Supplementary Figure Legends

Supplementary Fig. S1. Western blot of rabbit polyclonal anti-*Toxoplasma gondii* calcium-dependent protein kinase 1 (*Tg*CDPK1). (A) Rabbit antiserum raised to recombinant (r)*Tg*CDPK1 detects a band consistent with *Tg*CDPK1 in *T. gondii* lysates (T, 3 x 10^6 wild type *Toxoplasma*; C**,** 6 ng of purified CDPK1), but not in human host fibroblasts (F). Protein size marker lanes are labelled M. The r*Tg*CDPK1 antigen had the first 29 N- terminal

amino acid residues deleted relative to the wild-type enzyme, hence the lower size band of predicted molecular weight (54.73 versus 57.26 kDa). (B) Western blot of *T. gondii* lysates/anti-*Tg*CDPK1 showing the relationship between sample load and signal strength**.** A serial dilution of *T. gondii* parasite cells from 9 x 10^6 down to 0.3 x 10^6 and subsequent run of the respective lysate showed a linear relationship between sample load and signal strength in the immunoblot. C) Anti-*Tg*CDPK1 cross-reacts with *Sarcocystis neurona (Sn)*CDPK1. Numbers of parasites loaded per lane are indicated in millions. Sn-e, *S. neurona* extracellular cells; Sn-I, *S. neurona* intracellular cells; BT, bovine turbinate (epithelia) host cells for *S. neurona*; Tg, *T. gondii* tachyzoites. All lanes were run on the same 8% gel with equal exposure. The entire gel (top to bottom) is shown. Migration of CDPK1 is slightly slower for *S. neurona* compared with *T. gondii*, as expected according to the predicted molecular weights of 58.84 kDa and 57.26 kDa, respectively.

Supplementary Fig. S2. Amino acid sequence alignments. (A) Full sequence alignments of *Sarcocystis neurona* calcium-dependent protein kinase 1 (*Sn*CDPK1) SN3 (**SN3_02800495**; 524 bp) and SN1 (**SRCN_3314_SN1**, 519 bp), and *Toxoplasma gondii* (*Tg)*CDPK1, GenBank accession number **AAG53993**) (507 bp) with amino acids predicted as ATP binding sites are highlighted in turquoise. Based on the RNA-Seq data, *Sn*CDPK1 has an open reading frame of 1572 nucleotides encoding 524 amino acids. The transcript is 1925 bp. Similar to several other genes, differences in the lengths between the enzymes lie in the N-terminus. *Sn*CDPK1 sequences have longer N-termini compared with *Tg*CDPK1 (accession number **AAG53993**). (B) Sequence alignment of *Sn*CDPK1and *Tg*CPK1. *Sarcocystis neurona* CDPK1 is 524 amino acids while *Tg*CPK1 retrieved from ToxoDB (www. toxodb.org/toxo/)

(**TGME49_301440**) is 582 amino acids. The *Tg*CPK1 sequence from ToxoDB (**TGME49_301440**) has a 75 amino acid insertion not found in the *Tg*CDPK1 GenBank entry **AAG53993**. Highlighted in red is the region where additional differences are present. No putative conserved domains were detected in this region. (C) Sequence alignment of SN3 *Sn*CDPK1 and *Tg*CPK1 (**TGME49_301440**). *Sn*CDPK1 and *Tg*CDPK1 sequences retrieved from ToxoDB are 589 and 582 amino acids, respectively. There are additional exons in the middle of each protein that contribute to this sequence difference, 65 and 75 amino acids (yellow or red). *Sn*CDPK1 and *Tg*CDPK1sequences on ToxoDB were annotated through automation and might not have been manually curated. These regions were not found in the transcript retrieved from RNA- Seq data. Hence, we believe these exons in the middle are incorrect and not part of the CDPKs as shown by PCR and nucleotide sequence analysis of cDNA.