

Table S1: Cloning primers used in this study

Primer Name (restriction site)	Primer Sequence (restriction site underlined)	Cloning Vector (research objective)
Functional Expression of <i>TgCK</i> and <i>TgEK</i> in <i>E. coli</i>		
<i>TgCK-F (NdeI)</i>	CTCCATATGCAGGTACTCGCGTGTGTT	<i>pET22b</i> ⁺ (<i>TgCK</i> -6xHis in Rosetta strain)
<i>TgCK-R (HindIII)</i>	CTCA <u>AGCTT</u> TCAGGCCGGAAAGAGT	
<i>TgCK-wo-HP-F (NdeI)</i>	CTC <u>ATCC</u> ATATGTC <u>CCCCTTC</u> CAGGC <u>GCTGGCT</u>	<i>pET28b</i> ⁺ (6xHis- <i>TgCK</i> _S in Rosetta strain)
<i>TgCK-R (BglII)</i>	CT <u>CAGATCTTC</u> AC <u>TTTC</u> GAGGCCGGAAAGAGTCC	
<i>TgEK-F (NcoI)</i>	CT <u>CCC</u> ATGCC <u>AGCAAGGCAGAGAGAAC</u>	<i>pET28b</i> ⁺ (<i>TgEK</i> -6xHis in Rosetta strain)
<i>TgEK-R (HindIII)</i>	CT <u>CAAGCTT</u> GAACGACA <u>ATGC</u> GGGACT	
Functional Expression of <i>ScCK1</i> and <i>ScEK1</i> in <i>S. cerevisiae</i>		
<i>ScCK1-F (NotI)</i>	CTCG <u>GGGCCG</u> CATGGTACAAGAAC <u>TCACGTCCA</u>	<i>pESC-Ura</i> (<i>ScCK1</i> in KS106 strain)
<i>ScCK1-R (NotI)</i>	CTCG <u>GGGCCG</u> CTTACAAATA <u>ACTAGTATCGAGGAAC</u> TT	
<i>ScEK1-F (SpeI)</i>	CT <u>CACTAGT</u> ATGTAC <u>ACCAATTATTCA</u> CTTAC	<i>pESC-Ura</i> (<i>ScEK1</i> in KS106 strain)
<i>ScEK1-R (BglII)</i>	CT <u>CAAGATCTT</u> AAAAAATA <u>AGTTTAGTGCTAAG</u>	
Subcellular Localization of <i>TgCK</i> and <i>TgEK</i> in <i>T. gondii</i>		
<i>TgCK-Term-F (HindIII)</i>	CT <u>CAAGCTT</u> CTGGAA <u>ATTGGAGTCACGC</u>	Step # 1 for expressing <i>TgCK</i> -HA under the <i>pTgCK</i> promoter in tachyzoites
<i>TgCK-Term-R (NheI)</i>	CT <u>CGCTAG</u> CCA <u>AGCAGAAGTCGGATATTAGCG</u>	
<i>TgCK-Prom-F (Apal)</i>	CT <u>CGGGCCC</u> GG <u>CAGGTGGTTTGCTTC</u>	Step # 2 for expressing <i>TgCK</i> -HA under the <i>pTgCK</i> promoter in tachyzoites
<i>TgCK-Prom-R (HindIII)</i>	CT <u>ACTGAA</u> G <u>CTTA</u> A <u>ACTCTCGAAC</u>	
<i>TgCK-ORF-F (HindIII)</i>	GT <u>ATTCAAGCTT</u> CA <u>GTCAGTAGCACCAAC</u>	Step # 3 for expressing <i>TgCK</i> -HA under the <i>pTgCK</i> promoter in tachyzoites
<i>TgCK-ORF-HA-R (HindIII)</i>	CT <u>CAAGCTT</u> CA <u>AGCGTAATCTGGAACATCGTATGGGTA</u> CT <u>TTCCGAGCCGGAAAGAG</u>	
<i>TgCK-Prom-HP-Ty1-F (NheI)</i>	CT <u>CTCTGCTAGCCTGGATAAA</u> A <u>ACCCGATGCTACAAATC</u>	Step # 1 for expressing <i>TgCK</i> -Ty1 under the <i>pTgCK</i> promoter in tachyzoites
<i>TgCK-Prom-HP-Ty1-R (Apal)</i>	CT <u>CTCTGGGCCCATCGAGCGGGCTCTGGTGTGGAC</u> CT <u>UAGCGCCTGAA</u> GGGGAC <u>GC</u>	
<i>TgCK-ORF-Term-F (Apal)</i>	CT <u>CTCTGGGCCCGCTCTTGTTCTGGTGGC</u>	Step # 2 for expressing <i>TgCK</i> -Ty1 under the <i>pTgCK</i> promoter in tachyzoites
<i>TgCK-ORF-Term-R (Apal)</i>	CT <u>CTCTGGGCCCAAGCAGAAGTCGGATATTAGCG</u>	
<i>TgCK_S-F (SbfI)</i>	CT <u>CATCCCTGCAGGCC</u> CC <u>CTTCAGGC<u>GCTGGCT</u></u>	<i>pTKO</i> (For expressing <i>TgCK</i> _S -myc in tachyzoites)
<i>TgCK_S-myc-R (PacI)</i>	CT <u>CATCTTA</u> AT <u>TA</u> ACT <u>AGAGGTCTCTCGGAA</u> AT <u>CAAC</u> TT <u>CTGTTCC</u> TT <u>CGAGCCGGAAAGAGTCCA</u>	
<i>TgEK-F (NcoI)</i>	CT <u>CCC</u> ATGCC <u>AGCAAGGCAGAGAGAAC</u>	<i>pNTP3</i> (For expressing <i>TgEK</i> -HA in tachyzoites)
<i>TgEK-HA-R (PacI)</i>	CT <u>CTTA</u> AT <u>TA</u> AT <u>CAAGCGTAATCTGGAACATCGTATGGG</u> TAGAAC <u>GA</u> AA <u>ATGC</u> GGGACT	
Promoter Displacement of <i>TgCK</i> in <i>T. gondii</i>		
<i>TgCK-PD-5'UTR-F (NdeI)</i>	CT <u>CATCC</u> AT <u>GGGATG</u> A <u>AGTGTGTGGTCTG</u>	<i>pDT7S4</i> (promoter displacement of <i>TgCK</i> in the TaTi- <i>Δku80</i> strain)
<i>TgCK-PD-5'UTR-R (NdeI)</i>	CT <u>CATCC</u> AT <u>GTGTAA</u> ACT <u>AGGC</u> GACT <u>ACACAGC</u>	
<i>TgCK-PD-3'UTR-F (BglII)</i>	CT <u>CATCAGATCT</u> AT <u>GCAGGTACTCGCGTGTG</u>	<i>pDT7S4</i> (promoter replacement of <i>TgCK</i> in the TaTi- <i>Δku80</i> strain)
<i>TgCK-PD-3'UTR-R (AvrII)</i>	CT <u>CATCC</u> TA <u>GGGAAC</u> GGG <u>TACTCCATCAGGTAGT</u>	
<i>TgCK-PD-5'Scr-F</i>	CATTCCGAGGC <u>GGATAAA</u>	Screening for 5'-crossover in transgenic TaTi- <i>Δku80</i> strain
<i>TgCK-PD-5'Scr-R (DHFR-R)</i>	CGGGTT <u>GAATG</u> CA <u>AGGTT</u>	
<i>TgCK-PD-3'Scr-F (DHFR-F)</i>	CT <u>CTCTTT</u> CG <u>AGGGATCAG</u>	Screening for 3'-crossover in transgenic TaTi- <i>Δku80</i> strain
<i>TgCK-PD-3'Scr-R</i>	ACAA <u>ACCTGTCTGCACCG</u>	
Conventional Knockout of <i>TgCK</i> in <i>T. gondii</i>		
<i>TgCK-KO-5'UTR-F (HindIII)</i>	CT <u>CAAGCTT</u> CG <u>TAGGATAGAAGCGAGTCGTT</u>	<i>p2854</i> (conventional knockout of <i>TgCK</i> in the <i>HXPRT</i> strain)
<i>TgCK-KO-5'UTR-R (NheI)</i>	CT <u>CGCTAG</u> CC <u>GCTAGGAGGTCAA</u> TT <u>GC</u>	
<i>TgCK-KO-3'UTR-F (NotI)</i>	CT <u>CGGGCCG</u> CT <u>CGGATAACACAGTGGAA</u> CT <u>TGG</u>	<i>p2854</i> (conventional knockout of <i>TgCK</i> in the <i>HXPRT</i> strain)
<i>TgCK-KO-3'UTR-R (NotI)</i>	CT <u>CGGGCCG</u> CT <u>CACACCAAA</u> AG <u>AGGGCCG</u>	
<i>TgCK-KO-5'Scr-F</i>	CCT <u>CGTTCTAGA</u> AAA <u>AGGCTGC</u>	Screening of 5'-crossover in transgenic <i>HXPRT</i> strain
<i>TgCK-KO-5'Scr-R</i>	AT <u>GCAAGGTT</u> CG <u>GCTGTC</u>	
<i>TgCK-KO-3'Scr-F</i>	CGA <u>ATCCAGATGGAGATGGCTGTC</u>	Screening of 3'-crossover in transgenic <i>HXPRT</i> strain
<i>TgCK-KO-3'Scr-R</i>	AGA <u>ATGCGAGTGT</u> CG <u>CAA</u>	

Quantitative PCR of <i>TgCK</i> transcript		
<i>TgCK</i> -Ex1-F	CTTTGTTCTGGTGGCCAGT	EST encoded by the exon 1 of full-length choline kinase
<i>TgCK</i> -Ex1-R	CCCGATAAACGACTGCACTT	
<i>TgCK</i> -Ex6-F	TCTTCTCCGTCCACTTGACC	EST encoded by the exon 6 of full-length choline kinase
<i>TgCK</i> -Ex6-R	GTCCACCATAGCCTTGGAAA	
<i>TgElf1α</i> -F	AGTCGACCACTACCGGACAC	Housekeeping gene for normalizing the expression of ESTs from choline kinase
<i>TgElf1α</i> -R	CTCGGCCTTCAGTTATCCA	
GFP expression in <i>E. coli</i>		
<i>TgCK</i> -Ex1.1-F (<i>XcmI</i>)	CTCCCCACCGGTCACCTGGATGCAGGTACTCGCGTGTGT	A fragment of exon 1 of choline kinase until the 3 rd ATG (1-729 bp)
<i>TgCK</i> -Ex1.1-R (<i>NsiI</i>)	<u>CTCATGCATATCCCCTAAAGTTGAACACGC</u>	
<i>TgCK</i> -Ex1.2-F (<i>XcmI</i>)	CTCCCCACCGGTCACCTGGATGCAGGTACTCGCGTGTGT	A fragment of exon 1 of choline kinase until the 2 nd ATG (1-492 bp)
<i>TgCK</i> -Ex1.2-R (<i>NsiI</i>)	<u>CTCATGCATGGGGTTGCTGCCAGTGCG</u>	
<i>TgGRA2</i> -F (<i>XcmI</i>)	CTCCCCACCGGTCACCTGGCTCTCAGCTGCGCGAGG	A minimal promoter element of the dense granule 2 (Gra2) protein
<i>TgGRA2</i> -R (<i>NsiI</i>)	<u>CTCATGCATCTGCTTGATTCTCAAAGAACACAG</u>	
GFP-F (<i>NsiI</i>)	CTCATGCATAAAGGAGAAGAACCTTTCACTGGA	ORF of the green fluorescent protein
GFP-R (<i>PacI</i>)	<u>CTCTTAATTATTATTTGTATAGTTCATCCATGCCA</u>	