## SUPPLEMENTARY



## This file contains six supplementary figures and two supplementary tables.

**Supplementary figure 1. cryo-EM reconstruction of** *P. falciparum* 80S complexes, related to Figure 1 (A-E) FSC curves for (A) nrt80S-E, (B) nrt80S-P, (C) nrt80S-P-E, (D) nrt80S-empty, and (E) rt80S-A/P-P/E state respectively. (F) FSC curves for nrt80S-E state complex, using RELION auto-refinement (blue) and statistical movie processing (red) methods. Resolutions reported were based on the "gold standard" protocol along with the FSC=0.143 criterion, and involved soft masking and high-resolution noise substitution (18). (G) Representative electron micrograph showing *P. falciparum* 80S particles. (H) Density map of nrt80S-E state after further statistical movie processing. Map is colored according to local resolution calculated from ResMap (19). Shown are the central slice and the front view of the map.



**Supplementary figure 2. Movements of L1 stalk regions.** A close-up view of the region boxed on the 80S ribosome (left) shows the relative movements of L1 stalk among the following states: (i) rt80S-A/P-P/E (purple), (ii) nrt80S-E (blue), and (iii) nrt80S-P (cyan). The model of each state was generated by using MDFF. Angles of rotation from (i) to (ii) and from (ii) to (iii) are indicated by arrows. The movements were measured by using UCSF Chimera (25), based on residues 2732-2747 and 2801-2817 of 28S rRNA (approximately H76, L1 stalk 318 region).



Supplementary figure 3. Positions of rRNA expansion segments on solvent sides of *P. falciparum* 40S (left) and 60S (right) subunits. The largest rRNA expansion segments (painted in various colors) were identified according to the most recent published models.



**Supplementary figure 4. Absence of RACK1 on** *P. falciparum* **40S subunits.** Homology models of 40S crystal structure (PDB ID: 3U5B, 3U5C) (7) were rigid-body fitted using UCSF Chimera (25). Missing RACK1 protein is shown in red. 40S subunit is painted in yellow, 60S subunit in blue.



**Supplementary figure 5. Predicted models for ES10S, ES6BS and helix 16 using the 3dRNA software.** 10 predicted models generated from 3dRNA (26) were rigid fitted into nrt80S-P map (shows as mesh) using UCSF Chimera (25). The one with highest cross-correlation is highlighted in color: (A) ES10S, orange, (B) ES6BS, pink and (C) helix 16, purple. Lower-CC models are colored in grey.



Supplementary figure 6. Major clashes at the inter-subunit interface of the published *P. falciparum* ribosome subunit models (5). The published models, PDB 3J79 and 3J7A, were saved as a single combined 80S molecule (no hydrogen atoms included) and then evaluated in UCSF Chimera (25). The identification of major clashes was based on VDW radii, using the default criteria in the 'Find Clashes/Contacts' function in UCSF Chimera, with cutoff value 0.6Å and allowance value 0.4Å. (A) Clash atoms at the interface are shown as spheres and colored in red. (B) Most of the contact atoms are clustered at the tRNA CCA end, bridge B2a, B4, and regions of eukaryotic-specific bridges eB14 and eB12 (see Suppl. Table 2). Pseudo-bonds between these contact atoms are colored in light grey. The inter-subunit bridges assignments are based on Ben-Shem *el al's* work (7).

Model number	ES10S	ES6BS	Helix 16
1	0.60	0.71	0.83
2	0.60	0.72	0.83
3	0.61	0.72	0.83
4	0.56	0.73	0.83
5	0.57	0.76	0.83
6	0.66	0.77	0.83
7	0.66	0.77	0.84
8	0.65	0.76	0.83
9	0.57	0.77	0.83
10	0.57	0.77	0.82

Supplementary table 1. Cross-correlations between 3dRNA-predicted and experimental cryo-EM density map for nrt80S-P. The predicted models were rigid-body fitted into the individual domains of the segmented nrt80S-P density map. The segmentation of nrt80S-P density map and measurement of cross-correlation (CC) were both done in UCSF Chimera (25). The highest CC values are highlighted in bold.

40S subunit	60S subunit	
(PDB 3J7A)	(PDB 3J79)	
18S rRNA (h20)	28S rRNA (h34)	
18S rRNA (ES6ES)	eL19	
18S rRNA (h27)	eL41	
18S rRNA (h44)	28S rRNA (h69)	
18S rRNA (h44)	eL41	
18S rRNA (h45)	28S rRNA (h69)	
18S rRNA (h45)	eL41	
E-tRNA (CCA end)	28S rRNA (h68)	
E-tRNA (CCA end)	28S rRNA (H88)	
E-tRNA (CCA end)	eL44	
uS15	28S rRNA (h34)	
uS15	eL30	
eS8	28S rRNA (h63)	

Supplementary table 2. Major clashes at the inter-subunit interface of the published *P. falciparum* ribosome subunit models (5). The 256 contact atoms between the published *P. falciparum* 40S subunit and 60S subunit can be grouped into certain rRNA helices and proteins. For the identification of the contacts/clashes see Suppl. fig. 6. Zero contacts were found in the MDFF-fitted nrt80S-E, nrt80S-P and rt80S-P/E models. The assignment of the helices is based on the secondary structure in Wong *et al.*'s work (5).