Table S1. PCR cycling conditions for PCR-restriction fragment length polymorphism (PCR-RFLP) based *Toxoplasma gondii* genotyping.

Multiplex Multilocus Nested PCR-RFLP Protocol			**Singleplex Nested PCR-RFLP Protocol**			
1st step: multiplex PCR			1st step: singleplex PCR			
Initial Denaturation	95 °C, 4 min		95 °C, 4 min			
Denaturation	94 °C, 30 s		94 °C, 30 s			
Annealing	55 °C, 1 min		55 °C, 1 min			
Elongation	72° C, 2 min		72 °C, 2 min			
No. of cylces	30		35			
Final elongation	72 °C, 5 min		72 °C, 5 min			
2nd step: nested PCR			2nd step: nested PCR			
Template	1.5 µl of 1:2 diluted PCR product of multiplex PCR		1.5 µl of undiluted PCR product of singleplex PCR			
Marker	Apico	all other genetic markers[a]	SAG3	BTUB, GRA6	Apico	all other genetic markers[b]
Initial Denaturation	95 °C, 4 min	95 °C, 4 min	95 °C, 5 min	95 °C, 5 min	95 °C, 4 min	95 °C, 4 min
Denaturation	94 °C, 30 s	94 °C, 30 s	94 °C, 60 s	94 °C, 60 s	94 °C, 30 s	94 °C, 30 s
Annealing	55 °C, 1 min	60 °C, 1 min	60 °C, 1 min	55 °C, 1 min	55 °C, 1 min	60 °C, 1 min
Elongation	72 °C, 2 min	72 °C, 1.5 min	72 °C, 1 min	72 °C, 1 min	72 °C, 2 min	72 °C, 1.5 min
No. of cylces	35	35	39	39	39	39
Final elongation	na	na	72 °C, 5 min	72 °C, 5 min	72 °C, 5 min	72 °C, 5 min

Notes: DreamTaq DNA polymerase (Thermo Fisher Scientific, USA) was used in singleplex and nested PCRs, while FastStart DNA Polymerase (Roche, Switzerland) was used in multiplex PCR. [a]SAG1, 5′-SAG2, 3′-SAG2, SAG3, BTUB, GRA6, c22-8, c29-2, L358, PK1, alt.SAG2; [b]SAG1, 5′-SAG2, 3′-SAG2, c22-8, c29-2, L358, PK1, alt.SAG2; na, not applied. .