# Epitopes of anti-RIFIN antibodies and characterization of *rif*-expressing *Plasmodium falciparum* parasites by RNA sequencing

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## Supplementary Figure Legends

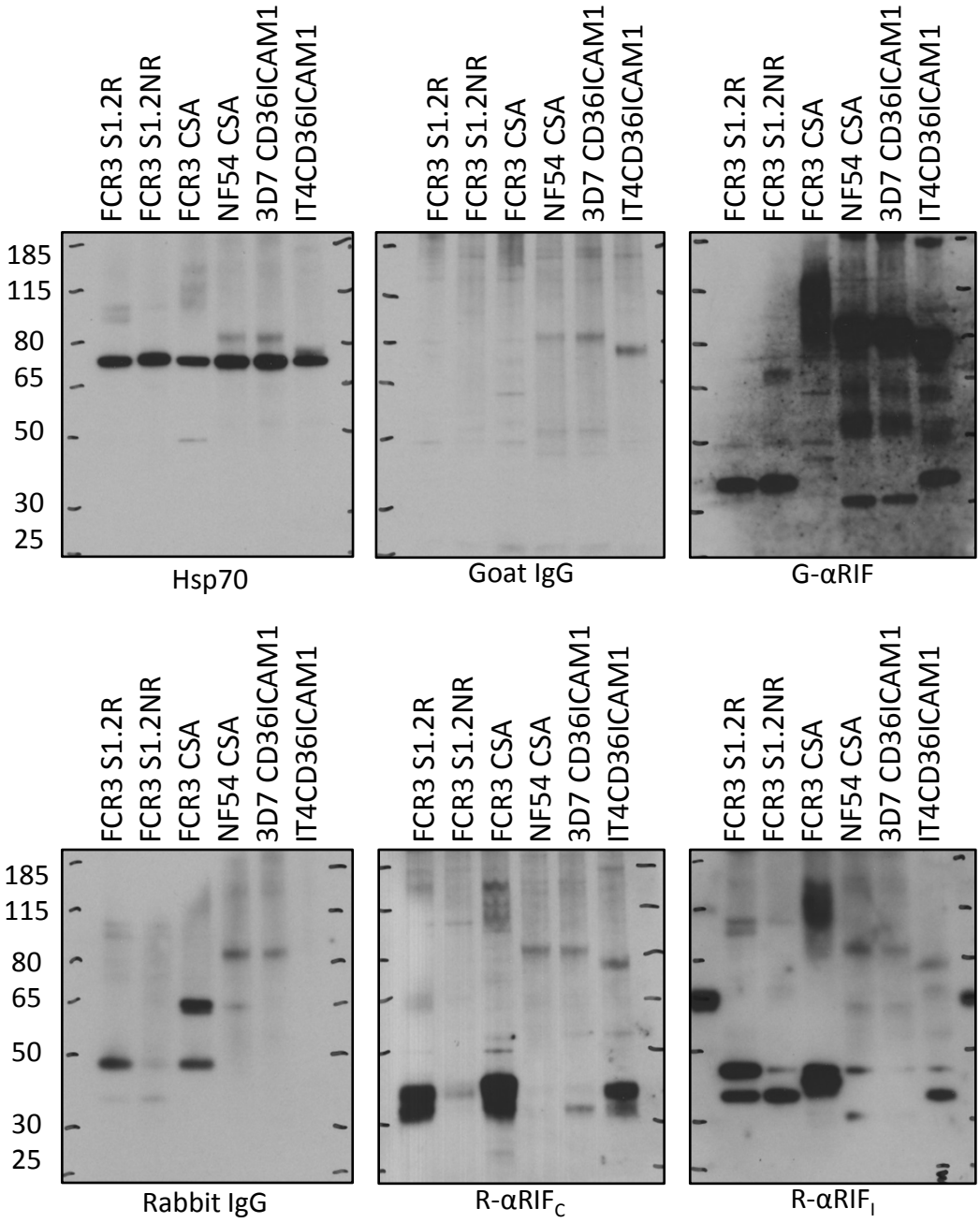
Supplementary Figure S1. Full length western blots of RIFINs in multiple parasite strains. SDS-extracted parasite lysates of (A) S1.2R, S1.2NR, FCR3CSA, NF54CSA, 3D7CD36ICAM1 and IT4CD36ICAM1 were run on SDS-PAGE (lanes 1-6 respectively), as well as SDS-extracted lysates of (B) PAvarO and R29 (lanes 1 and 2 respectively). Blots were transferred to nitrocellulose membrane and blotted with rabbit anti-PfHsp70 (1:2000), non-immune rabbit IgG (10 µg/ml), RαRIF$_C$ rabbit IgG (10 µg/ml), RαRIF$_I$ rabbit IgG (10 µg/ml), non-immune goat IgG (10 µg/ml) and GαRIF goat IgG (10 µg/ml). Corresponding HRP-conjugated secondary anti-rabbit and anti-goat IgG antibodies were used together with ECL reagent for detection. Description of the antigens used for immunizations of RαRIF$_C$, RαRIF$_I$ and GαRIF are described in Supplementary Table S1.

Supplementary Figure S2. Epitope region mapping of anti-RIFIN antibodies with epitope regions highlighted. The reactivity for IgG purified from (A) RαRIF$_C$, (B) RαRIF$_I$ and (C) GαRIF are shown here against peptides from the RIFIN protein PF3D7_0100400 (left panels) and the corresponding epitope regions (right panels, black bars). Y-axis shows mean median fluorescence intensity of duplicate spots and X-axis shows the peptide number from the N terminus to the C-terminus. RαRIF$_C$ was immunized with the C-terminus, RαRIF$_I$ with the indel region and GαRIF with the full length PF3D7_0100400 protein.

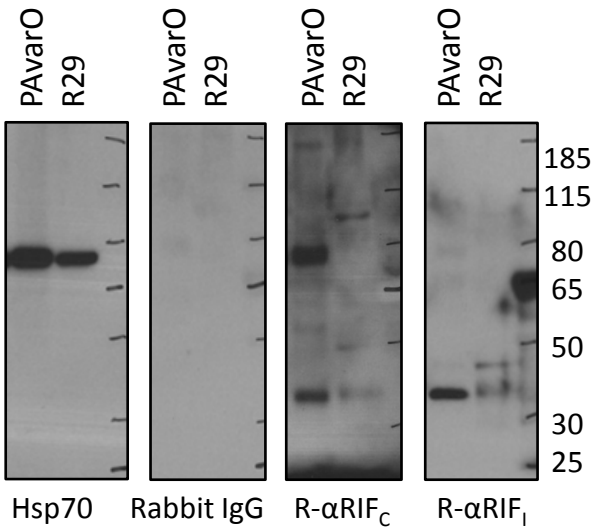
Supplementary Figure S3. Alignment of RIFIN amino acid sequences. Full length sequences of PF3D7_0100400, PFIT_bin05750 (dominant *rif* of S1.2R), PFIT_0835500 (dominant *rif* of IT4CD36ICAM1) and PFIT_bin00500 (dominant *rif* of FCR3CSA) were aligned by Clustal Omega and the three predicted transmembrane (TM) regions highlighted in pink. Amino acid sequences of PF3D7_0100400 and PFIT_bin05750 were used previously in Goel et al. (2015).

Supplementary Figure S1

A.

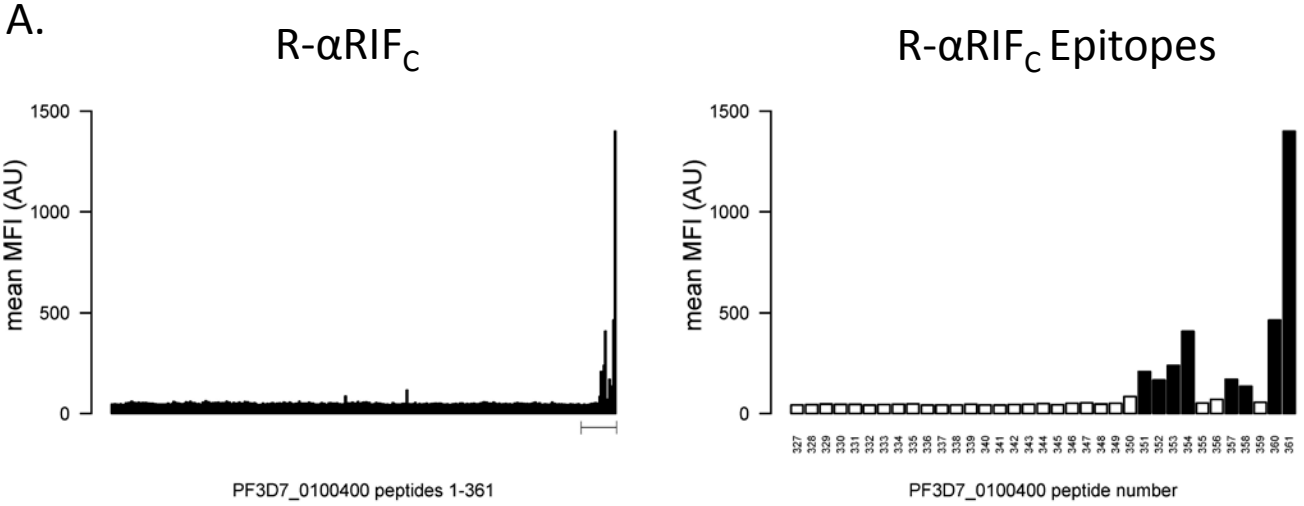



B.

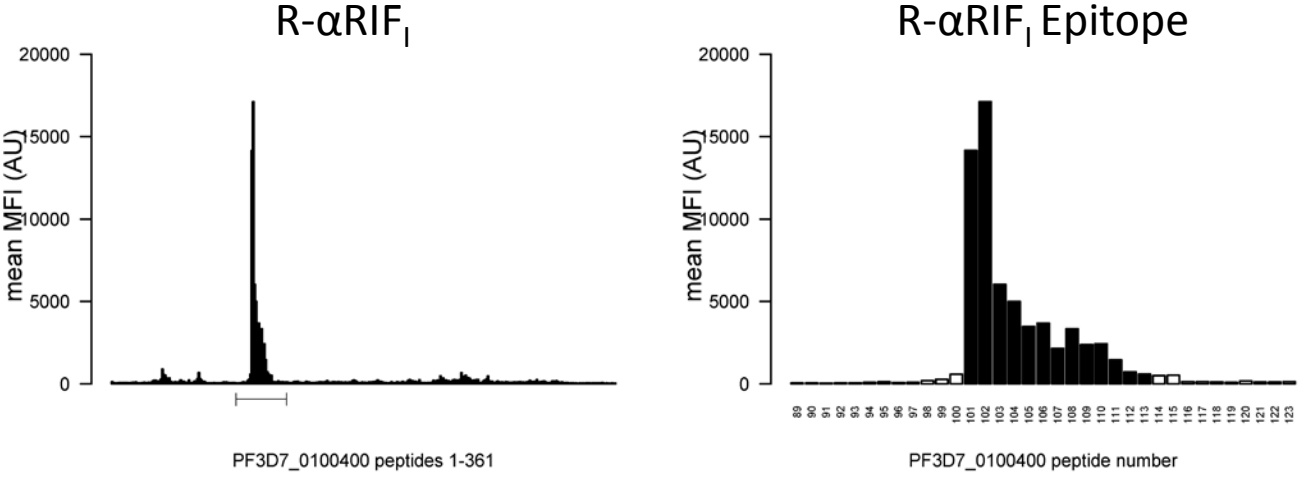


Supplementary Figure S2

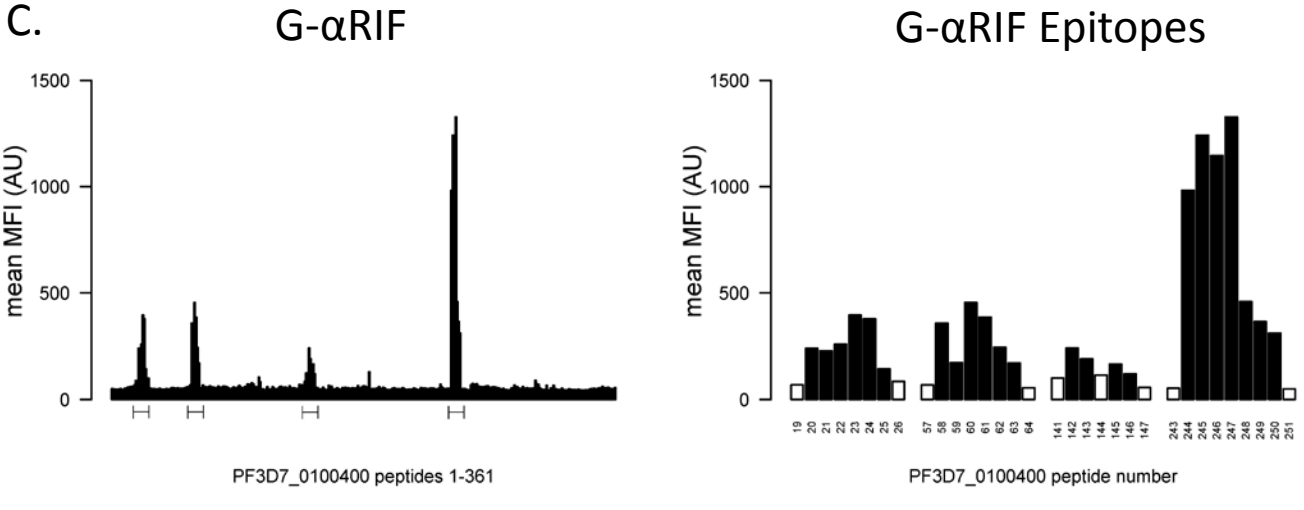
A.




B.




C.



# Supplementary Figure S3