Supplemental Information

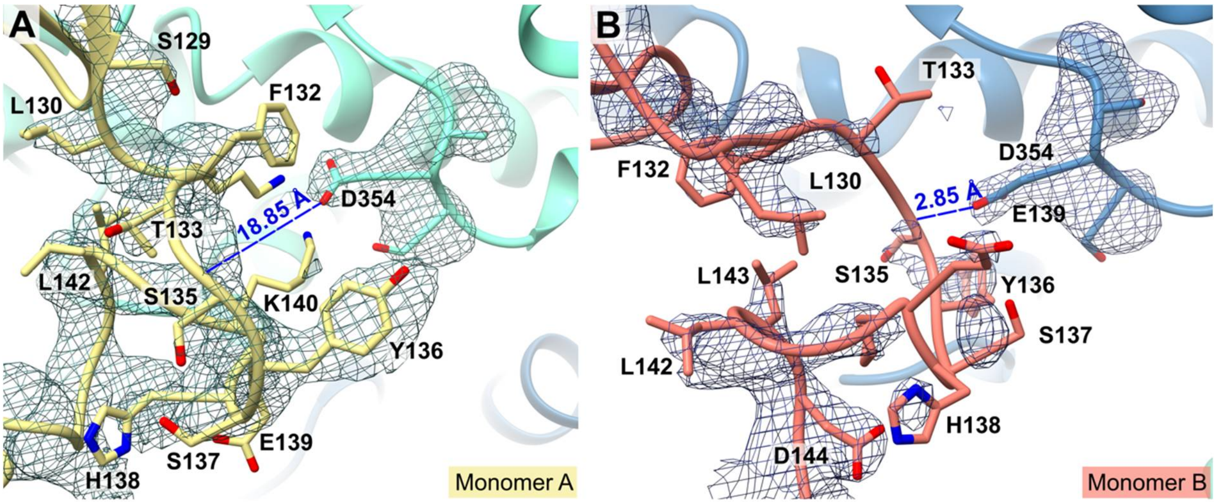


Figure S1. Electron density of the *Plasmodium*-specific insertion (P-insert) and structural comparison with *Eimeria tenella* hexokinase (*Et*HK).

(A) Close-up of the P-insert from monomer A. The final omit map covers the P-insert and residues S353-S355 and is contoured at 2.5 σ. The distance between S135 and D354 is 18.85 Å. **(B)** Close-up of the P-insert from monomer B. The final omit map covers the P-insert and residues S353-S355 and is contoured at 2.4 σ. The distance between residue S135 (N) and D354(OD2) is 2.85 Å.

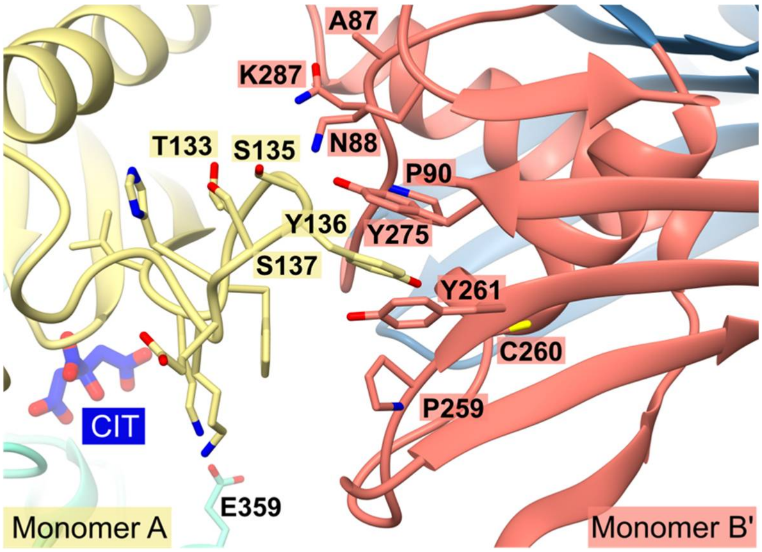


Figure S2. P-insert residues interact via hydrophobic interactions with the neighboring tetramer. Close-up of the P-insert from monomer A. Residues G134-Y136 interact with residues N87-P90, Y275, K287, and C260-Y261 of subunit B'.

CIT, citrate

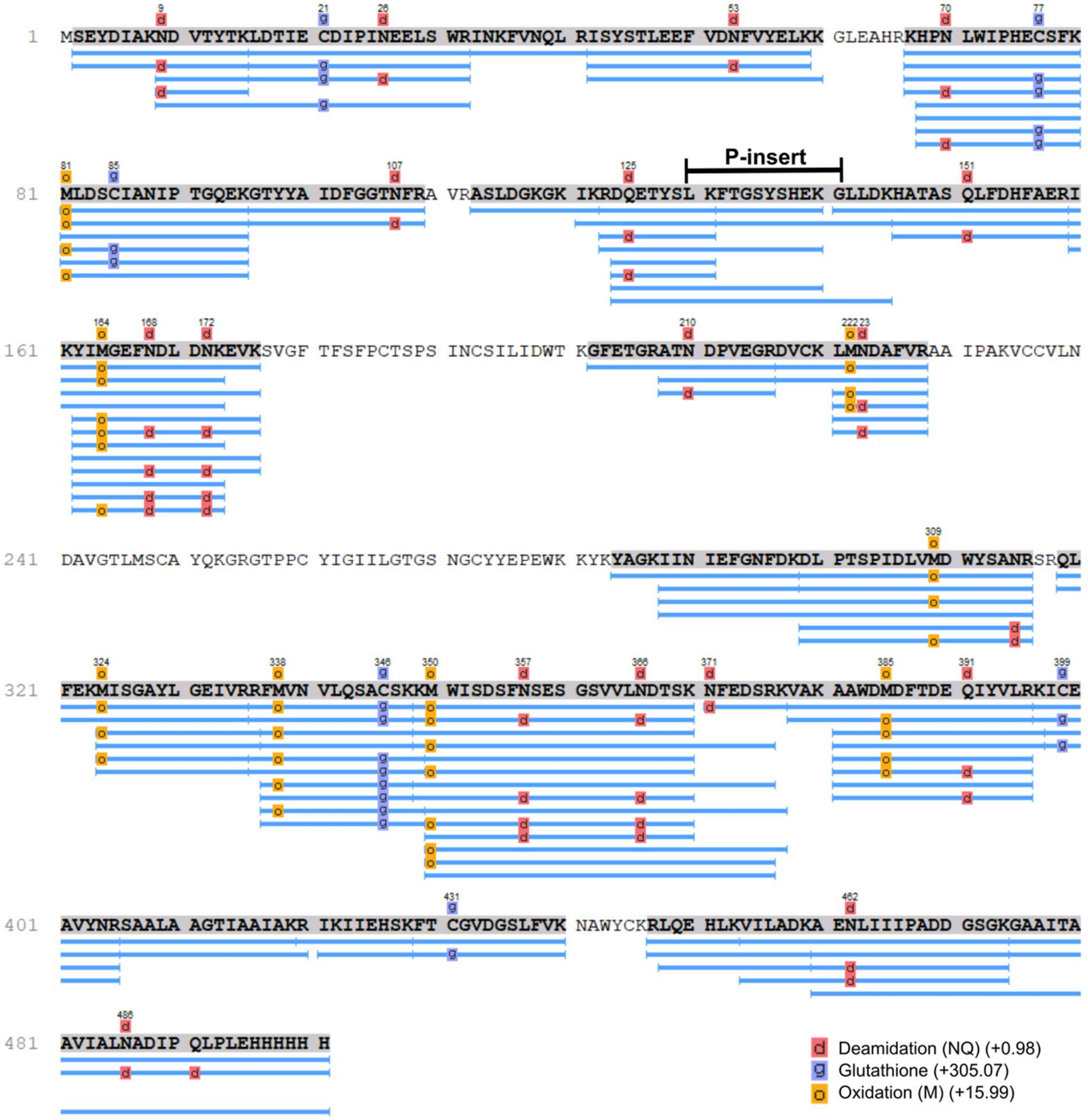


Figure S3. Protein coverage of S-glutathionylated *Pf*HK after mass spectrometry analysis. Modified protein samples were analyzed with the software PEAKS. Peptides found in mass spectrometry analysis are shown in blue lines under the respective amino acids (one-letter code). S-glutathionylated cysteines are indicated with blue squares (C21, C77, C85, C346, C399, C431) and were detected by a shift of mass (+305.07 Da).

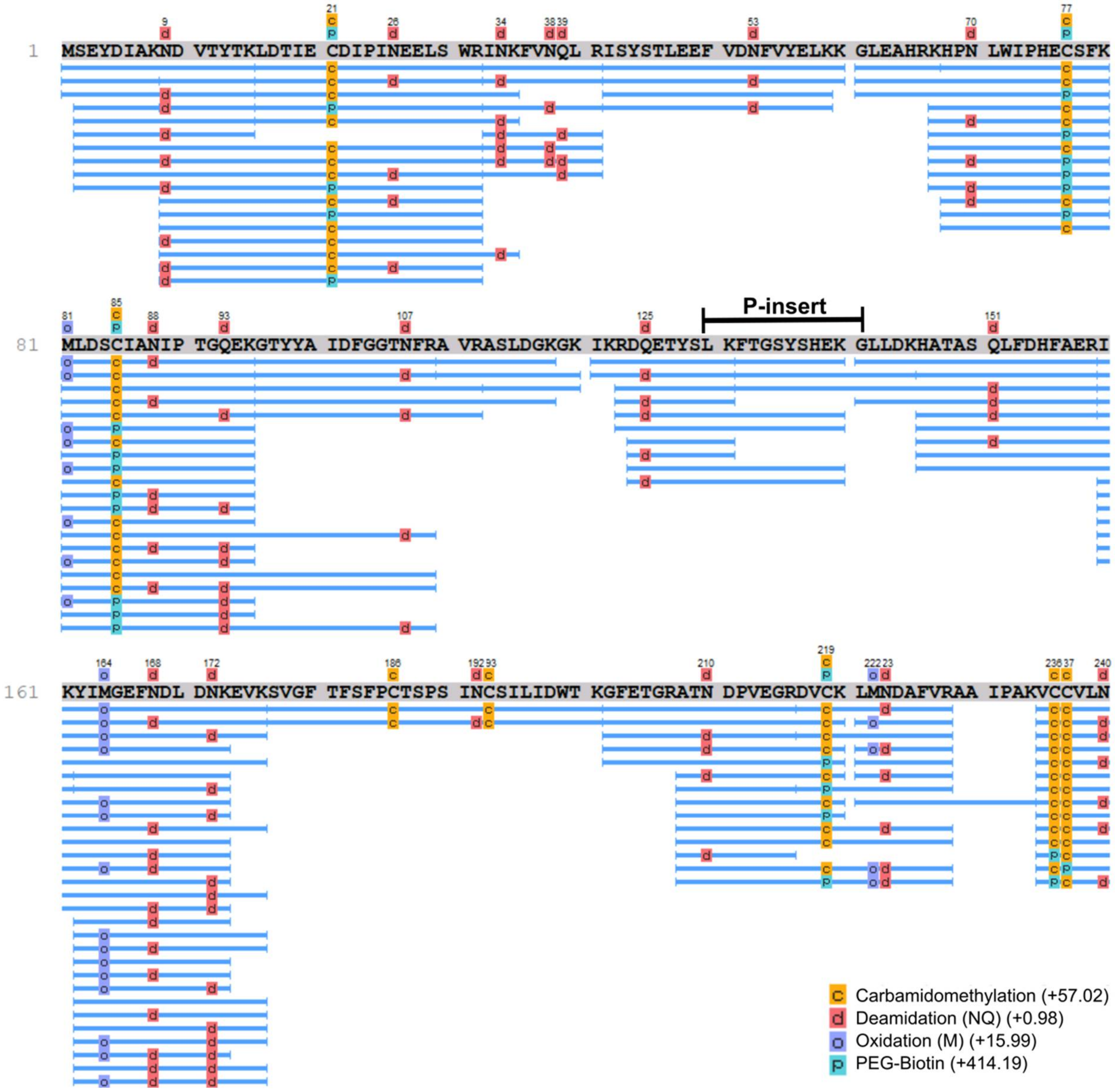


Figure S4 (a). Protein coverage of S-nitrosated *Pf*HK after mass spectrometry analysis.

Modified protein samples were analyzed with the software PEAKS. Peptides found in mass spectrometry analysis are shown in blue lines under the respective amino acids (one-letter code). S-nitrosated (biotinylated for the analysis) cysteines are indicated with green squares (C21, C77, C85, C219, C236, C237, C249, C260, C273, C346, C399, C431) and were detected via a shift of mass (+414.19 Da).

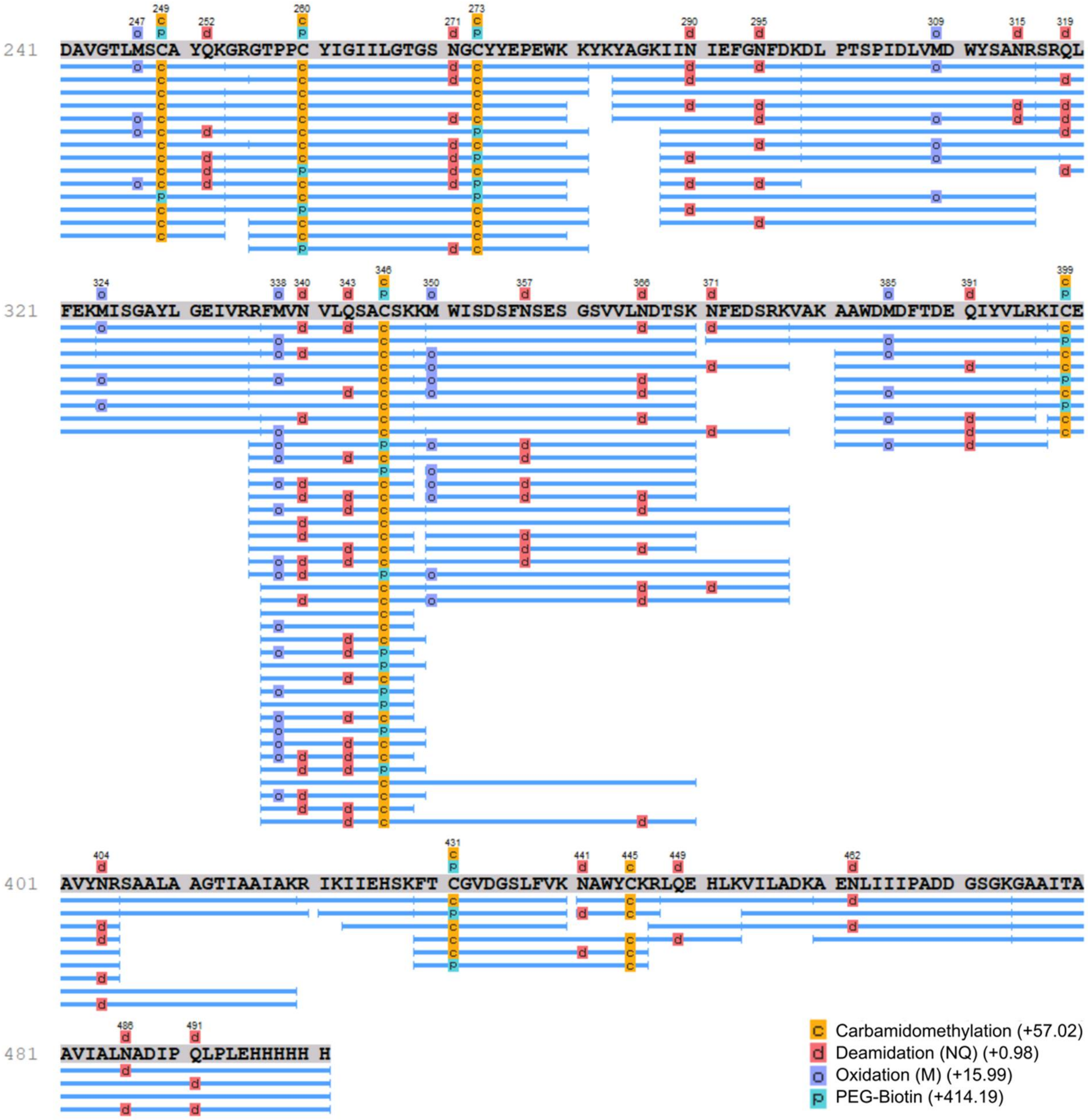


Figure S4 (b). Protein coverage of S-nitrosated *Pf*HK after mass spectrometry analysis.

Modified protein samples were analyzed with the software PEAKS. Peptides found in mass spectrometry analysis are shown in blue lines under the respective amino acids (one-letter code). S-nitrosated (biotinylated for the analysis) cysteines are indicated with green squares (C21, C77, C85, C219, C236, C237, C249, C260, C273, C346, C399, C431) and were detected via a shift of mass (+414.19 Da).

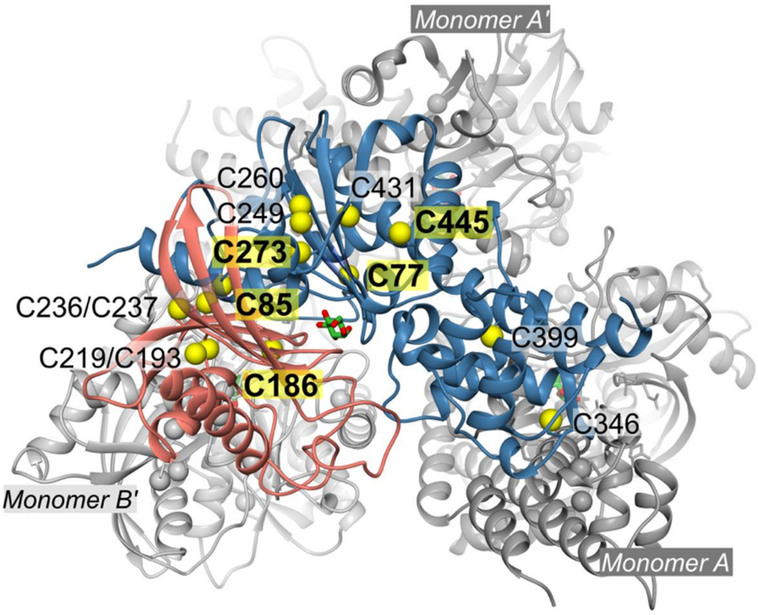
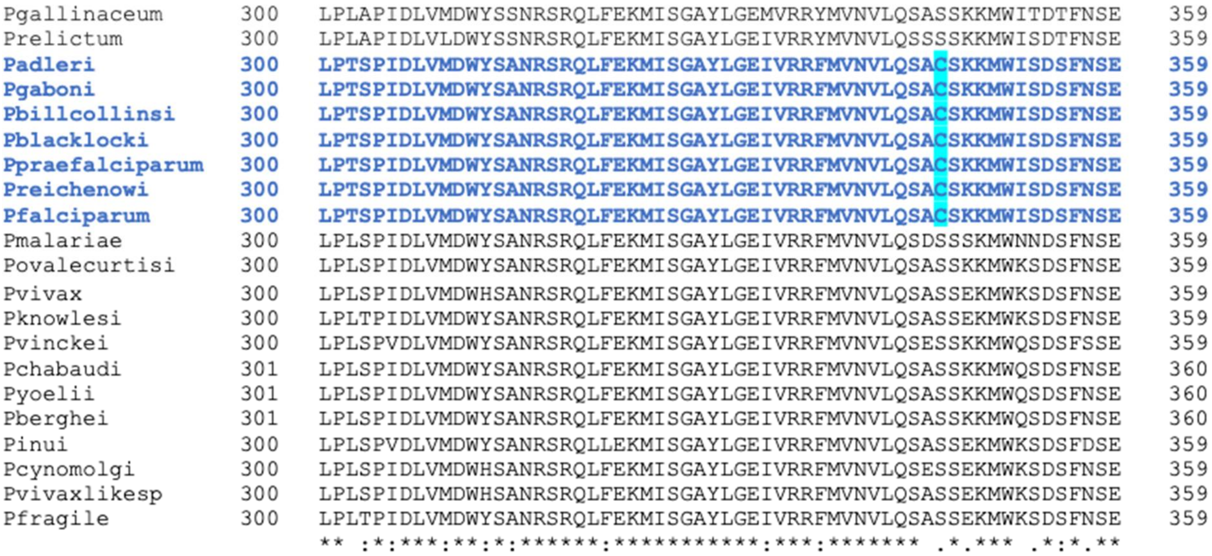


Figure S5. Distribution of cysteines in *Pf*HK monomer B.

The *Pf*HK monomer contains 15 cysteines, but only a few are probably involved in regulating the enzymatic activity via oxPTMs. No electron density could be detected for C21. C77 and C186 are located near the active site, C193 and C346 are part of the tetrameric interface. The active site is marked by GLC, coloured in green.



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Padleri           300    LPTSPIDLVMDWYSANRSRQLFEKMISGAYLGEIVRRFMVNVLQSACSKKMWISDSFNSE    359
Pgaboni           300    LPTSPIDLVMDWYSANRSRQLFEKMISGAYLGEIVRRFMVNVLQSACSKKMWISDSFNSE    359
Pbillcollinsi     300    LPTSPIDLVMDWYSANRSRQLFEKMISGAYLGEIVRRFMVNVLQSACSKKMWISDSFNSE    359
Pblacklocki       300    LPTSPIDLVMDWYSANRSRQLFEKMISGAYLGEIVRRFMVNVLQSACSKKMWISDSFNSE    359
Ppraefalciparum   300    LPTSPIDLVMDWYSANRSRQLFEKMISGAYLGEIVRRFMVNVLQSACSKKMWISDSFNSE    359
Preichenowi       300    LPTSPIDLVMDWYSANRSRQLFEKMISGAYLGEIVRRFMVNVLQSACSKKMWISDSFNSE    359
Pfalciparum       300    LPTSPIDLVMDWYSANRSRQLFEKMISGAYLGEIVRRFMVNVLQSACSKKMWISDSFNSE    359
Pmalariae         300    LPLSPIDLVMDWYSANRSRQLFEKMISGAYLGEIVRRFMVNVLQSDSSSKMWNNDSFNSE    359
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Pchabaudi         301    LPLSPIDLVMDWYSANRSRQLFEKMISGAYLGEIVRRFMVNVLQSASSKKMWQSDSFNSE    360
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Pfragile          300    LPLTPIDLVMDWYSANRSRQLFEKMISGAYLGEIVRRFMVNVLQSASSEKMWKSDSFNSE    359
                         ** :*:***:**:*:******:***********:***:******* .*.*** .*:*.**
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Figure S6. Multiple sequence alignment of different *Plasmodium* species.

C346 is conserved among *P. falciparum* and six other *Plasmodium* species pathogenic to chimpanzees and gorillas. Sequences were analyzed using Clustal Omega Multiple Sequence Alignment.