Supporting Information

Inhibition of GNNQQNY prion peptide aggregation by trehalose: a mechanistic view

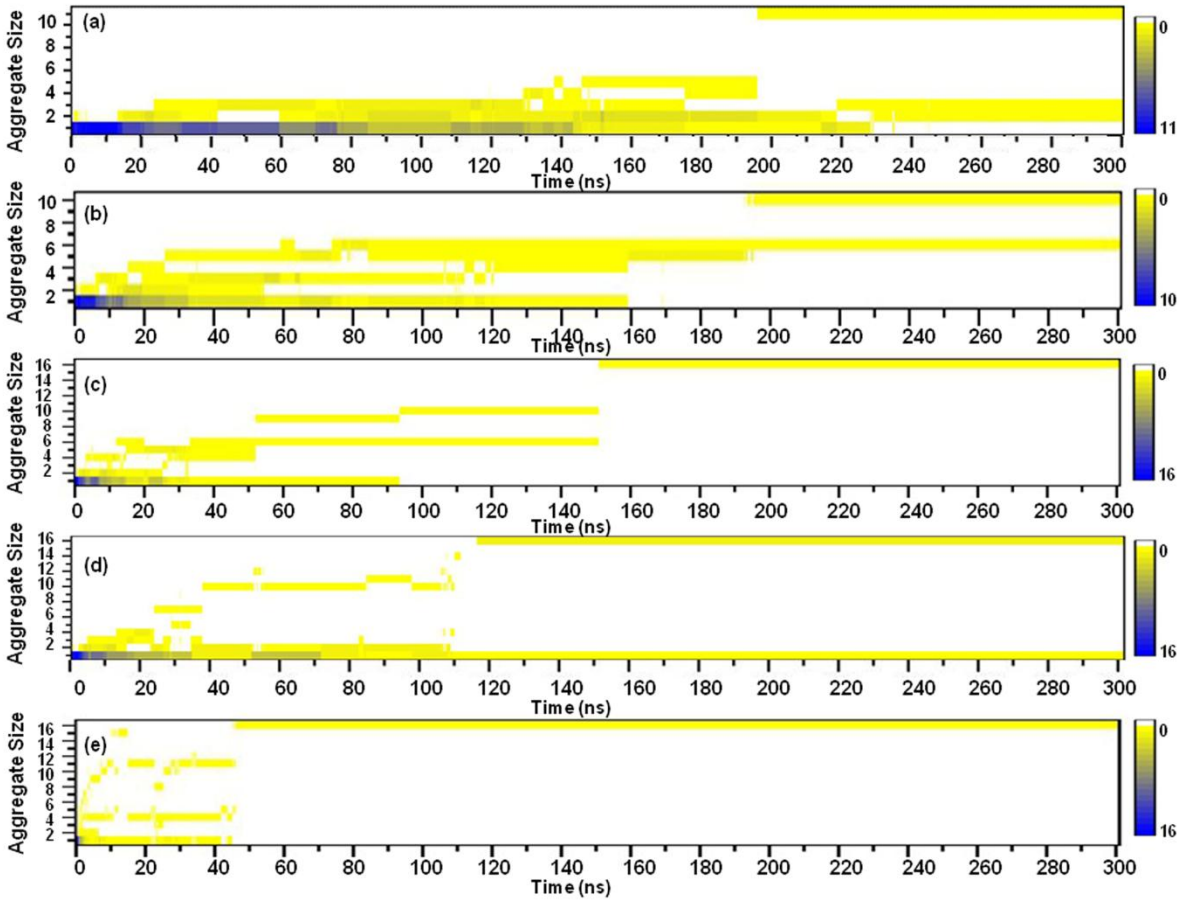
Nidhi Katyal, Shashank Deep

Indian Institute of Technology-Delhi, India

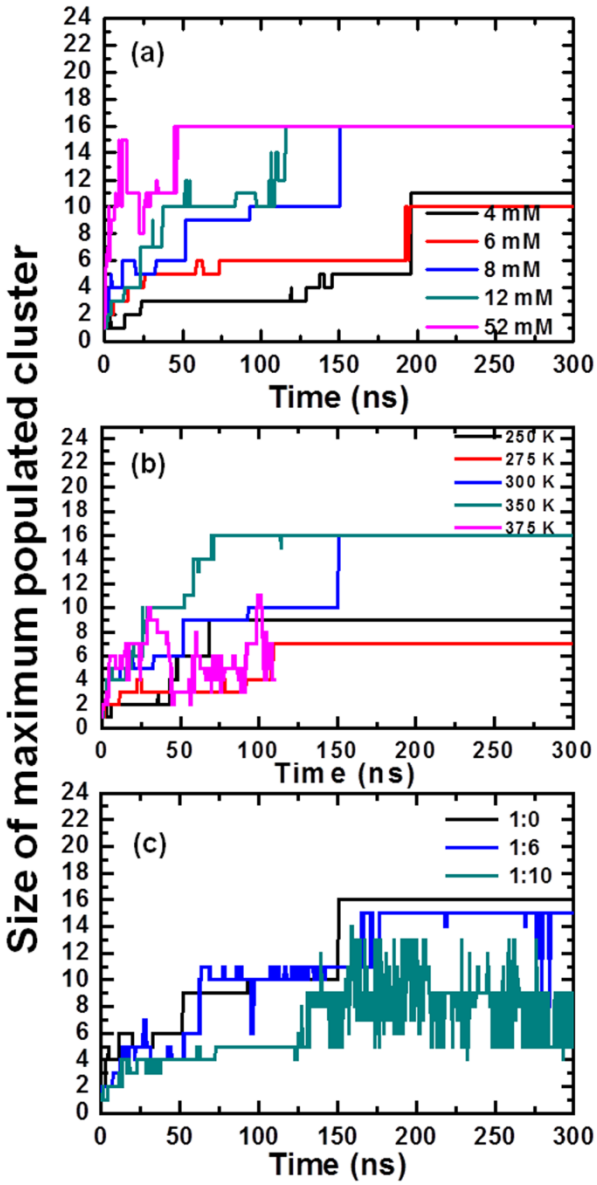
Email:sdeep@chemistry.iitd.ac.in

S.I Table 1: Conditions used for various MD simulations of peptide:

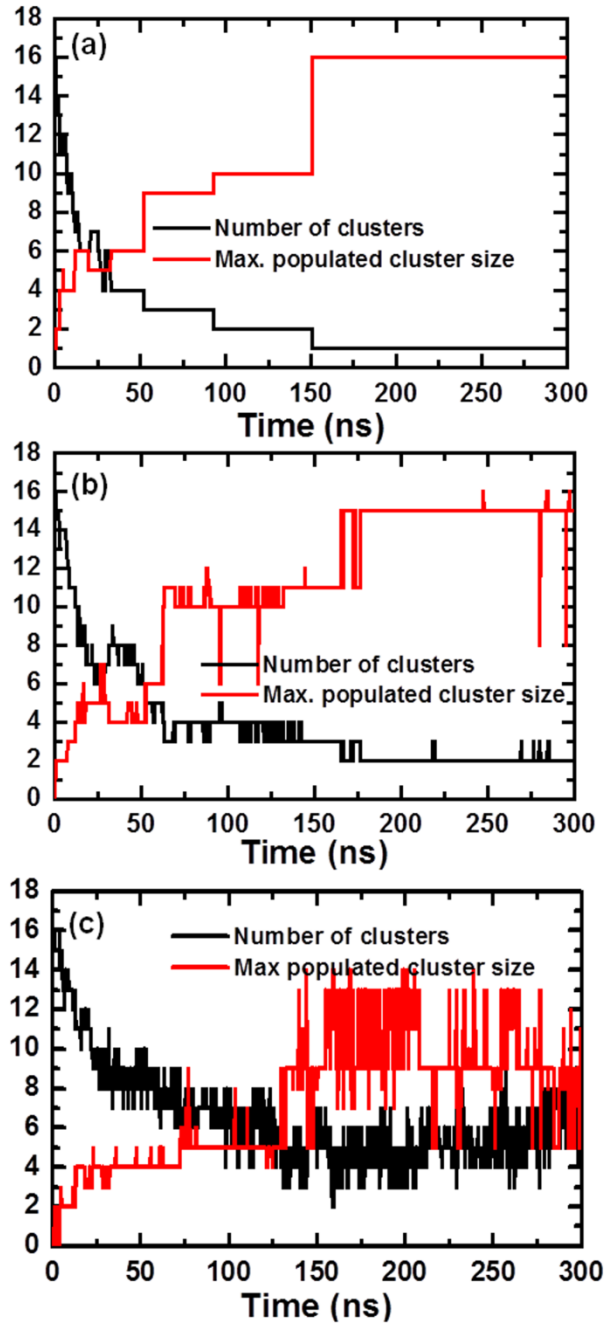
Temperature (K)	Peptide concentration (mM)	Number of peptide molecules	Number of trehalose molecules	Number of water molecules	Box length (Å)	Simulation length (ns)
250	8	16	0	90655	139.58	300
300	4	16	0	228260	191.6	300
	6	16	0	135708	161.02	300
	8	16	0	69217	128.85	300
	8	16	32	68172	128.37	300
	8	16	96	66445	127.75	300
	8	16	166	64506	127.04	300
	12	16	0	72742	131.01	300
	52	16	0	16460	82.23	300
350	8	16	0	90655	142.89	300
375	8	16	0	90655	144.33	112



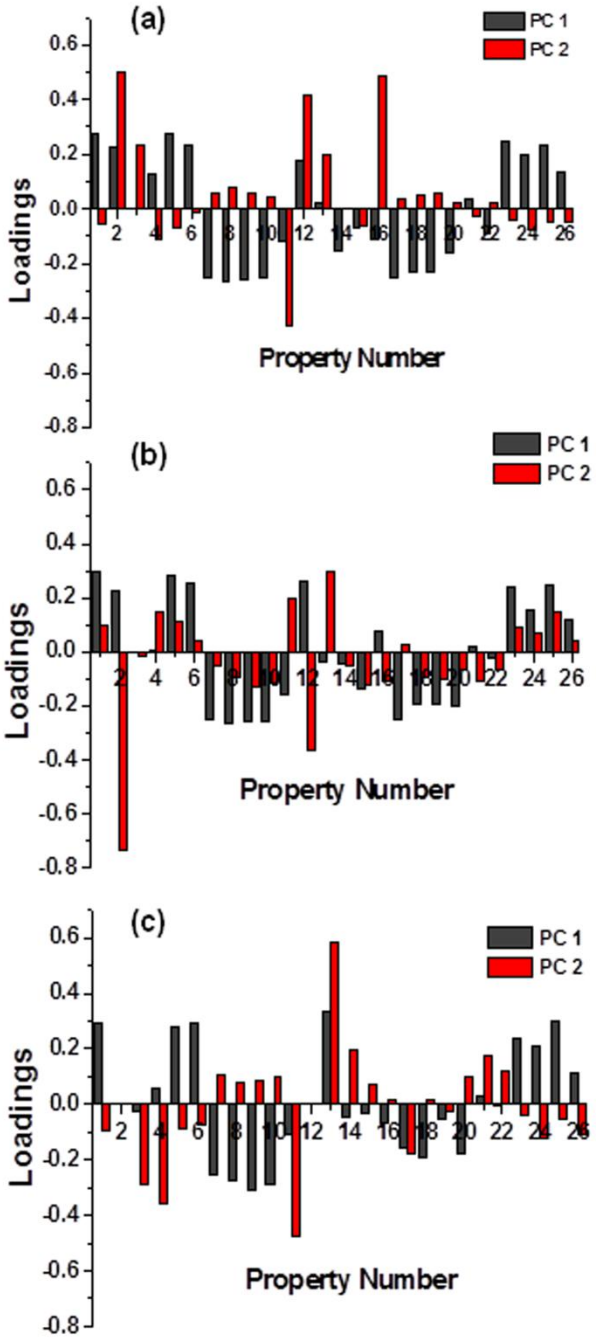
SI Fig 1: Time evolution of aggregate size at different peptide concentrations (a) 4 mM (b) 6 mM (c) 8 mM (d) 12 mM and (e) 52 mM. Color bar on the right represents number of peptides.



SI Fig 2: Time evolution of the size of the maximum populated cluster at different (a) peptide concentrations (b) temperatures and (c) peptide:trehalose molar ratio keeping peptide concentration fixed at 8 mM.

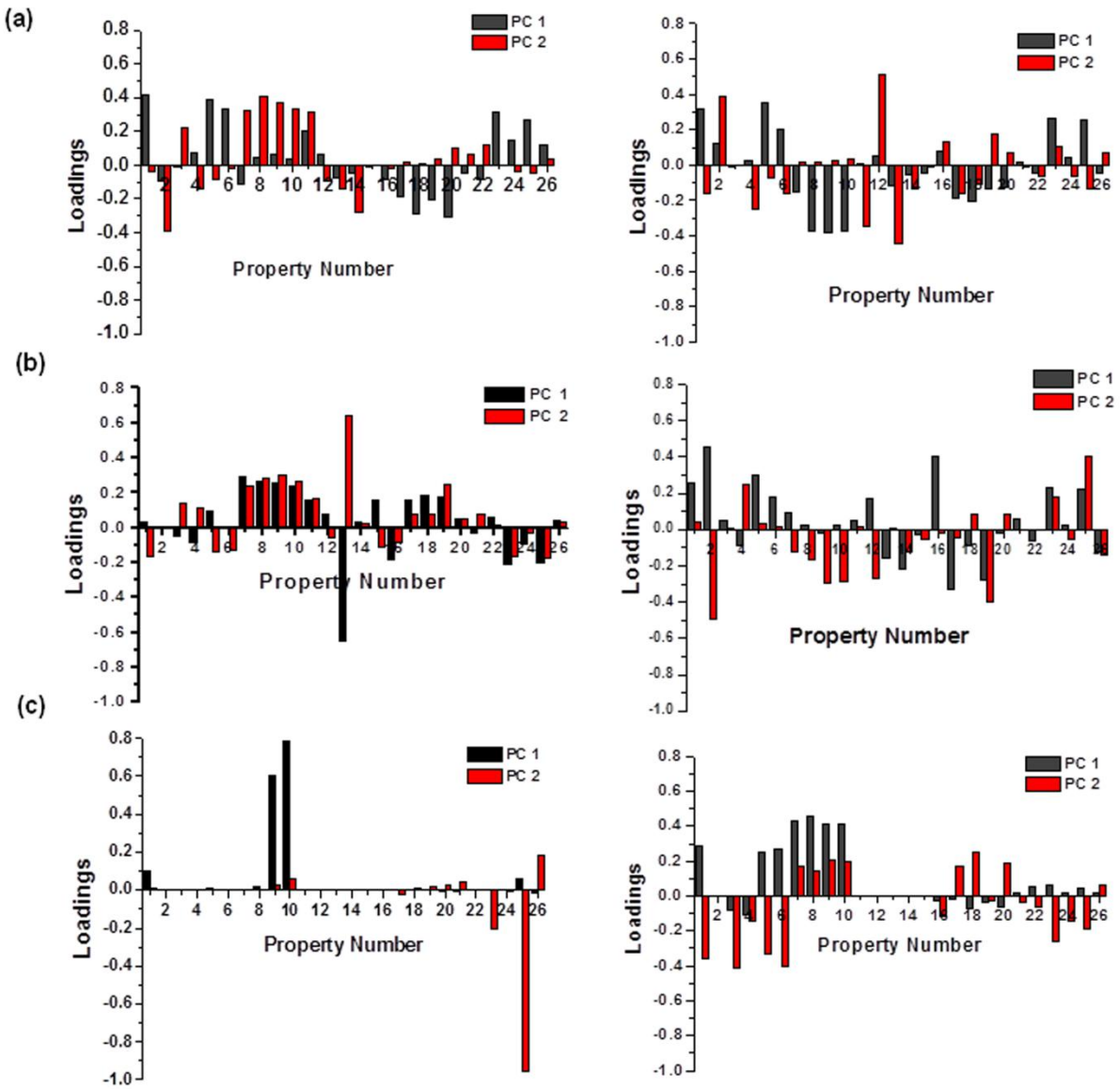


SI Fig 3: Simultaneous time evolution of number of clusters and maximum populated cluster size at 8 mM peptide concentration in the presence of following peptide:trehalose molar ratio: (a) 1:0 (b) 1:6 and (c) 1:10.



SI Fig 4: Bar graph representing loadings of 26 different properties for first two principal components in the presence of 8 mM peptide concentrations at different peptide:trehalose molar ratio: (a) 1:0 (b) 1:6 and (c) 1:10. Properties are: (1) Interchain contacts, (2) Beta-sheet contacts, (3) Polar order parameter P1, (4) Nematic order parameter P2, (5) Polar atom contacts, (6) Non-polar atom contacts, (7) Hydrophobic solvent accessible surface area, (8) Hydrophilic solvent accessible surface area, (9) Polar atom contacts with water, (10) Non-

polar atom contacts with water, (11) Coil content, (12) Beta-sheet content, (13) Beta-bridge content, (14) Bend content, (15) Turn content, (16) End to end distance, (17) Inter-mainchain coulombic energy, (18) Inter-mainchain Lennard Jones energy, (19) Inter-sidechain coulombic energy, (20) Inter-sidechain Lennard Jones energy, (21) Intra-mainchain coulombic energy, (22) Intra-mainchain Lennard Jones energy, (23) Mainchain-water coulombic energy, (24) Mainchain-water Lennard Jones energy, (25) Sidechain-water coulombic energy, (26) Sidechain-water Lennard Jones energy.

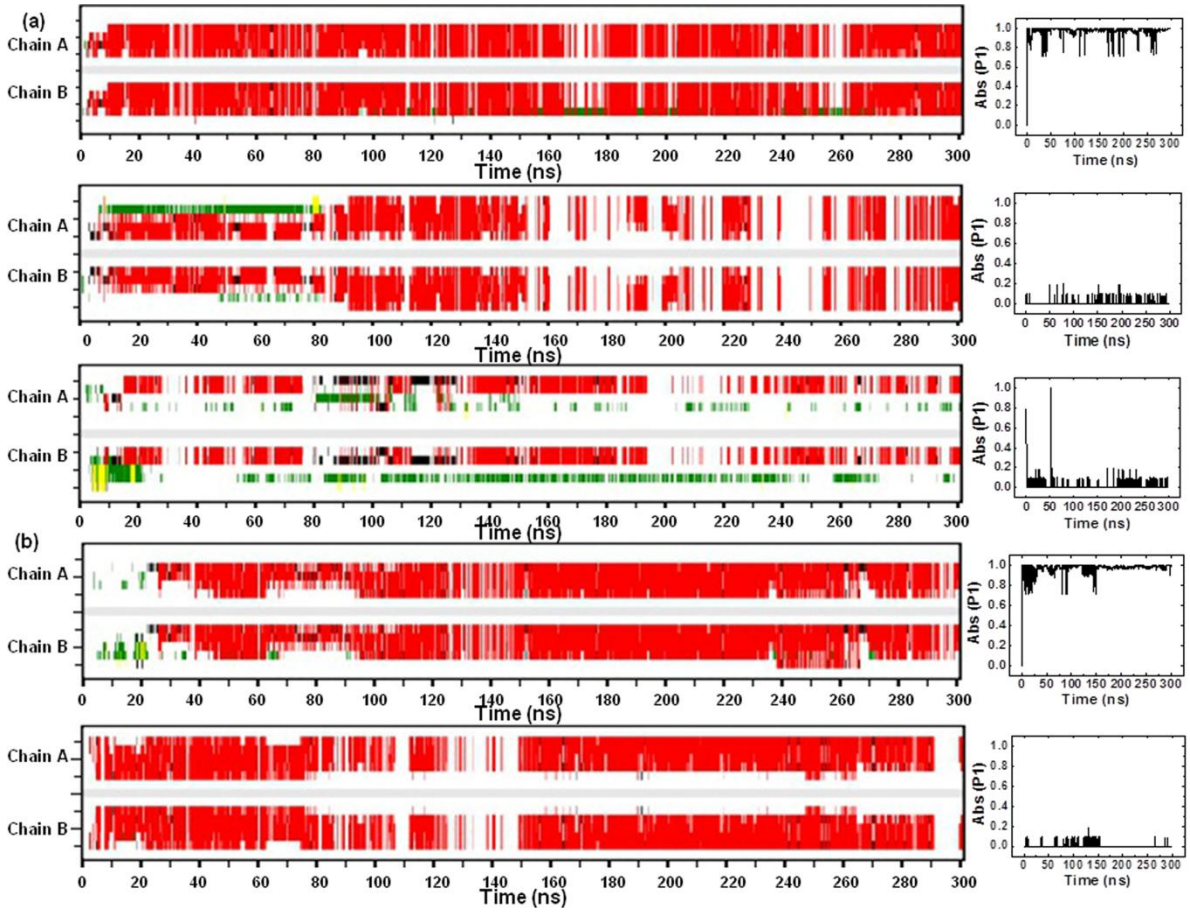


SI Fig 5: Bar graph representing loadings of 26 different properties for first two principal components in the presence of 8 mM peptide concentrations at different peptide:trehalose

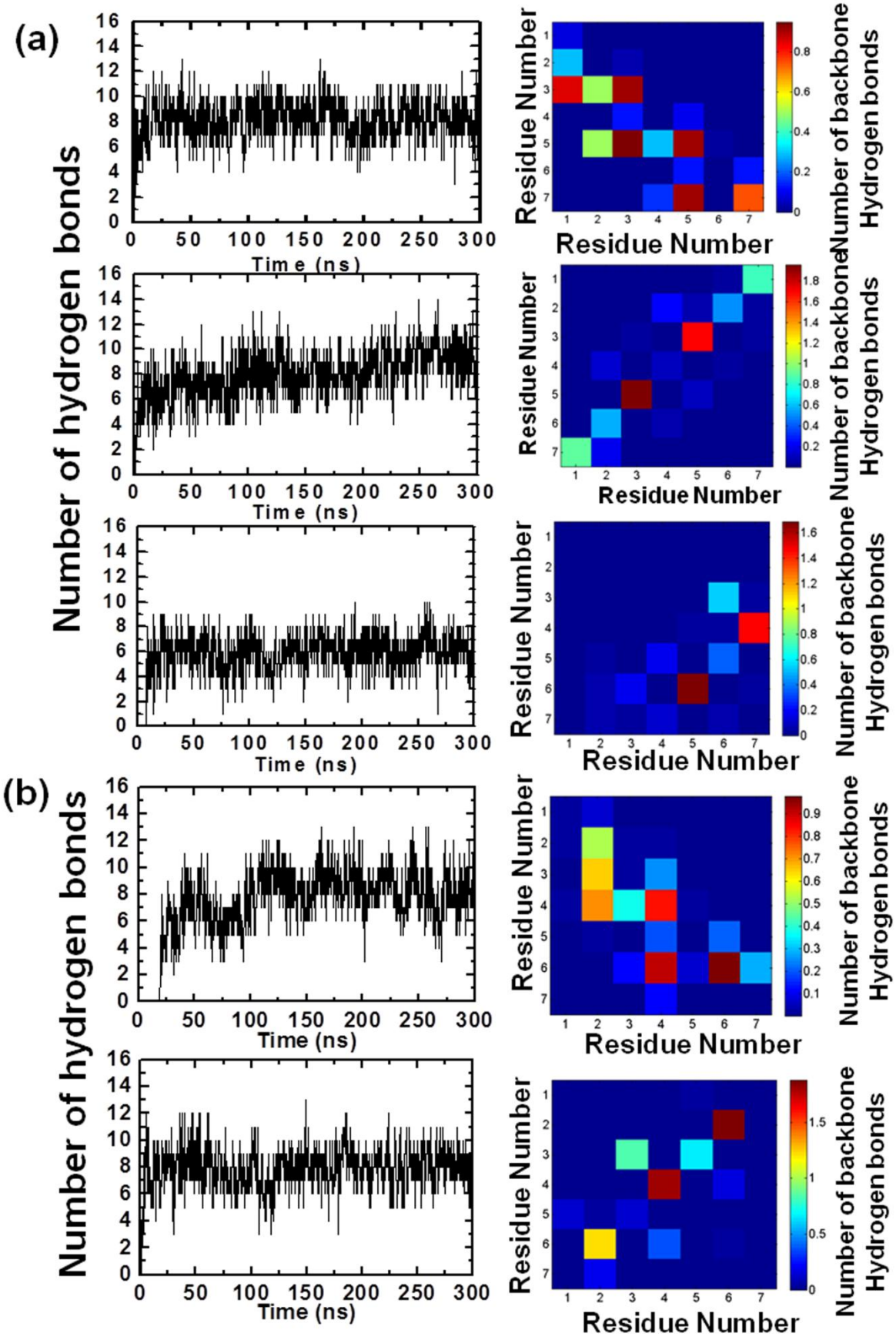
molar ratio: (a) 1:0 (b) 1:6 and (c) 1:10. Left panel represents that during the first lag, and right panel: during the second lag. Labels and Property numbers are same as described in SI Fig 4.

SI Table 2: Cumulative percentage of total variance captured by first two principal components

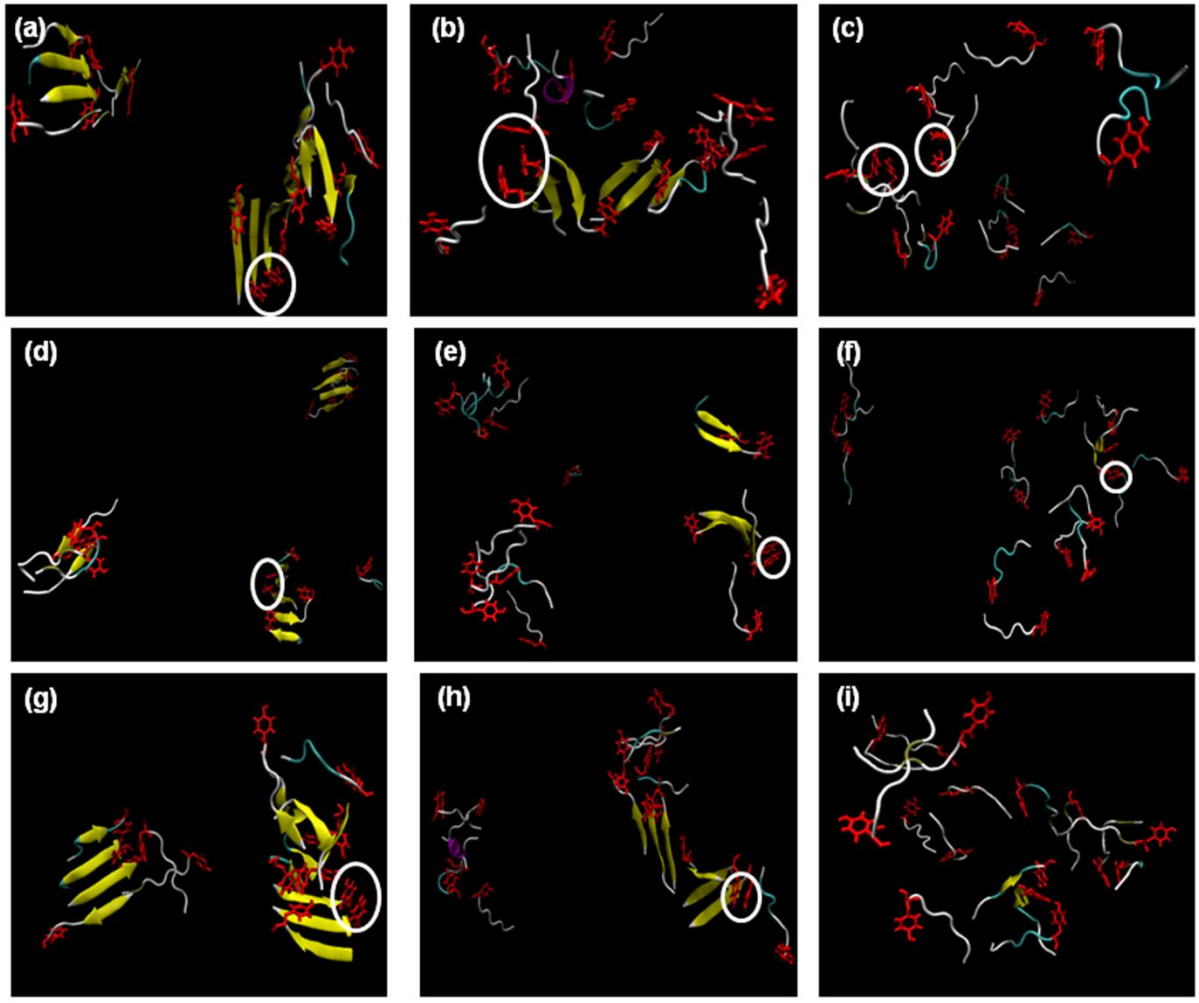
Protein: trehalose	Lag 1		Lag 2		Entire duration	
	PC 1	PC 1 + PC 2	PC 1	PC 1 + PC 2	PC 1	PC 1 + PC 2
1:0	25.21	45.01	19.62	32.48	55.51	63.17
1:6	33.41	48.61	22.52	34.39	56.30	64.62
1:10	25.21	45.01	19.62	32.48	48.21	57.18



SI Fig 6: Secondary structure (as calculated by dssp) along with absolute value of polar order parameter for stable dimers at 8 mM peptide concentration at (a) 1:0 (b) 1:6 peptide:trehalose molar ratio.


(a)
(b)
Number of hydrogen bonds
Time (ns)
Residue Number
Number of backbone Hydrogen bonds

SI Fig 7: Left panel represents time variation of number of hydrogen bonds between stable dimers at 8 mM peptide concentration for (a) 1:0 (b) 1:6 peptide:trehalose molar ratio. Right panel denotes residue-residue hydrogen bond interactions between them averaged during the time they are in beta sheet.



SI Fig 8: Most populated conformers at 8 mM peptide concentration in the presence of different peptide:trehalose molar ratio 1:0, 1:6 and 1:10 respectively during the entire course of simulation (a),(b),(c); during lag 1 (d),(e),(f); and during lag 2 (g),(h),(i). Figures are drawn in VMD[41].

SI Table 3: Transition matrix representing the total number of transitions from state i (row) to state j (column) where i and j represents the maximum populated oligomeric size at

different concentrations. Transitions highlighted in italics represent condensation of oligomers; the term given in bracket below them denotes the size of the oligomers that fuse together:

(a) 1:0 peptide:trehalose molar ratio

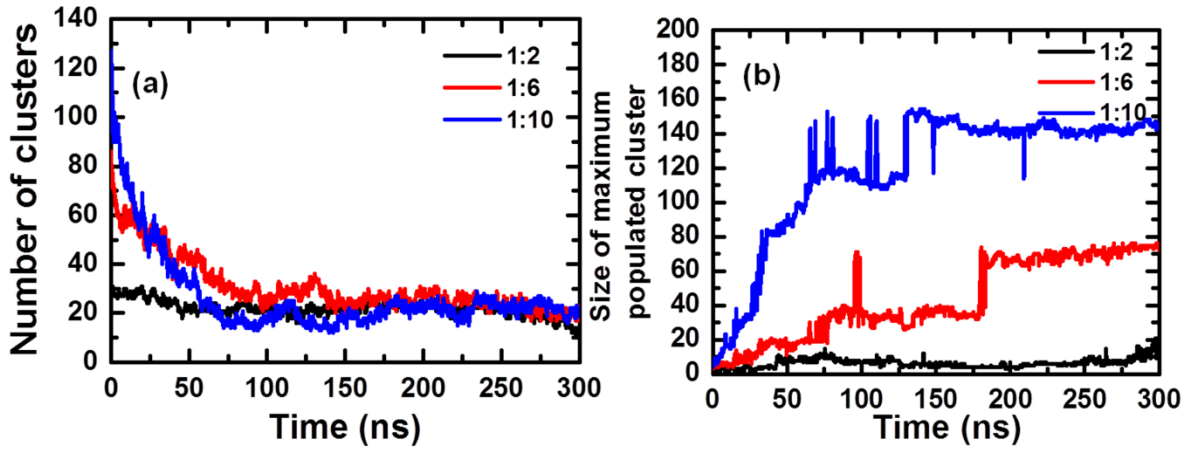
	1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**	**13**	**14**	**15**	**16**
1		1														
2			1													
3				1												
4					1	*1 (4+2)*										
5				1		1										
6					1				*1 (4+5)*							
7																
8																
9										1						
10																*1 (10+6)*
11																
12																
13																
14																
15																
16																.

(b) 1:6 peptide:trehalose molar ratio

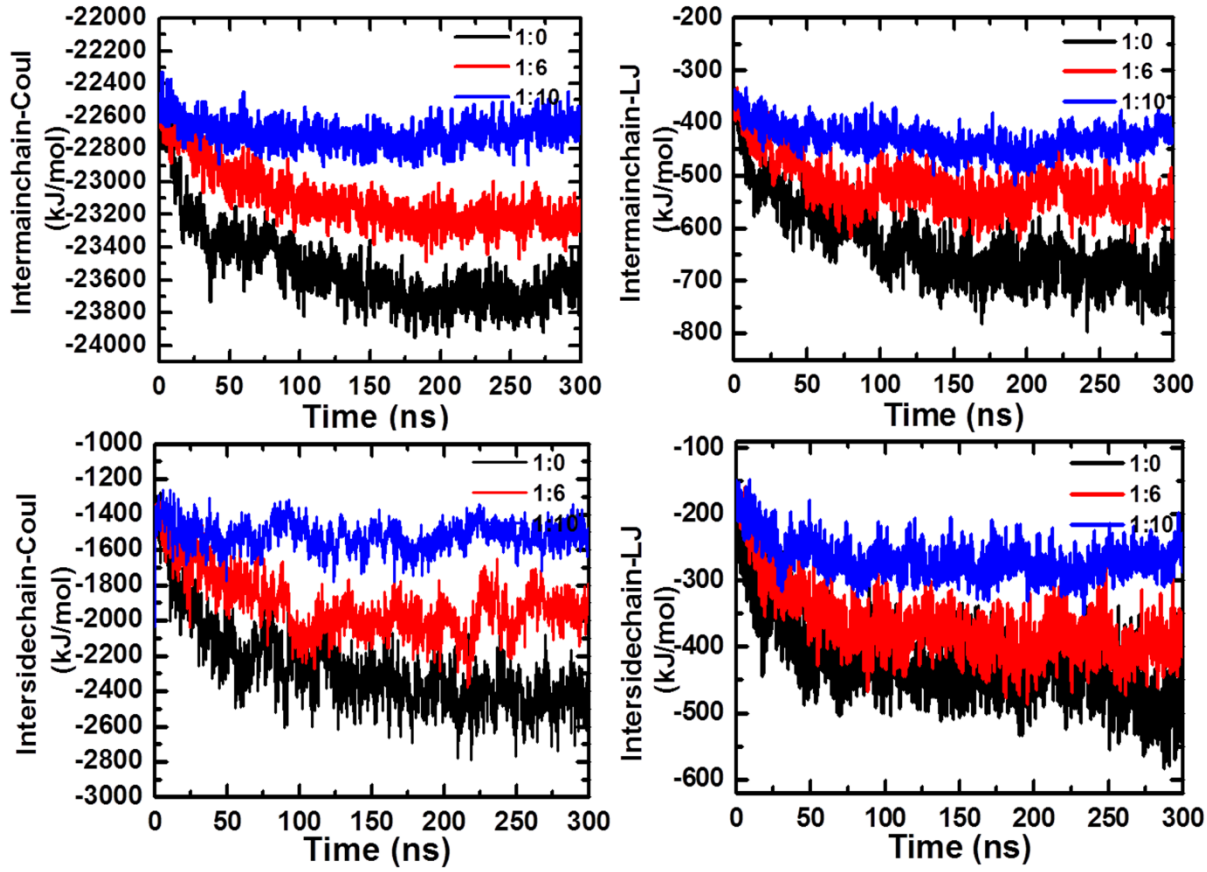
	1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**	**13**	**14**	**15**	**16**
1		1														
2			1													
3				1												
4					8	*2* *(4+2)*										
5				8		2	*1* *(5+2)*									
6				1	3					*4* *(6+4)*						
7						1										
8															*2* *(8+7)*	
9																
10						2					28					
11						1				26		3			*4* *(11+4)*	
12											3					
13																
14																
15								2			3					4
16															4	

(c) 1:10 peptide:trehalose molar ratio

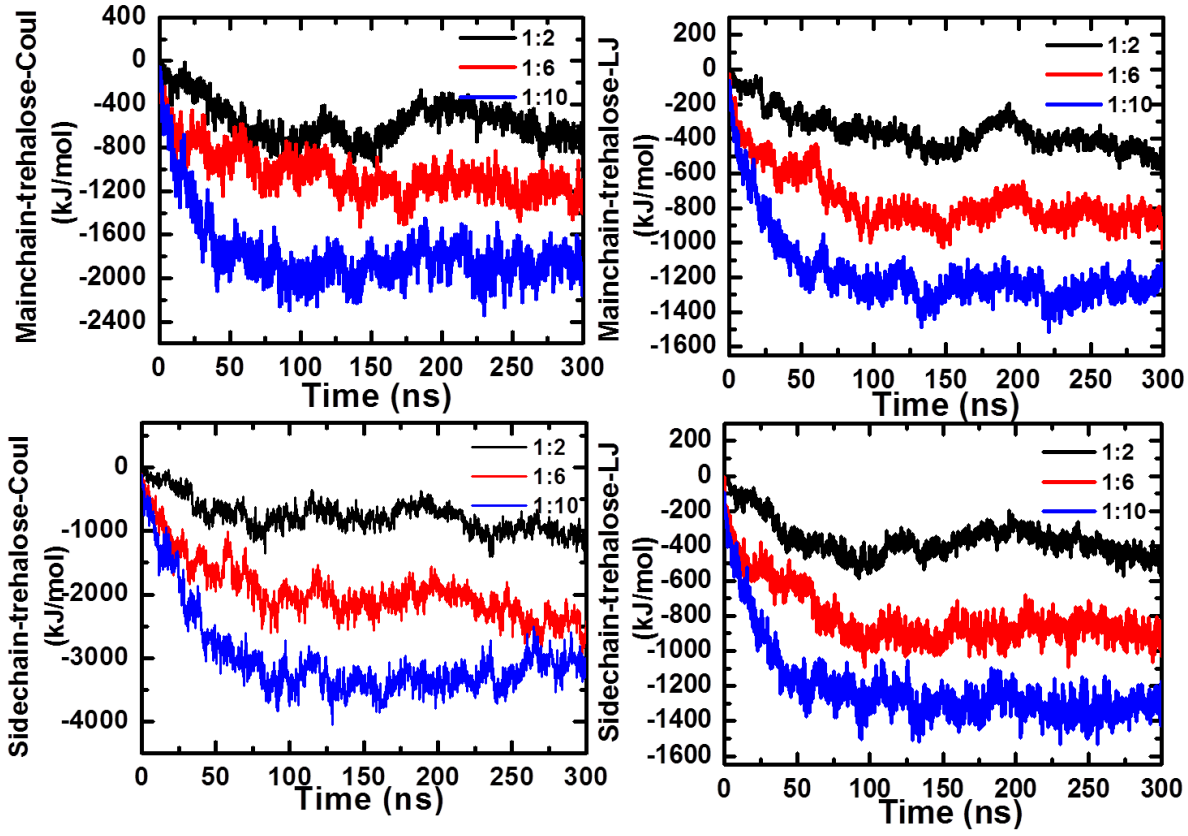
	1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**	**13**	**14**	**15**	**16**
1		3														
2	2		2	*2 (2+ 2)*												
3		2		7	*1 (3 + 2)*											
4		1	8		8											
5				8		6	*3 (4+ 3)*	*3 (5 +3)*	*18 (4 +5)*							
6					7		1									
7					2	2			*7 (7 +2)*				*1 (7+6)*			
8					1				21		*2 (8+ 5- 2)*	*2 (8+2+ 2)*	*2 (4+8+1)*			
9					1 8		5	23		4	*5 (9+ 2)*	*1 (9+4- 1)*	*36 (9+4),(9 +2+2)*	*3 (9+ 5)*		
10									5					*1 (10 +4)*		
11									6	1		6	10			
12							1	1	1	1	2		11	*1 (12 +2)*		
13					1		2	1	36		13	6		5		
14									2		1	3	4			
15																
16																



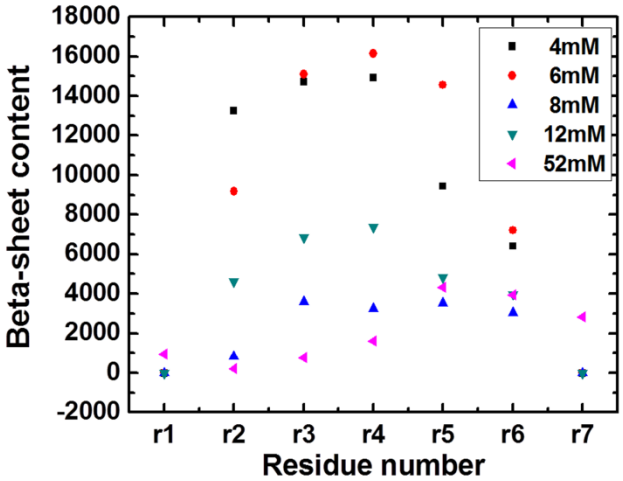
SI Fig9: Time variation of (a) number of clusters of trehalose (b) size of maximum populated trehalose cluster, formed at different peptide:trehalose molar ratio.



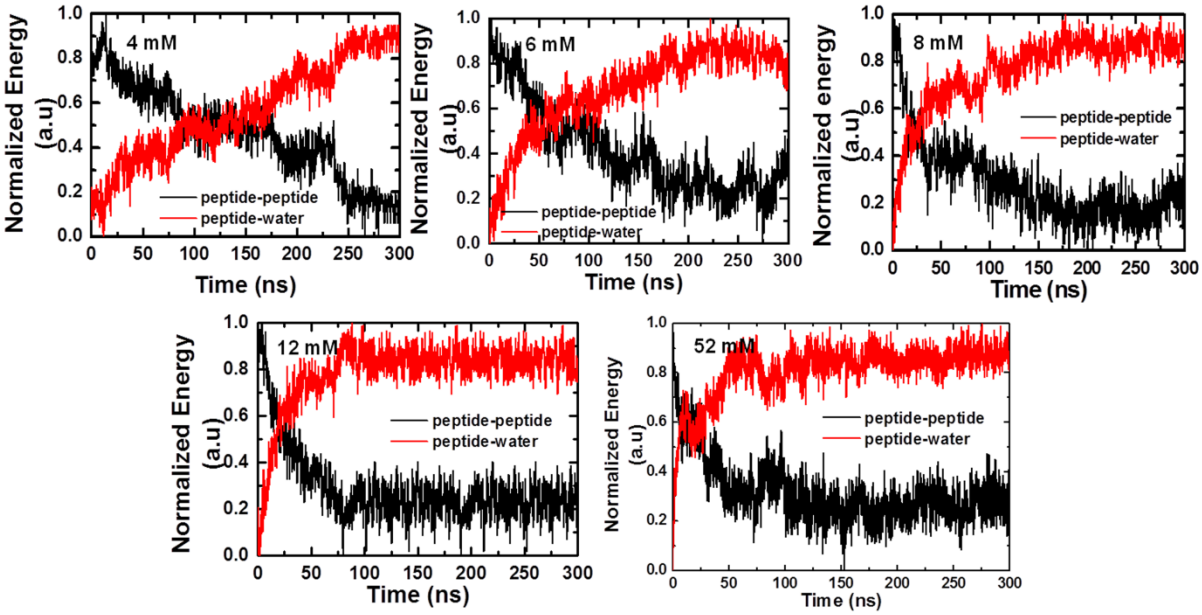
SI Fig 10: Time variation of interchain mainchain and sidechain coulombic and Lennard Jones Interaction energies at 8 mM peptide concentration in the presence of different peptide:trehalose molar ratio.



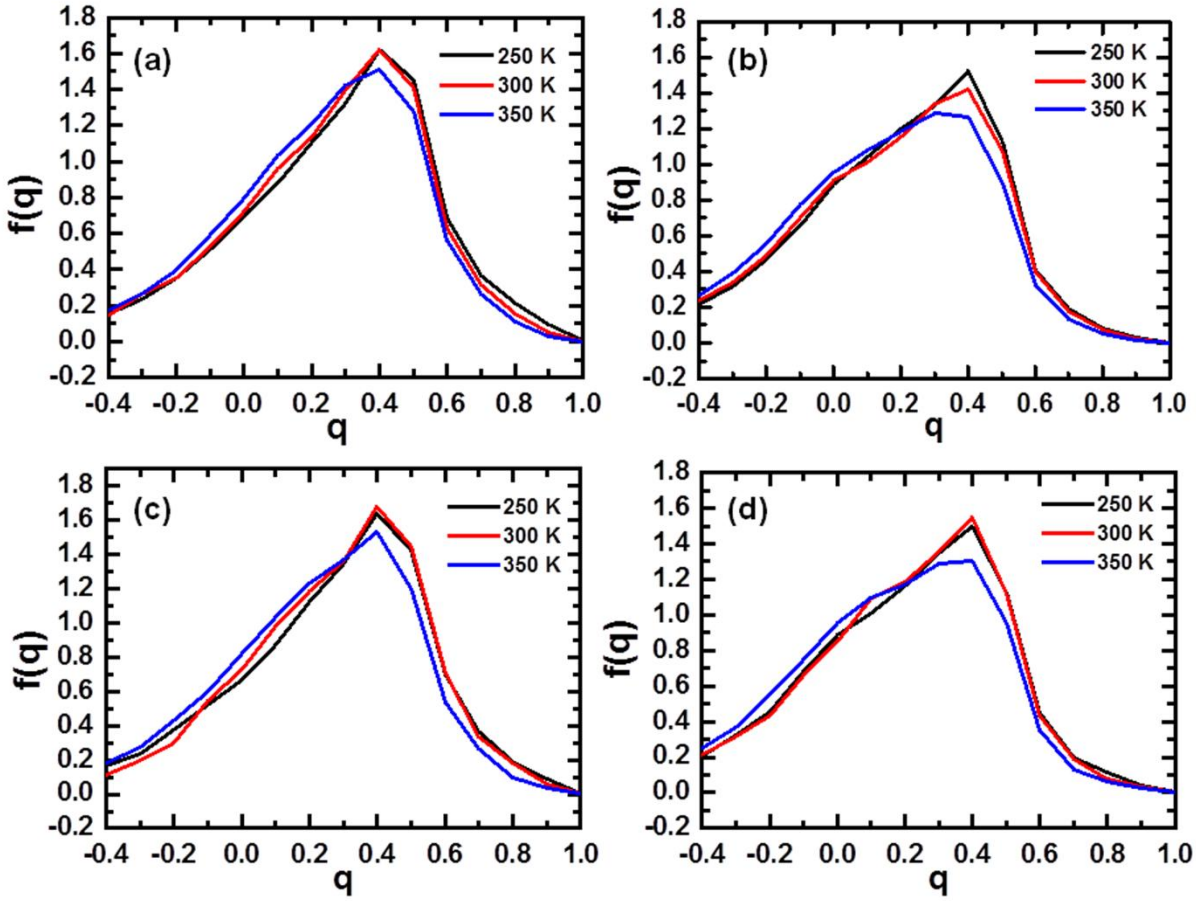
SI Fig 11: Time variation of mainchain and sidechain coulombic and Lennard Jones Interaction energies of peptide molecules at 8 mM peptide concentration with trehalose molecules in the presence of different peptide:trehalose molar ratio.



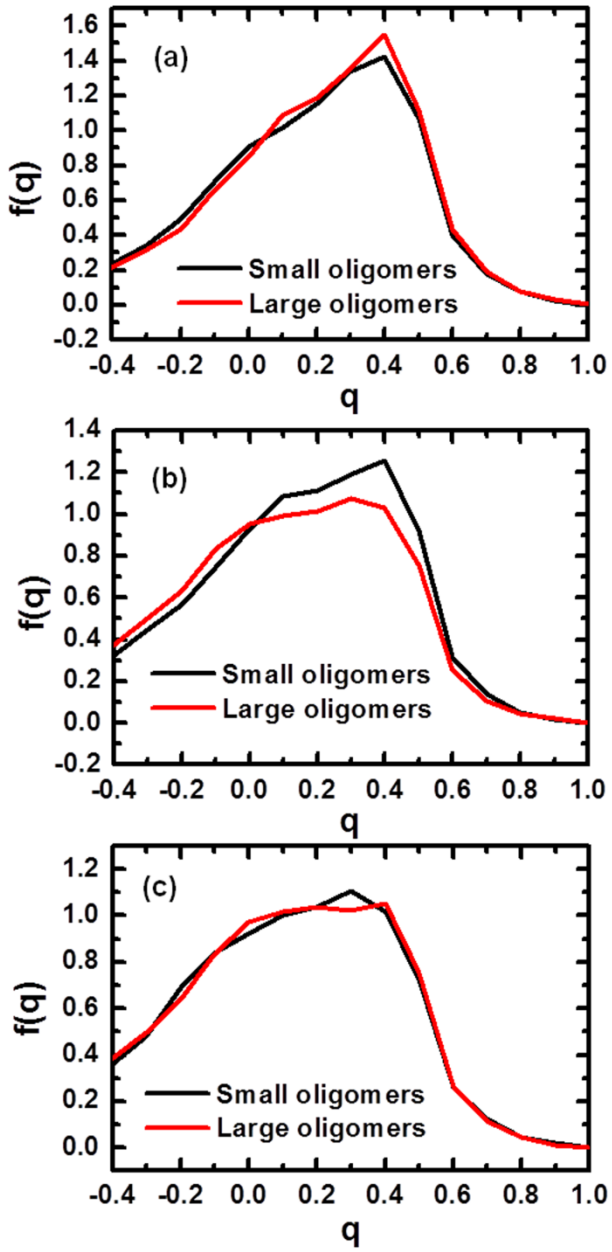
SI Fig 12: Beta-sheet content of different residues.



SI Fig 13: Time evolution of normalized interpeptide mainchain (peptide-peptide) and mainchain-water (peptide-water) Coulombic interaction energy at different peptide concentrations at 300 K.



SI Fig 14: Tetrahedral orientational order parameter (qtet) of (a) bulk water molecules (b) local water molecules during small oligomer formation, and (c)bulk water molecules (d) local water molecules during large oligomer formation, at different temperatures for 8 mM peptide concentration.



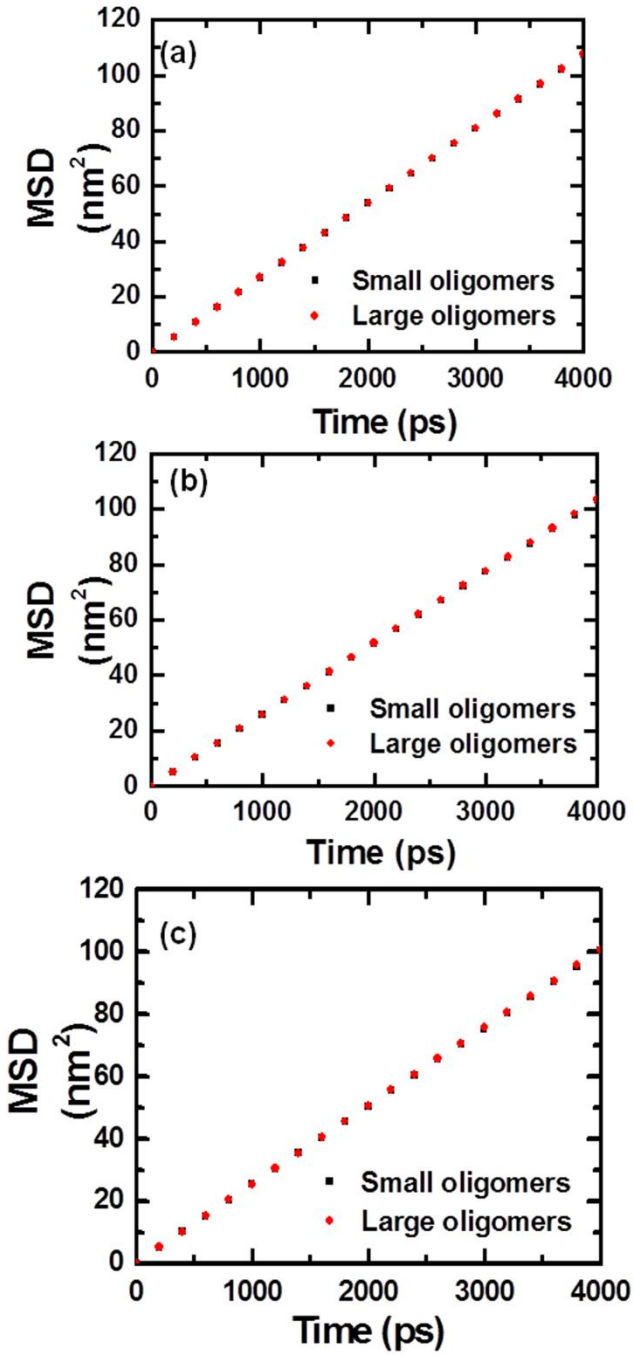
SI Fig 15: Tetrahedral orientational order parameter (qtet) of local water molecules at 8 mM peptide concentration, 300 K during two different time zones in the presence of different peptide:trehalose molar ratio: (a) 1:0 (b) 1:6 (c) 1:10.

SI Table 4: Mean number of water molecules forming hydrogen bonds with peptides with n atoms involved in bonding.

Peptide: Trehalose molar ratio		Total	n=1	n=2	n=3	n=4
1:6	Small oligomers	339±17.56	306.17±33.09 (90%)	42.67±4.27 (13%)	2.2±0.89 (0.6%)	o
	Large oligomers	258.2±23.16	223.5±21.25 (87%)	35.5±18.22 (14%)	5±1.41 (2%)	1
1:10	Small oligomers	279.8±5.63	250.67±10.89 (90%)	29.67±4.13 (11%)	3±1.09 (1%)	1
	Large oligomers	288.6±7.27	250.67±9.99 (87%)	31.17±4.83 (11%)	3.2±1.3 (1%)	1

SI Table 5: Mean number of trehalose molecules forming hydrogen bonds with peptides with n atoms involved in bonding.

Peptide: Trehalose molar ratio		Total	n=1	n=2	n=3	n=4
1:6	Small oligomers	29.5±4.59	11±1.79 (37%)	6.17±2.40 (21%)	5.83±1.72 (20%)	3.00±1.73 (10%)
	Large oligomers	39.67±2.73	11±1.79 (28%)	10±3.89 (25%)	9.83±2.32 (25%)	5.00±2.19 (13%)
1:10	Small oligomers	73.17±2.71	27.67±4.63 (38%)	20.83±5.84 (28%)	11.67±4.37 (16%)	5.00±2.37 (7%)
	Large oligomers	70.30±4.80	27.00±4.82 (38%)	20.5±3.15 (29%)	10.5±2.34 (15%)	5.2±2.17 (7%)



SI Fig 16: MSD of total water molecules at 8 mM during two different time zones zones in the presence of peptide:trehalose molar ratio: (a) 1:0 (b) 1:6 (c) 1:10.