## **Supplementary Materials:**

Proteins	Vmax (n/mol/min/mg)	<i>K</i> <sub>m</sub> (μ <b>M</b> )
DnaK	14.33 (± 1.4) *	179.1 (± 3.7)
DnaK+ PfHsp40	19.65 (± 0.81)	354.6 (± 8.1)
PfHsp70-1	22.45 (± 1.8)	370.8 (± 3.8)
PfHsp70-1+PfHsp40	34.50 (± 0.62)*	34.17 (± 0.5)
KPf	27.6 (± 0.22)	29.58 (± 2.2)
KPf + PfHsp40	42.6 (± 2.1)**	25.65 (± 0.23)

## Table S1. ATPase kinetics of DnaK, PfHsp70-1 and KPf

Table legends: The respective *Vmax* and *Km* values are shown. Standard deviations shown represent three independent assays made using separate protein purification batches. Statistical significance of data was validated by one way ANOVA test (p < 0.05 \*) and (p < 0.01\*\*).

## Table S2. Comparative ATP binding affinities for the Hsp70 proteins

Protein	ATP
	$K_D/\mu M$ (± standard deviation)
KPf	0.38 (± 0.9)*
PfHsp70-1	3.52 (± 1.1)
DnaK	83.80 (± 6.6)

The table shows equilibrium constant ( $K_D$ ) values representing the ATP binding affinities for DnaK, KPf, and PfHsp70-1. The ligand was the immobilized protein mounted onto HTE chip surface, while ATP represented the analyte which was injected at a flow rate of 100 µl/min. The standard deviations shown in parenthesis were obtained from three independent assays and validated using one way ANOVA test (p<0.05\*).





The three-dimensional models for Hsp70s were compared by superimposing them. The comparison structure of the nucleotide binding domain (NBD) and the substrate binding domain (SBD) for (**a**) DnaK (green) versus KPf (red); (**b**) DnaK (green) versus PfHsp70-1(Blue) and also shown is an insert representing top view showing domain orientation (**c**) PfHsp70-1 (blue) versus KPf (red) and also shown is an insert representing top view of the model. The SBD is subdivided into the peptide binding ( $\beta$ -SBD) and the lid ( $\alpha$ -SBD). Models were generated using (2KHO) [33] as template.



**Figure S2. Expression and purification of recombinant Hsp70s.** The respective recombinant proteins were expressed in *E. coli* XL1 Blue cells, purified using affinity chromatography and were analysed with SDS-PAGE (12 %): (a) DnaK; (b) PfHsp70-1; (c) KPf and (d) PfHsp40. Various lanes are denoted as follows: M–Page ruler (Thermo Scientific) in kDa; C—total extract for cells transformed with a neat pQE30 plasmid as control; 0–total extract of cells transformed with pQE30/PfHsp70-1/KPf/DnaK/PfHsp40 prior to IPTG induction; 1 - 6 - total cell lysate obtained hourly up to 6 h post induction; P, S; pellet and soluble fractions obtained from the total lysate of cells transformed with pQE30/PfHsp70-1/KPf/DnaK/PfHsp40, respectively; F- flow though, W-wash fraction; E-eluted fraction.



**Figure S3.** The relative affinities of DnaK, KPf and PfHsp70-1 for the various peptides were determined in the presence or absence of 5 mM ATP/ADP. Comparative relative affinities for peptides: (a) NRLLTG and NRNNTG (b) ALLLMYRR and ANNNMYRR; (c) GFRVVLMYRF and GFRNNNMYRF are depicted as bar graphs. The error bars shown were generated from three assays conducted using independent Hsp70 protein preparations.



**Figure S4: Co-expression and purification of PfAdoMetDC.** The PfAdoMetDC recombinant proteins were coexpressed with different chaperone sets in *E. coli* BL21 Star (DE3) cells and were analysed by SDS-PAGE (12 %) and Western blotting. Various co-expression chaperone sets were as follows: (a) DnaK+DnaJ; (b) KPf+DnaJ; (c) PfHsp70+DnaJ; (d) DnaK+DnaJ+GroEL; (e) KPf+DnaJ+GroEL; (f) PfHsp70+DnaJ+GroEL. Purification of PfAdoMetDC purified following expression with the various chaperone combinations is shown: (g); (h). Various lanes are denoted as follows: M–Page ruler (Thermo Scientific) in kDa; 0 – 5 total cell lysate obtained 0-5 hours IPTG induction; Note, induction with AHT was done 1 hour after IPTG induction. "+C" represents the positive control (purified PfAdoMetDC).



Figure S5. Combined tryptophan and tyrosine florescence spectra of PfAdoMetDC

Shown are the respective intrinsic combined tryptophan and tyrosine fluorescence signals of PfAdoMetDC coexpressed with: (a) Hsp70-DnaJ; (b) Hsp70-DnaJ-GroEL chaperone, respectively. PfAdoMetDC was analysed either in the absence of the substrate, SAM (c) and in the presence of SAM (d), respectively. The chaperone sets are denoted as: PfHsp70+DnaJ (J+Pf); PfHsp70+DnaJ+GroEL (J+Pf+EL); KPf+DnaJ (J+KPf); KPf+DnaJ+GroEL (J+KPf+EL). PfAdoMetDC produced in the absence of supplementary chaperones is represented by 'NC'.