

How Do Thermophilic Proteins and Proteomes Withstand High Temperature?

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Supporting Material

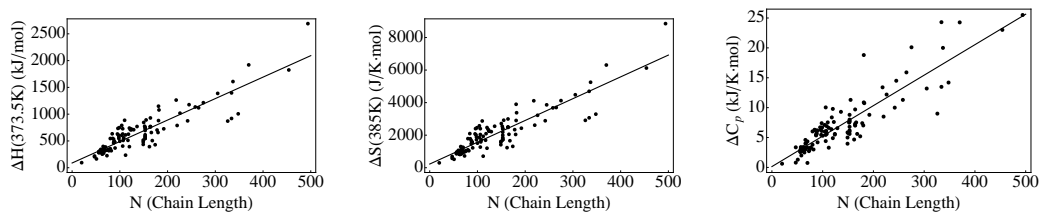


Figure 1: Change in enthalpy, entropy and specific heat upon unfolding for 116 different proteins. Solid lines show the best linear fit and filled circles show data. Table 1 in this supplemental material lists all these proteins. Based on the regression analysis we find $\Delta H(373.5) = (4.0N + 88)$ kJ/mol, $\Delta S(385) = (13.39N + 226)$ J/K-mol and $\Delta C_p = 0.051N + 0.14$ kJ/K-mol.

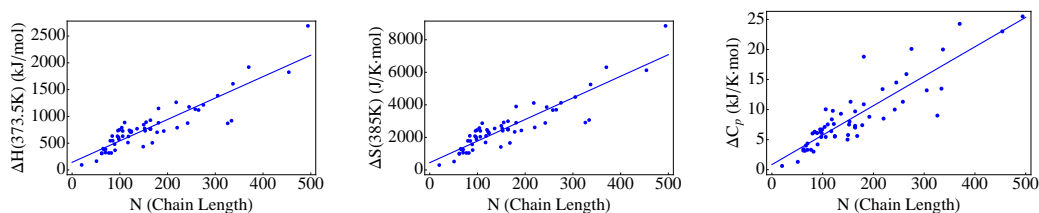


Figure 2: Change in enthalpy, entropy and specific heat upon unfolding for mesophiles is linearly dependent on the chain length N of the protein.

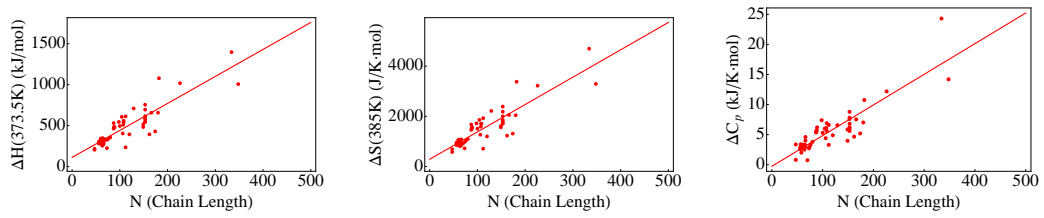


Figure 3: Change in enthalpy, entropy and specific heat upon unfolding for thermophiles is linearly dependent on the chain length N of the protein.

Table 1: Thermodynamic Parameters of the proteins

Protein	PDB	N _{res}	$\Delta H(T_m)$ ($\frac{\text{kJ}}{\text{mol}}$)	$\Delta S(T_m)$ ($\frac{\text{J}}{\text{mol K}}$)	ΔC_p ($\frac{\text{kJ}}{\text{mol K}}$)	T_m (K)	$\Delta H(373.5)$ ($\frac{\text{kJ}}{\text{mol}}$)	$\Delta S(385)$ ($\frac{\text{J}}{\text{mol K}}$)
S16 (Thermo) (1)	3BN0	112	270.0	702.6	3.3	384.3	234.4	708.6
BPTI (2)	5PTI	58	317.0	840.8	2.0	377	310.0	883.0
CheY (Thermo) (3)	1TMY	120	395.3	1056.9	4.9	374	392.8	1198.8
Sso7 (4)	1SSO	62	274.0	736.6	2.7	372	278.1	829.3
T4 lysozyme (K5I/K39V mutant) (5)		47	213.4	573.6	3.4	372	218.4	689.2
MGMT (6)	1MG1	174	419.0	1127.6	5.2	371.6	428.9	1311.8
Albumin (K5I Mutant) (7)	1PRB	47	200.8	544.3	0.8	369	204.4	578.0
Cold Shock Tm (4)	1G6P	66	259.0	705.7	4.6	367	288.9	926.0
Ribosomal L30E (8)	1H7M	100	459.0	1251.4	5.3	366.8	494.5	1508.0
Tendamistat (2)	3AIT	74	307.0	838.8	2.9	366	328.8	985.6
Stefin A (2)	1CYV	98	473.0	1300.2	7.4	363.8	544.8	1719.3
Sac7d (4)	1AZQ	66	245.0	673.6	3.0	363.7	274.4	844.4
TRP Repressor (2)	2OZ9	107	448.0	1233.1	6.1	363.3	510.2	1587.0
Parvalbumin (2)	5CPV	108	500.0	1377.4	5.6	363	558.8	1706.9
Ubiquitin (2)	1UBQ	76	308.0	848.5	3.3	363	342.7	1042.7
HistidineBSt (9)		88	414.2	1146.4	5.7	361.3	484.1	1510.5
ONC (10)	1ONC	104	530.0	1469.0	6.0	360.8	606.2	1858.5
HT c552 (11)	1AYG	80	305.9	848.5	3.8	360.5	355.3	1098.4
B1 of Protein G (2)	1PGB	56	258.0	715.7	2.6	360.5	291.8	886.0
Thioredoxin (2)	2TRX	108	447.3	1242.5	6.9	360	540.5	1705.8
β -Lactoglobulin (12)		162	325.9	907.5	4.7	359.1	393.1	1232.4
RNaseH (Thermo) (4)	1RIL	166	548.1	1526.7	7.5	359	657.3	2053.2
Eglin C (13)	2TEC	70	312.7	871.5	0.8	358.8	323.7	924.4
OMTKY3 (2)	2OVO	56	240.0	670.0	2.6	358.2	279.8	857.6

Protein	PDB	N _{res}	$\Delta H(T_m)$ ($\frac{\text{kJ}}{\text{mol}}$)	$\Delta S(T_m)$ ($\frac{\text{J}}{\text{mol K}}$)	ΔC_p ($\frac{\text{kJ}}{\text{mol K}}$)	T _m (K)	$\Delta H(373.5)$ ($\frac{\text{kJ}}{\text{mol}}$)	$\Delta S(385)$ ($\frac{\text{J}}{\text{mol K}}$)
SPCI (14)		180	548.1	1531.4	7.0	357.9	657.6	2043.9
Metmyoglobin (rat) (15)		153	473.0	1324.9	6.2	357	575.3	1793.1
Metmyoglobin (raccoon) (15)		153	465.0	1308.8	6.8	355.3	588.8	1854.7
Histidine Bh (9)		87	364.0	1024.5	5.4	355.3	463.0	1461.2
Subtilisin Inhibitor (2)	3SSI	226	795.5	2239.4	12.2	355.21	1018.4	3221.1
Metmyoglobin (horse) (15)	1YMB	153	548.0	1545.0	7.8	354.7	694.6	2184.3
Alkaline Proteinase Inhibitor (16)	1JIW	106	320.5	904.1	4.4	354.5	404.2	1267.7
Metmyoglobin (opossum) (15)	1MBO	153	435.0	1230.6	5.6	353.5	547.0	1708.6
Myoglobin (whale) (2)	1MBO	153	575.0	1628.4	8.8	353.1	754.5	2389.6
Btk (4)	1AWX	64	196.0	555.2	3.1	353	259.6	824.2
Iso2 Cytochrome (17)	1YEA	112	473.2	1342.8	6.6	352.4	612.7	1927.6
B2 of Protein G (2)	1PGX	56	238.0	675.4	2.9	352.4	299.2	931.6
Metmyoglobin (armadillo) (15)	1YMB	153	417.0	1184.7	6.0	352	546.0	1722.3
Sem5 (18)	1KFZ	60	271.0	769.9	3.3	352	342.0	1065.6
SRBP (19)	1RBP	182	836.8	2384.0	10.8	351	1078.7	3378.0
Cold Shock Bc (20)	1C9O	66	245.0	700.2	4.0	349.9	339.4	1082.6
Lysozyme (chicken) (21)	1HEL	129	550.3	1575.7	6.6	349.2	709.7	2216.0
Histidine St (9)	1Y4Y	87	397.5	1140.3	5.4	348.6	531.0	1672.6
Bergerac D48G mutant (22)	1BK2	57	225.0	648.0	3.1	347.2	306.7	969.2
Rhodopsin (bovine) (23)	1U19	348	630.0	1815.6	14.2	347	1006.3	3291.2
Phycocyanin (Thermo) (24)		334	753.0	2170.0	24.3	347	1396.9	4695.2
CI2 (2)	1COA	64	280.0	807.4	2.5	346.8	346.8	1069.2
Alpha 3D (25)	2A3D	73	150.6	435.1	2.7	346.2	224.9	724.0
HistidineBS (9)	1SPH	88	322.2	931.2	5.6	346	475.1	1525.0
Bergerac SHH mutant (22)	2OAW	65	230.6	667.8	3.3	345.3	323.4	1026.1
Tec (4)	1GL5	63	169.0	491.3	2.9	344	254.6	817.8

Protein	PDB	N _{res}	$\Delta H(T_m)$ ($\frac{\text{kJ}}{\text{mol}}$)	$\Delta S(T_m)$ ($\frac{\text{J}}{\text{mol K}}$)	ΔC_p ($\frac{\text{kJ}}{\text{mol K}}$)	T _m (K)	$\Delta H(373.5)$ ($\frac{\text{kJ}}{\text{mol}}$)	$\Delta S(385)$ ($\frac{\text{J}}{\text{mol K}}$)
Fyn (4)	1SHF	64	233.0	678.1	3.3	343.6	331.7	1053.5
Barstar (2)	1BTA	89	292.0	851.6	6.2	342.9	481.7	1569.5
glutamate dehydrogenase domain II (26)	1B26	149	301.2	879.3	5.9	342.6	482.2	1562.8
Odorant Binding Protein (27)	1A3Y	149	391.2	1143.2	4.0	342.2	515.5	1611.0
Irk (4)	2RNA	57	178.0	520.5	3.4	342	285.1	923.1
Abl (4)	1BBZ	63	194.0	568.1	3.3	341.5	299.6	963.7
Metmyoglobin (carp) (15)		153	370.0	1084.1	7.6	341.3	614.7	1999.7
NPS (28)		89	282.0	827.5	6.1	340.8	481.5	1571.3
cytochrome b562 (29)	1QPU	106	393.3	1156.1	10.0	340.2	727.7	2398.3
ESBL (30)	4BLM	265	585.8	1722.9	15.9	340	1118.5	3699.3
Transferrin (Apo)	1BTJ	337	927.0	2731.3	20.0	339.4	1609.0	5252.6
Bergerac SHA mutant (22)		70	237.5	700.4	4.3	339.1	386.9	1251.9
RNaseH (Meso) (4)	2RN2	155	502.1	1481.1	11.3	339	892.0	2919.0
Bergerac-SH3 (22)	1SHG	62	197.0	581.1	3.2	339	306.8	985.9
α -Spectrin (4)	1SHG	62	197.0	581.1	3.4	339	314.3	1013.7
Lac Repressor Headpiece (2)	1LCD	51	118.0	349.1	1.3	338	164.2	518.3
PsbQ (31)		149	256.5	759.3	5.0	337.8	435.0	1413.2
T4 Lysozyme (Pseudo WT) (5)	1L63	164	581.0	1721.0	9.7	337.6	929.2	2995.4
Rnase B (32)	1RBB	124	520.0	1542.1	5.6	337.2	723.3	2284.5
Histidine Ec (33)	1POH	85	317.1	942.1	6.2	336.6	547.0	1779.1
maltose binding protein (34)	1OMP	370	1010.0	3006.0	24.3	336	1920.0	6309.6
Phycocyanin (Meso) (24)		334	414.0	1232.1	13.5	336	919.5	3067.0
Rnase A (4)	5RSA	124	500.0	1489.0	5.5	335.8	707.4	2241.0
α -Chymotrypsinogen (2)	2CGA	245	619.0	1847.8	14.5	335	1177.3	3864.1
Arabinose Binding Protein (2)	1ABE	305	840.0	2530.1	13.2	332	1387.8	4483.0
Endoglucanase 3 (35)	1H8V	218	707.1	2129.8	13.4	332	1263.2	4114.5

Protein	PDB	N _{res}	$\Delta H(T_m)$ ($\frac{\text{kJ}}{\text{mol}}$)	$\Delta S(T_m)$ ($\frac{\text{J}}{\text{mol K}}$)	ΔC_p ($\frac{\text{kJ}}{\text{mol K}}$)	T _m (K)	$\Delta H(373.5)$ ($\frac{\text{kJ}}{\text{mol}}$)	$\Delta S(385)$ ($\frac{\text{J}}{\text{mol K}}$)
Kunitz type soybean trypsin inhibitor (29)	1AVU	181	428.9	1291.7	10.9	332	880.3	2902.9
EIN (36)		258	665.3	2003.9	11.3	332	1134.3	3677.5
S16 (Meso) (1)		117	244.0	735.6	6.4	331.7	511.5	1689.3
Subtilisin BPN (2)	2ST1	275	370.0	1116.1	20.1	331.5	1214.2	4123.4
Orosomucoid (37)		181	350.0	1057.4	18.8	331	1149.0	3898.6
flavodoxin (29)	1FTG	168	264.0	799.3	5.6	330.3	506.2	1658.5
Lamba-Repressor 6-85 (29)	1LMB	80	284.5	861.6	6.0	330.2	545.4	1786.7
Cold Shock A (4)	1MJC	70	181.0	548.5	3.3	330	324.6	1057.2
TRP-engineered (38)	1L2Y	20	67.4	204.2	0.6	329.9	94.4	299.8
CheY (Meso) (3)	3CHY	120	294.8	898.9	9.8	328	740.3	2467.7
T4 lysozyme (K48[A]) (5)		164	430.0	1312.6	7.3	327.6	765.1	2491.2
T4 lysozyme (A42K) (5)		164	440.0	1343.9	7.0	327.4	762.7	2478.3
T4 lysozyme (S44[A]) (5)	1L63	164	442.0	1350.4	7.1	327.3	770.0	2503.2
ApoLipoprotein A1 (39)	2A01	242	410.0	1253.8	10.0	327	875.0	2886.7
Staphylococcus Nuclease (2)	1STN	136	337.0	1030.6	9.3	327	769.5	2549.1
CT-Acp (40)	2VH7	98	290.0	886.9	6.1	327	573.7	1882.9
ECAD2 (41)	1EDH	106	376.6	1151.7	5.4	327	629.6	2039.9
Hisactophilin (42)	1HCE	118	234.3	717.2	8.4	326.7	626.0	2091.5
Cold Shock B (4)	1CSP	67	193.0	590.9	4.0	326.6	380.6	1249.0
Barnase (2)	1RNB	110	529.7	1623.8	7.5	326.2	885.9	2871.8
IL-1 β (2)	6I1B	153	351.0	1076.7	8.0	326	731.0	2406.8
Acyl Carrier Protein (apo) (2)	1ACP	77	160.0	491.2	3.3	325.7	317.7	1044.0
α -sarcin (43)		150	569.0	1747.5	5.8	325.6	845.4	2714.4
Rnase T1 (4)	9RNT	104	442.3	1362.4	7.1	324.6	789.9	2575.8
Stefin B (2)	1STF	95	293.0	906.6	6.7	323.2	630.0	2078.9
N1 CBD of CenC (44)		152	391.4	1213.6	7.5	322.5	773.9	2542.2

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ApoCytochrome b5 (45)		90	149.3	464.4	4.2	321.5	367.7	1221.4
Rnase Sa (4)	1RGG	96	407.5	1268.0	6.3	321.4	734.7	2401.8
ADK (Yeast) (46)	1AKY	220	340.0	1060.2	8.5	320.7	788.8	2613.4
Flagellin Monomer (47)	1UCU	494	1338.9	4177.5	25.5	320.5	2690.4	8853.2
PA c551 (11)	451C	82	157.7	492.4	3.0	320.3	317.3	1044.3
Rnase Sa3 (4)		99	391.6	1223.0	6.7	320.2	748.2	2456.0
Type III Antifreeze (48)	1MSI	65	228.5	714.4	3.1	319.8	397.4	1298.2
Glycoprotein (AGP) (49)	3BX6	192	252.0	788.5	8.8	319.6	726.3	2426.8
CPS (28)		84	283.0	890.2	6.3	317.9	633.3	2096.7
YopM (50)	1JL5	454	538.9	1696.8	23.0	317.6	1825.3	6125.5
AdaC (6)	1SFE	178	284.0	896.5	7.4	316.8	703.6	2339.3
α -Lactalbumin (2)	1ALC	122	276.1	872.9	7.6	316.3	710.8	2366.7
Rnase Sa2 (4)		97	286.2	911.1	5.4	314.1	609.3	2018.4
SBP (Apo) (51)		326	308.0	990.4	9.0	311	870.5	2911.4

Mesophilic Species	ΔH^\ddagger	Γ	T_{Opt}	\bar{L}
B. Megaterium (52)	19.7	26	315.2	272
B. Subtilis (52)	16.9	30	314.5	294
E. Coli (53)	27.1	51	314 .1	301
L. Monocytogenes (54)	25.9	223	309.7	303
P. Aeruginosa (52)	21.6	101	311.9	328
P. Fluorescens (52)	28.9	166	311.7	340
Thermophilic Species	ΔH^\ddagger	Γ	T_{Opt}	\bar{L}
B. Acidocaldarius (55)	15.6	107	339.2	306
M . Thermoautotrophicus (56)	29.8	81	341.2	281
T. Aquaticus (52)	19.1	3	347	284
T. Brockii (57)	48.8	22	345.3	304
T. Neopolitana (58)	28.4	1	348	313
T. Thermophilus (59)	10.6	2	346.9	299

Table 2: Fitted Parameters from Proteome Analysis: ΔH^\ddagger , Γ , and T_{Opt} are activation barrier, essential proteins, and optimum growth temperature, respectively, calculated from fitting equation 11 (in the manuscript) to experimental data. \bar{L} is average length of all the proteins found within a given proteome.

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