



Figure S1. Threonine Hsp70 mutants display lower V_{max} and increased K_M for ATP hydrolysis compared to wild type using the artificial NR substrate. This experiment was performed exactly as in Fig. 4, except the natural substrate prSSU was replaced with the artificial substrate NRLLLTG at 100 nM.

Supplemental Table 1. Primers used in the study.

Primer name	Sequence (5'-3')
pTYB12-HSP70F	ACTAGTGAAAAAGTTGTTCGGCATTGATCTT
pTYB12-HSP70R	GGATCCTCACGTTGAATCAGTGAAGTC
QCTVF	CTTGGCGGGGGCGTCTTTGATGTTTC
QCTVR	GAAACATCAAAGACGCCCCCGCCAAG
QCTEF	GACCTTGGCGGGGGCGAATTTGATGTTTCAGTC
QCTER	GACTGAAACATCAAATTCGCCCCCGCCAAGGTC
PpGrpE-1 NdeI_F	TTCGTCCATATGAGCAGTCATTACAGGCGT
PpGrpE-1 SalI_R	TTCGTTGTCGACTCACACATTCGCAAGGGC
PpDnaJ EcoRI_F	GAATTCTGTGGTTATTCATCAAGCACA
PpDnaJ XhoI_R	CTCGAGTCACCTGGAAGTCCA
G1	GGTGGACCATCGGTGTTTTTACTG
G2	CCTCCGCAGTCGCTGTAATA
R1	TTGGCGTAATCATGGTCATAGCTGT
R2	CAACGTCGTGACTGGGAAAACCCT

Supplemental Table 2. Constructs

Use	Construct	Plasmid name	Insert or PCR product	Primers	Template	Plasmid backbone	Cloning method
Bacterial overexpression	pTYB12-Hsp70	pTYB12-Hsp70	Hsp70-2	pTYB12-HSP70F/ pTYB12-HSP70R	pART7-Hsp70	pTYB12	SpeI /BamHI
	pTYB12-TV	pTYB12-TV	-	QCTVF/QCTVR	pTYB12-Hsp70	pTYB12	Quikchange
	pTYB12-TE	pTYB12-TE	-	QCTEF/ QCTER	pTYB12-Hsp70	pTYB12	Quikchange
	pTYB12-DnaJ	pTYB12-DnaJ	DnaJ	PpDnaJ EcoRI_F/ PpDnaJ XhoI_R	cDNA	pTYB12	EcoRI/XhoI
	pTYB12-CGE	pTYB12-CGE	CGE	PpGrpE-1 NdeI_F/PpGrp E-1 SalI_R	cDNA	pTYB12	NdeI/SalI
Moss transformation	pBJIR4-TV	pBJIR4-TV	-	QCTVF/QCTVR	pBJIR4	pCR2.1-TOPO	Quikchange
	pBJIR4-TE	pBJIR4-TE	-	QCTEF/QCTER	pBJIR4	pCR2.1-TOPO	