

Table 2

Kinetic and thermodynamic parameters for the folding of proteins that fold with three-state kinetics.

Protein	Reference	PDB code	Contact order	Chain length	Structure	Denaturant	$\Delta G_{U-F}^{H_2O}$ (kcal mol <sup>-1</sup> )	$m$ (kcal mol <sup>-1</sup> M <sup>-1</sup> )	$[D]_{50\%}$ (M)	Temp. (°C)	$k_F^{H_2O}$ (s <sup>-1</sup> )	$\Delta G_{U-I}^{H_2O}$ (kcal mol <sup>-1</sup> )	$k_U^{H_2O}$ (s <sup>-1</sup> )	$m^{I-F}$ (kcal mol <sup>-1</sup> M <sup>-1</sup> )	$\beta_T$
Ubiquitin wild type	[24]	1UBQ	15.1	67	$\alpha/\beta$	GdnHCl	7.2	2.0	3.7	25	350	1.5	0.0012	0.82	0.59
Ubiquitin wild type + 0.4 M Na <sub>2</sub> SO <sub>4</sub>							10.2	2.2	4.6		900	3.1	$9 \times 10^{-5}$	1.05	0.52
Barstar	[54]	1BTA	12.2	89	$\alpha$	Urea	4.8	1.3	3.9	25	31	1.9	0.068	0.17	0.87
<sup>a</sup> CD2, pH 7.0	[55]	1HNG	17.5	98	$\beta$ -sandwich	GdnHCl				25	6.0	0.8	$5.0 \times 10^{-4}$	1.60	<sup>b</sup> 0.68
<sup>a</sup> CD2, pH 4.5						GdnHCl	8.7	5.0	1.7	25	14	1.8	$1.0 \times 10^{-4}$	1.89	0.62
Barnase	[56]	1BNI	11.4	110	$\alpha/\beta$	Urea	10.5	2.3	4.6	25	13	3.2	$1.1 \times 10^{-4}$	0.27	0.88
<sup>c</sup> Suc 1				113	$\alpha/\beta$	Urea	7.2	1.7	4.4	25	65	1.6	0.0001	0.78	0.54
Lysozyme (hen egg white)	[57]	1HEL	10.8	129	$\alpha/\beta$	GdnHCl				20	4	2.7	$6.2 \times 10^{-7}$	0.73	0.75
Lysozyme (hen egg white)	[58]	1HEL				GdnHCl	13.5	2.1	3.8	25	3.5	4.9	$5.0 \times 10^{-5}$	0.43	0.80
CheY	[59]	3CHY	9.0	129	$\alpha/\beta$	Urea	5.2	1.6	3.3	25	2.7	<sup>d</sup> nd	0.012	0.47	0.71
<sup>e</sup> p16				148	$\alpha$	Urea	3.1	1.7	1.9	25	33	1.5	1.4	0.08	0.95
GroEL apical domain (191-345)	[60]	1JON	15.7	154	$\alpha/\beta$	Urea	5.6	2.0	2.8	25	2.3	3.8	0.004	0.45	0.78
Ribonuclease H ( <i>Escherichia coli</i> ), pH 5.5	[61]	2RN2	12.4	155	$\alpha/\beta$	Urea	99.9	2.1	4.7	25	0.6	3.6	$1.69 \times 10^{-5}$	0.42	0.80
Ribonuclease H ( <i>E. coli</i> ), pH 5.5	[62]	2RN2	12.4	155	$\alpha/\beta$	GdnHCl	9.5	5.2	1.8	25	4.1	4.8	$3.7 \times 10^{-5}$	1.91	0.63
N-terminal domain from PGK	[58]	1PHP	11.5	175	$\alpha/\beta$	GdnHCl	8.4	7.6	1.1	25	9.5	5.2	0.03	1.24	0.84
<sup>h</sup> C-terminal domain from PGK	[63]	1PHP	8.0	219	$\alpha/\beta$	GdnHCl	13.6	13	1.0	25	0.03	3.4	$7.6 \times 10^{-10}$	7.1	0.45

<sup>a</sup>Some care must be taken in comparing these proteins as the values are calculated using denaturant activity not concentration of denaturant. <sup>b</sup>Calculated assuming the equilibrium  $m$  value does not vary significantly with pH. <sup>c</sup>F. Rousseau, J.W.H. Schymkowitz, M. Sánchez del Pino and L.S. Itzhaki, unpublished observations. <sup>d</sup>Not determined as it requires knowledge of the *cis-trans* isomerisation ratio in the unfolded state. <sup>e</sup>L. Itzhaki, personal

communication. <sup>f</sup>Average of values obtained from CD and fluorescence experiments. <sup>g</sup>Calculated from the fit of the kinetic data to a three-state model. <sup>h</sup>The folding of the C-terminal domain of PGK is monitored in the presence of the N-terminal domain (fluorescence probe is only in C-terminal domain) – the domains act independently.

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