary Parasitology* 159, 112-120.

Bhoora, R., Quan, M., Franssen, L., Butler, C.M., van der Kolk, J.H., Guthrie, A.J., Zwegarth, E., Jongejan, F., Collins, N.E., 2009. Development and evaluation of real-time PCR assays for the quantitative detection of *Babesia caballi* and *Theileria equi* infections in horses from South Africa. *Veterinary Parasitology* doi:10.1016/j.vetpar.2009.11.011.

Bhoora, R., Quan, M., Zwegarth, E., Guthrie, A.J., Prinsloo, S.A., Collins, N.E., 2010. Sequence heterogeneity in the gene encoding the rhoptry-associated protein-1 (RAP-1) of *Babesia caballi* isolates from South Africa. *Veterinary Parasitology* doi:10.1016/j.vetpar.2010.01.009.



APPENDICES



Appendix A Quantitative PCR results obtained for the ten-fold dilution series from the *in vitro*-cultured South African *B. caballi* 502 and *T. equi* WL isolates

***B. caballi* 502**

%PE		8.1	0.81	0.081	0.0081	0.00081	0.000081	0.0000081					
Log%PE		0.9084850	0.0915150	-1.0915150	-2.0915150	-3.0915150	-4.0915150	-5.0915150		Slope	Intercept	Efficiency	Amplification
REP1	1	19.66	23.06	26.54	29.82	33.19	37.53	Negative		-3.34	22.90	0.99	1.99
	2	19.23	22.81	26.38	29.46	33.34	37.37	Negative		-3.52	22.60	0.92	1.92
	3	19.50	22.76	26.06	30.67	33.39	37.68	Negative		-3.58	22.75	0.90	1.90
	4	19.38	22.82	25.95	29.43	32.74	Negative	Negative		-3.26	22.62	1.03	2.03
REP2	5	19.25	22.74	26.08	29.35	32.76	35.97	Negative		-3.28	22.56	1.02	2.02
	6	19.25	22.78	25.94	29.30	32.28	37.68	Negative		-3.48	22.43	0.94	1.94
	7	19.30	22.83	26.07	29.29	32.82	37.20	Negative		-3.34	22.54	0.99	1.99
REP3	8	19.02	22.56	25.87	29.07	32.71	37.50	36.42		-3.13	22.54	1.09	2.09
	9	18.97	22.63	25.93	29.17	33.33	35.09	Negative		-3.26	22.43	1.03	2.03
	10	19.32	22.96	26.18	29.36	33.23	35.55	37.21		-3.03	22.86	1.14	2.14
REP4	11	19.11	22.77	26.09	29.14	32.67	35.69	Negative		-3.25	22.51	1.03	2.03
	12	19.24	22.72	26.00	29.24	33.46	Negative	Negative		-3.43	22.52	0.96	1.96
	13	19.39	22.80	25.98	29.57	32.31	35.56	Negative		-3.17	22.64	1.07	2.07
REP5	14	19.24	22.81	26.13	29.19	32.42	37.20	Negative		-3.42	22.49	0.96	1.96
	15	19.22	22.72	25.93	29.47	33.30	35.32	Negative		-3.25	22.58	1.03	2.03
	16	19.30	22.75	26.01	29.42	32.91	36.61	36.91		-3.08	22.76	1.11	2.11
REP6	17	19.22	22.76	25.94	29.37	33.45	36.31	Negative		-3.40	22.54	0.97	1.97
	18	19.23	22.70	25.96	29.45	32.70	37.12	Negative		-3.45	22.46	0.95	1.95
	19	19.29	22.70	25.91	29.32	32.61	Negative	Negative		-3.26	22.52	1.03	2.03
REP7	20	19.21	22.70	26.01	29.26	32.24	37.11	Negative		-3.41	22.43	0.97	1.97
	21	19.20	22.70	26.09	29.21	32.26	37.29	Negative		-3.43	22.43	0.96	1.96
	22	19.26	22.66	26.01	29.62	32.88	37.36	37.60		-3.22	22.69	1.05	2.05
REP8	23	19.17	22.62	26.03	29.44	32.60	37.64	Negative		-3.53	22.40	0.92	1.92
	24	19.07	22.70	25.90	29.29	32.69	Negative	Negative		-3.31	22.43	1.00	2.00
	25	19.32	22.71	25.94	29.34	32.88	37.18	Negative		-3.46	22.49	0.95	1.95
REP9	26	19.22	22.70	25.95	29.35	32.78	36.24	Negative		-3.34	22.50	0.99	1.99
	27	19.26	22.64	25.93	29.08	32.70	35.40	Negative		-3.20	22.50	1.05	2.05
	28	19.12	22.61	25.80	29.42	32.61	34.93	Negative		-3.16	22.47	1.07	2.07
REP10	29	19.03	22.60	25.84	29.33	33.23	35.37	Negative		-3.29	22.43	1.01	2.01
	30	19.03	22.59	25.90	29.45	32.81	35.56	Negative		-3.28	22.43	1.02	2.02
MEAN		19.23	22.73	26.01	29.40	32.84	36.52	37.03		-3.37	22.52	0.98	1.98



T. equi WL

%PE		13.8	1.38	0.138	0.0138	0.00138	0.000138	0.0000138					
Log %PE		1.1398791	0.1398791	-0.8601209	-1.8601209	-2.8601209	-3.8601209	-4.8601209		Slope	Intercept	Efficiency	Amplification
	1	19.03	22.79	26.46	29.53	32.96	37.68	Negative		-3.32	23.20	1.00	2.00
REP1	2	19.01	22.80	26.38	29.60	32.57	35.55	Negative		-3.39	23.15	0.97	1.97
	3	18.76	22.70	26.36	29.84	32.61	Negative	Negative		-3.48	23.05	0.94	1.94
	4	18.87	22.71	26.49	29.40	32.89	35.62	Negative		-3.35	23.11	0.99	1.99
REP2	5	18.82	22.72	26.35	29.70	32.69	Negative	Negative		-3.47	23.07	0.94	1.94
	6	18.58	22.79	26.23	29.73	33.24	39.99	Negative		-4.05	22.91	0.76	1.76
	7	18.88	22.52	26.59	29.65	32.78	38.54	Negative		-3.49	23.08	0.93	1.93
REP3	8	19.00	22.87	26.51	29.65	32.91	34.65	Negative		-3.19	23.26	1.06	2.06
	9	18.77	22.72	26.48	29.65	32.87	35.02	Negative		-3.28	23.12	1.02	2.02
	10	18.72	22.69	26.35	29.55	32.89	37.90	Negative		-3.71	22.97	0.86	1.86
REP4	11	18.88	22.68	26.40	29.85	32.22	36.14	Negative		-3.38	23.09	0.98	1.98
	12	18.70	22.83	26.46	29.90	Negative	Negative	39.03		-3.35	23.16	0.99	1.99
	13	18.95	22.58	26.31	29.48	33.21	35.28	Negative		-3.34	23.09	0.99	1.99
REP5	14	18.72	22.75	26.40	29.44	32.87	35.96	Negative		-3.42	23.04	0.96	1.96
	15	18.49	22.72	26.25	29.46	33.46	35.84	Negative		-3.49	22.95	0.93	1.93
	16	18.84	22.75	26.45	29.85	33.15	36.48	Negative		-3.51	23.14	0.93	1.93
REP6	17	18.84	22.76	26.38	29.73	32.84	35.78	Negative		-3.38	23.12	0.98	1.98
	18	18.79	22.68	26.29	29.68	33.00	36.06	Negative		-3.45	23.06	0.95	1.95
	19	18.89	22.60	26.58	29.73	33.32	37.32	Negative		-3.64	23.12	0.88	1.88
REP7	20	18.87	22.80	26.44	29.74	33.19	35.74	Negative		-3.39	23.18	0.97	1.97
	21	18.75	22.66	26.49	29.62	32.57	36.21	Negative		-3.44	23.04	0.95	1.95
	22	18.98	22.93	26.56	29.72	32.87	37.84	Negative		-3.64	23.20	0.88	1.88
REP8	23	19.02	22.81	26.62	29.79	32.98	36.44	Negative		-3.45	23.24	0.95	1.95
	24	18.82	22.74	26.33	29.87	33.06	35.21	38.49		-3.24	23.19	1.04	2.04
	25	19.06	22.88	26.75	30.03	33.01	35.33	Negative		-3.29	23.37	1.02	2.02
REP9	26	18.96	22.87	26.57	29.88	32.89	35.94	Negative		-3.38	23.25	0.98	1.98
	27	18.99	22.89	26.57	29.80	32.95	36.53	Negative		-3.46	23.25	0.95	1.95
	28	19.36	22.86	26.41	29.65	32.97	36.46	Negative		-3.40	23.32	0.97	1.97
REP10	29	18.93	23.55	26.44	31.22	32.57	Negative	Negative		-3.50	23.53	0.93	1.93
MEAN		18.87	22.78	26.44	29.75	32.91	36.33	38.76		-3.33	23.21	1.00	2.00

Appendix B Comparison of test results obtained for the National Yearling Sale 2006 samples.

Sample	cELISA	IFAT	<i>In vitro</i> culture	<i>T. equi</i> 18S rRNA TaqMan qPCR	<i>T. equi</i> <i>ema-1</i> TaqMan qPCR (Ueti et al., 2003)	<i>T. equi</i> <i>ema-1</i> Taqman MGB qPCR
EQ1	-	-	-	-	-	-
EQ2	+	+	+	-	-	-
EQ3	+	+	+	30.00	30.21	27.31
EQ4	+	+	+	33.61	33.27	31.18
EQ5	+	+	+	29.57	28.38	26.35
EQ6	+	+	+	28.85	-	26.88
EQ7	+	+	+	28.57	28.94	25.3
EQ8	+	+	+	28.55	-	28.76
EQ9	+	+	+	31.34	31.28	28.5
EQ10	+	+	+	29.4	-	29.56
EQ11	+	+	+	28.42	29.48	25.55
EQ12	+	+	+	29.68	36.42	26.7
EQ13	+	+	+	30.11	29.25	26.4
EQ14	-	-	-	-	-	-
EQ15	-	-	-	-	-	-
EQ16	-	-	-	-	-	-
EQ17	+	+	+	31.98	38.29	29.51
EQ18	-	-	-	-	-	-
EQ19	+	+	+	29.65	27.83	27.46
EQ20	+	+	+	32.4	-	31.23
EQ21	+	+	+	32.2	32.28	28.88
EQ22	-	-	-	-	-	-
EQ23	-	-	-	-	-	-
EQ24	+	+	+	30.85	30.31	27.96
EQ25	+	+	+	28.58	-	25.92
EQ26	+	+	+	28.75	28.45	26.02
EQ27	+	+	+	28.83	28.5	25.74
EQ28	+	+	+	27.85	27.3	24.39
EQ29	-	-	-	-	-	-
EQ30	+	-	+	29.42	29.22	26.73
EQ31	+	+	+	30.27	29.7	27.53
EQ32	+	+	+	29.08	37.97	28.96
EQ33	-	-	+	31.51	33.9	28.84
EQ35	+	-	+	28.3	31.19	26.02
EQ37	+	+	+	31.54	-	29.94
EQ38	+	+	+	33.34	-	33.3
EQ39	+	+	+	32.54	31.97	29.59
EQ40	+	+	+	28.67	29.7	25.71
EQ41	+	+	+	31.52	31.41	28.15
EQ42	+	+	+	27.98	27	24.85
EQ43	+	+	+	31.95	-	27.76
EQ44	+	+	+	29.05	-	26.72
EQ45	+	+	+	29.45	30.46	27.17
EQ46	+	+	+	28.99	-	26.32
EQ47	+	+	+	32.48	39.34	29.28
EQ48	+	+	+	31.03	36.03	28.79
EQ49	+	+	+	29.42	26.28	27.21
EQ50	-	+	+	31.21	-	29.28
EQ51	+	+	+	29.48	-	26.73
EQ52	+	+	+	30.98	30.08	27.97
EQ53	+	+	+	31.05	-	28.92
EQ54	-	+	+	32.04	-	29.85
EQ55	-	-	-	-	-	-



Sample	cELISA	IFAT	<i>In vitro</i> culture	<i>T. equi</i> 18S rRNA TaqMan qPCR	<i>T. equi</i> ema-1 TaqMan qPCR (Ueti et al., 2003)	<i>T. equi</i> ema-1 Taqman MGB qPCR
EQ56	+	+	+	28.29	28.4	25.56
EQ57	+	+	+	29.03	29.52	25.25
EQ58	-	-	-	-	-	-
EQ59	+	+	+	29.98	-	27.4
EQ60	+	+	+	32.85	-	31.01
EQ61	+	+	+	34.93	-	33.76
EQ62	+	+	+	27.16	27.43	24.59
EQ63	+	+	+	-	36.52	-
EQ64	+	+	+	30.93	-	28.62
EQ65	+	+	+	29.33	27.95	27.82
EQ66	+	+	+	32.05	28.94	29.48
EQ67	+	+	+	32.79	-	31.78
EQ68	+	+	+	31.09	30.3	28.9
EQ69	+	+	+	30.12	30.71	27.75
EQ70	+	+	-	32.11	-	34.77
EQ71	+	+	-	35.15	-	32.12
EQ72	+	+	+	33.03	33.57	30.68
EQ73	+	+	+	31.96	31.69	28.95
EQ74	+	+	+	32.53	36.93	30.39
EQ75	+	+	+	32.51	36.68	32.6
EQ76	+	+	+	30.43	29.58	27.51
EQ77	+	+	+	31.9	31.56	29.46
EQ78	+	+	+	29.8	29.27	26.84
EQ79	+	+	+	32.52	-	30.35
EQ80	+	+	+	29.51	29.39	26.85
EQ81	+	+	-	32.55	34.83	29.91
EQ82	+	+	+	29.21	36.16	26.17
EQ83	+	+	+	33.39	32.74	27.87
EQ84	+	+	+	29.77	28.85	26.58
EQ85	+	+	+	33.39	34.09	32.21
EQ86	+	+	+	33.18	34.47	30.21
EQ87	+	+	+	32.91	-	30.77
EQ88	+	+	+	29.95	29.37	27.01
EQ89	+	+	+	29.91	33.94	24.8
EQ90	+	+	+	30.56	35.32	27.78
EQ91	+	+	+	31.37	30.45	26.88
EQ92	-	-	-	-	-	-
EQ93	+	+	+	29.59	28.81	26.82
EQ94	-	+	+	31.25	33.22	28.66
EQ95	+	+	+	32.44	31.08	29.86
EQ96	+	+	+	33.28	31.8	29.68
EQ97	-	-	-	-	-	-
EQ98	+	+	+	28.63	28.18	25.9
EQ99	-	-	-	-	-	-
EQ100	+	+	+	28.97	28.72	26.57
EQ101	+	+	+	30.86	30.56	27.93
EQ102	+	+	+	28.73	28.34	25.85
EQ103	+	+	+	-	-	-
EQ104	+	+	+	28.97	27.79	25.17
EQ105	+	+	+	29.89	28.59	24.57
EQ106	+	+	+	31.37	31.66	27.53
EQ107	-	-	-	37.45	-	-