

Supplementary Material:

Spatial association with PTEX complexes defines regions for effector export into *Plasmodium falciparum*-infected erythrocytes

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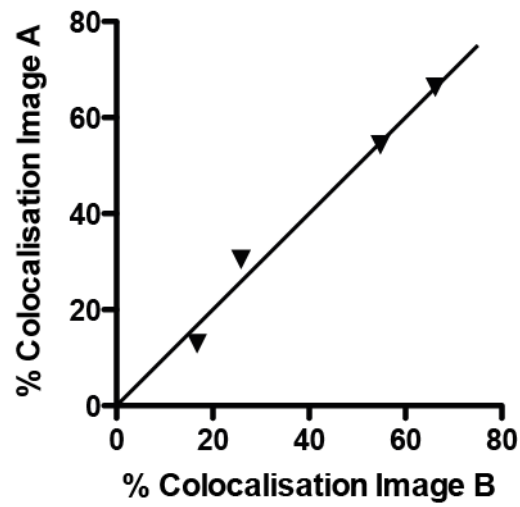
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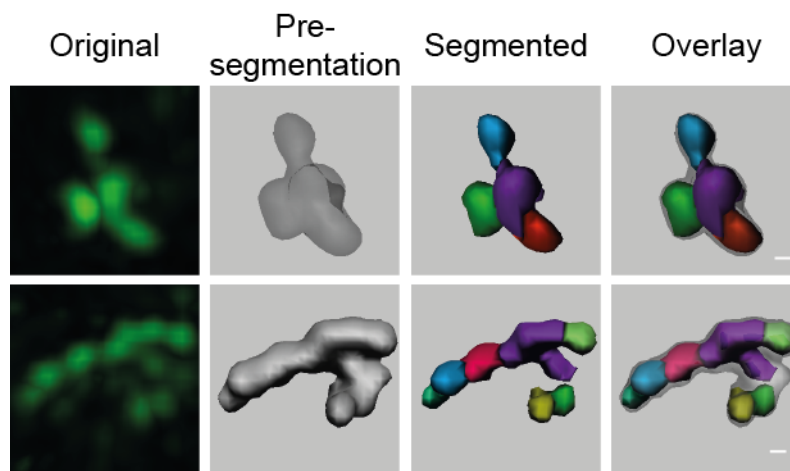
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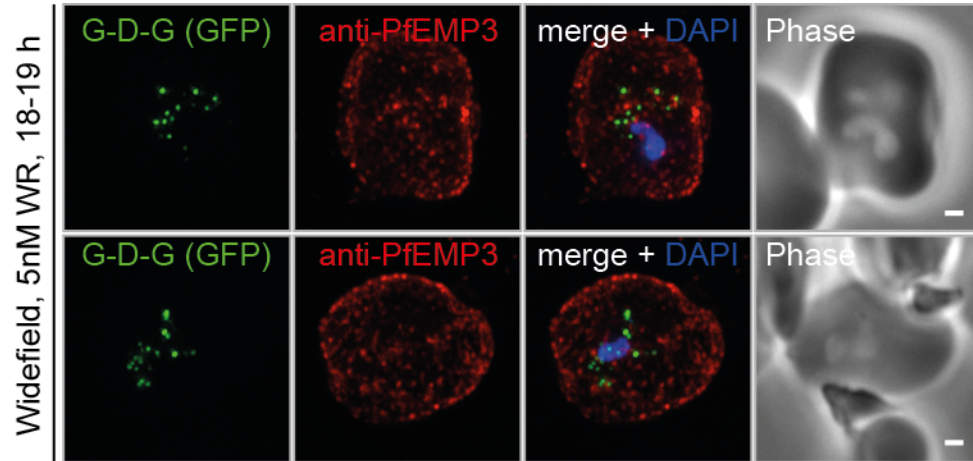
Supplementary Figure S1. Colocalisation analysis is robust upon repeated analysis of individual parasites.

Comparison of “% colocalisation” values between two independent measurements of four separate parasites from the HSP101HA <10 min time point showed consistency of results, as shown by proximity to the $y=x$ line.



Supplementary Figure S2. Upon manual evaluation EXP2 segmentation protocols identified and split regions as expected.

Automated imaging analysis was used to identify large “presegmentation” EXP2 clusters and “segmented” EXP2 foci using a region-growing algorithm. An example of the extensive manual evaluation of the regions identified by the automated protocols showed that isolation and region splitting occurred in line with observed fluorescent foci from original data. Scale = 100 nm.



Supplementary Figure S3. Native protein export occurs as normal upon the addition of WR.

Widefield deconvolution imaging of GBP130-DHFR-GFP expressing parasites grown in the presence of 5nM WR, showed export of native proteins is unimpeded by WR addition. Green = GFP, Red = PfEMP3, Blue = DAPI (nucleus).

Scale = 1 μ m.

Supplementary Table S1. EXP2 size and distribution analysis statistics, as depicted graphically in Figure 3D.

Timepoint	n	Pre-Segmentation Mean \pm SEM ($\times 10^{-3}$) μm^3	Segmented Mean \pm SEM ($\times 10^{-3}$) μm^3
<12 min	19	26.1 \pm 2.7	9.2 \pm 0.3
60-90 min	27	16.6 \pm 1.4	8.7 \pm 0.4
11-12 h	37	9.2 \pm 0.5	7.1 \pm 0.3
18-19 h	22	11.8 \pm 0.7	7.6 \pm 0.5