Electronic Supplementary Material (ESI) for RSC Advances.

Supplementary Information: Mutation-induced change in chignolin stability from π-turn to α-turn

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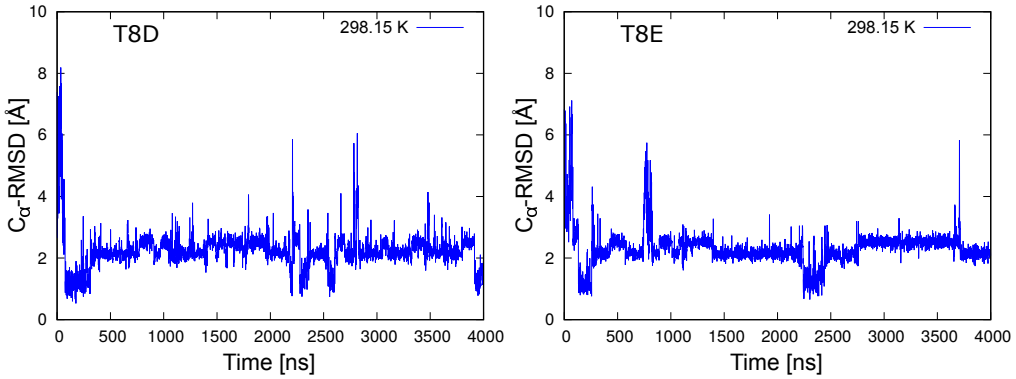

Figure S1 (Supplementary Information)

Time series of C_α–RMSD of chignolin mutants (acidic side chain) from a NMR structure of original chignolin.

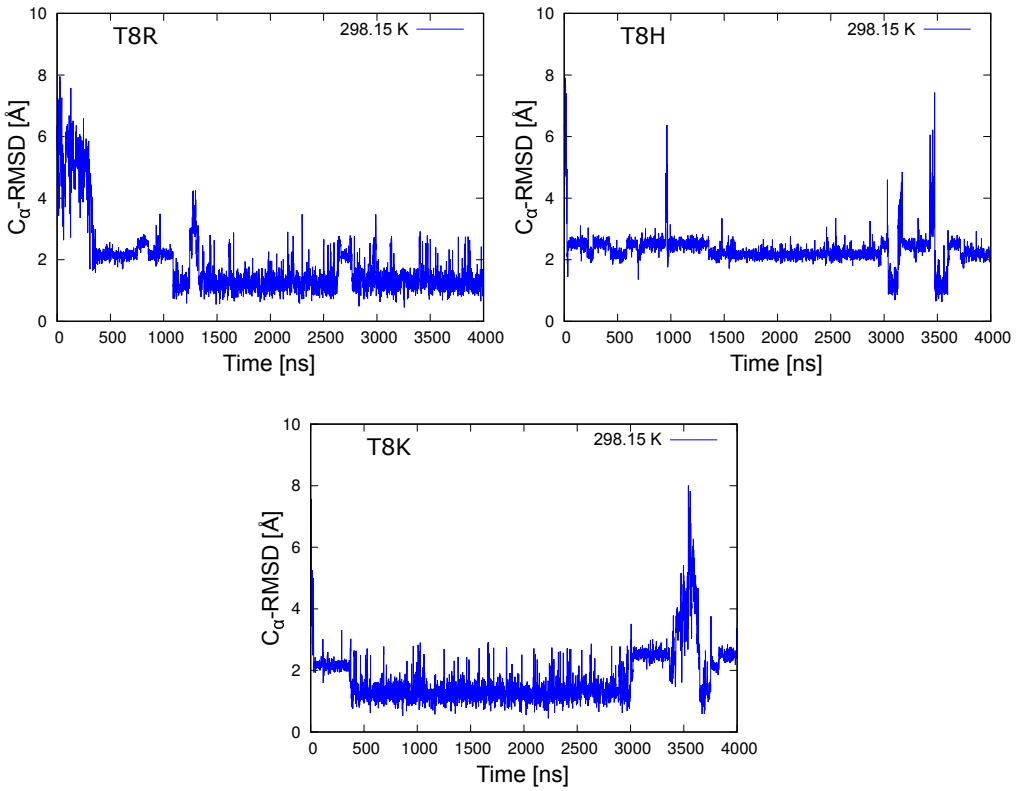

Figure S2 (Supplementary Information)

Time series of C_α–RMSD of chignolin mutants (basic side chain) from a NMR structure of original chignolin.

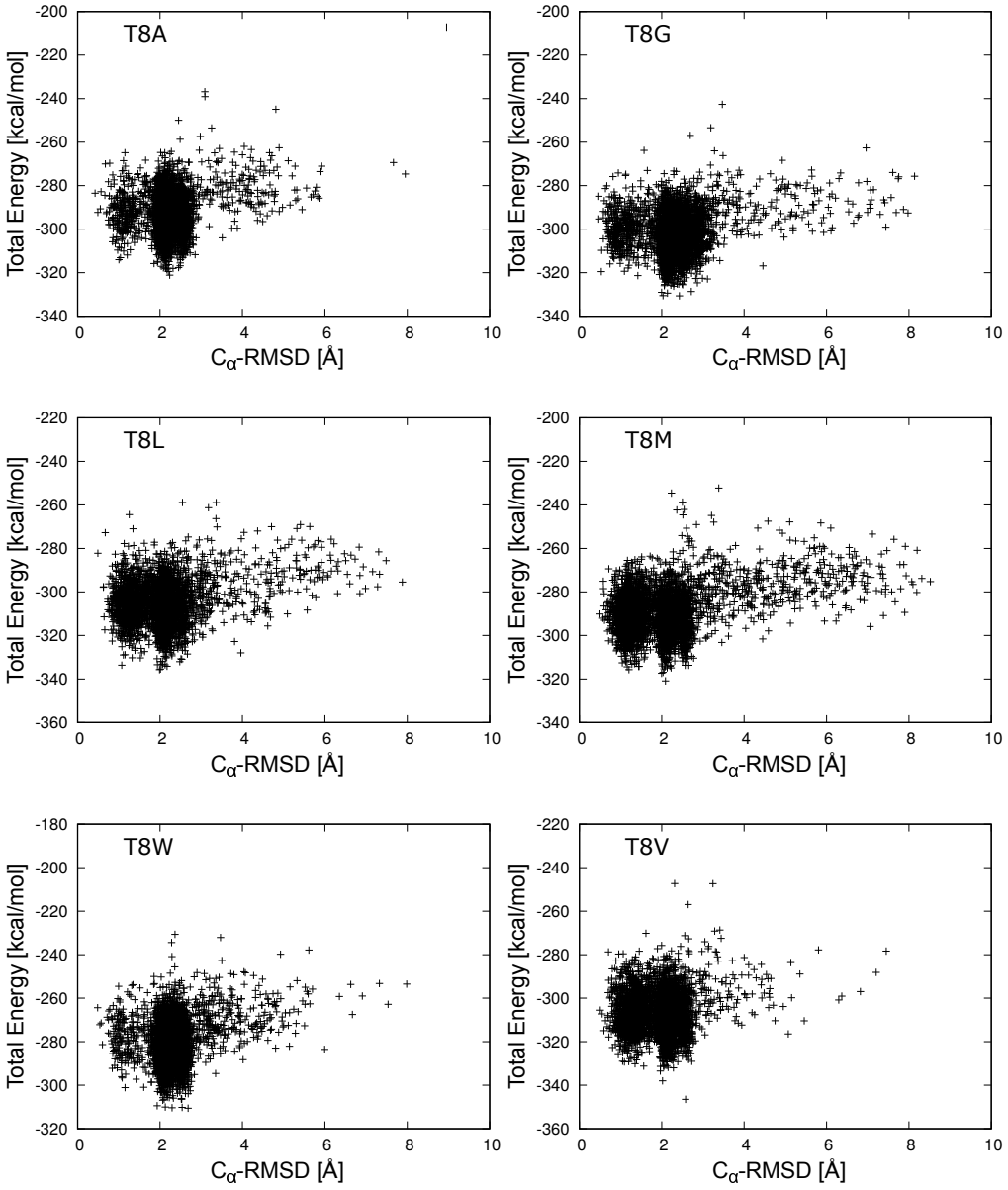

Figure S3 (Supplementary Information)

Total energy of chignolin mutants (hydrophobic side chain) as a function of C_α–RMSD.

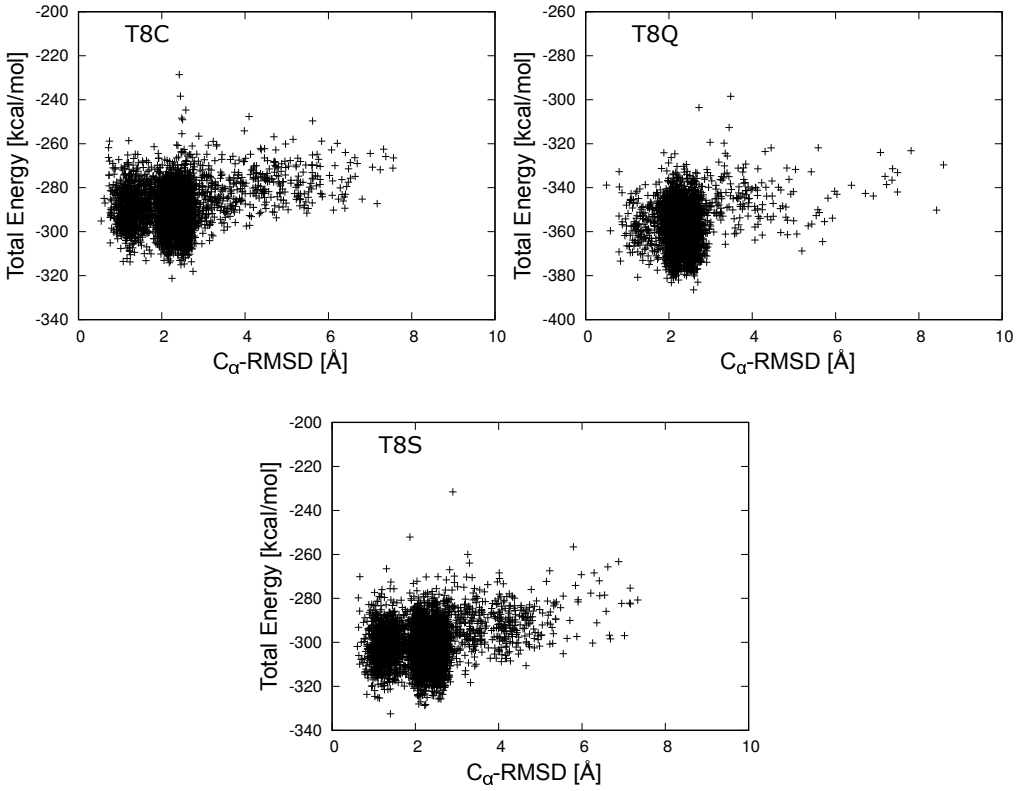

Figure S4 (Supplementary Information)

Total energy of chignolin mutants (polar uncharged side chain) as a function of C_α–RMSD.

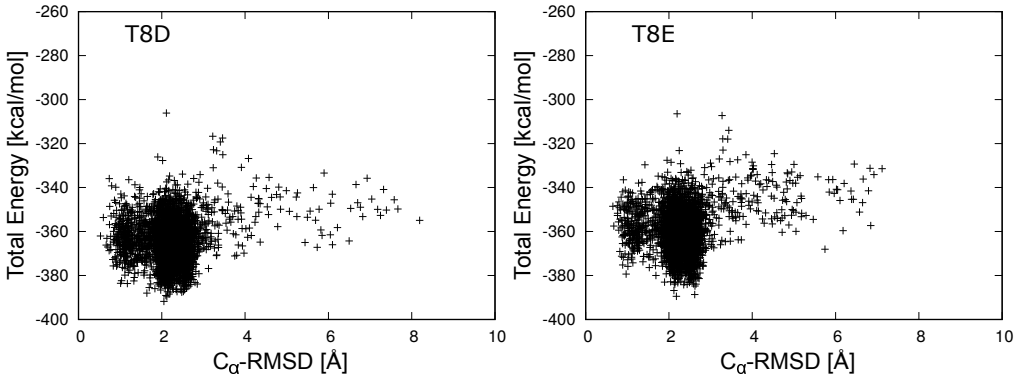

Figure S5 (Supplementary Information)

Total energy of chignolin mutants (acidic side chain) as a function of C_α–RMSD.

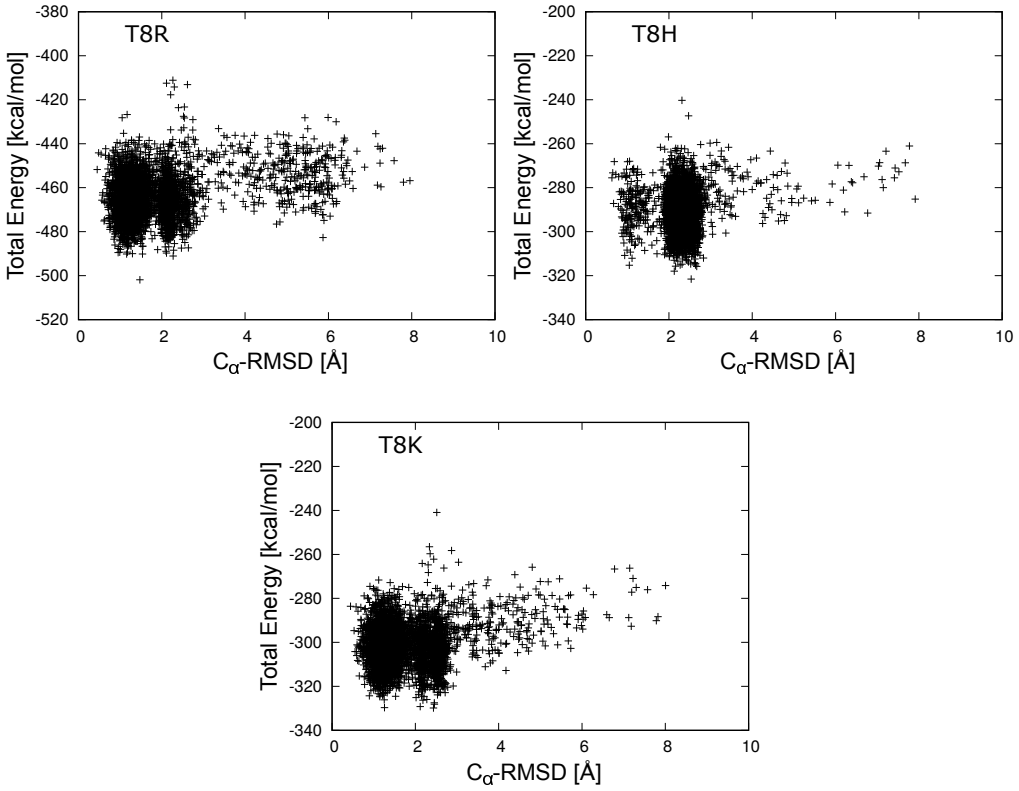

Figure S6 (Supplementary Information)

Total energy of chignolin mutants (basic side chain) as a function of C_α–RMSD.

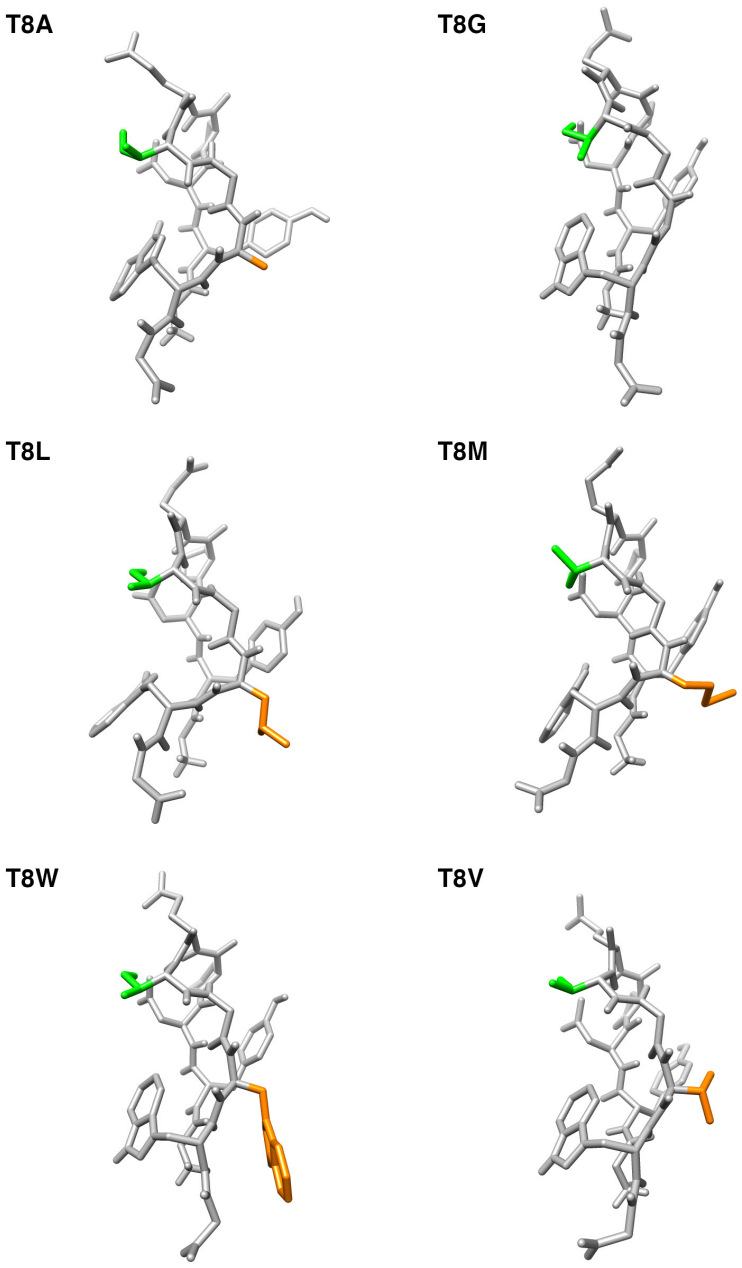

Figure S7 (Supplementary Information)

The lowest total energy structures of mutants (hydrophobic side chain). The green and orange regions indicate the side chains of Thr6 and the 8th amino acid.

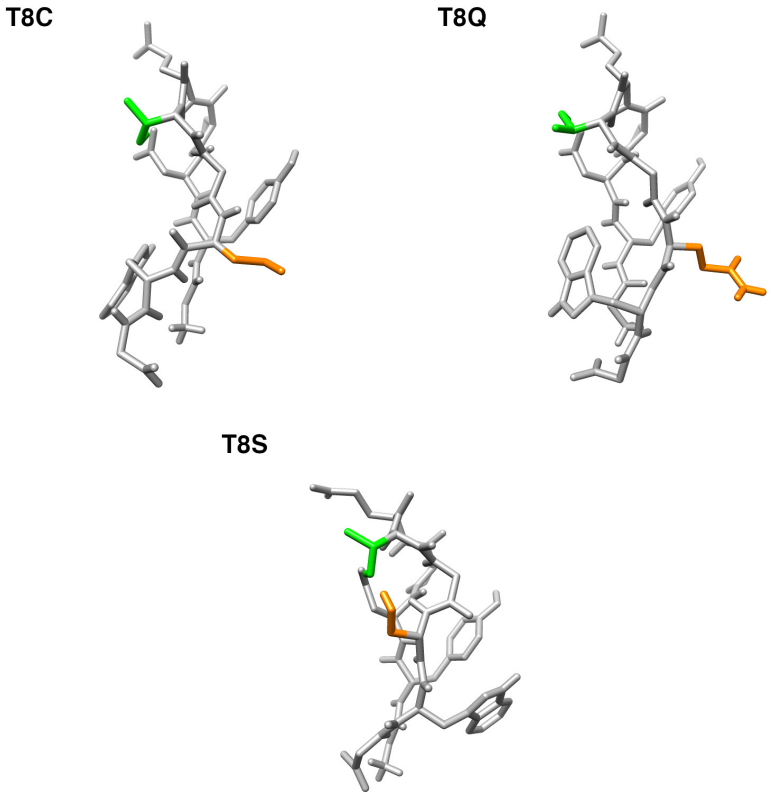

Figure S8 (Supplementary Information)

The lowest total energy structures of mutants (polar uncharged side chain). The green and orange regions indicate the side chains of Thr6 and the 8th amino acid. T8S mutant forms the native structure.

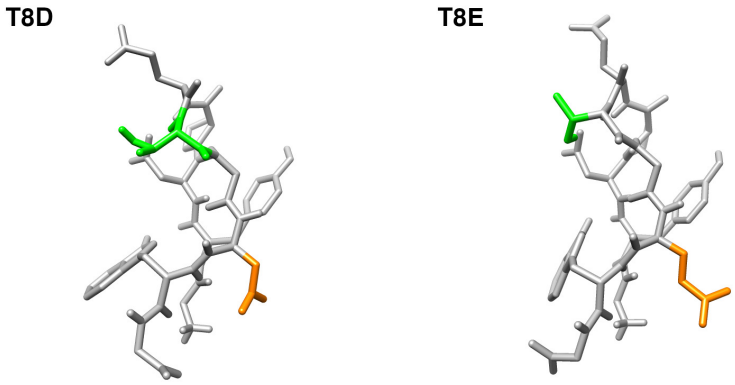

Figure S9 (Supplementary Information)

The lowest total energy structures of mutants (acidic side chain). The green and orange regions indicate the side chains of Thr6 and the 8th amino acid, respectively.

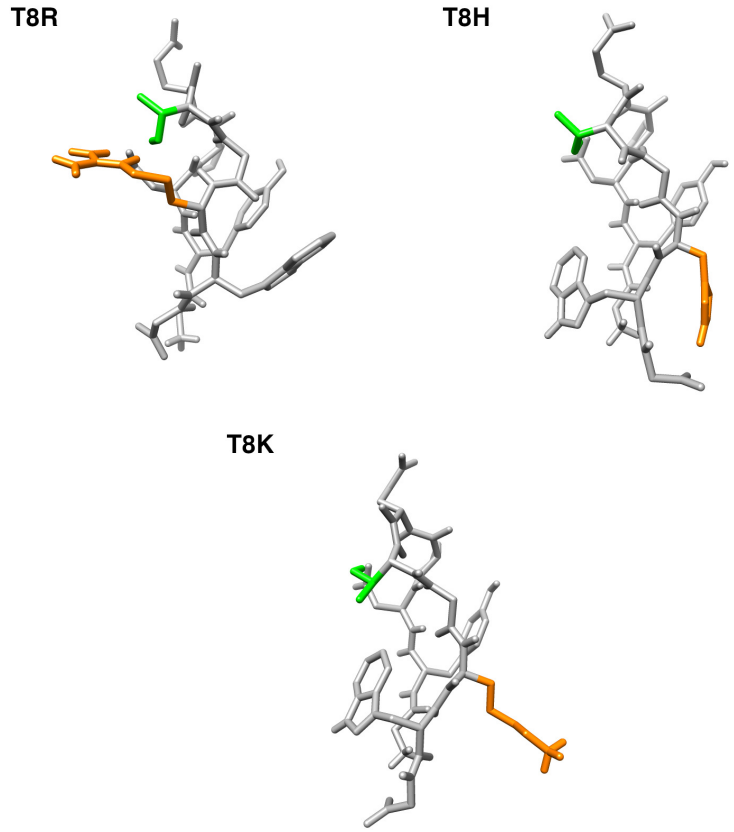

Figure S10 (Supplementary Information)

The lowest total energy structures of mutants (basic side chain). The green and orange regions indicate the side chains of Thr6 and the 8th amino acid, respectively. T8R mutant forms the native structure.

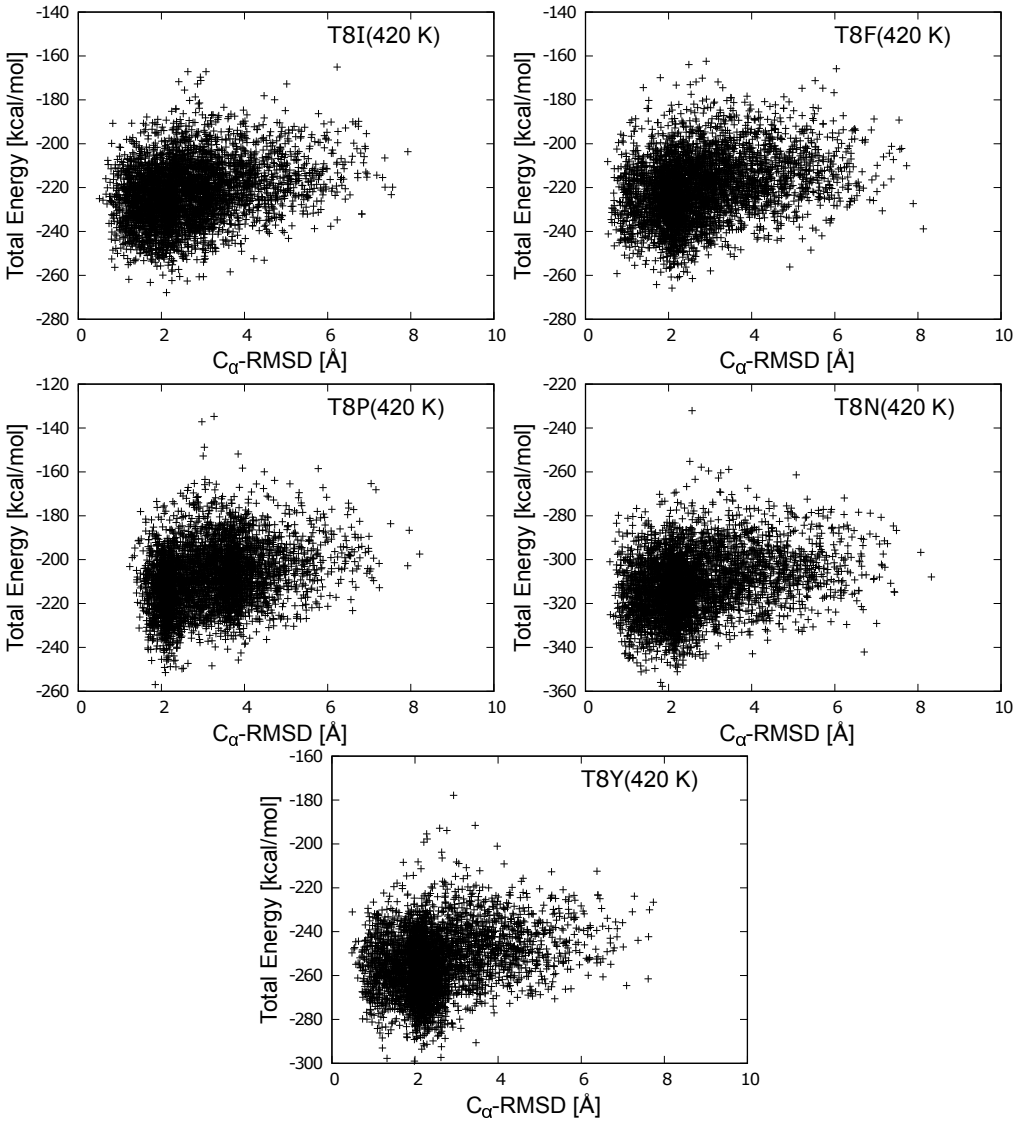

Figure S11 (Supplementary Information)

Total energy values of the chignolin mutants as a function of the C_α–RMSD at 420 K.

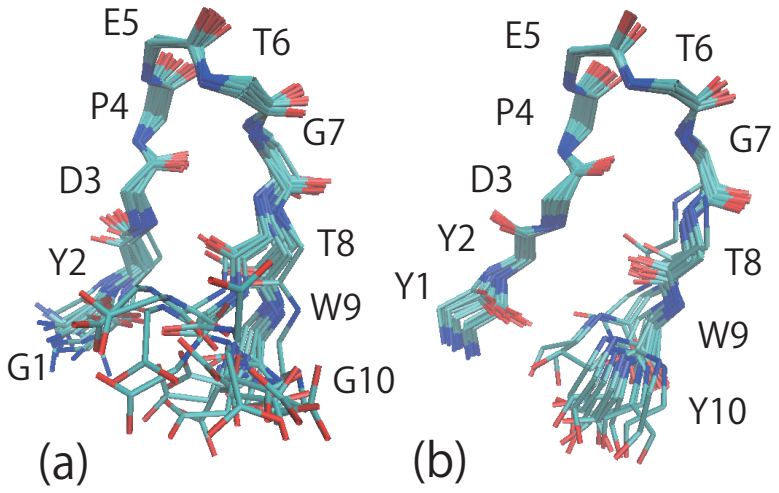

Figure S12 (Supplementary Information)

The backbone structures of chignolin (a) and CLN025 (b) obtained by the previous NMR experiments (PDBID: 1UAO and 2RVD). The structures are fitted from Asp 3 to Gly7.

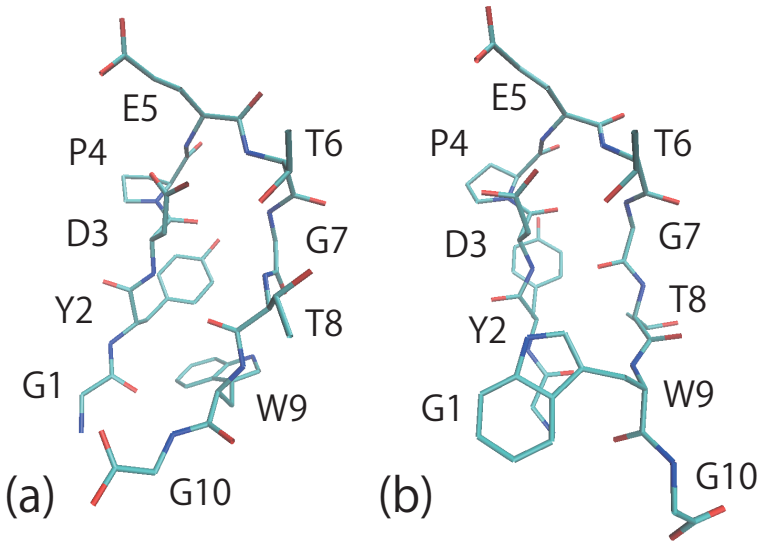

Figure S13 (Supplementary Information)

The structures without hydrogen atoms of native (a) and misfolded (b) states obtained by simulations. The structures are fitted from Asp 3 to Gly7.

Table S1 (Supplementary Information)

Average values of total energy, G, of each state of mutants with neutral side chain. Energy unit is kcal/mol. The value in parenthesis is the occurrence rate.

Mutant	native	misfolded
T8A	-291.8 ± 8.6 (7.0)	-293.7 ± 8.5 (85.2)
T8G	-299.7 ± 8.2 (9.9)	-304.3 ± 8.8 (68.0)
T8I	-280.6 ± 0.2 (0.1)	-287.7 ± 9.1 (74.2)
T8L	-305.2 ± 8.9 (22.7)	-307.6 ± 8.9 (58.9)
T8M	-290.3 ± 8.6 (39.6)	-292.7 ± 8.9 (40.0)
T8F	–	-288.0 ± 8.8 (92.7)
T8P	–	-274.3 ± 8.5 (94.4)
T8W	-277.5 ± 9.3 (4.6)	-282.0 ± 9.1 (83.8)
T8V	-306.1 ± 8.9 (27.9)	-308.9 ± 9.0 (58.5)
T8N	–	-375.3 ± 8.8 (92.6)
T8C	-289.0 ± 9.1 (16.8)	-291.1 ± 8.6 (65.0)
T8Q	-357.4 ± 8.9 (2.9)	-358.6 ± 8.7 (87.7)
T8S	-301.6 ± 8.9 (20.5)	-303.3 ± 8.7 (60.1)
T8Y	-297.8 ± 0.0 (0.0)	-308.3 ± 8.6 (83.0)

Table S2 (Supplementary Information)

Average values of total energy, G, of each state of mutants with chraged side chain. Energy unit is kcal/mol. The value in parenthesis is the occurrence rate.

Mutant	native	misfolded
T8D	-362.7 ± 8.7 (9.5)	-365.9 ± 8.8 (77.9)
T8E	-357.1 ± 8.6 (6.5)	-361.3 ± 8.6 (83.0)
T8R	-464.8 ± 9.0 (58.4)	-466.1 ± 9.5 (22.2)
T8H	-288.9 ± 9.5 (4.7)	-291.6 ± 8.8 (88.9)
T8K	-302.1 ± 8.9 (58.4)	-304.0 ± 9.2 (23.3)

Table S3 (Supplementary Information)

Chemical shifts for HN. The unit is ppm. The values from CLN025 to T8P(cis) in the present works are listed. The values of CLN025 (pre) and Chignolin (pre) are extracted from the previous NMR experiments ((PDBID:2rv_cs.str) and (BMRB code: 5649)), respectively.

	CLN025	Chignolin	T8P (trans)	T8P (cis)	CLN025 (pre)	Chignolin (pre)
Y2	8.75	8.77	8.60	8.60	8.56	8.81
D3	8.45	8.13	8.44	8.44	8.34	8.15
E5	8.19	8.24	8.59	8.66	8.14	8.25
T6	7.27	7.30	8.09	7.96	7.25	7.28
G7	8.35	8.38	8.24	8.00	8.30	8.4
residue 8	7.27	7.36	–	–	7.23	7.35
W9	8.66	8.87	8.17	8.69	8.50	8.94
residue 10	8.43	8.12	7.82	8.17	8.37	8.15

Table S4 (Supplementary Information)

Chemical shifts of H_α. The unit is ppm. The values from CLN025 to T8P(cis) in the present works are listed. The values of CLN025 (pre) and Chignolin (pre) are extracted by the previous NMR experiments ((PDBID:2rv_cs.str) and (BMRB code: 5649)), respectively.

	CLN025	Chignolin	T8P (trans)	T8P (cis)	CLN025 (pre)	Chignolin (pre)
residue 1	4.14	–	–	–	4.16	–
Y2	4.50	4.12	4.57	4.57	4.51	4.10
D3	4.94	4.89	4.88	4.88	4.95	4.89
P4	4.07	4.05	4.33	4.33	4.06	4.05
E5	4.16	4.14	4.28	4.43	4.16	4.14
T6	4.44	4.36	4.36	4.43	4.35	4.37
G7-1	3.73	3.78	4.00	2.76	3.73	3.77
G7-2	4.01	4.00	4.04	2.76	4.02	4.00
residue 8	4.43	4.61	4.32	4.23	4.45	4.62
W9	5.02	4.80	4.78	4.89	5.02	4.79
residue 10-1	4.35	3.93	3.77	3.82	4.39	3.94
residue 10-2	–	3.73	3.62	3.82	–	3.73