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Abstract

Using CRISPR-Cas9 and HALO-ligand FACS sorting to overcome the limitation of a single selection marker in *P. berghei*, we generated parasite lines expressing multiple fluorescent tags to study protein–protein interactions in live cells. We examined essential RNA-binding proteins involved in early gametocyte development (GD1, FD1, FD4) alongside translational repression complex components DOZI and CITH. Many of these proteins appear to relocalise from P-body-like granules to stress granules under moderate starvation stress. Because these complexes are insoluble and not easily characterised by co-immunoprecipitation, we applied TurboID proximity labelling combined with a biotin “bio-lock” system for temporal control, enabling analysis of complex composition across developmental stages and stress conditions. This work provides new tools for protein studies in Plasmodium and advances understanding of gametocytogenesis and parasite-specific adaptations of conserved RNA regulatory mechanisms.