



C

All values in the table were measured with saturating RPL41A mRNA

	ATPase k_{cat} , min^{-1}	K_m^{ATP} , μM
4A•4G•4E	2.4 ± 0.1	250 ± 10
TC, 40S, 4A•4G•4E	2.7 ± 0.2	280 ± 40
3, 4A•4G•4E	3.2 ± 0.7	200 ± 40
Complete PIC	8.2 ± 0.3	330 ± 20
Components missing from Complete PIC	-4A	$\sim 0.10 \pm 0.01$ ND
	-40S	4.5 ± 0.1 150 ± 1
	-2	4.7 ± 0.1 210 ± 5
	-3	3.3 ± 0.2 180 ± 20
	-3g, -3i	3.9 ± 0.3 170 ± 10
	-2, -40S	4.1 ± 0.1 150 ± 1
	-3, -40S	3.0 ± 0.1 150 ± 1
	-4B	8.0 ± 0.1 480 ± 10
	-5	8.0 ± 0.4 350 ± 10

D

All values in the table were measured with saturating RPL41A mRNA

	ATPase k_{cat} , min^{-1}	K_m^{ATP} , μM
4A	0.58 ± 0.08	2500 ± 400
Complete PIC -eIF4G•4E	3.5 ± 0.3	1600 ± 200
Components missing from Complete PIC	-40S, -4G•4E	0.55 ± 0.01 1800 ± 100
	-2, -4G•4E	0.67 ± 0.02 1900 ± 400
	-3, -4G•4E	0.62 ± 0.02 1600 ± 300
	-3g, -3i, -4G•4E	0.67 ± 0.07 1500 ± 300

Figure 2 – figure supplement 1.

Figure 2 – Figure supplement 1. Control and data tables for the ATPase experiments. **(A)**

Controls for NADH enzyme-coupled microplate ATPase assay. The decrease in absorbance of 340 nm light was dependent on the presence of ATPase activity (5 μM eIF4A, 0.5 μM eIF4G•4E, together referred to as eIF4A•4G•4E or eIF4F), $\text{ATP}\cdot\text{Mg}^{2+}$, and a pyruvate kinase (900-1400 units/mL)/lactate dehydrogenase (600-1000 units/mL) mixture from rabbit muscle used as a 250x stock solution. **(B)** ATPase activity as a function of uncapped *RPL41A* mRNA. Solid, black squares: eIF4A•4G•4E (no PIC), as in Figure 2B (k_{cat} of $4.3 \pm 0.1 \text{ min}^{-1}$ and K_m^{RNA} of $150 \pm 20 \mu\text{M}$). Solid black circles: "Complete PIC," as in Figure 2B (k_{cat} of $8 \pm 1 \text{ min}^{-1}$ and K_m^{RNA} of $120 \pm 40 \mu\text{M}$). **(C-D)** Summary of all k_{cat} and K_m^{ATP} values in the presence of eIF4G•4E (panel C, red) and absence of eIF4G•4E (panel D, blue). Data in B-D are mean values ($n \geq 2$) and error is reported as average deviation of the mean.