SUPPLEMENTAL INFORMATION

1 **Supplemental Table S1. Predicted apicoplast localization.**

		PlasmoAP Prediction					PlasMit Prediction	
Protein ID	**Species**	**Signal Peptide Value**	**Decision**	**Apicoplast Targeting Peptide Value**	**Decision**	**Targeted to the Apicoplast**	**Jury**	**Strict**
PF14_0061	*Plasmodium falciparum 3D7*	3/4	+	5/5	++	YES	99% non - mito	n/a
PFTANZ_05018	*Plasmodium falciparum Tanzania*	3/4	+	5/5	++	YES	99% non - mito	n/a
PFFVO_04677	*Plasmodium falciparum Vietnam Oak-Knoll*	3/4	+	5/5	++	YES	99% non - mito	n/a
PRCDC_1405700	*Plasmodium reichenowi*	2/4	0	5/5	++	NO?	99% non - mito	n/a
PKH_13050	*Plasmodium knowlesi strain H*	4/4	++	5/5	++	YES	99% non - mito	n/a
PVX_086110	*Plasmodium vivax Sal1*	3/4	+	3/5	-	NO?	mito 91%	failed
C922_03803	*Plasmodium inui San Antonio I*	0/4	-	2/5	-	NO	99% non-mito	n/a
PYO3222	*Plasmodium yoelii 17XNL*	3/4	+	5/5	++	YES	99% non-mito	n/a
PY17X_1038200	*Plasmodium yoelii 17X*	3/4	+	5/5	++	YES	99% non-mito	n/a
YYE_02192	*Plasmodium vinckei*	4/4	++	5/5	++	YES	99% non-mito	n/a
PBANKA_103580	*Plasmodium berghei ANKA*	4/4	++	5/5	++	YES	99% non-mito	n/a
PCHAS_103660	*Plasmodium chaubadi*	4/4	++	5/5	++	YES	99% non-mito	n/a
PCYB_135000	*Plasmodium cynmolgi Strain B*	0/4	-	0/5	-	NO	99% non-mito	n/a
PFIT_1407400	*Plasmodium falciparum IT*	3/4	+	5/5	++	YES	99% non-mito	n/a

Supplemental Table S2. Conditions trialled for detection of PfPPR1 *in P. falciparum 3D7* lysate by Western Blot

Problem	**Conditions trialled**
Transfer efficiency	iBLOT (dry transfer), wet transfer, temperature of wet transfer. Checked transfer efficiency by staining the membrane with Ponceau and by staining the gel post-transfer with coomassie blue. Wet transfer at room temperature best.
Background in western blot images	Tested preimmune and postimmune serum, background also present in preimmune serum
Blocking solution to decrease background	1, 2 and 5% (w/v) BSA and 5 and 10% (w/v) non-fat milk powder tested. 10% skim milk powder lowest background.
Increase specificity	Included 0.5 M NaCl in 1xTBS + tween washes. Did not make any difference.
Primary antibody detection	Purified primary antibody. Compared different dilutions of purified and non-purified primary antibody (1:200, 1:500, 1:1000, 1:2000). Decided upon 1:1000 dilution
Secondary antibody detection	Tested for specificity of anti-rabbit AlexaFluor-568 antibody by performing a western blot on purified PPR and plasmodium lysate with no primary antibody. No non-specific binding observed.

Supplemental Table S3. Primers used in this study

Primer	Sequence
clpC FWD	CCTTTATATGGAGCTCGT
clpC FWD + T7	GCCATAATACGACTCACTATAGCCTTTATATGGAGCTCGT
clpC REV	CTGATATTTCTTTTAATTTTATTC
KDPPR-Fwd	GTCTGACAGTCTGTCTCTATGGTGCGCCCGTTGTCGAGACAACT CTGTC
KDPPR-Rev	GGAAAGAGAAGAGAGAAGGACGGCGCATGCTAGCAGATCTGG TTGAAGAC
LSU rRNA + T7 FWD	GCCATAATACGACTCACTATAG GTGTATAATTCCTAATAAGTTGA
LSU rRNA FWD	GTGTATAATTCCTAATAAGTTGA
LSUrRNA + T7 FWD	GCCATAATACGACTCACTATAGGTGTATAATTCCTAATAAGTT GA
LSUrRNA FWD	GTGTATAATTCCTAATAAGTTGA
PF11_0264 minus leader + T7 FWD	GCCATAATACGACTCACTATAGAAGGACACACATATTATTGAT
PF11_0264 minus leader FWD	AAGGACACACATATTATTGATAAAAAG
PF11_0264 REV	TCAGCTAAAAAAGTAAAGACTATCC
PF14_0061 minus leader +T7 FWD	GCCATAATACGACTCACTATAGGACAACTTTCTGAACG
PF14_0061 minus leader FWD	GACAACTTTCTGAACG
PF14_0061 REV	ATAGTTTTCGGTTTCGG
PF14_0061_Nterm Fwd	AAGTTCTGTTTCAGGGCCCGGACAACTTTCTGAACG
PF14_0061_Nterm Rev	ATGGTCTAGAAAGCTTTAATAGTTTTCGGTTTCGG
pGEM FWD	TGCAAGGCGATTAAGTTGGGT
pGEM REV	TGTGGAATTGTGAGCGGATAAC
rpl2 REV	TCCACCTCCTTTATTATAAATAG
rpoB REV	GAATACATGTTTTATATAATCC
rps2 REV	TATGAACCAATTATTTTAGGTA
tufA + T7 FWD	GCCATAATACGACTCACTATAGAATGTACAAAAAGTAGCTATA CCT
tufA FWD	AATGTACAAAAAGTAGCTATACCT

A

Plasmodium

The sequence name: **5866954**
Per-protein P-value for being PPR: **2.0E-42**
Probability for being PPR: **100.00%**

Repeat	Begin	Alignment	End	P-value
PPR	167	-LAFNAAMSAVEKKGCLSTMLDLIGTMKSKNIKPDL--	201	2.6e-08
PPR	202	-VSYKLVLSLCDKYHLVDTAEILFEEMIESDKINPN--	236	6.3e-09
PPR	238	-EIYAIMISCYAKTGNGYKAIELFEKLRNDPFVEEM--	272	1.5e-09
PPR	338	-SEYANVIYACNISNLYEQGIKYFEELLKSGKYMPS--	372	4.0e-07
PPR	374	-FVFENIFDLLSKNGDYEKSLEYYNNLKNDPNFKKN--	408	2.5e-06
PPR	412	-NILNNLLKALSIHNKINVAEDIWNNEFDELLLTPN--	446	1.5e-04
PPR	448	-LSYQILLKIYSHIDNYEKAFKLFKEMQVNKLLNNK--	482	1.0e-09
PPR	485	-LPFIYTIESTKNCGIYNYAIYVLRVAKLLNFKAND--	519	1.0e-02
PPR	522	-MLYNNTMISCINSKKYDVIISLYAELINMQQKDTS--	556	1.1e-03
PPR	562	-NTLTFVLLAFKELNMKQDFINLKNIIIQRNYKLPP--	596	5.0e-04

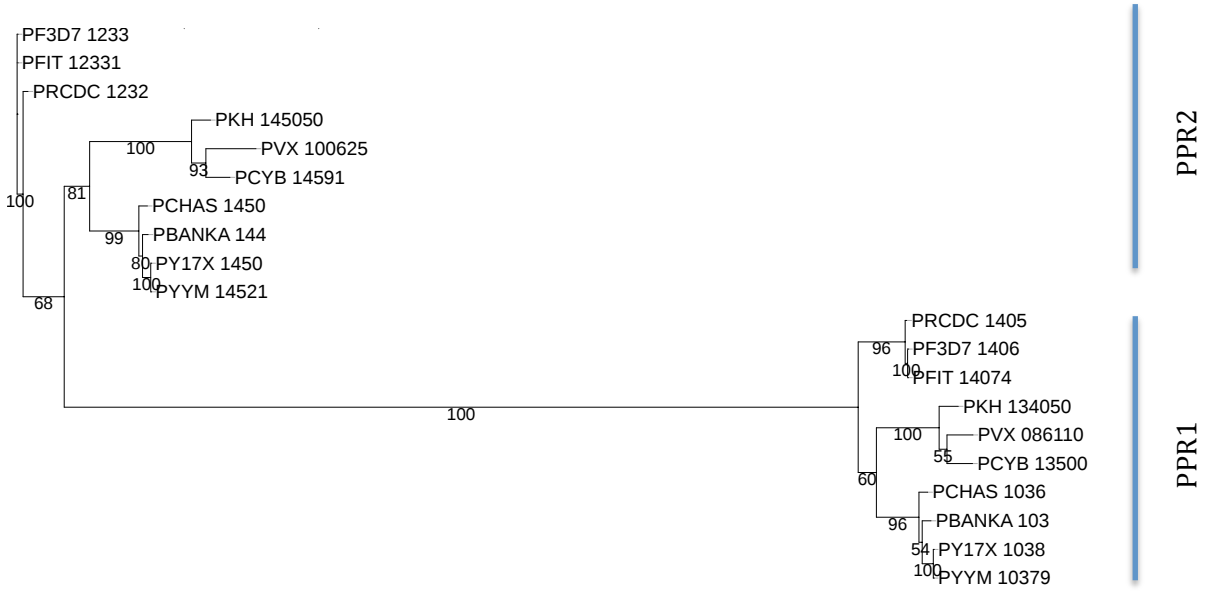
If you use TPRpred for your research, please cite:

> A Completely Reimplemented MPI Bioinformatics Toolkit with a New HHpred Server at its Core.
> Zimmermann L, Stephens A, Nam SZ, Rau D, Kübler J, Lozajic M, Gabler F, Söding J, Lupas AN, Alva V. J Mol Biol. 2018 Jul 20. S0022-2836(17)30587-9.

The sequence name: **Toxoplasma**
Per-protein P-value for being PPR: **1.3E-19**
Probability for being PPR: **100.00%**

Repeat	Begin	Alignment	End	P-value
PPR	336	-AAYNAALHACERQRDRPGALRIYAAMREKEIPIDV--	370	2.5e-09
PPR	371	-VTLHSLFTLLEAFADDTALLQILAQVDSADPESSM--	405	3.7e-03
PPR	411	-SPSSSLSPSPRSDSQASPTVSVTPSLLSLGISTCC--	445	3.0e-01
PPR	454	-VQLMERLKVLLRRNADKFLLNFTSVLPPDTADATA--	488	6.6e-01
PPR	496	-GVYVQLVVALTQEGRYEEALAYYEELKSLQRRFVE--	530	5.0e-07
PPR	585	-GAVNAALEACLYTGRLKQALLIYKEDVEFPEKQRL--	619	4.3e-03
PPR	646	-RACSQQRQAFALAQIWRDFEELQATVAETQNASPF--	680	8.4e-02
PPR	687	-PCVASAIQGFAACGLWTYALRLLLLLHKETGDARQ--	721	3.5e-05
PPR	726	-LLSDLLRDAREAAERSGAARETQSKMRSENEGERS--	760	7.2e-02
PPR	792	-EPYLAVLRACRDVGAWKPALGILRLLQQRHTQAKL--	826	6.4e-04

B



Supplemental Figure S1. PPR proteins in *Plasmodium* and *Toxoplasma.* A. PPR predictions for *Plasmodium falciparum* and *Toxoplasma gondii* PPR1. B. PPR1 and PPR2 proteins across the *Plasmodium* species. Mature (i.e. without targeting sequence) PPR1 (apicoplast) and PPR2 (mitochondrial) were aligned across selected *Plasmodium* species using ClustalW, and a phylogenetic tree inferred using PhyML.

Key:
PF3D7: *P. falciparum 3D7* (PF14_0061 and PF3D7_1233300),
PFIT: *P. falciparum* IT (PFIT_1407400 and PfIT_120038300),
PRCDC: *P. reichenowi*, (PRCDC_1232600 and PRCDC_1405700),
PKH: *P. knowlesi* strain H (PKH_13050 and PKNH_1452800),
PVH: *P. vivax (*PVX_086110 and PVX_100625),
PCYB: *P. cynmolgi* Strain B (PCYB_135000 and PCYB_145910),
PBANKA: *P.berghei* ANKA (PBANKA_103580 and PBANKA_1448000),
PY17X *P. yoelii yoelii* 17X (PY17X_1038200 and PY17X_1450500),
PYYM: *P. yoelii yoelii* YM (PYYM_1037900 and PYYM_1452100).

Supplemental Figure 2. Alignment of PPR1 proteins from *Plasmodium*, *Toxoplasma* and PPR10 from *Arabidopsis thaliana*. *Plasmodium* species as in Supplemental Figure 1.

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AthPPR10            ------------------------------------------------------------   0
Toxoplasma          HFAFASFEKAKGSEKCEPAKTPRPTVETADGEPLRRDPADPSSGSARVCGVHTSGAPLAT   60
PVX_086110          ------------------------------------------------------------   0
PKH_134050          ------------------------------------------------------------   0
C922_03803          ------------------------------------------------------------   0
PCYB_135000         ------------------------------------------------------------   0
PRCDC_1405700       ------------------------------------------------------------   0
PF3D7_1406400       ------------------------------------------------------------   0
PFFVO_05018         ------------------------------------------------------------   0
PFIT_1407400        ------------------------------------------------------------   0
PFTANZ_05018        ------------------------------------------------------------   0
PCHAS_103660        ------------------------------------------------------------   0
PY17X_1038200       ------------------------------------------------------------   0
PYO3222             ------------------------------------------------------------   0
PYYM_1037900        ------------------------------------------------------------   0
PBANKA_103580       ------------------------------------------------------------   0

AthPPR10            ------------------------------------------------------------   0
Toxoplasma          VPKANNESCEGKRHFRFRTNDEEGNHGNELEGRFEEPEEPEEPDRAAEGQEKRRRENSGK   120
PVX_086110          ------------------------------------------------------------   0
PKH_134050          ------------------------------------------------------------   0
C922_03803          ------------------------------------------------------------   0
PCYB_135000         ------------------------------------------------------------   0
PRCDC_1405700       ------------------------------------------------------------   0
PF3D7_1406400       ------------------------------------------------------------   0
PFFVO_05018         ------------------------------------------------------------   0
PFIT_1407400        ------------------------------------------------------------   0
PFTANZ_05018        ------------------------------------------------------------   0
PCHAS_103660        ------------------------------------------------------------   0
PY17X_1038200       ------------------------------------------------------------   0
PYO3222             ------------------------------------------------------------   0
PYYM_1037900        ------------------------------------------------------------   0
PBANKA_103580       ------------------------------------------------------------   0

AthPPR10            ------------------------------------------------------------   0
Toxoplasma          TPGRATGKRKSLNGYHASWLASAFNRHQASHIHSLRVSSFRSPLSSFLSSSHSSSLALCS   180
PVX_086110          ------------------------------------------------------------   0
PKH_134050          ------------------------------------------------------------   0
C922_03803          ------------------------------------------------------------   0
PCYB_135000         ------------------------------------------------------------   0
PRCDC_1405700       ------------------------------------------------------------   0
PF3D7_1406400       ------------------------------------------------------------   0
PFFVO_05018         ------------------------------------------------------------   0
PFIT_1407400        ------------------------------------------------------------   0
PFTANZ_05018        ------------------------------------------------------------   0
PCHAS_103660        ------------------------------------------------------------   0
PY17X_1038200       ------------------------------------------------------------   0
PYO3222             ------------------------------------------------------------   0
PYYM_1037900        ------------------------------------------------------------   0
PBANKA_103580       ------------------------------------------------------------   0
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AthPPR10          ------------------------------------------------------ASRRDELLRA   10
Toxoplasma        SSFSPFASSLLSPSPLFLNPASSASGRRVSLSNFLSSPSSSSPSVSASSSCSSSSSLLRA   240
PVX_086110        ------------------------------------------------------------   0
PKH_134050        ------------------------------------------------------------   0
C922_03803        ------------------------------------------------------------   0
PCYB_135000       ------------------------------------------------------------   0
PRCDC_1405700     ------------------------------------------------------------   0
PF3D7_1406400     ------------------------------------------------------------   0
PFFVO_05018       ------------------------------------------------------------   0
PFIT_1407400      ------------------------------------------------------------   0
PFTANZ_05018      ------------------------------------------------------------   0
PCHAS_103660      ------------------------------------------------------------   0
PY17X_1038200     ------------------------------------------------------------   0
PYO3222           ------------------------------------------------------------   0
PYYM_1037900      ------------------------------------------------------------   0
PBANKA_103580     ------------------------------------------------------------   0

AthPPR10          D-------------------------------------ITSLLKALELSGHWEWALA   30
Toxoplasma        LVACHSSPQTASVSLSPPRGSSFSSLSPSVLLRRSETRTQAPTLQAPLLSVRGS------   294
PVX_086110        ----------------------------------------------DSPPDGD------   7
PKH_134050        ----------------------------------------------AAIHDED------   7
C922_03803        ----------------------------------------------TATPDED------   7
PCYB_135000       ----------------------------------------------GATPDED------   7
PRCDC_1405700     ----------------------------------------------DNFLNEN------   7
PF3D7_1406400     ----------------------------------------------DNFLNEN------   7
PFFVO_05018       ----------------------------------------------DNFLNEN------   7
PFIT_1407400      ----------------------------------------------DNFLNEN------   7
PFTANZ_05018      ----------------------------------------------DNFLNEN------   7
PCHAS_103660      ----------------------------------------------NQNDDS------   6
PY17X_1038200     ----------------------------------------------NKNDDN------   6
PYO3222           ----------------------------------------------NKNDDN------   6
PYYM_1037900      ----------------------------------------------NKNDDN------   6
PBANKA_103580     ----------------------------------------------NRNDD-------   5

AthPPR10          LLRWAGKEGAADASALEMVVRALGREGQHDAVCALLDETPLPPGSRLDVRAYTTVLHALS   90
Toxoplasma        ---WPPDAGDDRET--EKRLSPLRI--DYRQAFE-LLENADPTGIKALTAAYNAALHACE   346
PVX_086110        ---ILKRKGAEKEV--EKSVVPLNM--DWVKVMN-LIYAS--RDVDATTLAFNAAMSAVE   57
PKH_134050        ---ILRKKGAQKEV--EKSVVPLNM--DWVKVMN-LIYAS--RDVDATTLAFNAAMSAVE   57
C922_03803        ---ILKKKGAQKEV--EQSVVPLNM--DWVKVMN-LIYAS--RDVDATTLAFNAAMSAVE   57
PCYB_135000       ---ILKKKGAQKEV--EKSVVPLNM--DWVKVMN-LIYAS--RDVDATTLAFNAAMSAVE   57
PRCDC_1405700     ---ILKKKSSEKEI--EQSLTPLNM--DWVKVMN-LIYSS--NDIDATTLAFNAAMSAVE   57
PF3D7_1406400     ---ILKKKSSEKEI--EQSLTPLNM--DWVKVMN-LIYSS--NDIDATTLAFNAAMSAVE   57
PFFVO_05018       ---ILKKKSSEKEI--EQSLTPLNM--DWVKVMN-LIYSS--NDIDATTLAFNAAMSAVE   57
PFIT_1407400      ---ILKKKSSEKEI--EQSLTPLNM--DWVKVMN-LIYSS--NDIDATTLAFNAAMSAVE   57
PFTANZ_05018      ---ILKKKSSEKEI--EQSLTPLNM--DWVKVMN-LIYSS--NDIDATTLAFNAAMSAVE   57
PCHAS_103660      ---ILKKKHLEKEI--EQSVIPLNM--DWVKVMN-LISSS--SDVNTTTLAFNAALSAVE   56
PY17X_1038200     ---ILKKKHSEKQI--EQSIIPLNI--DWIKVMN-LISSS--NDVNTTTLAFNAALSAVE   56
PYO3222           ---ILKKKHSEKQI--EQSIIPLNI--DWIKVMN-LISSS--NDVNTTTLAFNAALSAVE   56
PYYM_1037900      ---ILKKKHSEKQI--EQSIIPLNI--DWIKVMN-LISSS--NDVNTTTLAFNAALSAVE   56
PBANKA_103580     ---ILKKKYSEKEI--EQSIIPLNM--DWVKVMN-LISAS--NDVNTTTLAFNAALSAVE   55
                                 *  :  *    :   .   *       .    . *:.:.: * .

AthPPR10          RAGRYERALELFAELRRQGVAPTLVTYNVVLDVYGRMGRSWPRIVALLDEMR-AAGVEPD   149
Toxoplasma        RQRDRPGALRIYAAMREKEIPIDVVTLHSLFTLLEAFADDTA----LLQILAQVDSADPE   402
PVX_086110        KKGCLKSMLEVFEIMKKKNIKPDLVSYKLLLRLCANYHLGEHAEI-LFDEMVETDKLTPT   116
PKH_134050        KKGCLKSMLELFEVMKKKNIKPDLVSYKLLLRLCANYHLGDHAEI-LFDEMVETDKLTPT   116
C922_03803        KKGCLKSMLELFEIMKKKNIKPDLVSYKLLLRLCANYHLGEHAEI-LFDEMVETEKLTPT   116
PCYB_135000       KKGCLKSMLELFEIMKKKNIKPDLVSYKLLLRLCANYHLGEHAEI-LFDEMVETDKLTPT   116
PRCDC_1405700     KKGCLTTMLDLIGTMKSKNIKPDLVSYKLVLSLCDKYHLVDTAEI-LFEEMIESDKINPN   116
PF3D7_1406400     KKGCLSTMLDLIGTMKSKNIKPDLVSYKLVLSLCDKYHLVDTAEI-LFEEMIESDKINPN   116
PFFVO_05018       KKGCLSTMLDLIGTMKSKNIKPDLVSYKLVLSLCDKYHLVDTAEI-LFEEMIESDKINPN   116
PFIT_1407400      KKGCLSTMLDLIGTMKSKNIKPDLVSYKLVLSLCDKYHLVDTAEI-LFEEMIESDKINPN   116
PFTANZ_05018      KKGCLSTMLDLIGTMKSKNIKPDLVSYKLVLSLCDKYHLVDTAEI-LFEEMIESDKINPN   116
PCHAS_103660      KKGCLTSIIELFEIMKKKNIKPDLISYKLILTLCDKYHLAEYAEI-LFDEMTESDNIRPN   115
PY17X_1038200     KKGCLTSIIELFEIMKKKNIKPDLISYKLILSLCDKYHLAEYAEI-LFDEMVESDNIRPS   115
PYO3222           KKGCLTSIIELFEIMKKKNIKPDLISYKLILSLCDKYHLAEYAEI-LFDEMVESDNIRPS   115
PYYM_1037900      KKGCLTSIIELFEIMKKKNIKPDLISYKLILSLCDKYHLAEYAEI-LFDEMVESDNIRPS   115
PBANKA_103580     KKRCLTSIIELFEIMKKKNIKPDLISYKLILTLCDKYHLADYAEI-LFNEMVESDNIRPN   114
                  :       : :   :: : :   ::: : :: :            *:: :       *
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AthPPR10          --------------------------------GFTASTVIAACCRDGLVDEAVAFFEDLKA   178
Toxoplasma        SSMAFLSRSPSSSLSPSPRSDSQASPTVSVTPSLLSLGISTCCRAGNATAAVQLMERLKV   462
PVX_086110        --------------------------------YEIYALMISCFAKVGDGHRAVEFVEKLRS   145
PKH_134050        --------------------------------YEIYALMINCFAKVGDGHKAVEFLEKLRS   145
C922_03803        --------------------------------YEIYALMISCFAKVGDGHKAVEFLEKLRS   145
PCYB_135000       --------------------------------YEIYALMISCFAKVGDGHKAVEFVEKLRS   145
PRCDC_1405700     --------------------------------YEIYAIMISCYAKTGNGYKAIELFEKLRN   145
PF3D7_1406400     --------------------------------YEIYAIMISCYAKTGNGYKAIELFEKLRN   145
PFFVO_05018       --------------------------------YEIYAIMISCYAKTGNGYKAIELFEKLRN   145
PFIT_1407400      --------------------------------YEIYAIMISCYAKTGNGYKAIELFEKLRN   145
PFTANZ_05018      --------------------------------YEIYAIMISCYAKTGNGYKAIELFEKLRN   145
PCHAS_103660      --------------------------------YEIYSIMISCFSKVGDGHKAIEFLEKLRN   144
PY17X_1038200     --------------------------------YEIYSIMISCFSKVGDGHKAIEFLEKLRN   144
PYO3222           --------------------------------YEIYSIMISCFSKVGDGHKAIEFLEKLRN   144
PYYM_1037900      --------------------------------YEIYSIMISCFSKVGDGHKAIEFLEKLRN   144
PBANKA_103580     --------------------------------YEIYSIMISCFSKVGDGYKASEFLEKLRN   143
                                                     :  *   .: *    *  :.* *:

AthPPR10          RG---HAPCVVTYNALLQVFGKAGNYTEALRV--LGEMEQNG-------------------   215
Toxoplasma        LLRRNADKFLLNFTSVLPPDTADA-------------------------------------   486
PVX_086110        D------PLVGEVNNWG-EAGG----------------------VSGSGSADD---------   169
PKH_134050        D------PLVEGVNNWEVDGSGDGNGSAGRNVSNDGNVSNDGNVSDDGNVSDDGNVNRGE   199
C922_03803        D------PLVEEVNNWG-DRSGD---------------------TADRGDTAD------RGD   173
PCYB_135000       D------PLVEEVNNWGGDSSGVGDSSGG------GDSSGGGDRSGAGDSSGDG---DGS   190
PRCDC_1405700     D------PLVEEMRSLNITNSNDN--------------------KENSNDLET---------   172
PF3D7_1406400     D------PFVEEMRSLNITNTNDN--------------------KENSNDLQT---------   172
PFFVO_05018       D------PFVEEMRSLNITNTNDN--------------------KENSNDLQT---------   172
PFIT_1407400      D------PFVEEMRSLNITNTNDN--------------------KENSNDLQT---------   172
PFTANZ_05018      D------PFVEEMRSLNITNTNDN--------------------KENSNDLQT---------   172
PCHAS_103660      D------PFVENFKEL---TSKYD--------------------KEKSNRWED---------   168
PY17X_1038200     D------PFVENIKEL---DSKYD--------------------KEKSNRWEN---------   168
PYO3222           D------PFVENIKEL---DSKYD--------------------KEKSNRWEN---------   168
PYYM_1037900      D------PFVENIKEL---DSKYD--------------------KEKSNRWEN---------   168
PBANKA_103580     D------PFVENIKEL---NSKYD--------------------KEKSNRWDN---------   167
                         :

AthPPR10          ---------------------------------------------CQPDAVTYNELAGTYA-   231
Toxoplasma        ------------------------------------------------------------   486
PVX_086110        RQHPAWRDSFAEEATTEEGAEAEGSTYTNQFKEITKKIKHIERGSS-KIQYSEYANVIFA   228
PKH_134050        RGNYLWKDHFDEQAATEEDAKAEGNAYTNQFKDITKKIKHIEKGSS-KIQYSEYTNVIFA   258
C922_03803        GGNPAWKDAFAEEAPTEEGAKAEGKTYTNEFKEITKKIKQIEKGSS-KIQYSEYTNVIFA   232
PCYB_135000       GENRVWKDAFAEEAATEEGAKEEGNNYTNQFKEITKKIKHIEKGSS-KIQFSEYTNVIFA   249
PRCDC_1405700     --SIIHNNIEDNNNN-NNNN---NNIYDDKFKHISNKIKNVENYSG-KIQYSEYANVIYA   225
PF3D7_1406400     --SIIHNNMEDNNNN-NNNN--DNNIYDDKFKHISNKIKNVENCSG-KIQYSEYANVIYA   226
PFFVO_05018       --SIIHNNMEDNNNN-NNNNNNDNNIYDDKFKHISNKIKNVENCSG-KIQYSEYANVIYA   228
PFIT_1407400      --SIIHNNMEDNNNN-NNNNNNDNNIYDDKFKHISNKIKNVENCSG-KIQYSEYANVIYA   228
PFTANZ_05018      --SIIHNNMEDNNNN-NNNNN-DNNIYDDKFKHISNKIKNVENCSG-KIQYSEYANVIYA   227
PCHAS_103660      --TFAQVNS-------NNGEDNTPNYYNAQFKELTEKIKNVENNNN-KIQYSEYANVIFA   218
PY17X_1038200     --TFAQVNSKNDDNKINNGEDDSPNYYNTQFKELTEKIKNVENNNNNKIQYSEYANVIFA   226
PYO3222           --TFAQVNSKNDDNKINNGEDDSPNYYNTQFKELTEKIKNVENNNNNKIQYSEYANVIFA   226
PYYM_1037900      --TFAQVNSKNDDNKINNGEDDSPNYYNTQFKELTEKIKNVENNNNNKIQYSEYANVIFA   226
PBANKA_103580     --TFAQVKSKSEDNKINHGEDDSPNYYNTQFKELTEKIKNVENNN--KIQYSEYANVIFA   223

AthPPR10          --RAGFFEEAARCLD-TMASKGLLPNAFTYNTVMTAYGNVGKVDEALALFDQMKKTGFVP   288
Toxoplasma        ------------------TAAVAHPPTGVYVQLVVALTQEGRYEEALAYYEELKSLQRRF   528
PVX_086110        CNMSNLPEQGIKYFEELLNTGKYMPSTLLLESIFDLLAKNGNYEKCLDYYNKLKDDPNFK   288
PKH_134050        CNMSNLPEQGIKYFEELLNTGKYMPSTLLLESIFDLLAKNGKYEKCLDYYNKLKDDPNFK   318
C922_03803        CNMSNLPEQGIKYFEELLNTGKYMPSTLLLESIFDLLAKNGNYEKCLEYYNKLKDDPNFK   292
PCYB_135000       CNMSNLPEQGIKYFEELLNTGKYMPSTLLLESIFDLLAKNGKYEKSLEYYNKLKDDPNFK   309
PRCDC_1405700     CNISNLYEQGIKYFEELLKSGKYMPSIFVFENIFDLLSKNGDYEKSLEYYNNLKNDPNFK   285
PF3D7_1406400     CNISNLYEQGIKYFEELLKSGKYMPSIFVFENIFDLLSKNGDYEKSLEYYNNLKNDPNFK   286
PFFVO_05018       CNISNLYEQGIKYFEELLKSGKYMPSIFVFENIFDLLSKNGDYEKSLEYYNNLKNDPNFK   288
PFIT_1407400      CNISNLYEQGIKYFEELLKSGKYMPSIFVFENIFDLLSKNGDYEKSLEYYNNLKNDPNFK   288
PFTANZ_05018      CNISNLYEQGIKYFEELLKSGKYMPSIFVFENIFDLLSKNGDYEKSLEYYNNLKNDPNFK   287
PCHAS_103660      CNISNLHEQGIKYFEELLNSTKYIPSTFIFENIFDLLGKNGNYEKALDYYNKIKDDPNFK   278
PY17X_1038200     CNMSNLHEHGIKYFEELLNSTKYIPSTFIFENIFNLLGKNGNYEKALDYYNKIKDDPNFK   286
PYO3222           CNMSNLHEHGIKYFEELLNSTKYIPSTFIFENIFNLLGKNGNYEKALDYYNKIKDDPNFK   286
PYYM_1037900      CNMSNLHEHGIKYFEELLNSTKYIPSTFIFENIFNLLGKNGNYEKALDYYNKIKDDPNFK   286
PBANKA_103580     CNISNLHEHGIKYFEELLNSTKYIPSTFIFENIFDLLGKNGNYEKALDYYNKIKDDPNFK   283
                                  :    *      :.    : *  ::.*  ::::*.
```

```
AthPPR10          NVNTYNLVLGMLGKKSRF----------TVMLEMLGEMSRSGCTPNRVTWNTMLAVCGKR   338
Toxoplasma        VEK-QQLLEREVARRAELVERELKNAPEQERKKLLGDIERQV-----------------   569
PVX_086110        -KA-I-------------------------------------------------------   291
PKH_134050        -KY-I-------------------------------------------------------   321
C922_03803        -KY-I-------------------------------------------------------   295
PCYB_135000       -KY-I-------------------------------------------------------   312
PRCDC_1405700     -KN-I-------------------------------------------------------   288
PF3D7_1406400     -KN-I-------------------------------------------------------   289
PFFVO_05018       -KN-I-------------------------------------------------------   291
PFIT_1407400      -KN-I-------------------------------------------------------   291
PFTANZ_05018      -KN-I-------------------------------------------------------   290
PCHAS_103660      -KY-I-------------------------------------------------------   281
PY17X_1038200     -KY-I-------------------------------------------------------   289
PYO3222           -KY-I-------------------------------------------------------   289
PYYM_1037900      -KY-I-------------------------------------------------------   289
PBANKA_103580     -KY-I-------------------------------------------------------   286

AthPPR10          GMEDYVTRVLEGMRSCGVELSRDTYNTLIAAYGRCGSRTNAFKMYNEMTSAGFTPCITTY   398
Toxoplasma        ------------------------------------------VEEQELLLEDYQPPIGAV   587
PVX_086110        ----------------------------------------------------NVNIL   296
PKH_134050        ----------------------------------------------------NVNIL   326
C922_03803        ----------------------------------------------------NVNIL   300
PCYB_135000       ----------------------------------------------------NVNIL   317
PRCDC_1405700     ----------------------------------------------------NVNIL   293
PF3D7_1406400     ----------------------------------------------------NVNIL   294
PFFVO_05018       ----------------------------------------------------NVNIL   296
PFIT_1407400      ----------------------------------------------------NVNIL   296
PFTANZ_05018      ----------------------------------------------------NVNIL   295
PCHAS_103660      ----------------------------------------------------NVNIL   286
PY17X_1038200     ----------------------------------------------------NVNIL   294
PYO3222           ----------------------------------------------------NVNIL   294
PYYM_1037900      ----------------------------------------------------NVNIL   294
PBANKA_103580     ----------------------------------------------------NVNIL   291
                                                                         :

AthPPR10          NALLNVLSRQGDWSTAQSIVSKMRTKGFKPNEQSYSLLLQCYAKGGNVAGIAAIENEVYG   458
Toxoplasma        NAALEACLYTGRLKQALLIYK-------------------------------------   608
PVX_086110        NNILKALSVQGKINIIEDLWK-------------------------------------   317
PKH_134050        NNILKALCVHGKINIIEQVWK-------------------------------------   347
C922_03803        NNILKALSVHAKSNIIEDVWK-------------------------------------   321
PCYB_135000       NNILKALTVHGKINIIEQVWK-------------------------------------   338
PRCDC_1405700     NNLLKTLSIHNKINVAEDIWN-------------------------------------   314
PF3D7_1406400     NNLLKALSIHNKINVAEDIWN-------------------------------------   315
PFFVO_05018       NNLLKALSIHNKINVAEDIWN-------------------------------------   317
PFIT_1407400      NNLLKALSIHNKINVAEDIWN-------------------------------------   317
PFTANZ_05018      NNLLKALSIHNKINVAEDIWN-------------------------------------   316
PCHAS_103660      NNLLKSLSLSNKINIIENIWN-------------------------------------   307
PY17X_1038200     NNILKSLSLSNKINIIENIWN-------------------------------------   315
PYO3222           NNILKSLSLSNKINIIENIWN-------------------------------------   315
PYYM_1037900      NNILKSLSLSNKINIIENIWN-------------------------------------   315
PBANKA_103580     NNILKSLSLSNKINIIENIWN-------------------------------------   312
                  *  *:        .    : .

AthPPR10          SGAVFPSWVILRTLVIANFKCRRLDGMETAFQEVKARGYNPDLVIFNSMLSIYAKNGMYS   518
Toxoplasma        EDVEFPEKQRL-----------------------------------------------R   620
PVX_086110        N-------------------------------------------------------------   318
PKH_134050        N-------------------------------------------------------------   348
C922_03803        N-------------------------------------------------------------   322
PCYB_135000       N-------------------------------------------------------------   339
PRCDC_1405700     N-------------------------------------------------------------   315
PF3D7_1406400     N-------------------------------------------------------------   316
PFFVO_05018       N-------------------------------------------------------------   318
PFIT_1407400      N-------------------------------------------------------------   318
PFTANZ_05018      N-------------------------------------------------------------   317
PCHAS_103660      N-------------------------------------------------------------   308
PY17X_1038200     N-------------------------------------------------------------   316
PYO3222           N-------------------------------------------------------------   316
PYYM_1037900      N-------------------------------------------------------------   316
PBANKA_103580     N-------------------------------------------------------------   313
                  .
```

```
AthPPR10          KATEVFDSIKRSGLSPDLITYNSLMDMYAKCSESWEAEKILNQLKCSQTMK----PDVVS   574
Toxoplasma        AGEALDQDLAPSKAAPTLRTFELLLRACSQQRQAFALAQIWRDFEELQAT-VAETQNASP   679
PVX_086110        -------EFDELMLTPNEISYGYMLKVYSIVDD---YEKAFKLFKEMQMKKMLSNKNIIP   368
PKH_134050        -------EFDELMLTPNAISYAYMLNAYSIVDD---YEKAFKLFKEMQMKKMLNNKNIIP   398
C922_03803        -------EFDDLMLTPNAISYAYMLNVYSIMDD---YEKAFKLFKEMQMKKLLNNKNIIP   372
PCYB_135000       -------EFDDLMLTPNAISYGFMLNAYSIVDD---YEKAFKLFKEMHMKKLLNKKNIIP   389
PRCDC_1405700     -------EFDELLLTPNNLSYQILLKIYSHIDN---YEKAFKLFKEMQINKLLNNKNILP   365
PF3D7_1406400     -------EFDELLLTPNNLSYQILLKIYSHIDN---YEKAFKLFKEMQVNKLLNNKNILP   366
PFFVO_05018       -------EFDELLLTPNNLSYQILLKIYSHIDN---YEKAFKLFKEMQVNKLLNNKNILP   368
PFIT_1407400      -------EFDELLLTPNNLSYQILLKIYSHIDN---YEKAFKLFKEMQVNKLLNNKNILP   368
PFTANZ_05018      -------EFDELLLTPNNLSYQILLKIYSHIDN---YEKAFKLFKEMQVNKLLNNKNILP   367
PCHAS_103660      -------EYDELLLTHNSVSYQIMLNVYGNIDD---YEKAFKLFKEMQIKKMLNKKNILP   358
PY17X_1038200     -------EYDELLLVQNSVSYQIMLNVYSNIDD---YEKAFKLFKEMQMKKMLNKKNILP   366
PYO3222           -------EYDELLLVQNSVSYQIMLNVYSNIDD---YEKAFKLFKEMQMKKMLNKKNILP   366
PYYM_1037900      -------EYDELLLVQNSVSYQIMLNVYSNIDD---YEKAFKLFKEMQMKKMLNKKNILP   366
PBANKA_103580     -------EYDELLLTQNSVSYQIMLNVYSNIDD---YEKSFKLFKEMQMKKMLNKKNILP   363
                            .          ::  ::   .   :    :  . ::  :       :

AthPPR10          Y---------NTVINGFCKQGLVKEAQRVLS-----------------EMVADGMAPCA   607
Toxoplasma        FRLPSAAPCVASAIQGFAACGLWTYALRLLLLLHKETGDARQRHCELLSDLLRDAREAAE   739
PVX_086110        F---------VYTINAFKNCGIYNYAIYVLRVAKLLGV-SSED----LLKLYNDAMVACV   414
PKH_134050        F---------VYTIHAFKNCGIYNYAIYVLRVAKLLNV-SSED----LLKLYNDAMVACI   444
C922_03803        F---------VYTINAFKNCGIYNYAIYVLRVAKLLNV-SSED----LLKLYNDAMIACI   418
PCYB_135000       F---------VYTINAFKNCGIYNYAIYVLRVAKLLDV-FSED----LLKLYNNAMIACV   435
PRCDC_1405700     F---------IYTIESTKNCGIYNYAIYVLRVAKLLNF-KAND----LLMLYNNTMISCI   411
PF3D7_1406400     F---------IYTIESTKNCGIYNYAIYVLRVAKLLNF-KAND----LLMLYNNTMISCI   412
PFFVO_05018       F---------IYTIESTKNCGIYNYAIYVLRVAKLLNF-KAND----LLMLYNNTMISCI   414
PFIT_1407400      F---------IYTIESTKNCGIYNYAIYVLRVAKLLNF-KAND----LLMLYNNTMISCI   414
PFTANZ_05018      F---------IYTIESTKNCGIYNYAIYVLRVAKLLNF-KAND----LLMLYNNTMISCI   413
PCHAS_103660      F---------VYVLNSFKNCGIYSYSIYVLRIAKLIGI-VGKD----LLFLYNNAMITCV   404
PY17X_1038200     F---------VYTINSFKNCGIYNYSIYVLRIAKLIGI-VGKD----LLFLYNNAMISCI   412
PYO3222           F---------VYTINSFKNCGIYNYSIYVLRIAKLIGI-VGKD----LLFLYNNAMISCI   412
PYYM_1037900      F---------VYTINSFKNCGIYNYSIYVLRIAKLIGI-VGKD----LLFLYNNAMISCI   412
PBANKA_103580     F---------VYTINSFKNCGIYNYSIYVLRIAKLIGI-VGKN----ILFLYNNAMIACI   409
                  :          .:..    *: . :  :*                   :  :    .

AthPPR10          V-----------------------------------------------------TYHTLVG   615
Toxoplasma        RSGAARETQSKMRSENEGERSSEQKDGDARAAASESGDWERCLEGRFVDLGVEPYLAVLR   799
PVX_086110        NAK---KYDVVI--SLYAELITMQEKGAPS-----------------LEISISTLGFVLL   452
PKH_134050        NSK---KYEVVI--SLYAELVTMQEKGAPS-----------------LEINISTLGFVLL   482
C922_03803        SCK---KYDVVI--SLYAELITMQEKGAPS-----------------LEINISTLGFVLL   456
PCYB_135000       NCK---KYDVVI--SLYAELITMQEKGAPS-----------------LEINISTLGFVLL   473
PRCDC_1405700     NSK---KYDVII--SLYAELINMQQK-DTS-----------------FQININTLTFVLL   448
PF3D7_1406400     NSK---KYDVII--SLYAELINMQQK-DTS-----------------FQININTLTFVLL   449
PFFVO_05018       NSK---KYDVII--SLYAELINMQQK-DTS-----------------FQININTLTFVLL   451
PFIT_1407400      NSK---KYDVII--SLYAELINMQQK-DTS-----------------FQININTLTFVLL   451
PFTANZ_05018      NSK---KYDVII--SLYAELINMQQK-DTS-----------------FQININTLTFVLL   450
PCHAS_103660      NAK---KYDVII--SLYTELIALQEK-DTS-----------------LTININTLSFVLL   441
PY17X_1038200     NAK---KYDVII--SLYTELIALQEK-DTS-----------------LTININTLSFVLL   449
PYO3222           NAK---KYDVII--SLYTELIALQEK-DTS-----------------LTININTLSFVLL   449
PYYM_1037900      NAK---KYDVII--SLYTELIALQEK-DTS-----------------LTININTLSFVLL   449
PBANKA_103580     NAK---KYDVII--SLYTELIALQEK-DTS-----------------LIININTLSFVLL   446
                                                                          ::

AthPPR10          GYSSLEMFSEAREVIGYMVQHGLKPMELTYRRVV---------------------ESYCRA   655
Toxoplasma        ACRDVGAWKPALGILRLLQQRHTQAKLLHALGAARERREAERREQFQQRVRALRRRLSEA   859
PVX_086110        AFRELDMREDFTNLKNLIIQKNYKLTPL---------------------------------   480
PKH_134050        AFKELNMRDDFINLKNLIIQKNYKLTPL---------------------------------   510
C922_03803        AFKELNMREDFTNLKNLIIQKNYKLTPL---------------------------------   484
PCYB_135000       AFKELNMREDFTNLKNLIIQKNYKLTPL---------------------------------   501
PRCDC_1405700     AFKELNMKQDFINLKNIIIQRNYKLPPL---------------------------------   476
PF3D7_1406400     AFKELNMKQDFINLKNIIIQRNYKLPPL---------------------------------   477
PFFVO_05018       AFKELNMKQDFINLKNIIIQRNYKLPPL---------------------------------   479
PFIT_1407400      AFKELNMKQDFINLKNIIIQRNYKLPPL---------------------------------   479
PFTANZ_05018      AFKELNMKQDFINLKNIIIQRNYKLPPL---------------------------------   478
PCHAS_103660      AFKELKMKEDFSNLKNLILQKNYKLTPL---------------------------------   469
PY17X_1038200     AFKELKMKEDFLNLKNIILQKNYKLTPL---------------------------------   477
PYO3222           AFKELKMKEDFLNLKNIILQKNYKLTPL---------------------------------   477
PYYM_1037900      AFKELKMKEDFLNLKNIILQKNYKLTPL---------------------------------   477
PBANKA_103580     AFKELKMKEDFSNLKNLILQKNYKLTPL---------------------------------   474
                  .  .:   .    :   : *:  :   *
```

```
AthPPR10          KRFEEARGFLSEVSETDLD----------FDKKALEAYIE----DAQFGR----------   691
Toxoplasma        KRKTNGADCVGEAAEQQRPTSASPTGDSDPLLSRDMERFLGSLHNSVAVGRTPLLPEFPY   919
PVX_086110        --------CGQLVSEQPH----------------------------------------   490
PKH_134050        --------CGKLVNEQQND---------------------------------------   521
C922_03803        --------CGKLVNEEPEW---------------------------------------   495
PCYB_135000       --------CAKLVSEESDS---------------------------------------   512
PRCDC_1405700     --------CSKIFSETENY*--------------------------------------   487
PF3D7_1406400     --------CSKIFSETENY---------------------------------------   488
PFFVO_05018       --------CSKIFSETENY---------------------------------------   490
PFIT_1407400      --------CSKIFSETENY*--------------------------------------   490
PFTANZ_05018      --------CSKIFSETENY---------------------------------------   489
PCHAS_103660      --------CSKIINEPEDQ*--------------------------------------   480
PY17X_1038200     --------CSKVINEQES*---------------------------------------   487
PYO3222           --------CSKVINEQES----------------------------------------   487
PYYM_1037900      --------CSKVINEQES----------------------------------------   487
PBANKA_103580     --------CSKVINEP------------------------------------------   482
                          *

AthPPR10          ------------------------------------------------------------   691
Toxoplasma        EAYALTLGTMAAARAWDRVLAVSSEFFSRRDSGACGSASLGADGPAGEANEERKPARRAV   979
PVX_086110        ------------------------------------------------------------   490
PKH_134050        ------------------------------------------------------------   521
C922_03803        ------------------------------------------------------------   495
PCYB_135000       ------------------------------------------------------------   512
PRCDC_1405700     ------------------------------------------------------------   487
PF3D7_1406400     ------------------------------------------------------------   488
PFFVO_05018       ------------------------------------------------------------   490
PFIT_1407400      ------------------------------------------------------------   490
PFTANZ_05018      ------------------------------------------------------------   489
PCHAS_103660      ------------------------------------------------------------   480
PY17X_1038200     ------------------------------------------------------------   487
PYO3222           ------------------------------------------------------------   487
PYYM_1037900      ------------------------------------------------------------   487
PBANKA_103580     ------------------------------------------------------------   482

AthPPR10          ------------------------------------------------------------   691
Toxoplasma        GFGGERDEGGQPERGAEGERRDAPLEIRKSVHAYRLMALMHLGRHEEVEAERRSLIRLTE   1039
PVX_086110        ------------------------------------------------------------   490
PKH_134050        ------------------------------------------------------------   521
C922_03803        ------------------------------------------------------------   495
PCYB_135000       ------------------------------------------------------------   512
PRCDC_1405700     ------------------------------------------------------------   487
PF3D7_1406400     ------------------------------------------------------------   488
PFFVO_05018       ------------------------------------------------------------   490
PFIT_1407400      ------------------------------------------------------------   490
PFTANZ_05018      ------------------------------------------------------------   489
PCHAS_103660      ------------------------------------------------------------   480
PY17X_1038200     ------------------------------------------------------------   487
PYO3222           ------------------------------------------------------------   487
PYYM_1037900      ------------------------------------------------------------   487
PBANKA_103580     ------------------------------------------------------------   482

AthPPR10          ------------------------------------------------------------   691
Toxoplasma        MERRTRERRDARQREREGGEEKREEGEGSEEAFLPKWREETEHEEGEGGEDEGSHGIATL   1099
PVX_086110        ------------------------------------------------------------   490
PKH_134050        ------------------------------------------------------------   521
C922_03803        ------------------------------------------------------------   495
PCYB_135000       ------------------------------------------------------------   512
PRCDC_1405700     ------------------------------------------------------------   487
PF3D7_1406400     ------------------------------------------------------------   488
PFFVO_05018       ------------------------------------------------------------   490
PFIT_1407400      ------------------------------------------------------------   490
PFTANZ_05018      ------------------------------------------------------------   489
PCHAS_103660      ------------------------------------------------------------   480
PY17X_1038200     ------------------------------------------------------------   487
PYO3222           ------------------------------------------------------------   487
PYYM_1037900      ------------------------------------------------------------   487
PBANKA_103580     ------------------------------------------------------------   482
```

```
AthPPR10          ------------------------------------------------------------   691
Toxoplasma        VLEEAETWLMRGVTRERRTQGKDNIYPTPSIHETTPHTYEATHHSFPSPSKSSSASSSAS   1159
PVX_086110        ------------------------------------------------------------   490
PKH_134050        ------------------------------------------------------------   521
C922_03803        ------------------------------------------------------------   495
PCYB_135000       ------------------------------------------------------------   512
PRCDC_1405700     ------------------------------------------------------------   487
PF3D7_1406400     ------------------------------------------------------------   488
PFFVO_05018       ------------------------------------------------------------   490
PFIT_1407400      ------------------------------------------------------------   490
PFTANZ_05018      ------------------------------------------------------------   489
PCHAS_103660      ------------------------------------------------------------   480
PY17X_1038200     ------------------------------------------------------------   487
PYO3222           ------------------------------------------------------------   487
PYYM_1037900      ------------------------------------------------------------   487
PBANKA_103580     ------------------------------------------------------------   482

AthPPR10          ------------------------------------------------------------   691
Toxoplasma        SSASSSASSSASSSSFSSASSSSFSSSPLQPSFIAGSGGGRSMSLRDVRHLEGVSAMSEG   1219
PVX_086110        ------------------------------------------------------------   490
PKH_134050        ------------------------------------------------------------   521
C922_03803        ------------------------------------------------------------   495
PCYB_135000       ------------------------------------------------------------   512
PRCDC_1405700     ------------------------------------------------------------   487
PF3D7_1406400     ------------------------------------------------------------   488
PFFVO_05018       ------------------------------------------------------------   490
PFIT_1407400      ------------------------------------------------------------   490
PFTANZ_05018      ------------------------------------------------------------   489
PCHAS_103660      ------------------------------------------------------------   480
PY17X_1038200     ------------------------------------------------------------   487
PYO3222           ------------------------------------------------------------   487
PYYM_1037900      ------------------------------------------------------------   487
PBANKA_103580     ------------------------------------------------------------   482

AthPPR10          ------------------------------------------------------------   691
Toxoplasma        LKEGRGEETSVARRAEFLIGGGTKRKVCVQEEDRGDAVAEGEANHKSVCRDETGEEHSGR   1279
PVX_086110        ------------------------------------------------------------   490
PKH_134050        ------------------------------------------------------------   521
C922_03803        ------------------------------------------------------------   495
PCYB_135000       ------------------------------------------------------------   512
PRCDC_1405700     ------------------------------------------------------------   487
PF3D7_1406400     ------------------------------------------------------------   488
PFFVO_05018       ------------------------------------------------------------   490
PFIT_1407400      ------------------------------------------------------------   490
PFTANZ_05018      ------------------------------------------------------------   489
PCHAS_103660      ------------------------------------------------------------   480
PY17X_1038200     ------------------------------------------------------------   487
PYO3222           ------------------------------------------------------------   487
PYYM_1037900      ------------------------------------------------------------   487
PBANKA_103580     ------------------------------------------------------------   482

AthPPR10          ------------------------------------------------------------   691
Toxoplasma        SLDRFFAGLEPVSEREARAVAEAVERRRTEEHWRQEKMQSGVKRATGDREGMQPKRSADP   1339
PVX_086110        ------------------------------------------------------------   490
PKH_134050        ------------------------------------------------------------   521
C922_03803        ------------------------------------------------------------   495
PCYB_135000       ------------------------------------------------------------   512
PRCDC_1405700     ------------------------------------------------------------   487
PF3D7_1406400     ------------------------------------------------------------   488
PFFVO_05018       ------------------------------------------------------------   490
PFIT_1407400      ------------------------------------------------------------   490
PFTANZ_05018      ------------------------------------------------------------   489
PCHAS_103660      ------------------------------------------------------------   480
PY17X_1038200     ------------------------------------------------------------   487
PYO3222           ------------------------------------------------------------   487
PYYM_1037900      ------------------------------------------------------------   487
PBANKA_103580     ------------------------------------------------------------   482
```

```
AthPPR10          ------------------------------------------------------------   691
Toxoplasma        EGDKNDPGCQTAGNDPGGETGLRRREGERDGVNEEAGGREKGASGQIERE-REGEHDTEEA   1399
PVX_086110        ------------------------------------------------------------   490
PKH_134050        ------------------------------------------------------------   521
C922_03803        ------------------------------------------------------------   495
PCYB_135000       ------------------------------------------------------------   512
PRCDC_1405700     ------------------------------------------------------------   487
PF3D7_1406400     ------------------------------------------------------------   488
PFFVO_05018       ------------------------------------------------------------   490
PFIT_1407400      ------------------------------------------------------------   490
PFTANZ_05018      ------------------------------------------------------------   489
PCHAS_103660      ------------------------------------------------------------   480
PY17X_1038200     ------------------------------------------------------------   487
PYO3222           ------------------------------------------------------------   487
PYYM_1037900      ------------------------------------------------------------   487
PBANKA_103580     ------------------------------------------------------------   482

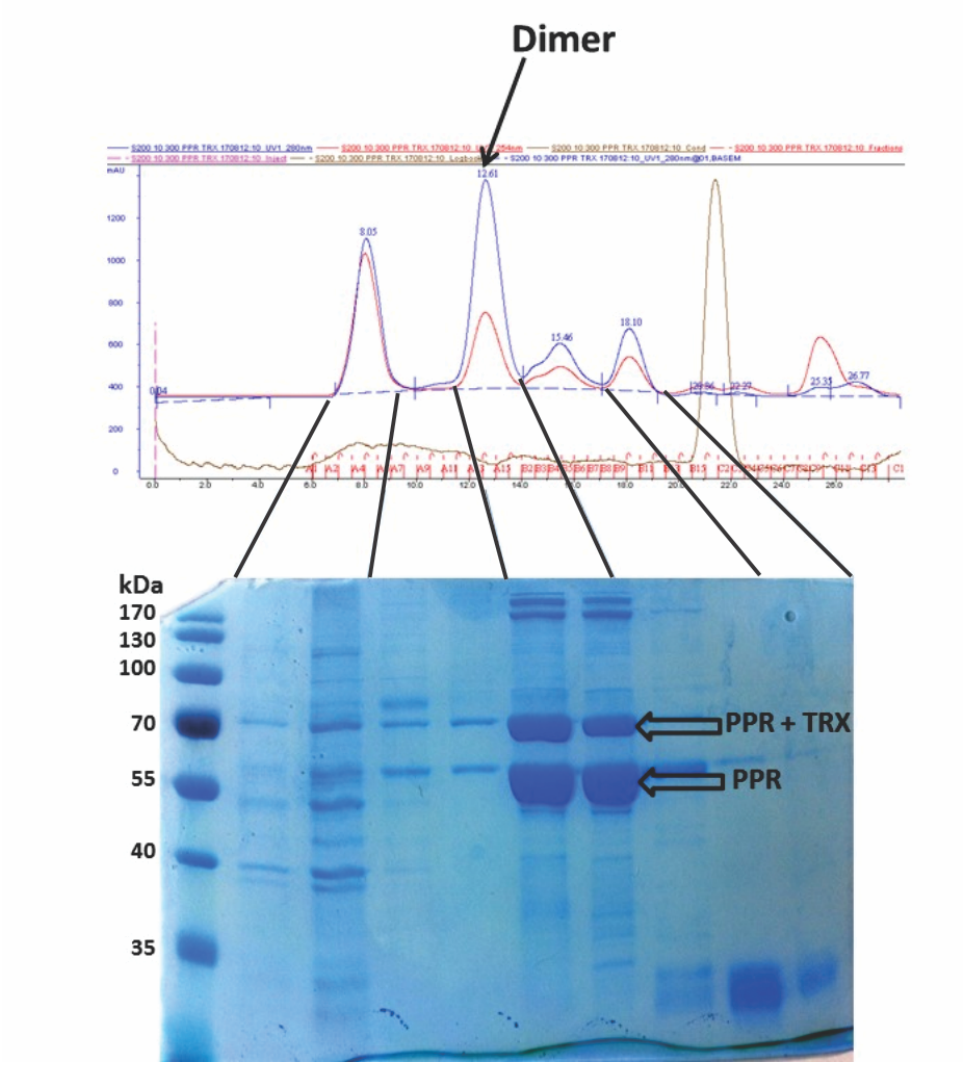
AthPPR10          ------------------------------------------------------------   691
Toxoplasma        RERLEEKEEEREKENSEREGRAIRAVSGERGLSVSAFLSSVLGPRRAVLESMREEPVLED   1459
PVX_086110        ------------------------------------------------------------   490
PKH_134050        ------------------------------------------------------------   521
C922_03803        ------------------------------------------------------------   495
PCYB_135000       ------------------------------------------------------------   512
PRCDC_1405700     ------------------------------------------------------------   487
PF3D7_1406400     ------------------------------------------------------------   488
PFFVO_05018       ------------------------------------------------------------   490
PFIT_1407400      ------------------------------------------------------------   490
PFTANZ_05018      ------------------------------------------------------------   489
PCHAS_103660      ------------------------------------------------------------   480
PY17X_1038200     ------------------------------------------------------------   487
PYO3222           ------------------------------------------------------------   487
PYYM_1037900      ------------------------------------------------------------   487
PBANKA_103580     ------------------------------------------------------------   482

AthPPR10          ------------------------------------------------------------   691
Toxoplasma        TVTEEPESLELHADAEAQLEAKQREGESRDRSGENTEASTERRNSPDREEEKQDPVTTAT   1519
PVX_086110        ------------------------------------------------------------   490
PKH_134050        ------------------------------------------------------------   521
C922_03803        ------------------------------------------------------------   495
PCYB_135000       ------------------------------------------------------------   512
PRCDC_1405700     ------------------------------------------------------------   487
PF3D7_1406400     ------------------------------------------------------------   488
PFFVO_05018       ------------------------------------------------------------   490
PFIT_1407400      ------------------------------------------------------------   490
PFTANZ_05018      ------------------------------------------------------------   489
PCHAS_103660      ------------------------------------------------------------   480
PY17X_1038200     ------------------------------------------------------------   487
PYO3222           ------------------------------------------------------------   487
PYYM_1037900      ------------------------------------------------------------   487
PBANKA_103580     ------------------------------------------------------------   482

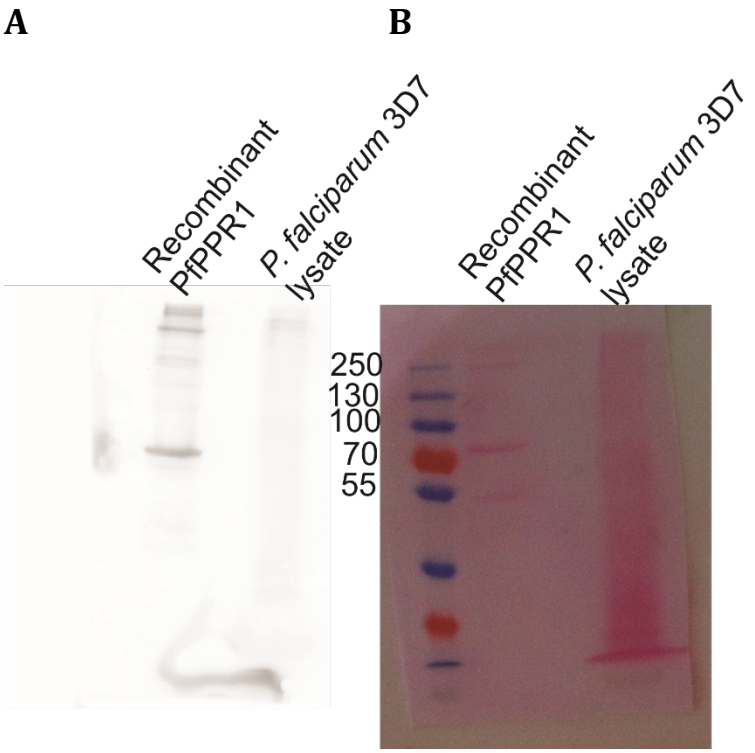
AthPPR10          ------------------------------------------------------------   691
Toxoplasma        QETDANTGGTLSQAGGRKSEAASRSESVETSLDLLLRLKHKTPAETQQLEDGPPRWLARR   1579
PVX_086110        ------------------------------------------------------------   490
PKH_134050        ------------------------------------------------------------   521
C922_03803        ------------------------------------------------------------   495
PCYB_135000       ------------------------------------------------------------   512
PRCDC_1405700     ------------------------------------------------------------   487
PF3D7_1406400     ------------------------------------------------------------   488
PFFVO_05018       ------------------------------------------------------------   490
PFIT_1407400      ------------------------------------------------------------   490
PFTANZ_05018      ------------------------------------------------------------   489
PCHAS_103660      ------------------------------------------------------------   480
PY17X_1038200     ------------------------------------------------------------   487
PYO3222           ------------------------------------------------------------   487
PYYM_1037900      ------------------------------------------------------------   487
PBANKA_103580     ------------------------------------------------------------   482
```

```
AthPPR10          ------------------------------------------------------------   691
Toxoplasma        SGTGEPGAEAVAGSRRGVHPPDGRAGVGIPGGGAGSQRQRQETRGQSAFLFAGDHGWGWP   1639
PVX_086110        ------------------------------------------------------------   490
PKH_134050        ------------------------------------------------------------   521
C922_03803        ------------------------------------------------------------   495
PCYB_135000       ------------------------------------------------------------   512
PRCDC_1405700     ------------------------------------------------------------   487
PF3D7_1406400     ------------------------------------------------------------   488
PFFVO_05018       ------------------------------------------------------------   490
PFIT_1407400      ------------------------------------------------------------   490
PFTANZ_05018      ------------------------------------------------------------   489
PCHAS_103660      ------------------------------------------------------------   480
PY17X_1038200     ------------------------------------------------------------   487
PYO3222           ------------------------------------------------------------   487
PYYM_1037900      ------------------------------------------------------------   487
PBANKA_103580     ------------------------------------------------------------   482

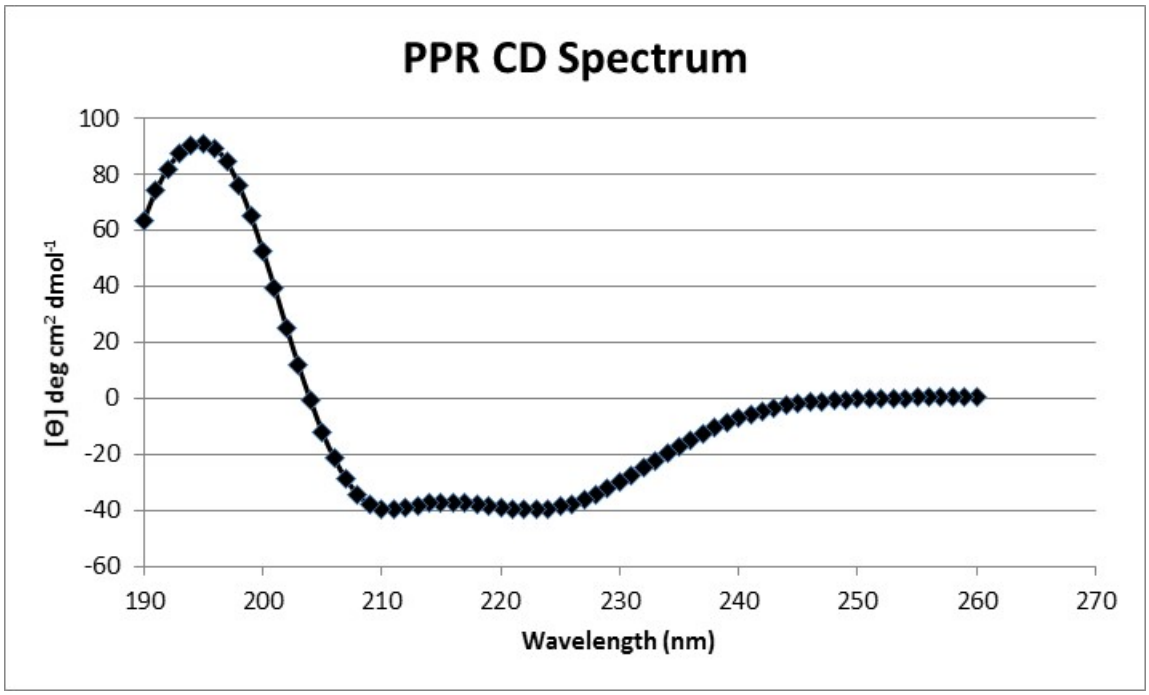
AthPPR10          ----------------------------------------  691
Toxoplasma        PHWEEKLDKARARLQRTNDESGETEENAKTETPWGMRPT  1678
PVX_086110        ----------------------------------------  490
PKH_134050        ----------------------------------------  521
C922_03803        ----------------------------------------  495
PCYB_135000       ----------------------------------------  512
PRCDC_1405700     ----------------------------------------  487
PF3D7_1406400     ----------------------------------------  488
PFFVO_05018       ----------------------------------------  490
PFIT_1407400      ----------------------------------------  490
PFTANZ_05018      ----------------------------------------  489
PCHAS_103660      ----------------------------------------  480
PY17X_1038200     ----------------------------------------  487
PYO3222           ----------------------------------------  487
PYYM_1037900      ----------------------------------------  487
PBANKA_103580     ----------------------------------------  482
```



Supplemental Figure S3. Gel filtration chromatography of His_6-TRX-PfPPR1 and corresponding SDS-PAGE gel. Blue line represents absorbance at 280 nm, red line absorbance at 260 nm and brown line conductivity. The trace for absorbance at 280 nm (blue line) shows four major peaks eluted from the S200 10/300 column. Based on calibration of the S200 10/300 column, the second peak corresponds to a molecular weight of approximately 135 kDa, and the estimated molecular weight of the His_6-TRX-*Pf*PPR1 dimer is 141.6 kDa. Spontaneous cleavage of the TRX-Histag is observed in the second peak, corresponding to PPR protein without the His_6-TRX tag (estimated molecular weight of 56.8 kDa) with a smaller amount in the third peak. The fourth peak corresponds to the His_6-TRX tag only (14 kDa).

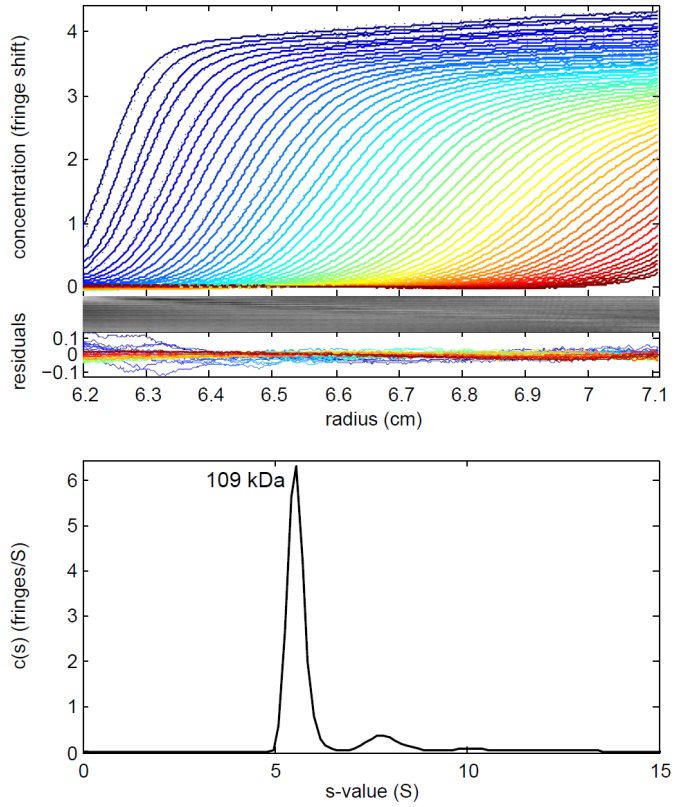


Supplemental Figure S4. Western blot of purified *Pf*PPR1 protein and *P. falciparum* 3D7 lysate using the purified polyclonal anti-*Pf*PPR1 antibody. Purified PfPPR1 protein was run on 10% SDS-PAGE gel along with *P. falciparum 3D7* cell lysate. After incubation with the purified *Pf*PPR1 polyclonal rabbit antibody and a secondary goat anti-rabbit antibody conjugated to HRP, no *Pf*PPR1 protein could be detected in the *P. falciparum* 3D7 lysate (Panel A).. The positive control (recombinant *Pf*PPR1) shows a band of the correct size (*Pf*PPR1 + TRX His_6 ~ 72.8 kDa) .The ponceau stained gel (Panel B) shows good transfer of proteins of all molecular weights. Size markers are shown in kDa.



Supplemental Figure S5. Circular Dichroism (CD) spectrum of purified *Pf*PPR1.

*Pf*PPR1 minus His-TRX tag at 25°C in 10 mM potassium phosphate pH 8.0, 50 mM Na fluoride. Spectrum is typical of that for an alpha-helical protein.

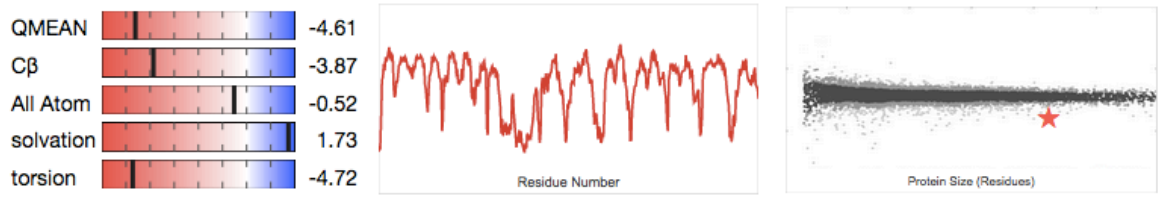


Supplemental Figure S6. Analytical ultracentrifugation (AUC) sedimentation velocity data for *Pf*PPR1 minus His_6-TRX tag. The residuals are from the fit with the continuous c(s) distribution model. Component sedimentation coefficient distribution for PPR at 1.8 mg/mL showing populations of dimeric (fitted mass of 109 kDa) and higher-order species, fitting to a uniform frictional ratio of Fk,w = 1.378. The r.m.s.d. was 0.016.

Supplementary Figure S7. SWISS-MODEL analysis of PPR1 from both *Toxoplasma* and *Plasmodium*.

Plasmodium

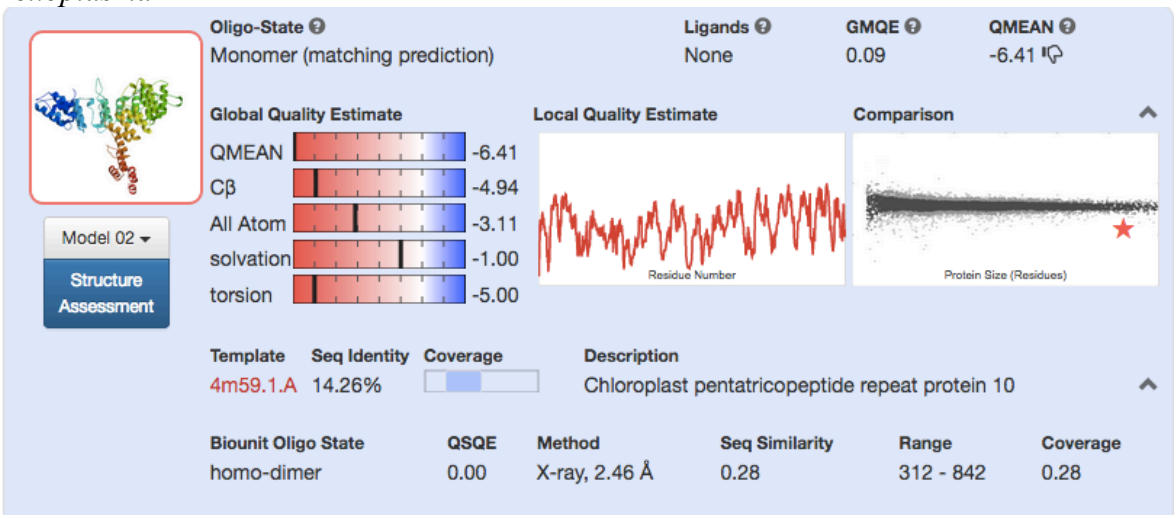
Model #01	File	Built with	Oligo-State	Ligands	GMQE	QMEAN
	PDB	ProMod3 Version 1.3.0.	monomer	None	0.39	-4.61



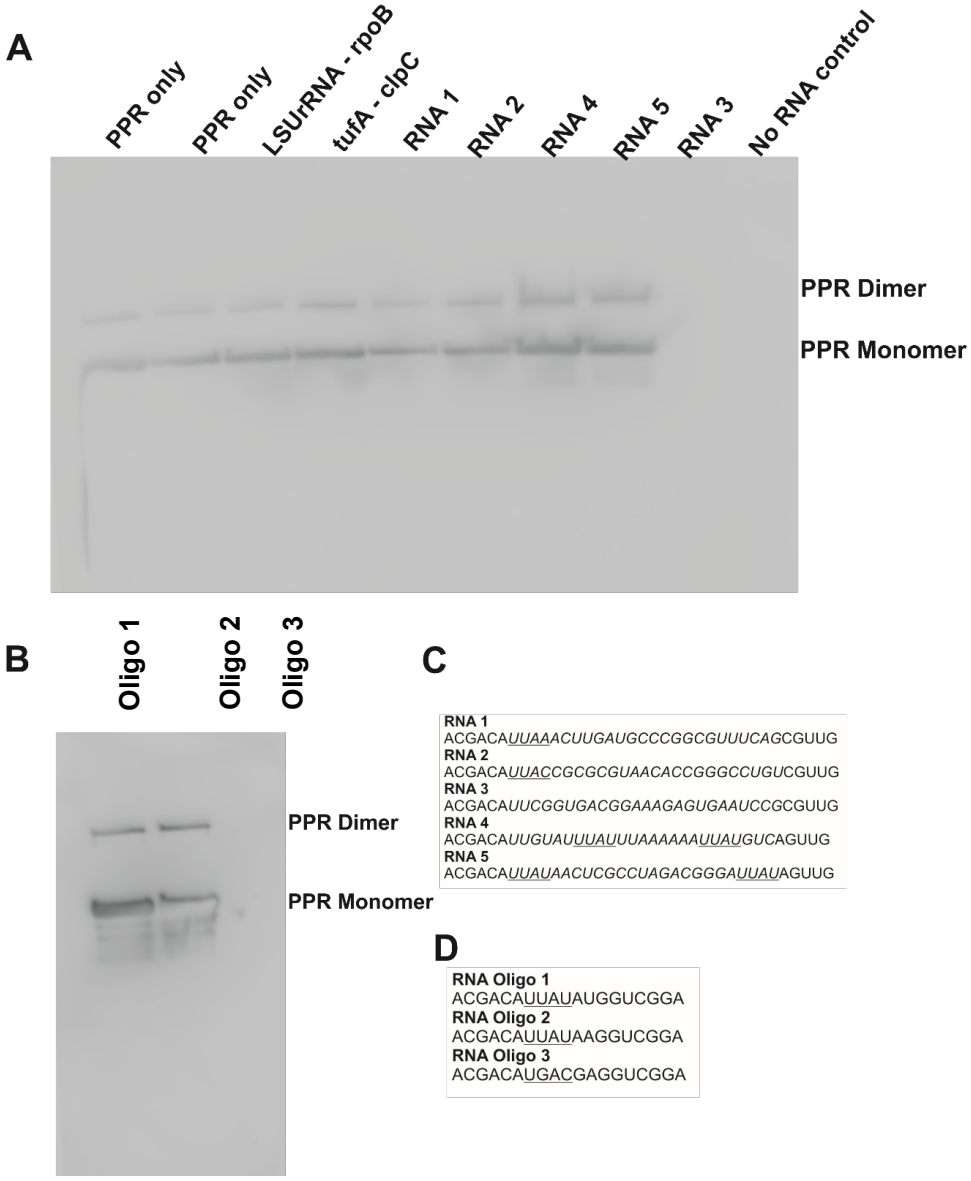
Template	Seq Identity	Oligo-state	Found by	Method	Resolution	Seq Similarity	Range	Coverage	Description
5i9f.1.A	20.00	monomer	HHblits	X-ray	2.19Å	0.31	160 - 584	0.63	pentatricopeptide repeat protein dPPR-U10

The template contained no ligands.

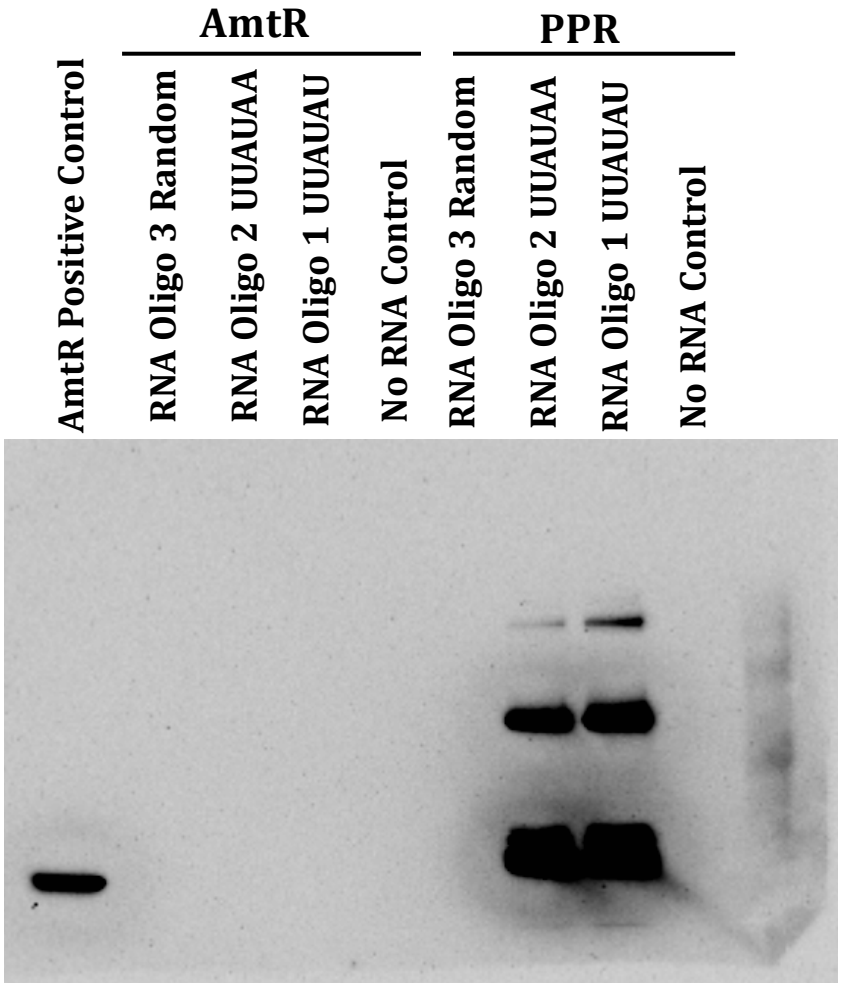
Toxoplasma



Supplemental Figure S8. *Pf*PPR1 – RNA pull down assays. (A) Five 150 nt RNA molecules (RNAs 1- 5, sequences as shown in Figure 3) and apicoplast RNA transcripts (*LSU rRNA – rpoB* and *tufA – clpC*) were used in a pull-down experiment. Biotinylated RNA was bound to streptavidin beads and used as 'bait' to pull down *Pf*PPR1 protein (with the TRX-His_6 tag removed). Bound *Pf*PPR1 was protein detected using a purified polyclonal anti-*Pf*PPR1 antibody from rabbit. Loading controls (PPR1 only) and a no RNA control reaction showed no non-specific binding in the absence of RNA. (B) The same pull down experiment using RNA oligonucleotides 1 – 3. The same result was obtained when the experiment was repeated (E). Details of RNA sequences are given in the legend to Figure 3, and specific sequences are shown (C and D).



E


AmtR
PPR
AmtR Positive Control
RNA Oligo 3 Random
RNA Oligo 2 UUAUAA
RNA Oligo 1 UUAUAU
No RNA Control
RNA Oligo 3 Random
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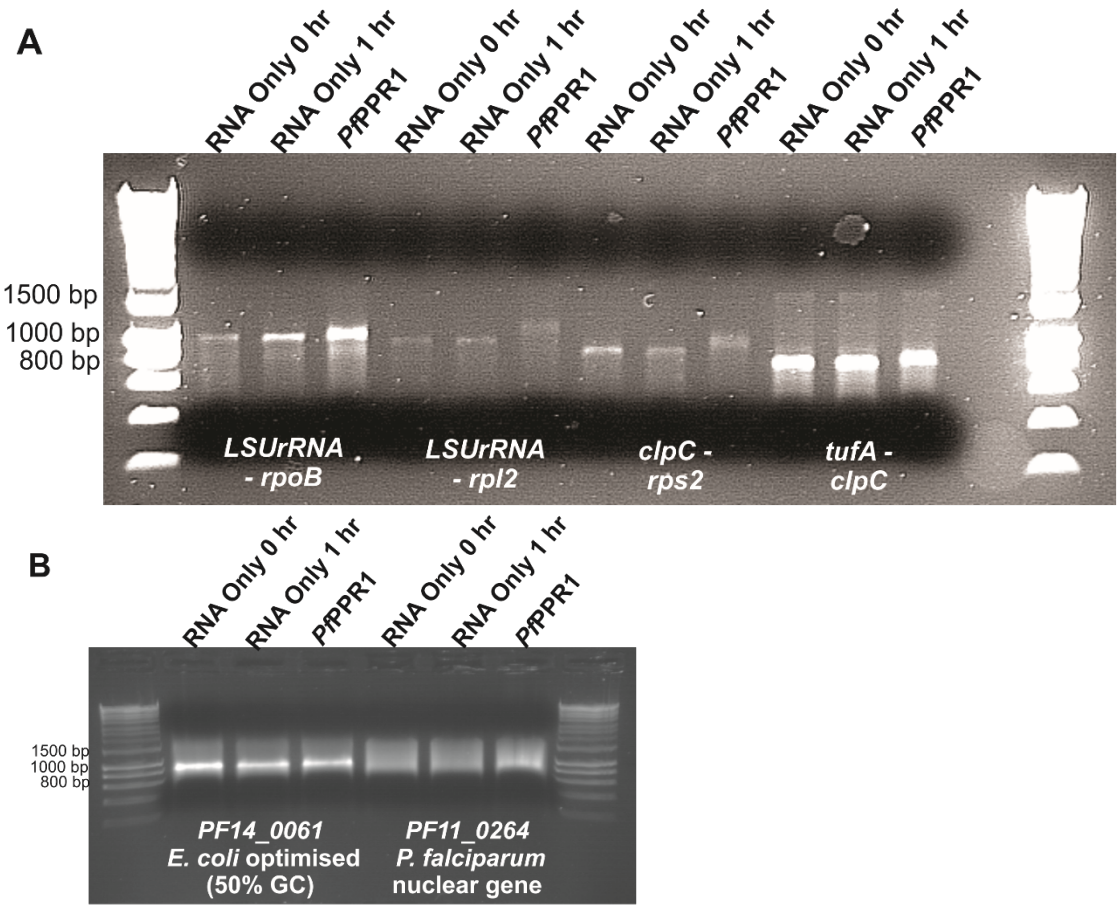
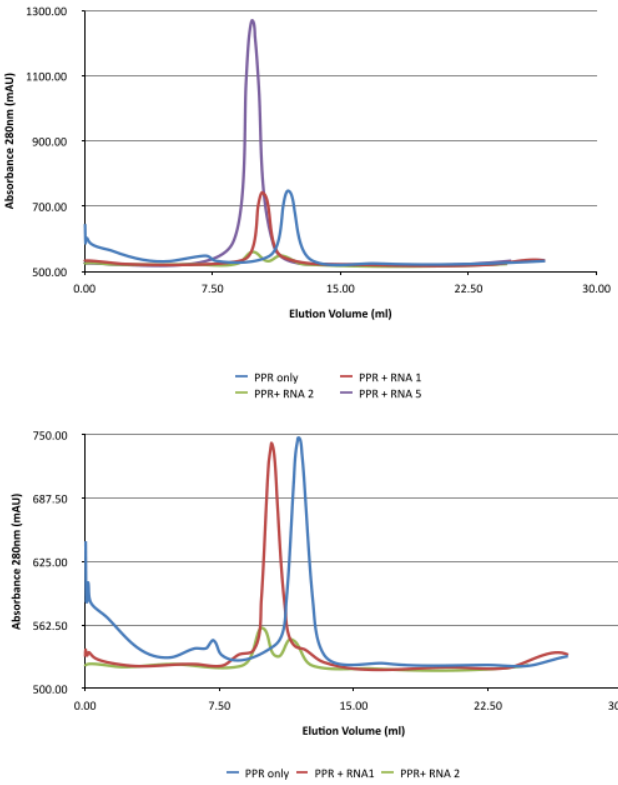
RNA Oligo 1 UUAUAU
No RNA Control



Supplemental Figure S9. *Pf*PPR1 shows specific binding to apicoplast RNA transcripts. *Pf*PPR1 binding to apicoplast RNA transcripts was tested in a gel shift experiment. Recombinant *Pf*PPR1 (minus TRX-His_6) causes a shift in the migration of *in vitro* transcribed apicoplast RNA molecules following incubation for one hour (Panel A). No shift is seen in the RNA if it is not bound to PPR1. Panel B shows that no shift is seen when PPR1 is incubated with *in vitro* transcribed RNA from a nuclear encoded *P. falciparum* gene (PF11_0264, or from an *E. coli* codon-optomized *P. falciparum* gene (PF14_0061) and a *P. falciparum 3D7* nuclear gene (PF11_0264).

Supplemental Figure S10 Gel filtration shows a change in elution profile when *Pf*PPR1 is bound to RNA. This is a repeat of the data shown in Figure 4, showing that the change in mobility following gel filtration is reproducible.



Expected Elution Volumes:
PPR TRX His_6 (143600 Da)
= 13.37 ml
RNA only (150 bases) 48750 Da
PPR + RNA
= 143600 Da + 48750 Da
= 192493.6 Da
= 12.76 ml

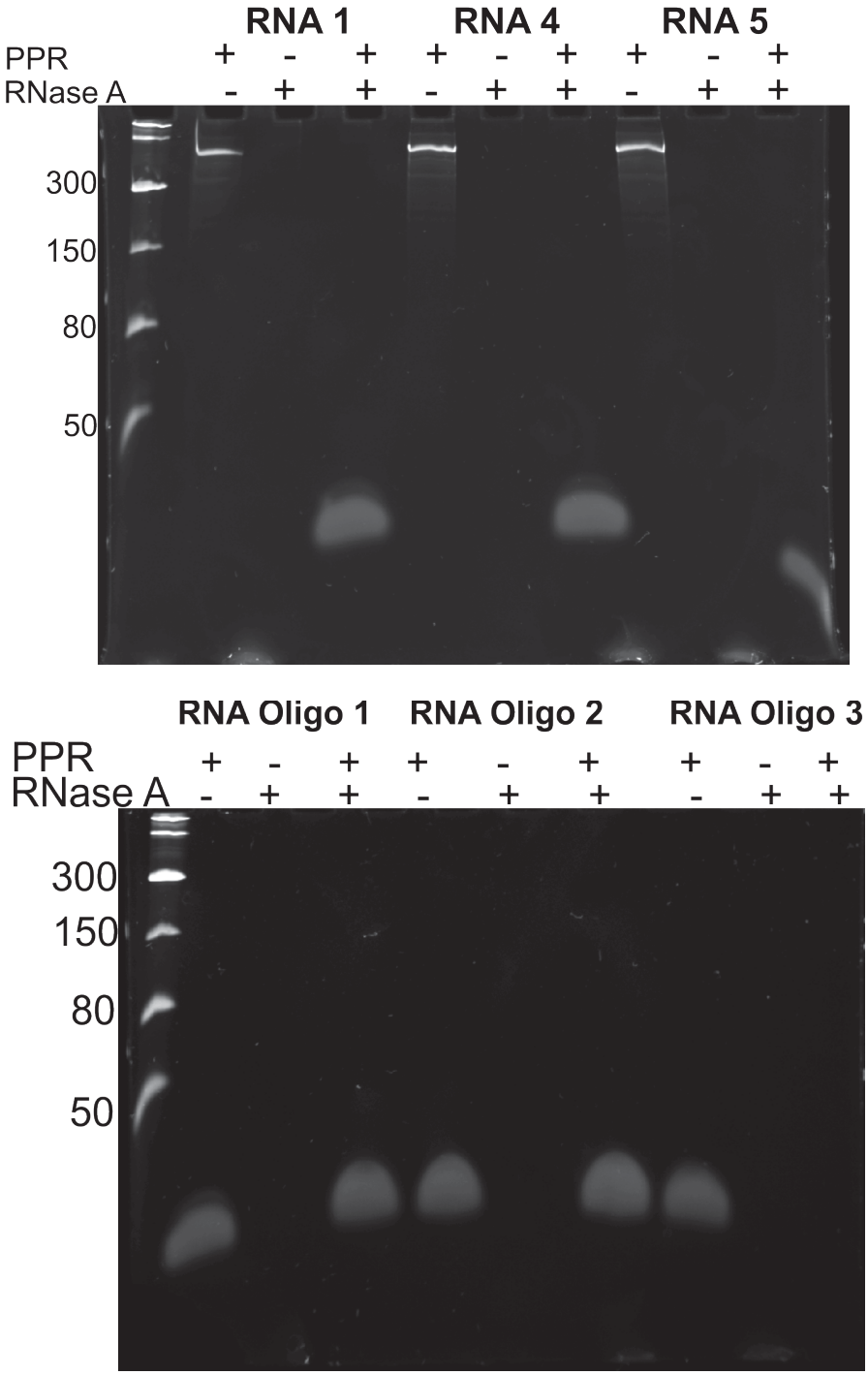
PPR (No RNA) Ve = 11.93 ml

RNA 1 = 10.42ml
ACGACAUUAAACUUGAUGCCCGGCGUUUCAGCGUUG

RNA2 = 9.87 & 11.50 ml
ACGACAUUACCGCGUAAUCACCGGGCCUGUCCGUUG

RNA5 = 9.81 ml
ACGACAUUAUAACUCGCCUAGACGGGAUUAUAGUUG

Supplemental Figure 11. Ribonuclease A protection assays. RNA transcripts 1, 4 and 5 and RNA oligos 1,2 and 3 were incubated in a 1:1 molar ratio with *Pf*PPR1 prior to treatment with RNase A. Samples were analyzed using a native acrylamide gel (ladder in nt). Experiments with no *Pf*PPR1 bound to RNA showed complete degradation by RNase A



Plasmid sequences used for *T. gondii* transformation.

pCRISPR/Cas9-GFP_PPR-sgRNA

392-1148 SAG1 Promoter
1149-5022 Cas9-GFP coding sequence
6178-6490 SAG1 Terminator
6508-7053 U6 Promoter
7054-7170 sgRNA (7060-7079 PPR protospacer)
7420-8087 Col E1 origin
8235-9095 AMP

```
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9061 ggaataaggg cgacacggaa atgttgaata ctcatactct tccttttca atattattga
9121 agcatttatc agggttattg tctcatgagc ggatacatat ttgaatgtat ttagaaaaat
9181 aaacaaatag gggttccgcg cacatttccc cgaaaagtgc cac

pPPR-mCherry_CAT

388-1441 1054 bp of 3' PPR sequence
1442-2152 mCherry coding sequence
2157-2469 SAG1 terminator
2482-2935 GRA1 promoter
2937-3602 CAT (chloramphenicol acetyltransferase) coding sequence
3607-3919 SAG1 terminator
4128-4795 Col E1 origin
4943-5803 AMP

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   1 ctaaattgta agcgttaata ttttgttaaa attcgcgtta aatttttgtt aaatcagctc
  61 atttttaac caataggccg aaatcggcaa aatcccttat aaatcaaaag aatagaccga
 121 gatagggttg agtggccgct acagggcgct cccattcgcc attcaggctg cgcaactgtt
 181 gggaagggcg tttcggtgcg ggcctcttcg ctattacgcc agctggcgaa aggggatgt
 241 gctgcaaggc gattaagttg ggtaacgcca gggttttccc agtcacgacg ttgtaaaacg
 301 acggccagtg agcgcgacgt aatacgactc actatagggc gaattggcgg aaggccgtca
 361 aggcctaggc gcgccaggtc tcatgccgga gggaatgcag ccgaagagat cggccgatcc
 421 ggagggcgac aagaacgatc ctggttgtca aacagcagga aacgatcccg ggggagagac
 481 aggactgaga cgacgagaag gagagagaga cggagtgaat gaagaagcgg gaggtagaga
 541 gaaaggcgca tcagggcaga tagaacgaga gcgagaagga gaacacgata cagaagaagc
 601 aagagagaga ctcgaagaga aagaagaaga gagagaaaag gagaacagtg aacgggaagg
 661 gagagcaata agagcagttt caggagagag aggattgtcg gtttctgctt tcttgagttc
 721 cgtcctcggc ccgcgtcgcg cggtgcttga gagcatgcgg gaagagccgg ttttggagga
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 841 ggcgaagcag agggaaggcg agagccgcga cagaagcggt gaaaacacag aagcctcaac
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 961 ccaggagaca gatgcgaaca ctgggggaac gctgtcgcag gcgggaggaa gaaagagcga
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