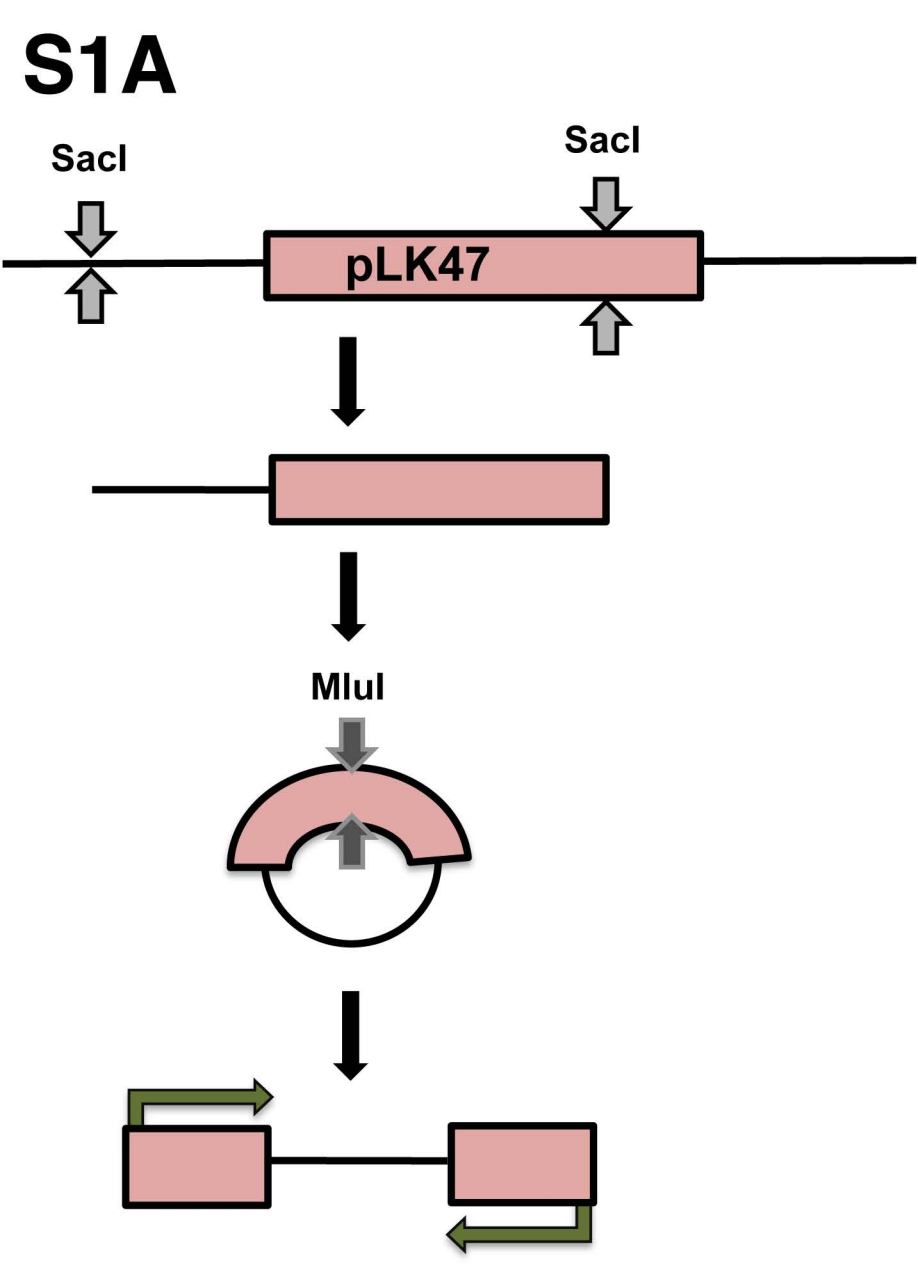

Figure S1. Insertional mutagenesis into Adaptin-3β slows replication in the presence of both MP-IV-1 and QQ-437 but does not kill tachyzoites. A. Method for insertional mutagenesis probe and identifying the gene disrupted. The flanking region in the first line before the SAC1 site is proportional to that in the second line. We indicate where the SC1 sites are by a different symbol from the MluI sites. We use these symbols in all appropriate lines to indicate where the process takes place. This illustrates that the SC1 is cut, recircularized, then cut with Mlu1 and then amplfied to find the flanking region.

S1B

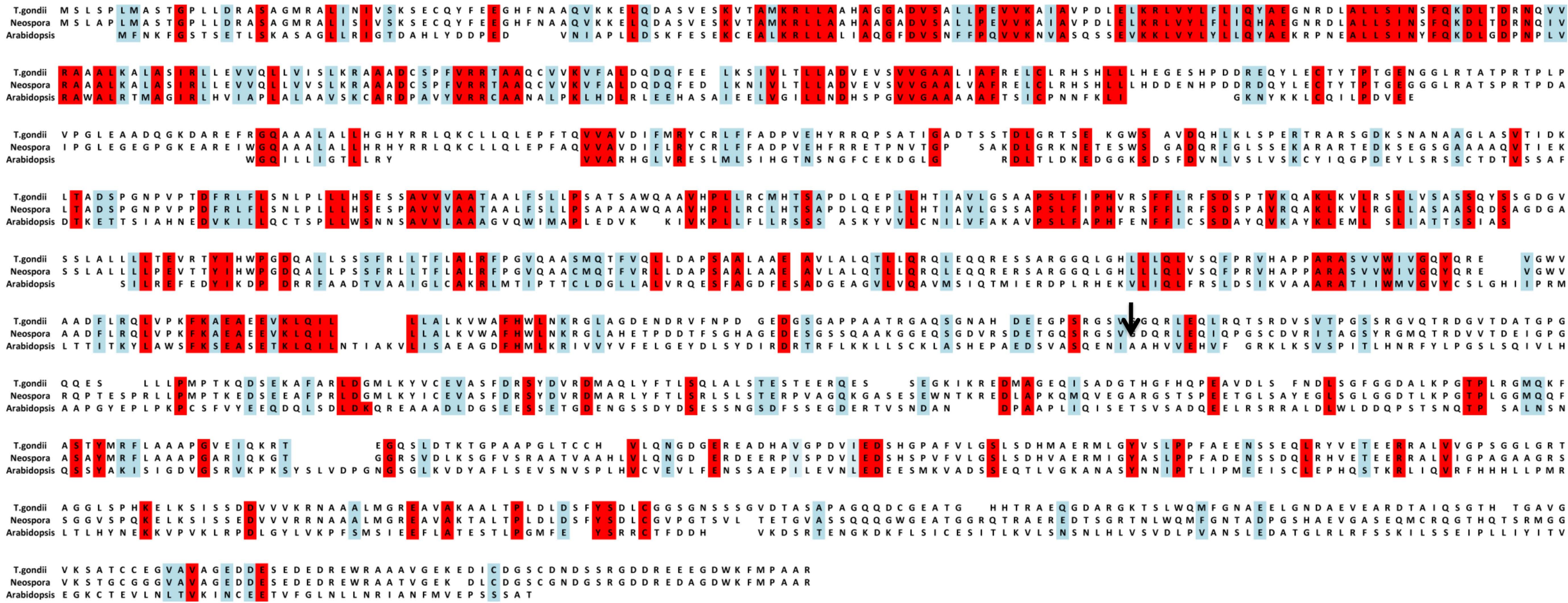

Figure S1. Insertional mutagenesis into Adaptin-3β slows replication in the presence of both MP-IV-1 and QQ-437 but does not kill tachyzoites. B. Location of insertional mutation at bp 1320 in the *T.gondii* Adaptin 3β. Alignment with *Neospora caninum* and *Arabidopsis thaliana* is shown.

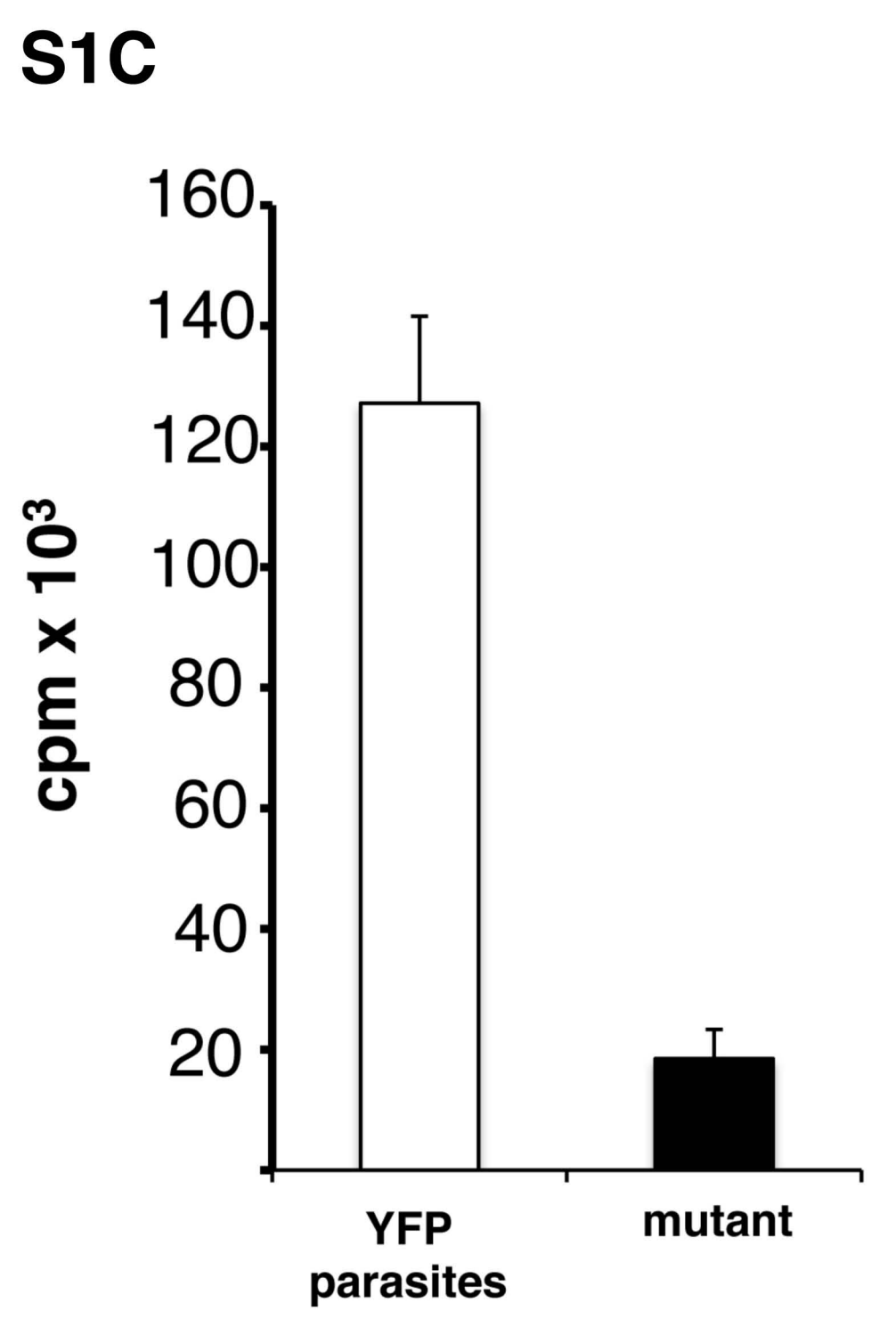

Figure S1. Insertional mutagenesis into Adaptin-3β slows replication in the presence of both MP-IV-1 and QQ-437 but does not kill tachyzoites. C. These insertional mutants grow slowly in the presence of both MP-IV-1 and QQ-437 but do form plaques (data not shown). Mean numbers of parasites per vacuole(8-16 in wild type cultures and 1-2 in mutant cultures) and comparison of replication of parasites in cultures infected with comparable inocula of 2000 wild type or 2000 mutant parasites demonstrate that replication is substantially less in the mutant compared with the wild type parasites, measured as fluorescence as CPM uptake of tritiated uracil in image C.

S2A

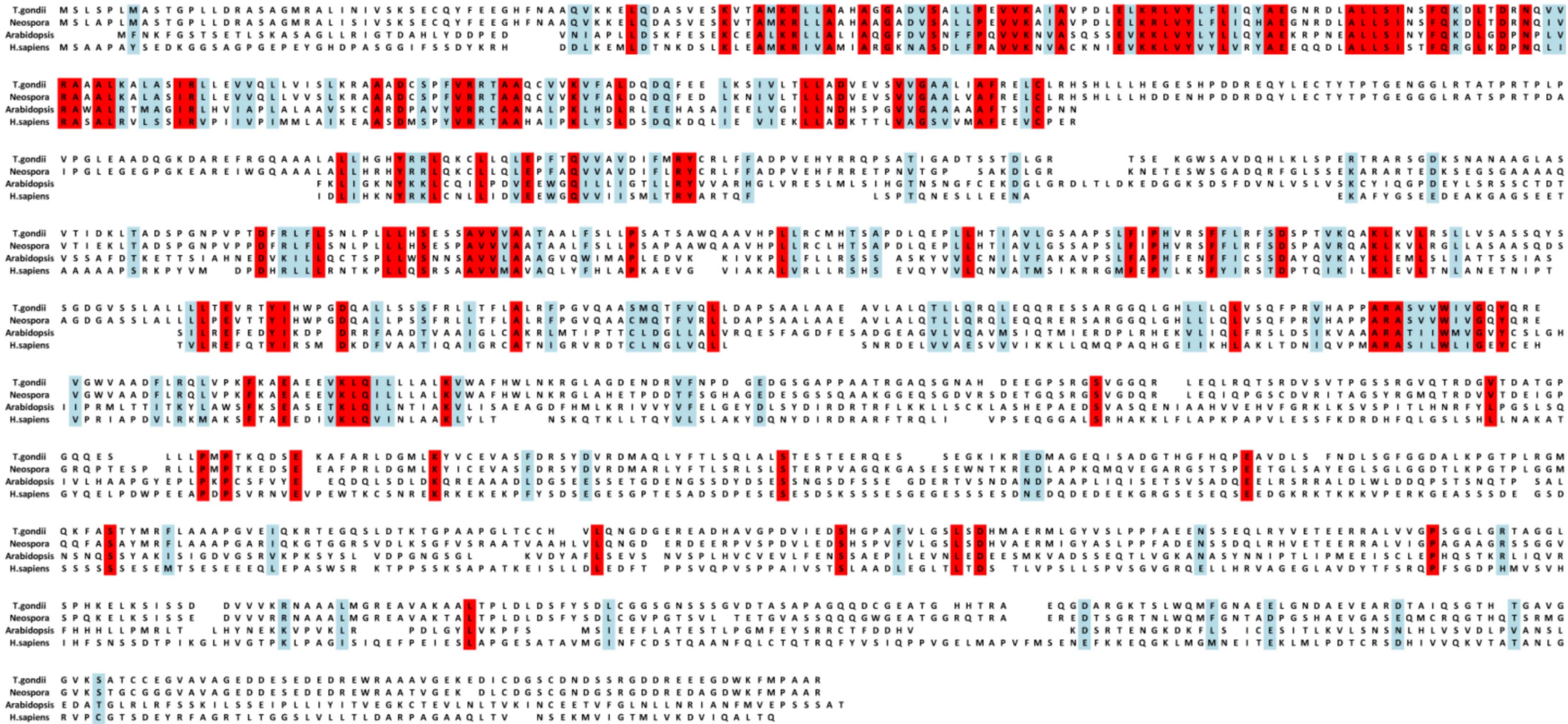

Figure S2. Multi-sequence Aligments of Adaptin-3βs. Figure shows similarities and differences of *T.gondii* Adaptin-3β to that of *Neospora caninum, Arabidopsis, Homo sapiens, Plasmodium yoeli, Leishmania*, and *Trypanosomes* with separate alignments of different species together**. A.** *T. gondii, Neospora caninum, Arabidopsis* and *Homo sapiens*.

S2B

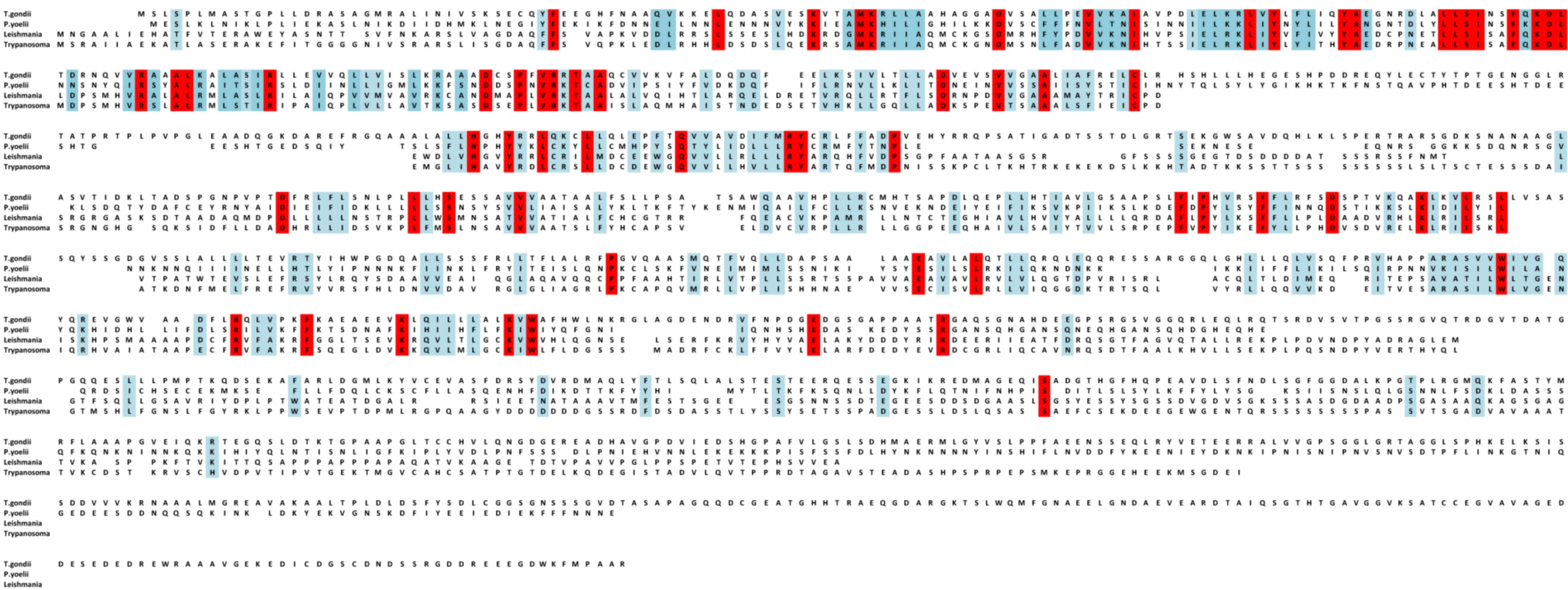

Figure S2. Multi-sequence Aligments of Adaptin-3βs. Figure shows similarities and differences of *T.gondii* Adaptin-3β to that of *Neospora caninum, Arabidopsis, Homo sapiens, Plasmodium yoeli, Leishmania*, and *Trypanosomes* with separate alignments of different species together**. B.** *T. gondii, Plasmodium yoelli. Leishmania donovani, Trypanosoma cruzii*.

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