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Protocol update

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An assay to quantify sexual commitment and stage conversion in the human malaria parasite *Plasmodium falciparum*

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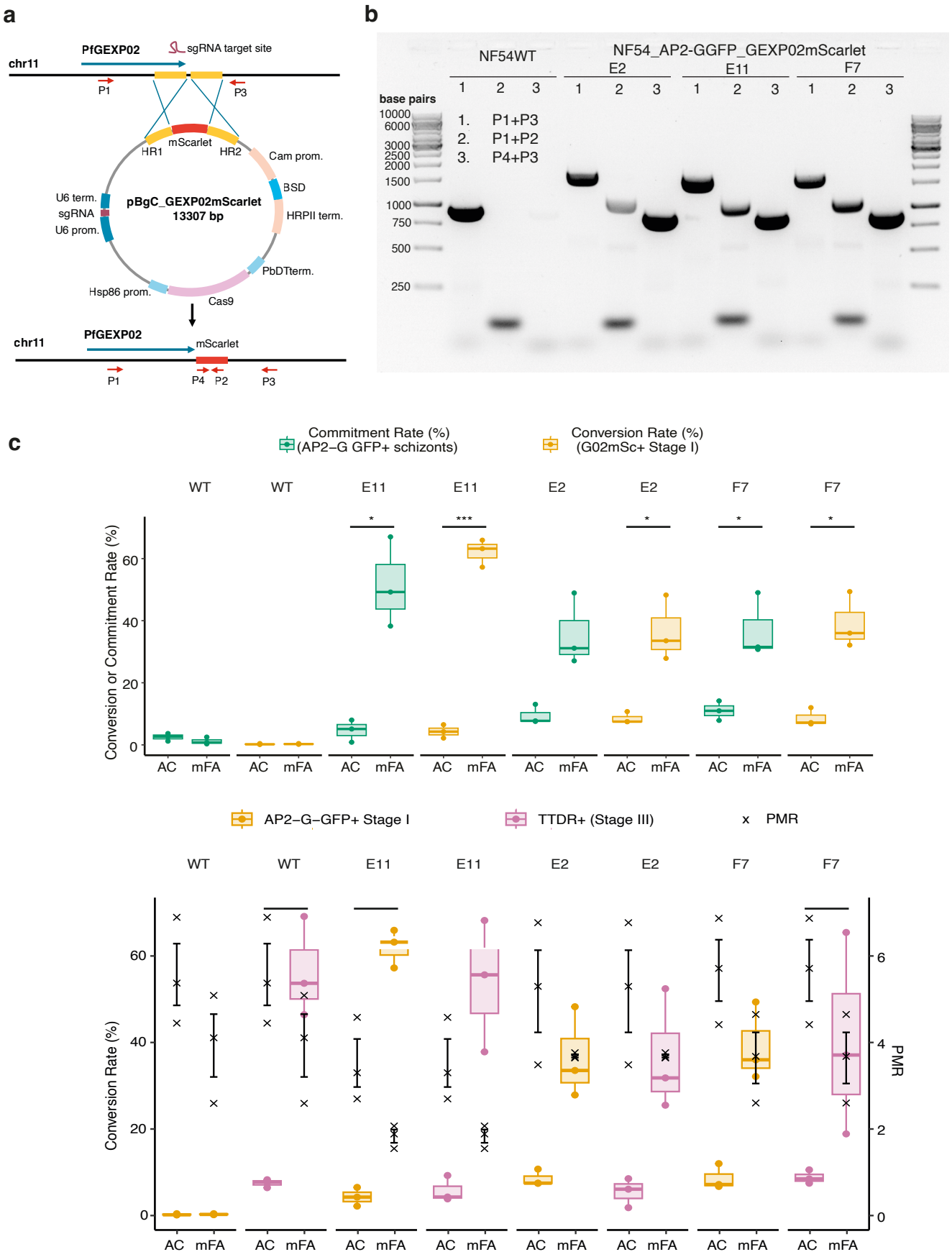
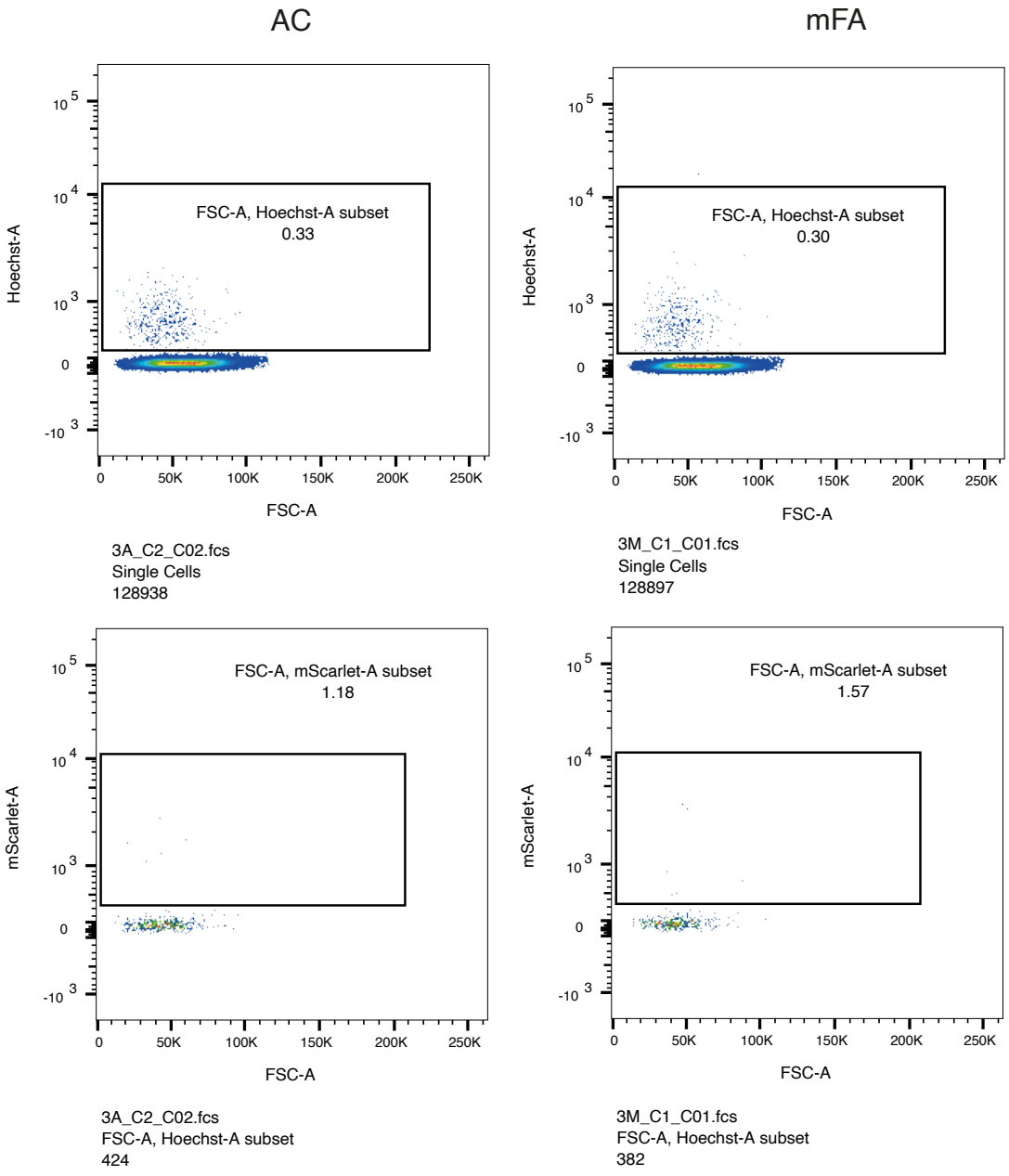
Fig S1



Supplementary Figure 1. Generation of the dual reporter line. a. Single plasmid CRISPR Cas9 editing of the endogenous gexp02 locus on chromosome 11 (chr) with the aid of homologous regions (HR1 and HR2) and a specific single guide RNA (sgRNA) resulted in the targeted addition of the gene encoding mScarlet at the 3' of the gexp02. The transfection was followed by selection with Blasticidin S conferred by the blasticidin S deaminase (BSD) resistance gene cassette b. Genotyping PCR on NF54 WT parasites versus three different clones of the NF54_AP2-G GFP_GEXP02mScarlet dual reporter line (E2, E11 and F7) showing successful gene editing. Clone F7 was used for all experiments presented in Figures 2-4. Reaction 1 (P1+P3) yields an 851bp fragment in WT parasites and a 1554 bp fragment in transgenic parasites. Reaction 2 (P1+P2) yields a 1105bp in transgenic parasites and Reaction 3 yields a 910 bp fragment in transgenic parasites. Bp: base pair. c. Sexual Commitment Conversion Assay on NF54 WT versus dual reporter line clones quantifying commitment, conversion and parasite multiplication rates (PMR). The top panel shows conversion versus commitment rates, and the bottom panels shows conversion rates versus PMR. Differences in all three measurements are compared between parasites grown in AlbumaxII medium supplemented with choline chloride (AC) and minimal fatty acid medium (mFA). Statistical significance between AC and mFA treatments was assessed using a student's t-test. **** p<0.0001, *** p<0.001, ** p<0.01. Source data for panel c is provided as Supplementary Data 1

Fig S2



Supplementary Figure 2. Quantification of background conversion rates by flow cytometry. Representative scatter plots showing the proportion of GEXP02-mScarlet positive cells present in the population at the time of induction. The plots in the upper panel show the percentage of iRBCs based on Hoechst staining. The plots in the lower panel show the subpopulation of GEXP02-mScarlet positive cells amongst all iRBCs in AlbumaxII medium supplemented with choline chloride (AC) versus minimal fatty acid medium (mFA).