Supplementary information

Figures

Suppl. Figure S1: iTRAQ sample preparation and analysis. Flow chart describes the workflow for the iTRAQ approach.

Suppl. Figure S2: Peptide distribution per protein for a subset of hits. Predicted protein orientation and peptide mapping for these hits is shown.

Suppl. Figure S3: Complete Western blot data. Entire Western blot membranes for the data in 6A are shown.

Suppl. Figure S4. 3D surface plot of Pf3D7_1310500 (green) and PfSBP1 (red) in untreated pRBCs (**A**), or pRBCs treated with trypsin (**B**), showing the pRBC surface (Spectrum LUT), and the specific protein localization (original colors). Scale for the x/y axis was 0.86, while scale for the z-axis was 0.97.

Tables

Suppl. Table S1: Peptide report from pRBC membrane proteomics against the Pf3D7 and Pf2004 database.

Suppl. Table S2: Protein report from pRBC membrane proteomics against the Pf3D7 and Pf2004 database.

Suppl. Table S3: Significant hits from pRBC membrane proteomics.

Suppl. Tables S4,S5: Peptide report from supernatant proteomics against the Pf3D7 and Pf2004 databases, respectively.

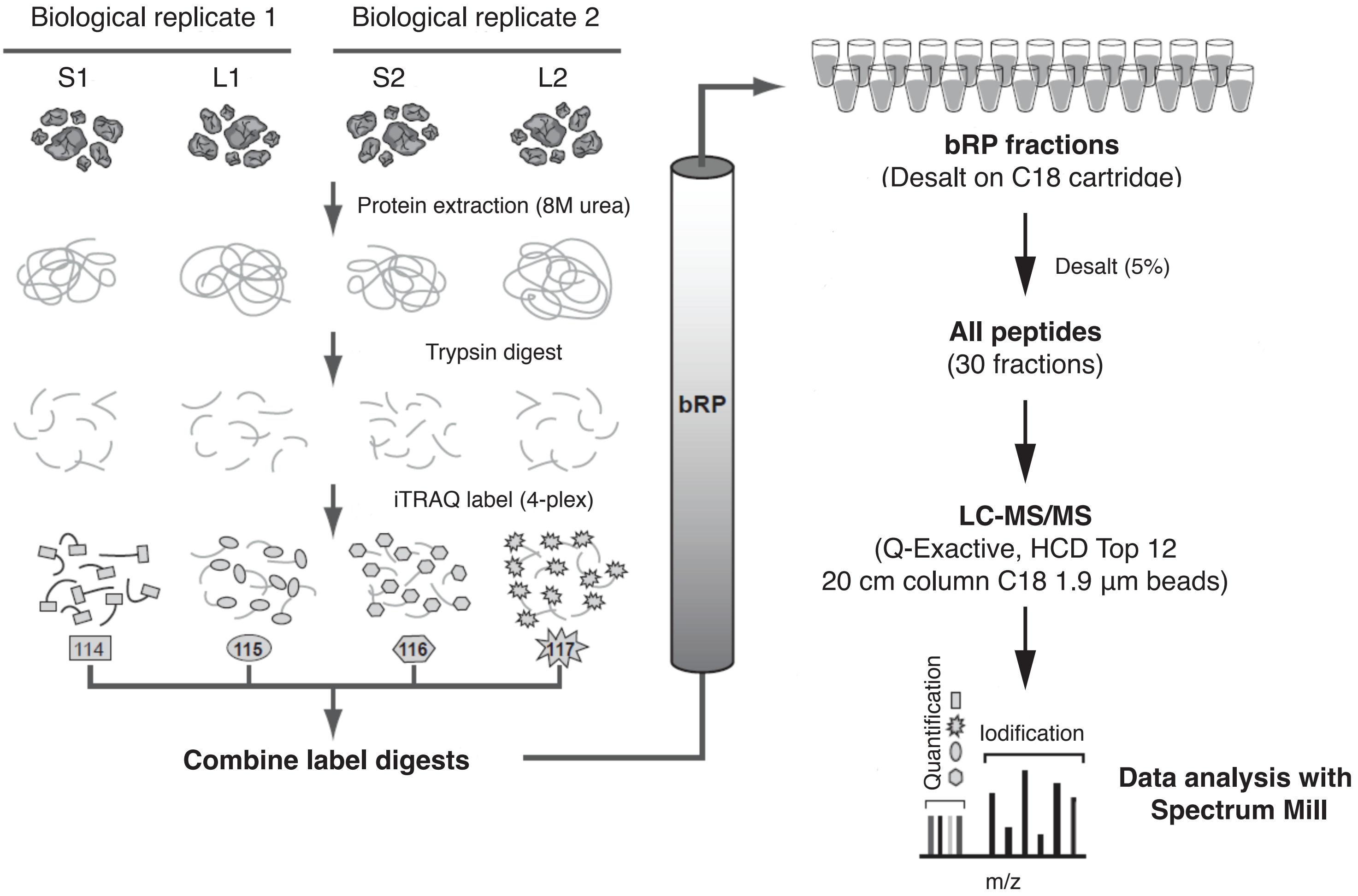
Suppl. Tables S6,S7: Protein report from supernatant proteomics against the Pf3D7 and Pf2004 databases, respectively.

Suppl. Table S8: Quantitative protein data from PEXEL search.

Suppl. Tables S9, S10: All significant hits from quantitative protein data against Pf3D7 and Pf2004 databases.

Suppl. Table S11: Significant hits from quantitative proteomics enriched in pRBC supernatant.

Figure S1



Biological replicate 1
Biological replicate 2
S1
L1
S2
L2
Protein extraction (8M urea)
Trypsin digest
iTRAQ label (4-plex)
114
115
116
117
Combine label digests
bRP
bRP fractions
(Desalt on C18 cartridge)
Desalt (5%)
All peptides
(30 fractions)
LC-MS/MS
(Q-Exactive, HCD Top 12
20 cm column C18 1.9 µm beads)
Quantification
Iodification
m/z
Data analysis with
Spectrum Mill

Figure S2

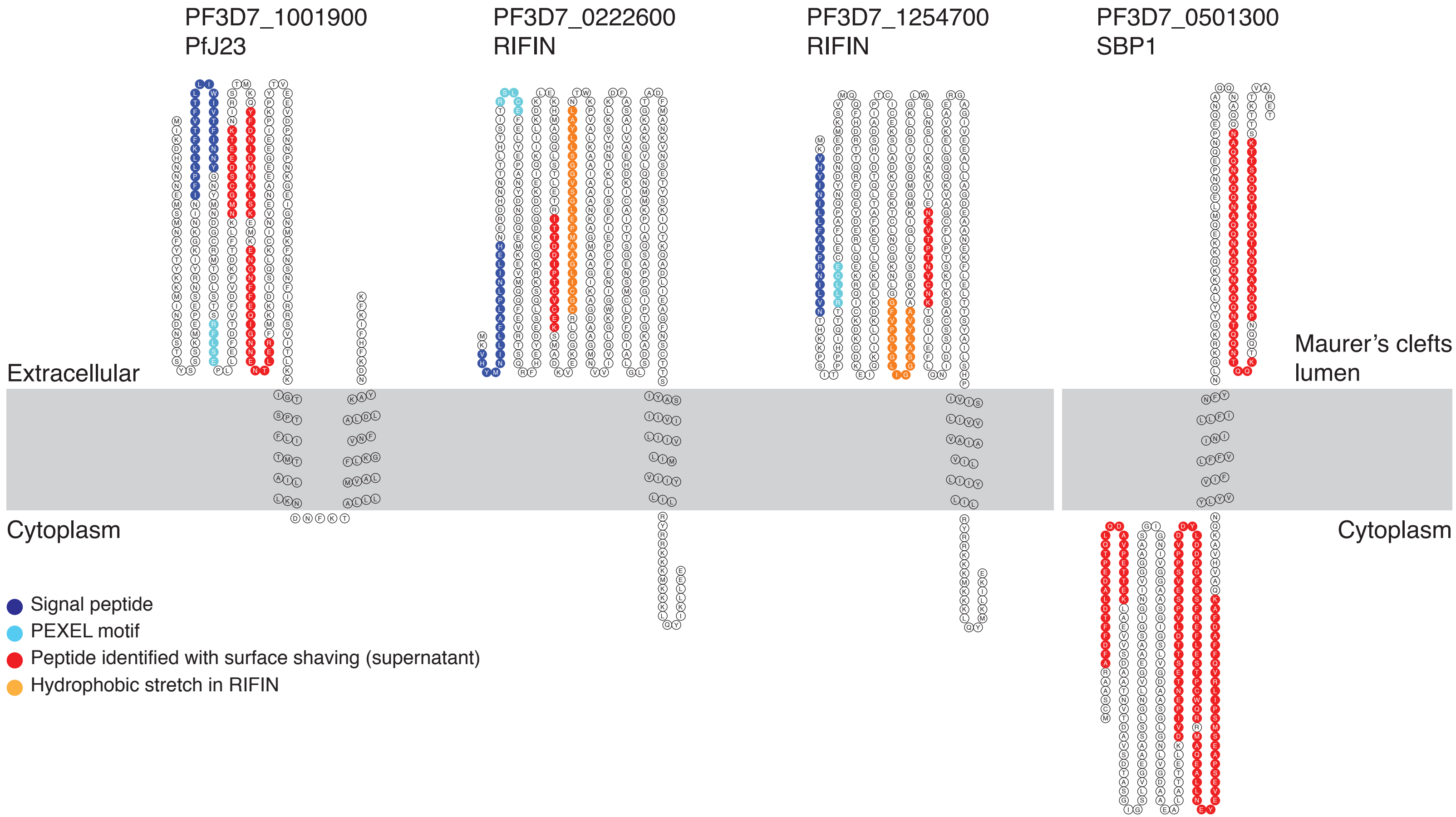

PF3D7_1001900
PfJ23
PF3D7_0222600
RIFIN
PF3D7_1254700
RIFIN
PF3D7_0501300
SBP1
Extracellular
Cytoplasm
Maurer's clefts lumen
Cytoplasm
● Signal peptide
● PEXEL motif
● Peptide identified with surface shaving (supernatant)
● Hydrophobic stretch in RIFIN

Figure S3

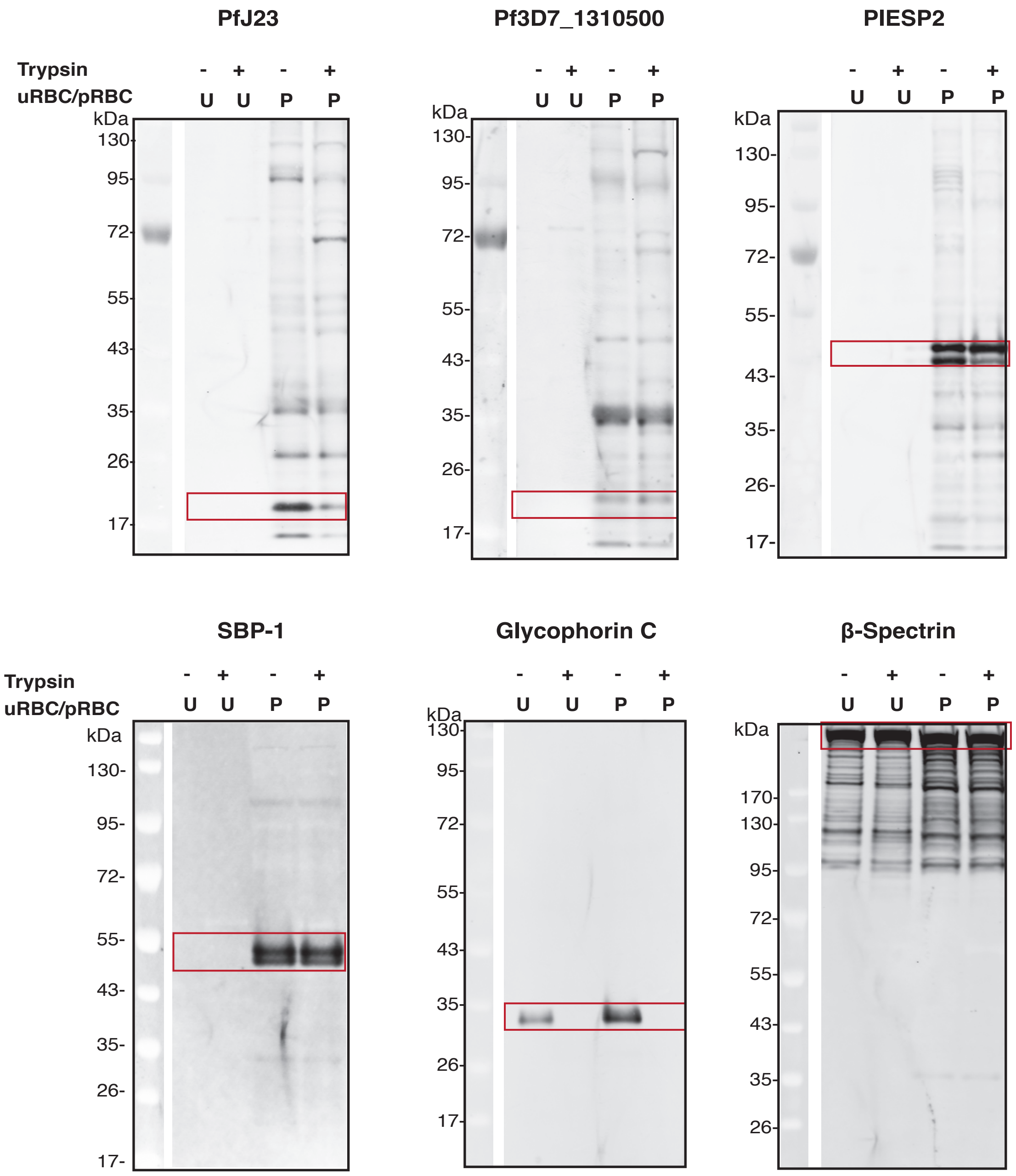

PfJ23
Trypsin
uRBC/pRBC
- + - +
U U P P
kDa
130-
95-
72-
55-
43-
35-
26-
17-
Pf3D7_1310500
- + - +
U U P P
kDa
130-
95-
72-
55-
43-
35-
26-
17-
PIESP2
- + - +
U U P P
kDa
130-
95-
72-
55-
43-
35-
26-
17-
SBP-1
Trypsin
uRBC/pRBC
- + - +
U U P P
kDa
130-
95-
72-
55-
43-
35-
26-
17-
Glycophorin C
- + - +
U U P P
kDa
130-
95-
72-
55-
43-
35-
26-
17-
β-Spectrin
- + - +
U U P P
kDa
170-
130-
95-
72-
55-
43-
35-
26-

Figure S4