Bioinformation by Biomedical Informatics Publishing Group *open access*

www.bioinformation.net **Hypothesis**

Structural features differentiate the mechanisms between 2S (2 state) and 3S (3 state) folding of homodimers.

Lei Li[1], Kannan Gunasekaran[2], Jacob Gah-Kok Gan[1], Cui Zhanhua[1], Paul Shapshak[3], Meena Kishore Sakharkar[1], and Pandjassarame Kangueane[1*]

[1]School of Mechanical and Aerospace Engineering, Nanyang Technological University, Singapore 639798; Basic Research Program, SAIC-Frederick, Inc., Laboratory of Experimental and Computational Biology, National Cancer Institute, Frederick, MD 21702, USA; [3]Dementia/HIV Laboratory, Elliot Building Room 2013, Department of Psychiatry and Beh Sci, University of Miami Miller Medical School, 1800 NW 10th Avenue, Miami, Florida 33136;

Pandjassarame Kangueane* - Email: mpandjassarame@ntu.edu.sg; * Corresponding author

received July 07, 2005; revised August 28, 2005; accepted September 2, 2005; published online September 2, 2005

Abstract:

The formation of homodimer complexes for interface stability, catalysis and regulation is intriguing. The mechanisms of homodimer complexations are even more interesting. Some homodimers form without intermediates (two-state (2S)) and others through the formation of stable intermediates (three-state (3S)). Here, we analyze 41 homodimer (25 `2S` and 16 `3S`) structures determined by X-ray crystallography to estimate structural differences between them. The analysis suggests that a combination of structural properties such as monomer length, subunit interface area, ratio of interface to interior hydrophobicity can predominately distinguish 2S and 3S homodimers. These findings are useful in the prediction of homodimer folding and binding mechanisms using structural data.

Keywords: homodimer; structural difference; 2 state; 3 state; stable intermediate; folding mechanism

Abbreviations:

2S	2 state homodimer
3S	3 state homodimer
3SMI	3 state homodimer with monomer intermediate
3SDI	3 state homodimer with dimer intermediate
ML	monomer length
B/2	subunit interface area
F_{hp}	fraction of interface to interior hydrophobicity

Background:

Equilibrium denatruation experiments (using temperature and chemical agents) are employed to analyze the unfolding of proteins. These studies are useful in understanding monomeric protein folding. Recently, such techniques have been used to study the mechanism of homodimer formation. **[1]** Dimer folding involves both intra-molecular and inter-molecular interaction, unlike monomer folding that involves only intra-molecular interaction. It is known that some dimers denature from native dimer to unfolded monomers with no thermodynamically stable intermediates, whereas others have folded intermediates during the process. **[1, 2, 3]** Based on the unfolding patterns, homodimers are known to exist in three different states. They are (1) two-state (2S), (2) three-state with dimeric intermediate (3SDI) and (3) three-state with monomeric intermediate (3SMI). 2S refers to $N_2 \leftrightarrow 2U$ mechanism, 3SDI refers to $N_2 \leftrightarrow I_2 \leftrightarrow 2U$ and 3SMI refers to $N_2 \leftrightarrow 2I \leftrightarrow 2U$, where N_2 is the native dimer state, I is the intermediate monomeric species, I_2 is the intermediate dimeric species, and U is unfolded monomeric state. 3SDI and 3SMI are commonly considered as three-state (3S). It is found that 2S interfaces are similar to protein cores and 3SMI interfaces resemble the monomer surfaces. **[4]** 2S and 3SMI dimerization were also studied by following the evolution of two identical 20-letter residue chains within the framework of a lattice model, using Monte Carlo simulations. **[5]** It is found that folding of 2S sequences depend on a significantly larger number of conserved amino acids than 3SMI sequences. The effects of the monomer and interface geometry on 2S and 3S association mechanism were also studied by the energetically minimally frustrated Gō model. **[6]** It is found that the native protein 3D structure is the major factor that governs the choice of binding mechanism.

Bioinformation by Biomedical Informatics Publishing Group *open access*

www.bioinformation.net **Hypothesis**

Mei and colleagues investigated the importance of 2S and 3S dimers using structural and folding data. **[2]** Apiyo and colleagues proposed (using 13 obligomers (multimers with permanent interfaces)) that small obligomers (molecular mass < 20 kDa) unfold through 2S. **[7]** On the other hand, large obligomers (molecular mass > 35 kDa) unfold through oligomeric intermediate (3SDI) and those with intermediate size unfold through monomeric intermediate (3SMI). Moreover, Levy and colleagues proposed (using 21 homodimers) that 2S and 3SMI dimers can be effectively classified based on the ratio of intra-molecular/inter-molecular contacts and interface hydrophobicity. **[6]** Here, we created an extended dataset of 41 homodimers (2S: 25; 3SDI: 6; and 3SMI: 10) to design a methodology for the discrimination of 2S, 3SDI and 3SMI dimers using 3D structural properties.

Methods:

Dataset creation

We created a dataset consisting of 41 homodimer complex structures (2S: 25; 3SDI: 5; and 3SMI: 10) from Protein Databank (PDB). **[8]** The unfolding pathways for these dimers observed using thermodynamic experiments were obtained from literature (Table 1). The selected homodimers are at least 40 residues per monomer.

Analyses of 2S and 3S homodimers

Interface area

The solvent accessible surface area (ASA) was computed using the program NACCESS. **[9]** The dimeric interface area (B) was calculated as ΔASA (change in ASA upon complex formation from monomer to dimer state). **[10]** We then calculated subunit interface area (B/2), due to the two-fold symmetry of homodimer complexes.

Interior, interface and exterior residues

Homodimer residues were classified into three categories (interior, interface and exterior) based on relative ASA. The percentage relative ASA was obtained by dividing the accessible surface area by the total surface area of a side-chain in an extended conformation in the tripeptide GXG. Exterior residues were defined as having a relative ASA > 5%, interior residues were defined as having a relative ASA < 5% and interface residues were defined satisfying the conditions ΔASA > 1Å^2 & relative ASA < 5%. The 5% cut-off was optimized elsewhere by Miller *et al*., **[11]**

Fraction of interface to interior Hydrophobicity (F_{hp})

F_{hp} (Fraction of interface to interior hydrophobicity) was defined by the equation $(H_{inf}-H_{ext})/(H_{int}-H_{ext})$, where H_{int} is interior hydrophobicity, H_{inf} is interface hydrophobicity and H_{ext} is exterior hydrophobicity. The individual hydrophobicity values were calculated using the equation $\Sigma n_i h_i/\Sigma n_i$, where n_i is the number of residue type i and h_i is hydrophobicity value (based on SES (solvent excluded surface) & SAS (solvent accessible surface)) of type I, as described elsewhere. **[12]**

Small and large homodimers

By definition, small homodimers were defined as those with ML (monomer length) less than the dataset mean length (185 residues). By definition, large homodimers were defined as those with ML larger than the dataset mean length (185 residues).

Homodimers with small and large B/2

By definition, homodimers with small B/2 were defined as those whose B/2 is less than the dataset mean B/2 (1424 Å^2). By definition, homodimers with large B/2 were defined as those whose B/2 is larger than the dataset mean B/2 (1424 Å^2).

Results:

Distribution of 2S and 3S in a Cartesian plane of monomer length and subunit interface area

Figure 1 shows the distribution of 2S and 3S in the Cartesian plane consisting of ML (monomer length) and B/2 (subunit interface area). It shows that 76% of small proteins form 2S and 60% of large proteins form 3S homodimers. Figure 1 also shows that 68% of 2S have large interface area and 45% of 3S have small interface area. 2S have ML in the range of 45-270 residues and 3S have ML in the range of 70-850 residues. However, 3SMI lie within 90-380 residues and 3SDI lie within 70-850 residues. 2S and 3S dimers have significantly different ML range ($p = 0.05$ in F test). Nonetheless, 2S and 3SMI have similar ML range ($p = 0.05$ in F test). The dataset mean ML is 185 residues. This lies between 2S mean (125 residues) and 3S mean (282 residues). Data also show that 2S and 3S ML means are different ($p < 0.05$). The mean ML for 3SDI is 405 and this is much greater than the mean ML for 2S (125) and 3SMI (208).

The B/2 range for 2S (650 - 2500 Å^2) and 3S (300 - 2317 Å^2) are overlapping and are not significantly different ($p = 0.21$). However, 3SMI and 3SDI are distinguished by the B/2 range ($p < 0.05$). 3SMI having small B/2 range (300-1550 Å^2) and 3SDI having large B/2 range (1350-2317 Å^2) are distinguished from each other. The dataset mean for B/2 is 1424Å^2, which lies between 2S mean (1509 Å^2) and 3S mean (1239 Å^2). Interestingly, the 3SMI mean (1068 Å^2) is close to 3S mean B/2 ($p = 0.25$) and 3SDI mean (1705 Å^2) is close to 2S mean B/2 ($p = 0.35$).

In Figure 1, the distribution of 2S and 3S were divided into four regions (G1 to G4) based on the dataset mean of ML and B/2. Entries in G1 are small proteins with large B/2 and entries in G4 are large proteins with small B/2 (refer to methodology section for definition of small and large proteins). However, entries in G2 are small proteins with small B/2 and those in G3 are large proteins with large B/2. This grouping shows 84% of homodimers in G1 are 2S and 66% of homodimers in G4 are 3S. Nevertheless, homodimers in G3 there are 44% 2S and 56% 3S. Homodimers in G2 have 67% 2S and 33% 3S. It should be observed that 3S in G2 are solely 3SMI. The results show that 2S and 3S are distinctly and prevalently distinguished in G1 and G4 but not as much in G2 and G3. The distribution of 2S and 3S in regions G1 to G4 provide insight to their structural preference in terms of ML and B/2.

Exterior, interior and interface hydrophobicity in 2S and 3S

Table 1 gives the hydrophobicity of interior, interface and exterior residues for 2S, 3SDI and 3SMI. It also gives the mean hydrophobicity of interior, interface and exterior residues for 2S, 3SDI and 3SMI in the dataset. Very small 2S (≤ 90 residues) have greater interface hydrophobicity compared to interior hydrophobicity. However, this is not true with very large 2S (> 90 residues). It is also interesting to observe that majority of 3SMI have less interface hydrophobicity compared to interior hydrophobicity. Nonetheless, this is not true with a majority of 3SDI. Table 1 shows that the mean interface hydrophobicity values satisfy a condition (2S > 3SDI > 3SMI). However, the mean interior hydrophobicity satisfy a different condition (2S > (3SDI = = 3SMI)). The ratio of interface to interior hydrophobicity is ~1 for 2S and 3SDI, while it is < 1 for 3SMI.

F_{hp} (Factor of interface to interior hydrophobicity) value in 2S and 3S

Figure 1, shows that 92% of entries in G1 have high F_{hp} value (> 0.5) and 83% of entries in G4 have low F_{hp} value (< 0.5). It also shows that 3S in G1 have high F_{hp} value and 2S in G4 have low F_{hp} value. Interestingly, 75% of entries in G2 have high F_{hp} value and 78% of entries in G3 have high F_{hp} value. Moreover, Figure 1 show that 91% 2S in G1 have high F_{hp} value and 75% 3S in G4 have low F_{hp} value. However, 100% 3S (2 entries) in G1 have high F_{hp} value and 100% 2S (2 entries) in G4 have low F_{hp} value. In G2, 75% of 2S have high F_{hp} value and 67% of 3S have high F_{hp} value. Nonetheless, 100% 3S have high F_{hp} value and 50% of 2S have high F_{hp} value in G3. The mean F_{hp} value for 2S and 3SDI is 1, while it is 0.5 for 3SMI. Thus, the distribution of 2S and 3S in the G1 to G4 regions is described.

Discussion:

The mechanism of homodimer folding and binding has been investigated using denaturation experiments. **[14-52]** 3 dimensional structures are also available for many of these homodimers with known folding and binding mechanisms (Table 1). The folding and binding homodimer data collected from the literature is classified into three 2S, 3SMI and 3SDI. The study of homodimer folding and binding using energy models is computational intensive and time consuming. Alternatively, study on their folding and binding using structural data is found useful. **[2]** Recently, Mei and colleagues documented the differences between 2S, 3SMI and 3SDI homodimers using 3S structure data. **[2]** The study provided structural insight to the mechanism of 2S and 3S folding. However, the analysis did not document parameters to differentiate 2S, 3SMI and 3SDI homodimers using structural data. In this study, we study an extended dataset of homodimer complexes to distinguish 2S and 3S homodimers using structural features. Results show that 76% of small proteins are 2S homodimers and 60% of large proteins are 3S homodimers. Thus, protein size plays an important role in determining the pathways of homodimer folding and binding. The result also shows that 68% of 2S have large subunit interface area and 45% of 3S have small subunit interface area. These observations suggest the importance of protein size and subunit interface area in determining the mechanism of homodimer formation.

Bioinformation by Biomedical Informatics Publishing Group *open access*

www.bioinformation.net **Hypothesis**

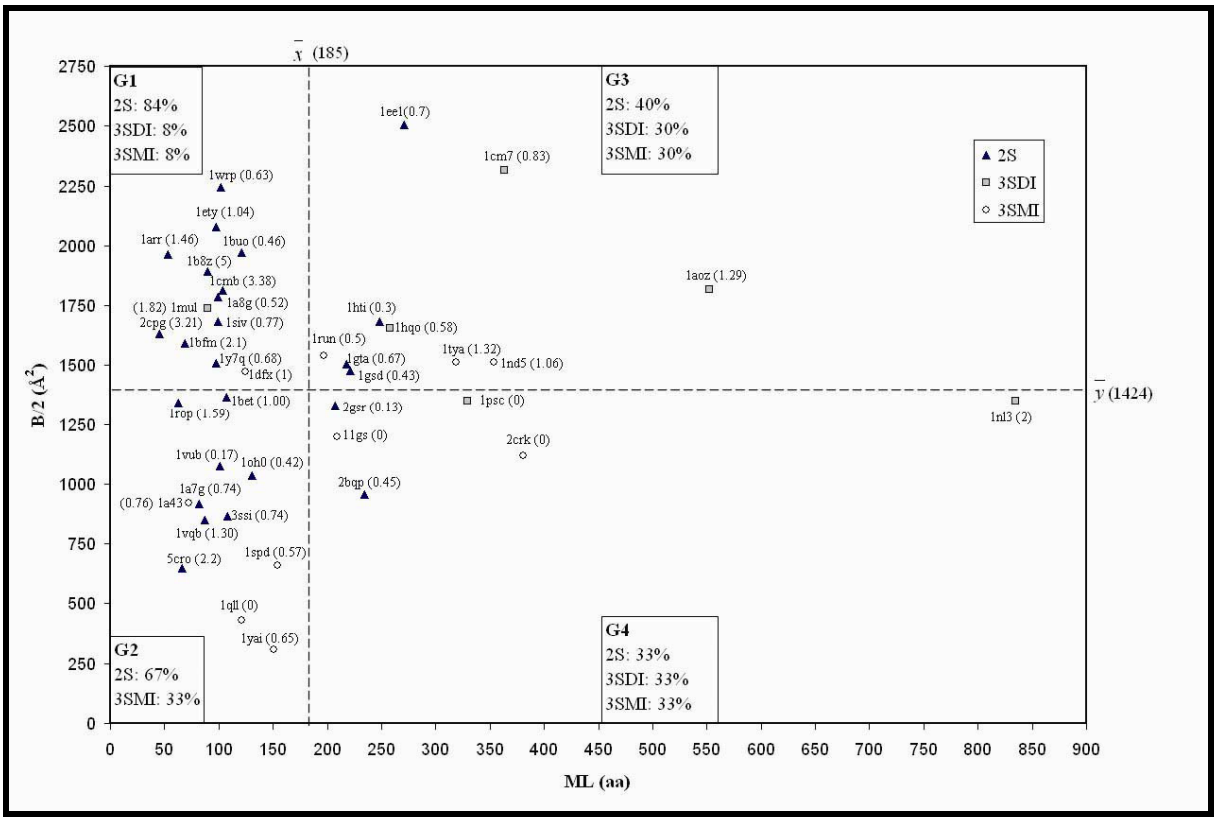

Figure 1: Correlation between monomer length (ML) and subunit interface area (B/2) for three groups of homodimers. 2S: two-state; 3SDI: three-state with dimeric intermediate; 3SMI: three-state with monomeric intermediate. The two dash lines through 185 aa and 1424Å^2 represent mean monomer length and mean B/2 for all homodimers, respectively. They classify the dimers into four regions (G1, G2, G3 and G4). The distributions of 2S, 3SDI and 3SMI dimers are given for each region. The value within parentheses is hydrophobicity factor (F_{hp}), calculated by the equation $(H_{inf} - H_{surf})/(H_{int} - H_{surf})$, where H_{inf} is interface hydrophobicity, H_{int} is interior hydrophobicity and H_{surf} is surface hydrophobicity.

The distribution of 2S and 3S in the G1 and G4 regions of Figure 1 show difference between them based on protein size, subunit interface area and F_{hp}. In G1, 84% dimers are 2S and 92% of dimers have high F_{hp} (> 0.5). Thus, entries with high F_{hp} are grouped in G1 and this region represents small proteins with large subunit interface area. Moreover, 91% of 2S in G1 have high F_{hp}. This implies that a majority of small proteins with large subunit interface area and high F_{hp} are 2S. 3S in G1 have high F_{hp} and this explains the presence of exceptional 3S entries in G1. Similarly, 66% of dimers are 3S and 83% of dimers have low F_{hp} (< 0.5) in G4. Thus, entries with low F_{hp} are grouped into G4 and this region represents large proteins with small subunit interface area. Furthermore, 75% 3S in G4 have low F_{hp}. 2S in G4 have low F_{hp} and this explains the presence of unusual 2S entries in G4. Entries in G2 and G3 have a mixture of 2S and 3S with low and high F_{hp} values. This is different to the distribution in G1 and G4. 100% 3S and 50% 2S in G3 have high F_{hp} and thus dimers in G3 are not distinguished by their folding mechanisms using structural parameters. The mean F_{hp} for 2S and 3SDI is 1, while it is 0.5 for 3SMI. The similarity between 2S and 3SDI in F_{hp} is interesting. It implies that binding after folding displayed by 3SMI resembles the association of protein-protein complexes. **[13]** However, the cooperative folding-binding displayed by 2S and 3SDI resembles a single-chain folding.

Bioinformation by Biomedical Informatics Publishing Group *open access*

www.bioinformation.net **Hypothesis**

Table 1: Dataset of homodimeric proteins divided into three groups according to their unfolding pathways

PDB ID	Chain	Protein name	Cofactors	Source	ML (aa)	B/2(Å^2)	Hydrophobicity			Reference
							H_{int}	H_{inf}	H_{surf}	
2S (25)										
2cpg	A&B	transcriptional repressor CopG	-	Streptococcus agalactiae	45	1632	0.37	0.68	0.23	[14]
1arr	A&B	arc repressor	-	Bacteriophage P22	53	1962	0.47	0.58	0.23	[15]
1rop	(Sym)	repressor of protein Rop	-	E. coli	63	1345	0.41	0.51	0.24	[16]
5cro	A&C	Cro repressor	-	Bacteriophage lambda	66	648	0.49	0.73	0.29	[17]
1bfm	A&B	Histone B	-	Methanothermus fervidus	69	1593	0.50	0.72	0.30	[18]
1a7g	(Sym)	E2 DNA-binding domain	-	HPV strain 16E2	82	918	0.6	0.52	0.29	[19]
1vqb	(Sym)	gene V protein	-	Bacteriophage f1	87	850	0.58	0.66	0.31	[20]
1b8z	A&B	histone-like protein HU	-	Thermotoga maritima	90	1894	0.26	0.67	0.23	[21]
1ety	A&B	FIS protein	-	E. coli	98	2079	0.49	0.5	0.25	[22]
1y7q	A&B	SCAN domain of ZNF 174	-	Homo sapiens	98	1508	0.67	0.54	0.26	[23]
1a8g	A&B	HIV-1 protease	-	HIV type 1	99	1785	0.63	0.49	0.33	[24]
1siv	A&B	SIV protease	-	SIV	99	1684	0.59	0.53	0.33	[24]
1vub	A&B	CcdB		E. coli	101	1074	0.51	0.36	0.33	[25]
1cmb	A&B	Met repressor	-	E. coli	104	1813	0.35	0.54	0.27	[26]
3ssi	(Sym)	subtilisn inhibitor	-	Streptomyces albogriseolus	108	866	0.51	0.46	0.32	[27]
1wrp	(Sym)	trp repressor	-	E. coli	108	2243	0.69	0.54	0.29	[28]
1bet	(Sym)	β-nerve growth factor	-	Mus musculus	107	1366	0.47	0.47	0.31	[29]
1buo	(Sym)	Btb domain from PLZF protein	-	Homo sapiens	121	1972	0.56	0.41	0.28	[30]
1oh0	A&B	ketosteroid isomerase	-	Pseudomonas putida	131	1036	0.49	0.41	0.31	[31]
2gsr	A&B	class π glutathione s-transferase	-	Sus scrofa	207	1331	0.5	0.3	0.27	[32]
1gsd	A&B	glutathione transferase A1-1	-	Homo sapiens	208	1477	0.57	0.4	0.27	[33]
1gta	(Sym)	glutathione transferase	-	Schistosoma japonica	218	1505	0.5	0.42	0.26	[34]
2bqp	A&B	pea lectin	Mn & Ca ion	Garden pea	234	955	0.49	0.37	0.27	[35]
1hti	A&B	triosephosphate isomerase	-	Homo sapiens	248	1685	0.54	0.35	0.27	[36]
1ee1	A&B	Nh(3)-dependent Nad(+) synthetase	-	Bacillus subtilis	271	2507	0.51	0.43	0.24	[37]
3SDI (6)										
1mul	(Sym)	histone-like protein hu-α	-	E. coli	90	1739	0.47	0.61	0.3	[38]
1hqo	A&B	Ure2 Protein	-	Saccharomyces cerevisiae	258	1656	0.54	0.44	0.3	[39]
1psc	A&B	parathion hydrolase	Cd ion	Brevundimonas diminuta	329	1353	0.5	0.3	0.3	[40]
1cm7	A&B	3-isopropylmalate dehydrogenase	-	E. coli	363	2317	0.5	0.47	0.28	[41]
1aoz	A&C	ascorbate oxidase	Cu ion	Green zucchini	552	1817	0.43	0.47	0.29	[42]
1nl3	A&B	SecA	-	Mycobacterium tuberculosis	835	1351	0.46	0.64	0.28	[43]

3SMI (10)									
1a43	(Sym)	C-terminal domain of HIV-1 capsid protein	-	HIV type 1	72	921	0.47	0.42	0.26 [44]
1qll	A&B	lysine-49 phospholipase A2	-	Bothrops jararacussu	121	432	0.38	0.17	0.27 [45]
1dfx	(Sym)	desulfoferrodoxin	Fe & Ca ion	Desulfovibrio desulfuricans	125	1472	0.44	0.44	0.29 [7]
1yai	B&C	cu, zn superoxide dismutase	Cu & Zn ion	Photobacterium leiognathi	151	309	0.48	0.41	0.28 [46]
1spd	A&B	cu, zn superoxide dismutase	Cu & Zn ion	Homo sapiens	154	658	0.49	0.4	0.28 [47]
1run	A&B	cAMP receptor protein	-	E. coli	197	1542	0.66	0.47	0.28 [48]
11gs	A&B	glutathione-s-transferase	-	Homo sapiens	209	1197	0.5	0.28	0.3 [49]
1tya	(Sym)	tyrosyl-tRNA synthetase	-	Bacillus stearothermophilus	319	1513	0.48	0.55	0.26 [50]
1nd5	A&B	prostatic acid phosphatase	-	Homo sapiens	354	1512	0.43	0.44	0.27 [51]
2crk	(Sym)	creatine kinase	-	Oryctolagus cuniculus	381	1119	0.46	0.18	0.25 [52]
Average for 2S					125	1509	0.51	0.50	0.28
SD					65	475	0.10	0.12	0.03
Average for 3SDI					405	1705	0.48	0.49	0.29
SD					259	358	0.04	0.14	0.01
Average for 3SMI					208	1067	0.48	0.38	0.27
SD					107	468	0.07	0.13	0.02

ML=monomer length; B/2 = subunit interface area. 2S=two-state; 3SDI=three-state with dimeric intermediate; 3SMI=three-state with monomeric intermediate. SIV = Simian immunodeficiency virus; HIV=Human immunodeficiency virus; HPV=Human papillomavirus; Ccdb = controller of cell division or death B protein; PLZF=promyelocytic leukemia zinc finger protein; FIS=factor for inversion stimulation. (sym) indicates that the dimer is generated from a single chain in the PDB by Protein Quaternary Structure Server (PQS)[53]. Interior hydrophobicity (H_{int}), interface hydrophobicity (H_{inf}) and surface hydrophobicity (H_{surf}) for each dimer were calculated, separately, by the single equation $\sum n_i h_i / \sum n_i$, where n_i is the number of residue type i and h_i is ASA hydrophobicity factor (based on SES (solvent excluded surface) & SAS (solvent accessible surface)) of residue type i from Pacios. **[12]**

Thus, we show that small homodimers with large interface area and high F_{hp} are prevalently 2S. Similarly, large homodimers with small interface area and low F_{hp} are prevalently 3S. Hence, it is possible to distinguish 2S and 3S dimers using 3D structural data. However, small homodimers with small interface area and large homodimers with large interface area are not significantly distinguished into 2S and 3S using structural parameters ML, B/2 and F_{hp}. It should be noted that the conclusion made in the report are based on a limited set of homodimers given in Table 1.

Conclusion:

The mechanisms of homodimer complexations have implications in drug discovery. However, elucidation of homodimer mechanism using unfolding experiments is difficult. Prediction of homodimer folding and binding using structural data has application in target validation. Here, we show that small proteins with large interface area and high F_{hp} form 2S. We also show that large proteins with small interface area and low F_{hp} form 3S. Therefore, it is feasible to differentiate 2S and 3S homodimers using structural data.

Acknowledgement:

Lei Li acknowledges Nanyang Technological University, Singapore for support.

References:

[1] K. E. Neet *et al., Protein Sci.* 3:2167 (1994) [PMID: 7756976]
[2] G. Mei, *et al., Febs J* 272:16 (2005) [PMID: 15634328]
[3] A. Mazzini, *et al., Biochim Biophys Acta* 1599:90 (2002) [PMID: 12479409]
[4] C. J. Tsai, *et al., Protein Sci* 6:1793 (1997) [PMID: 9300480]
[5] G. Tiana, *et al., Proteins* 49:82 (2002) [PMID: 12211018]
[6] Y. Levy, *et al., Proc Natl Acad Sci U S A* 101:511 (2004) [PMID: 14694192]
[7] D. Apiyo, *et al., Biochemistry* 40:4940 (2001) [PMID: 11305909]
[8] H. M. Berman, *et al., Nucleic Acids Res* 28:235 (2000) [PMID: 12037327]
[9] Hubbard, (1993). NACCESS, computer program, Department of Biochemistry and Molecular Biology, University College London, UK.
[10] S. Jones, *et al., Proc Natl Acad Sci U S A* 93:13 (1996) [PMID: 8552589]
[11] S. Miller, *et al., Nature* 328:834 (1987) [PMID: 3627230]
[12] L. F. Pacios, *J Chem Inf Comput Sci* 41:1427 (2001) [PMID: 11604044]
[13] L. Lo Conte, *et al., J Mol Biol* 285:2177 (1999) [PMID: 9925793]
[14] T. E. Wales, *et al., Protein Sci* 13:1918 (2004) [PMID: 15169951]
[15] M. E. Milla, *et al., Biochemistry* 33:1125 (1994) [PMID: 8110744]
[16] C. Steif, *et al., Biochemistry* 32:3867 (1993) [PMID: 8471599]
[17] R. Jana, *et al., J Mol Biol* 273:402 (1997) [PMID: 9344748]
[18] T. B. Topping, *et al., J Mol Biol* 342:247 (2004) [PMID: 15313621]
[19] Y. K. Mok, *et al., Protein Sci* 5:310 (1996) [PMID: 8745409]
[20] H. Liang, *et al., Biochemistry* 30:2772 (1991) [PMID: 2007116]
[21] J. Ruiz-Sanz, *et al., Eur J Biochem* 271:1497 (2004) [PMID: 15066175]
[22] T. B. Topping, *et al., J Mol Biol* 335:1065 (2004) [PMID: 14698300]
[23] J. R. Stone, *et al., J Biol Chem* 277:5448 (2002) [PMID: 11741982]
[24] S. K. Grant, *et al., Biochemistry* 31:9491 (1992) [PMID: 1390732]
[25] K. Bajaj, *et al, Biochem J* 380:409 (2004) [PMID: 14763902]
[26] C. M. Johnson, *et al., Biochemistry* 31:9717 (1992) [PMID: 1390748]
[27] A. Tamura, *et al., J Mol Biol* 249:636 (1995) [PMID: 7783216]
[28] L. M. Gloss, *et al., J Mol Biol* 312:1121 (2001) [PMID: 11580254]
[29] D. E. Timm, et al., *Biochemistry* 33:4667 (1994) [PMID: 8161524]
[30] X. Li, *J Biol Chem* 272:27324 (1997) [PMID: 9341182]
[31] D. H. Kim, *et al., Protein Sci* 10:741 (2001) [PMID: 11274465]
[32] H. W. Dirr, *et al., Biochem Biophys Res Commun* 180 :294 (1991) [PMID: 1930226]
[33] L. A. Wallace, *et al., Biochemistry* 37:5320 (1998) [PMID: 9548764]
[34] W. Kaplan, *et al., Protein Sci* 6:399 (1997) [PMID: 9041642]
[35] N. Ahmad, *et al., Biochemistry* 37:16765 (1998) [PMID: 9843447]
[36] V. Mainfroid, *et al.,J Mol Biol* 257:441 (1996) [PMID: 8609635]
[37] Z. W. Yang, *et al., Protein Sci* 13:830 (2004) [PMID: 14978314]
[38] J. Ramstein, *et al., J Mol Biol* 331:101 (2003) [PMID: 12875839]
[39] L. Zhu, *et al., J Mol Biol* 328:235 (2003) [PMID: 12684011]
[40] J. K. Grimsley, *et al., Biochemistry* 36:14366 (1997) [PMID: 9398154]

[41] C. Motono, *et al., Biochemistry* 38:1332 (1999) [PMID: 9930995]
[42] G. Mei, *et al., Biochemistry* 36:10917 (1997) [PMID: 9283082]
[43] S. M. Doyle, *et al., Biochemistry* 39:11667 (2000) [PMID: 10995234]
[44] M. G. Mateu, *J Mol Biol* 318:519 (2002) [PMID: 12051856]
[45] R. Ruller, *et al., Arch Biochem Biophys* 411:112 (2003) [PMID: 12590929]
[46] F. Malvezzi-Campeggi, *et al., Arch Biochem Biophys* 370:201 (1999) [PMID: 10510278]
[47] M. E. Stroppolo, *et al., Arch Biochem Biophys* 377: 215 (2000) [PMID: 10845696]
[48] J. Malecki, *et al., Eur J Biochem* 243: 660 (1997) [PMID: 9057829]
[49] A. Aceto, *et al., Biochem J* 285:241 (1992) [PMID: 1637306]
[50] Y. C. Park, *et al., J Biol Chem* 273:18052 (1998) [PMID: 9660761]
[51] P. Wojciak, *et al., Int J Biol Macromol* 32:43 (2003) [PMID: 12719131]
[52] Y. Liang, *et al., J Biol Chem* 278:30098 (2003) [PMID: 12771138]
[53] K. Henrick, *et al., Trends Biochem Sci* 23:358 (1998) [PMID: 9787643].

Edited by SANDEEP Kumar
Citation: Li *et al.,* Bioinformation 1(2): 42-49 (2005)