Electronic Supplementary Material (ESI) for Physical Chemistry Chemical Physics

Supporting Information for the Manuscript

"The Influence of the ΔK280 Mutation and N- or C- Terminal Extensions on the Structure, Dynamics, and Fibril Morphology of the Tau R2 Repeat"

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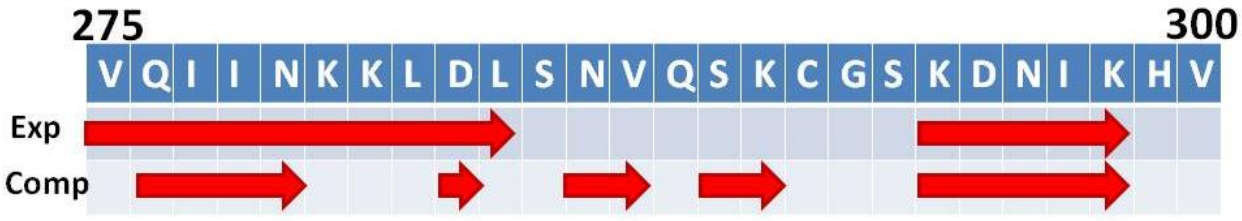


Figure S1: Schematic representations of the secondary structure (β-sheet indicated as arrow) of the wild-type tau R2 repeat proposed by experiment[1] and by proposed by our constructed model. The secondary structure proposed by simulations had been estimated by computing the ψ and Φ dihedral angles from the last 5 ns for each residue. Angles with the range of ψ = 90-180° and Φ = (-60)-(-120)° were considered as β-sheet structure.[2]

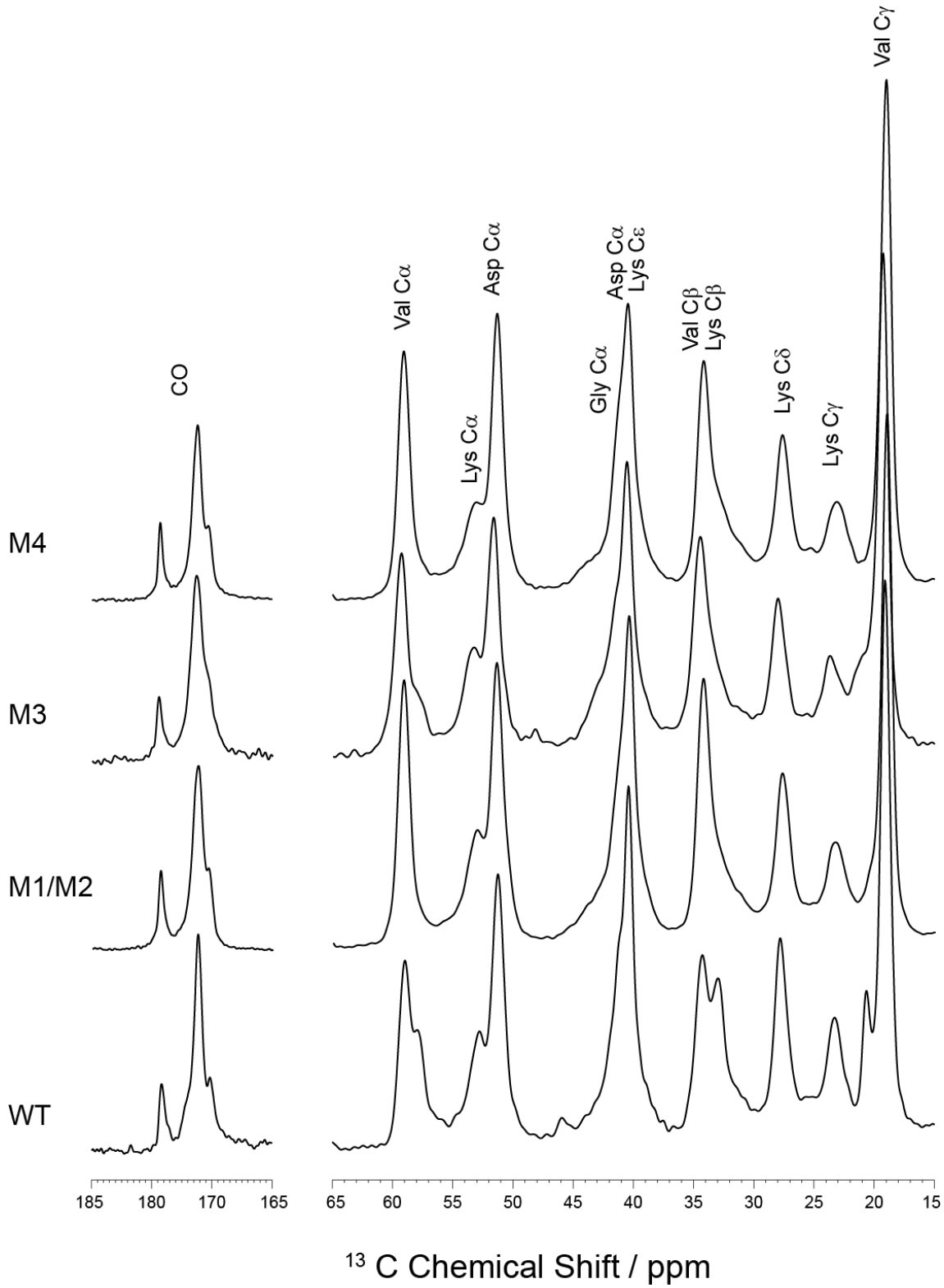


Figure S2: Proton-decoupled ^{13}C CP MAS NMR spectra of the four tau peptides recorded at a MAS frequency of 7 kHz and the temperature to 30°C. The amino acid assignment of the peaks is also given.

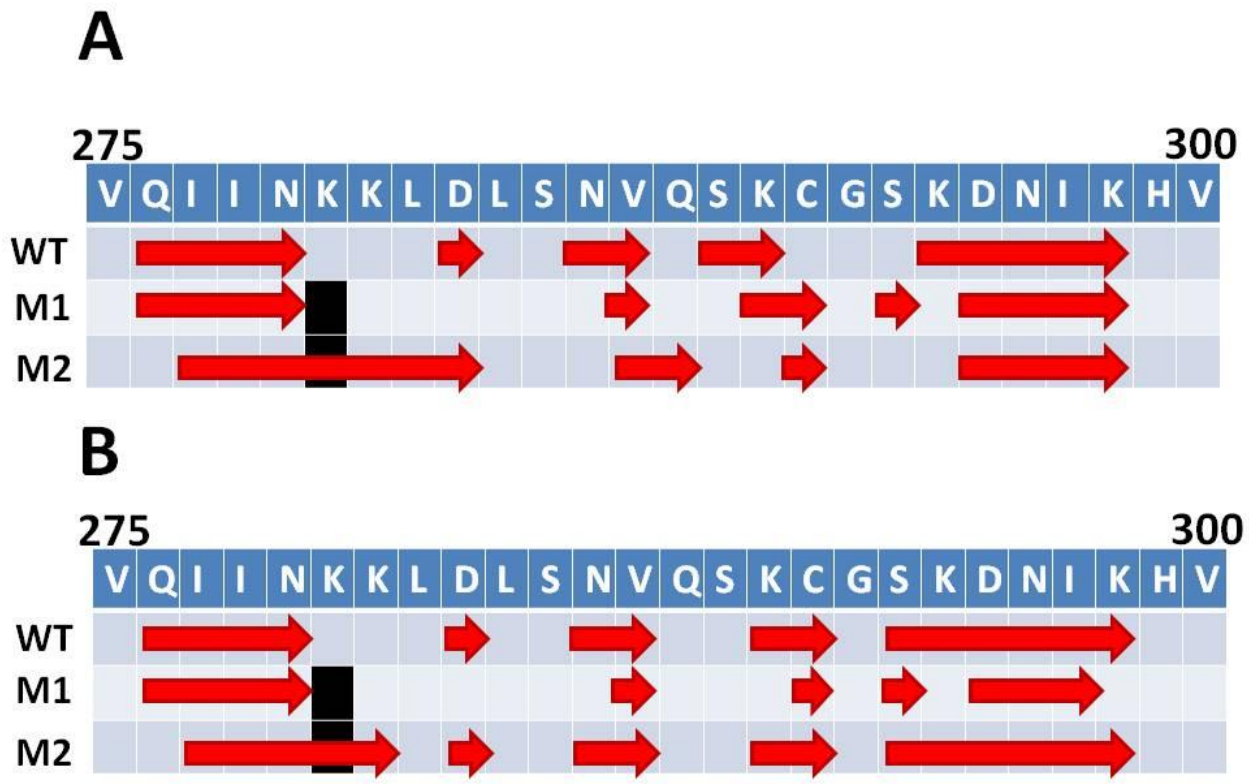


Figure S3: Schematic representations of the secondary structure (β-sheet indicated as arrow) of wild-type tau R2 repeat model and the ΔK280 mutated tau R2 repeat models M1 and M2. The secondary structure proposed by simulations had been estimated by computing the ψ and Φ dihedral angles from (A) the last 5 ns and (B) all 30 ns and for each residue. Angles with the range of ψ = 90-180° and Φ = (-60)-(-120)° were considered as β-sheet structure.[2]

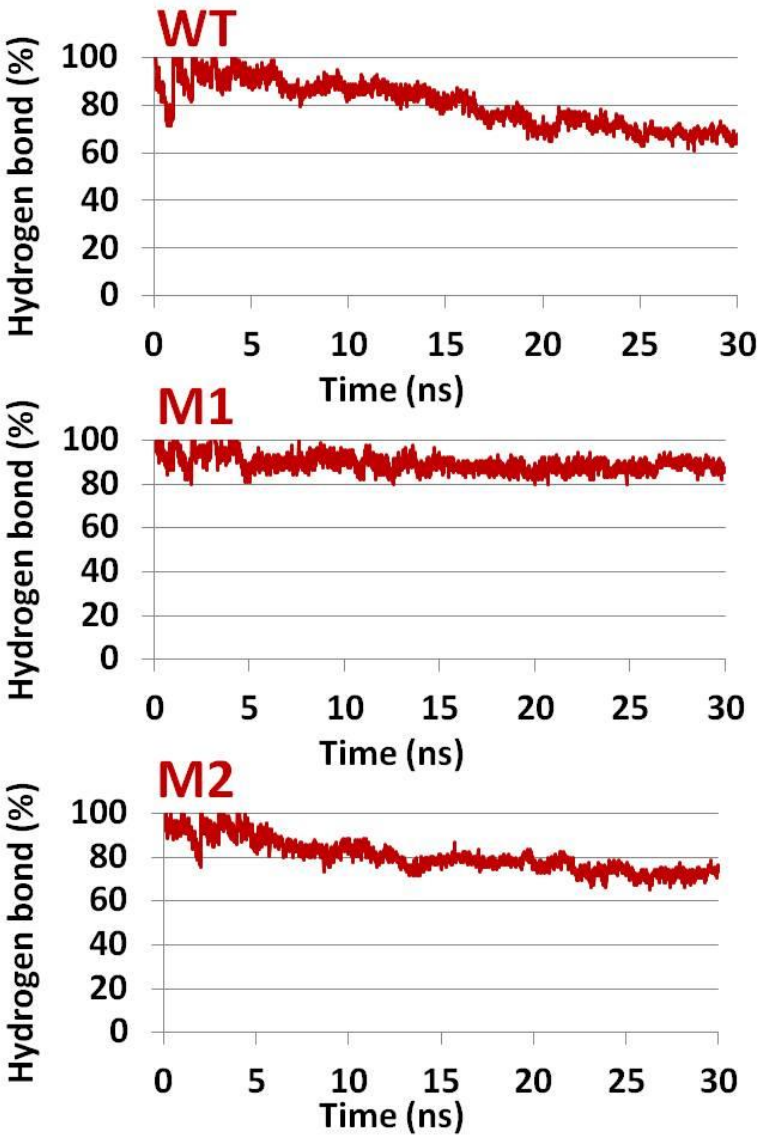


Figure S4: The fraction of the number of hydrogen bonds (in percentage) between all β-strands compared to the number in the initial oligomer for the WT tau R2 repeat model and the ΔK280 mutated tau R2 repeat models M1 and M2.

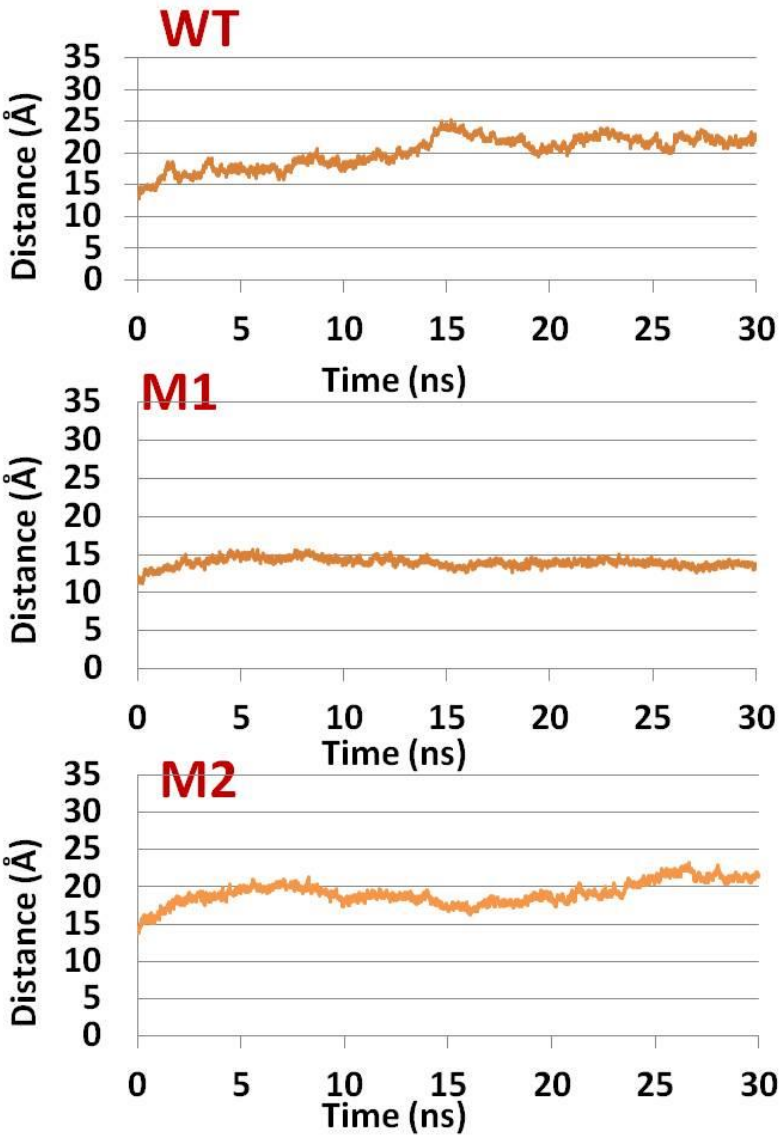


Figure S5: The averaged inter-sheet (Cα backbone-backbone) distances for WT, M1 and M2 models during the molecular dynamics (MD) simulations.

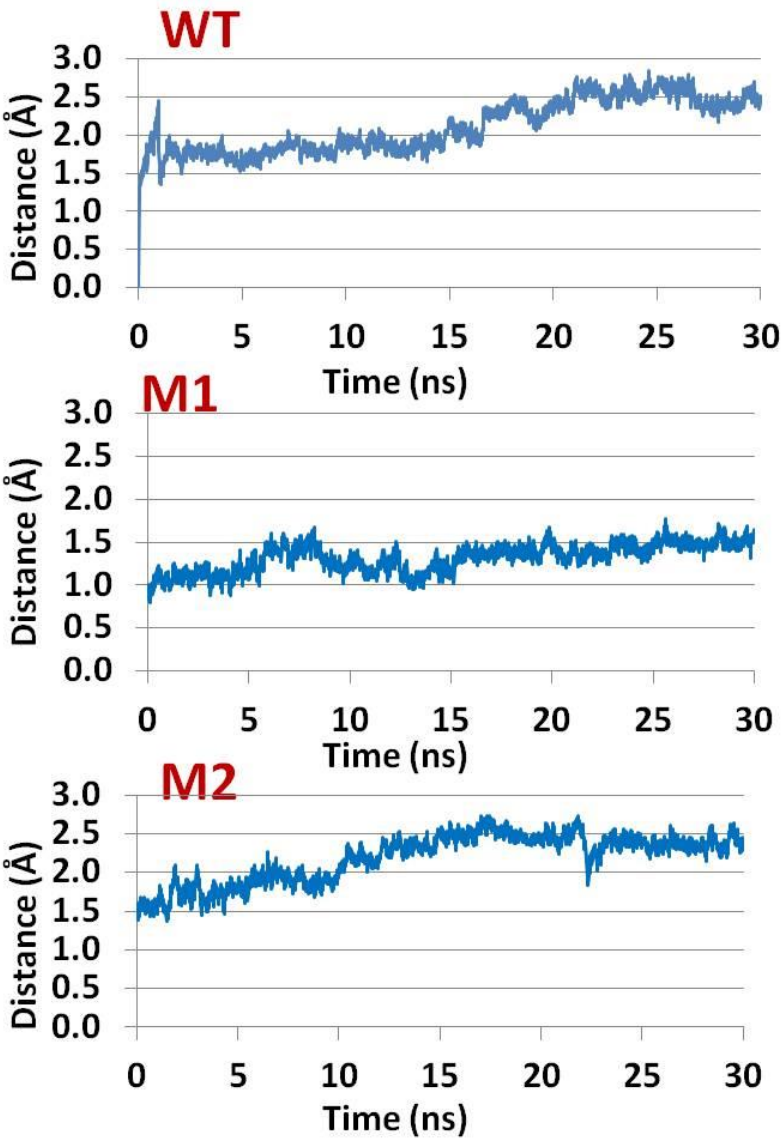


Figure S6: RMSDs of WT, M1 and M2 models.

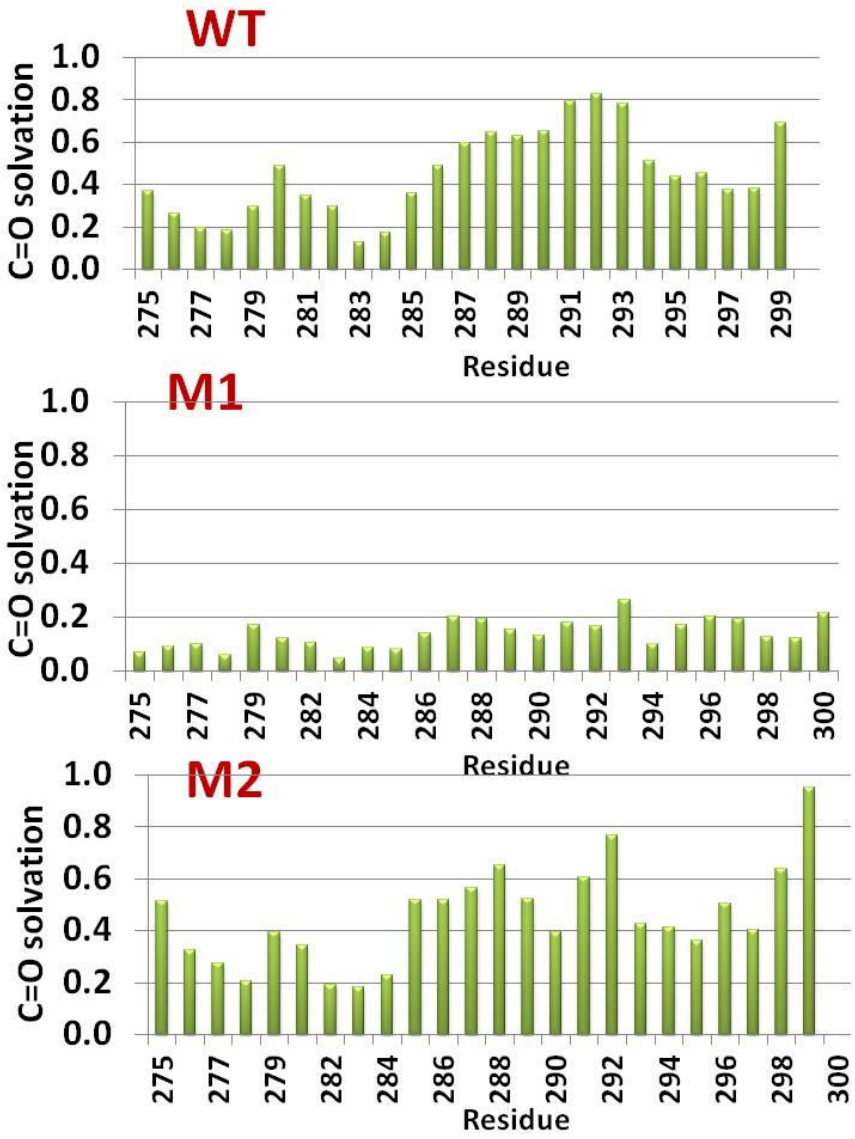


Figure S7: The average number of water molecules around each side chain Cβ carbon (within 4 Å) for WT, M1 and M2 models.

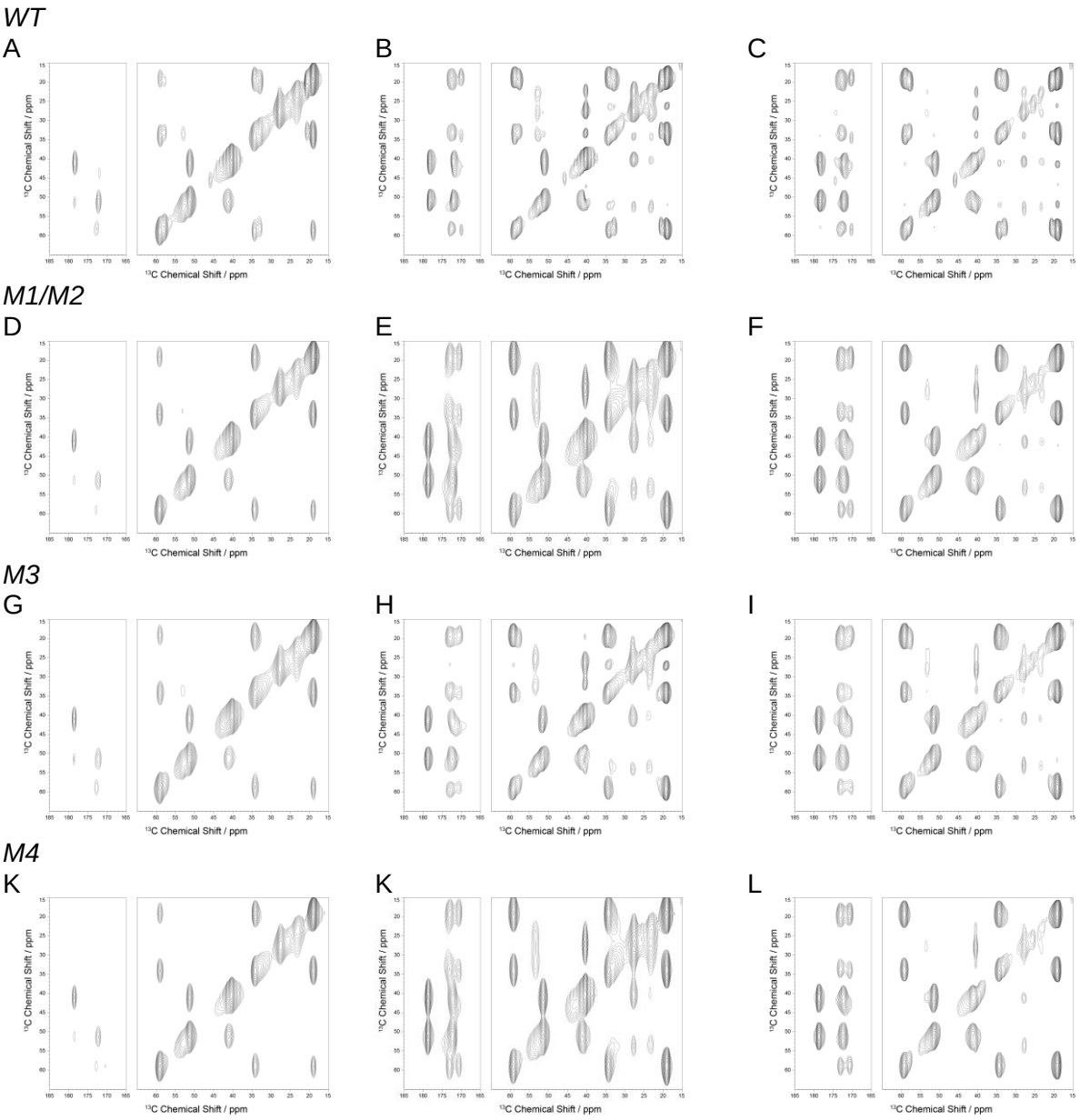


Figure S8: ^{13}C-^{13}C proton driven spin diffusion spectra of the four peptides at a MAS frequency of 7 kHz and a temperature of 30°C. The mixing times were 5 ms (A, D, G, K), 50 ms (B, E, H, K) and 600 ms (C, F, I, L).

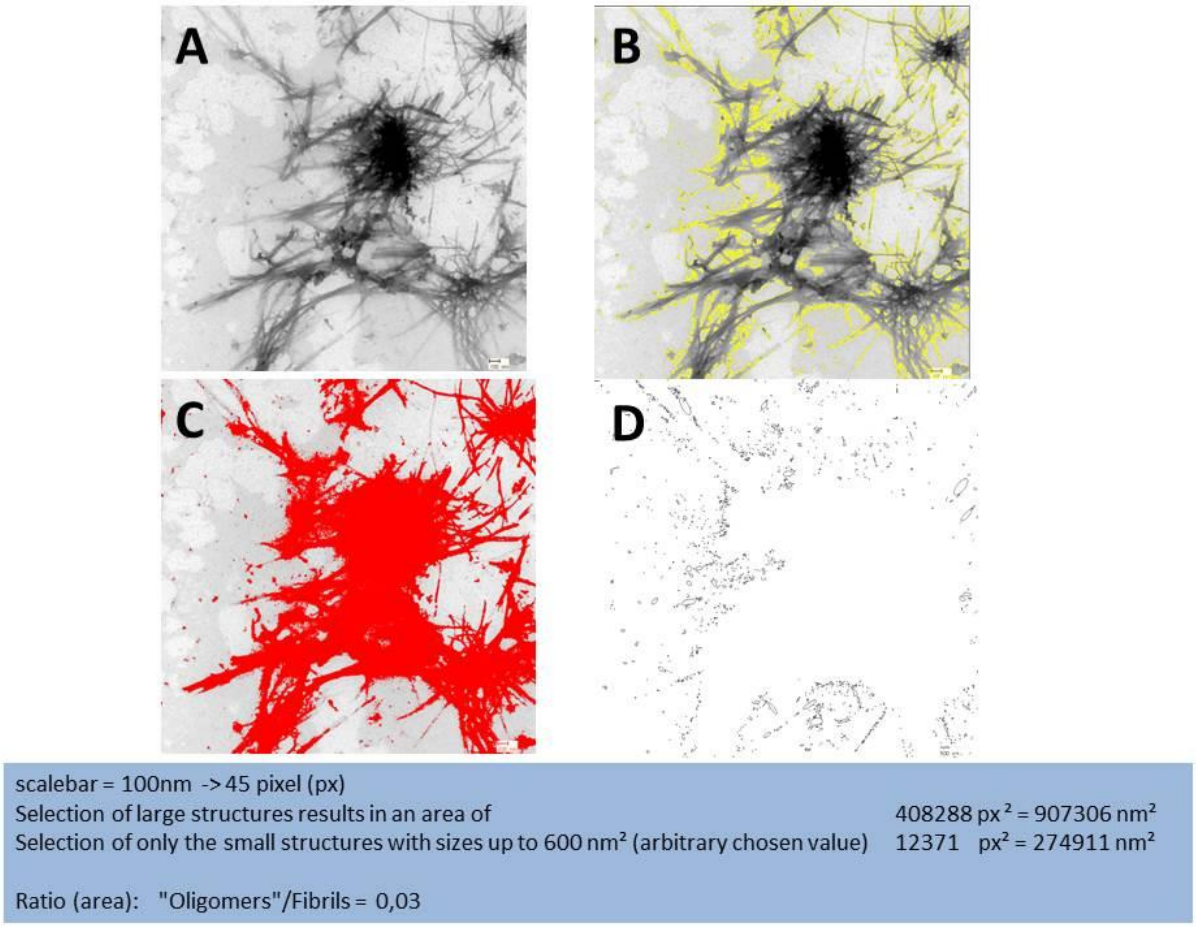


Figure S9: TEM images for the wild-type tau R2 repeat. (A) Original image, (B) selection of the large structures (fibrils), (C) selection of large structures (fibrils) and small aggregates (oligomers), (D) selection of only small aggregates (oligomers).

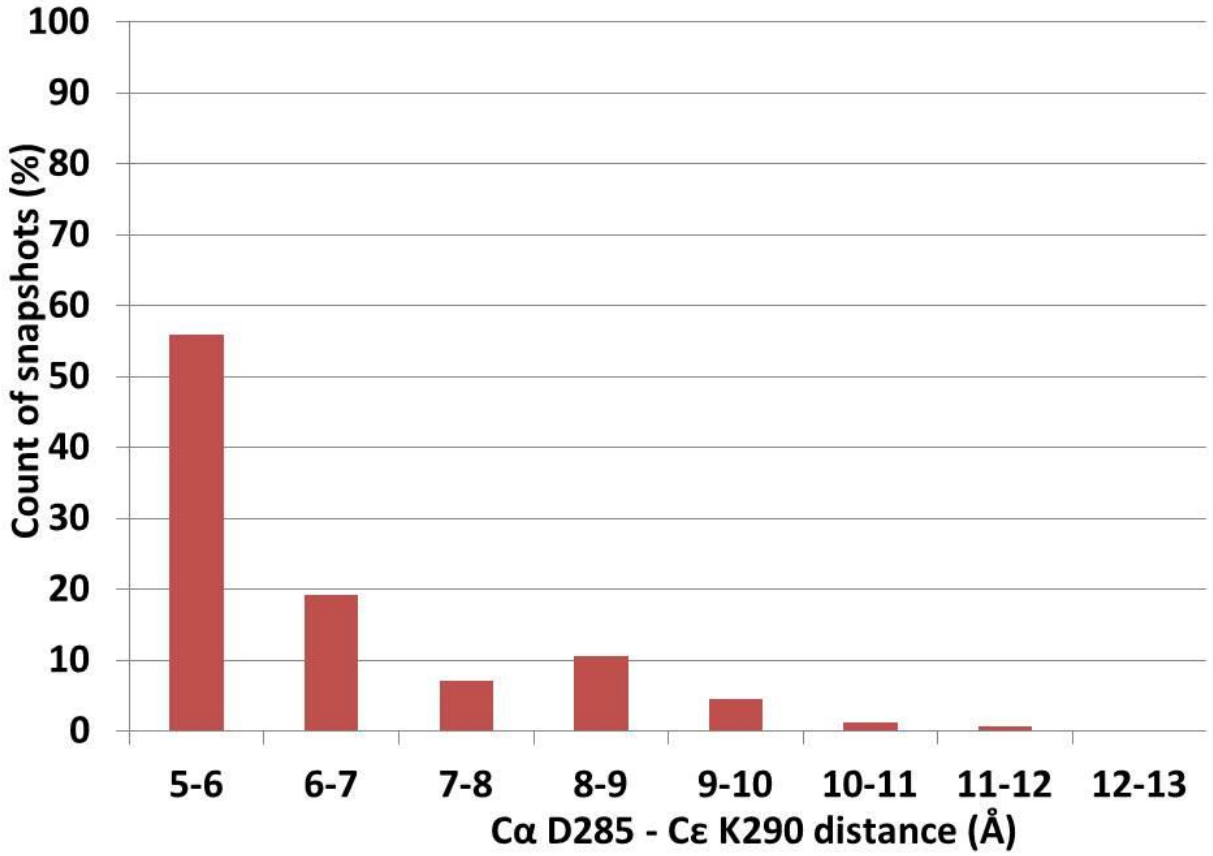
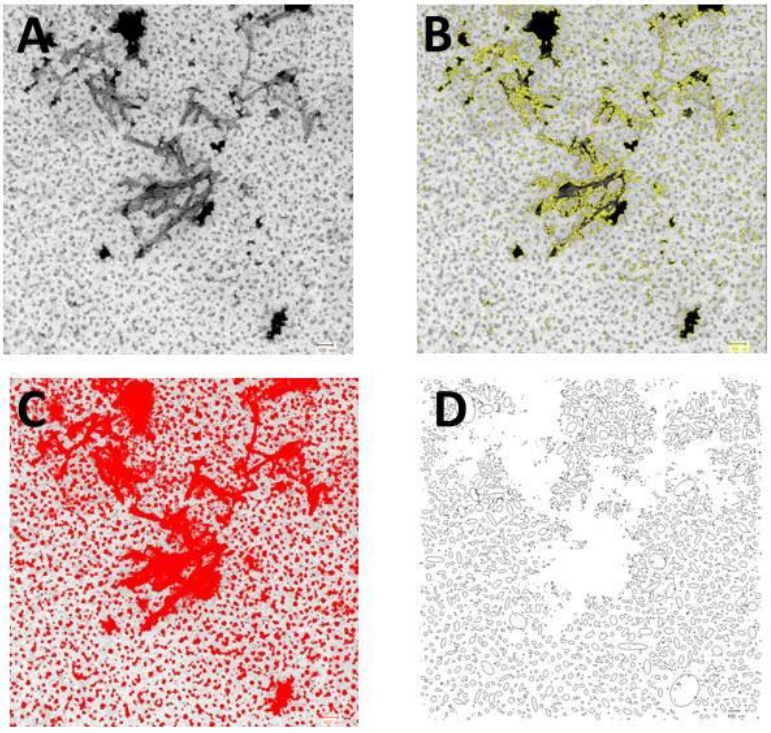


Figure S10: Cα D285-Cε K290 distances distribution in model M2 for all snapshots from MD simulations.



scalebar = 100nm -> 45 pixel (px)

Selection of large structures results in an area of	86827 px² = 192948 9nm²
Selection of only the small structures with sizes up to 600 nm² (arbitrary chosen value)	115266px² = 256146 nm²

Ratio (area): "Oligomers"/Fibrils = 1,3

Figure S11: TEM images for the ΔK280 mutated tau R2 repeat. (A) Original image, (B) selection of the large structures (fibrils), (C) selection of large structures (fibrils) and small aggregates (oligomers), (D) selection of only small aggregates (oligomers).

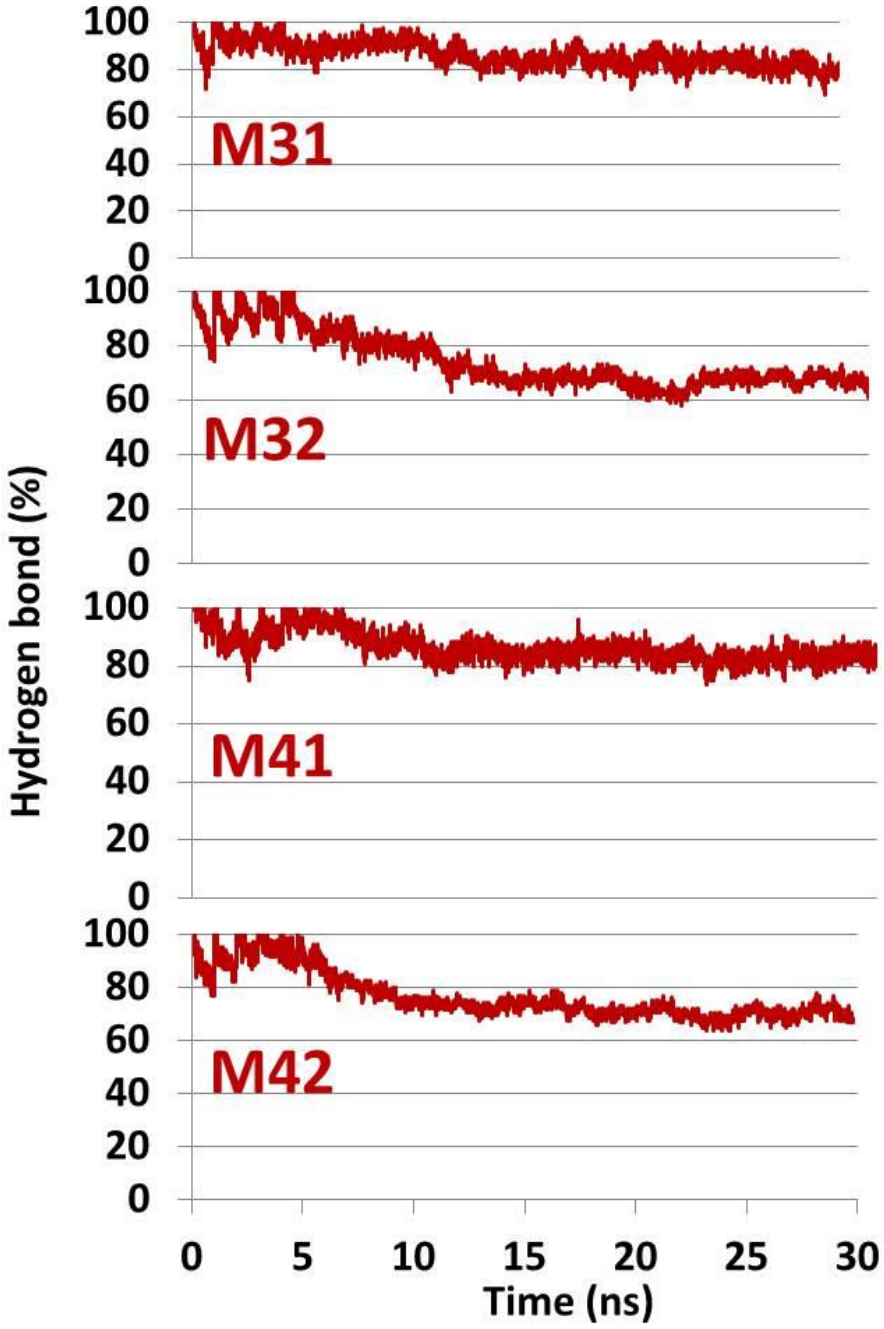


Figure S12: The fraction of the number of hydrogen bonds (in percentage) between all β-strands compared to the number in the initial oligomer for the extended ΔK280 mutated tau R2 repeat by one residue in the C-terminal models M31 and M32 and by the N-terminal: models M41 and M42.

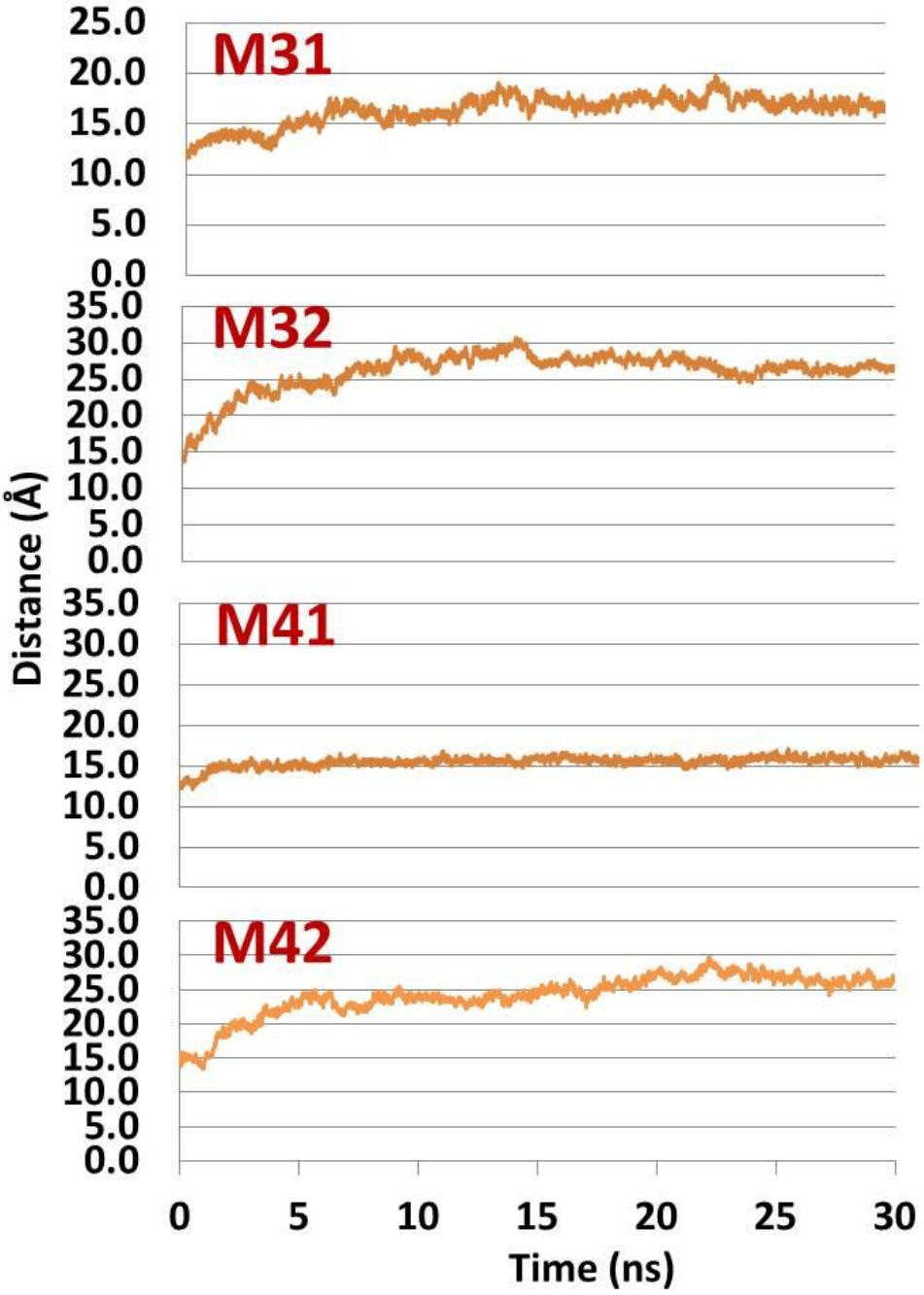


Figure S13: The averaged inter-sheet (Cα backbone-backbone) distances for M31, M32, M41 and M42 models during the MD simulations.

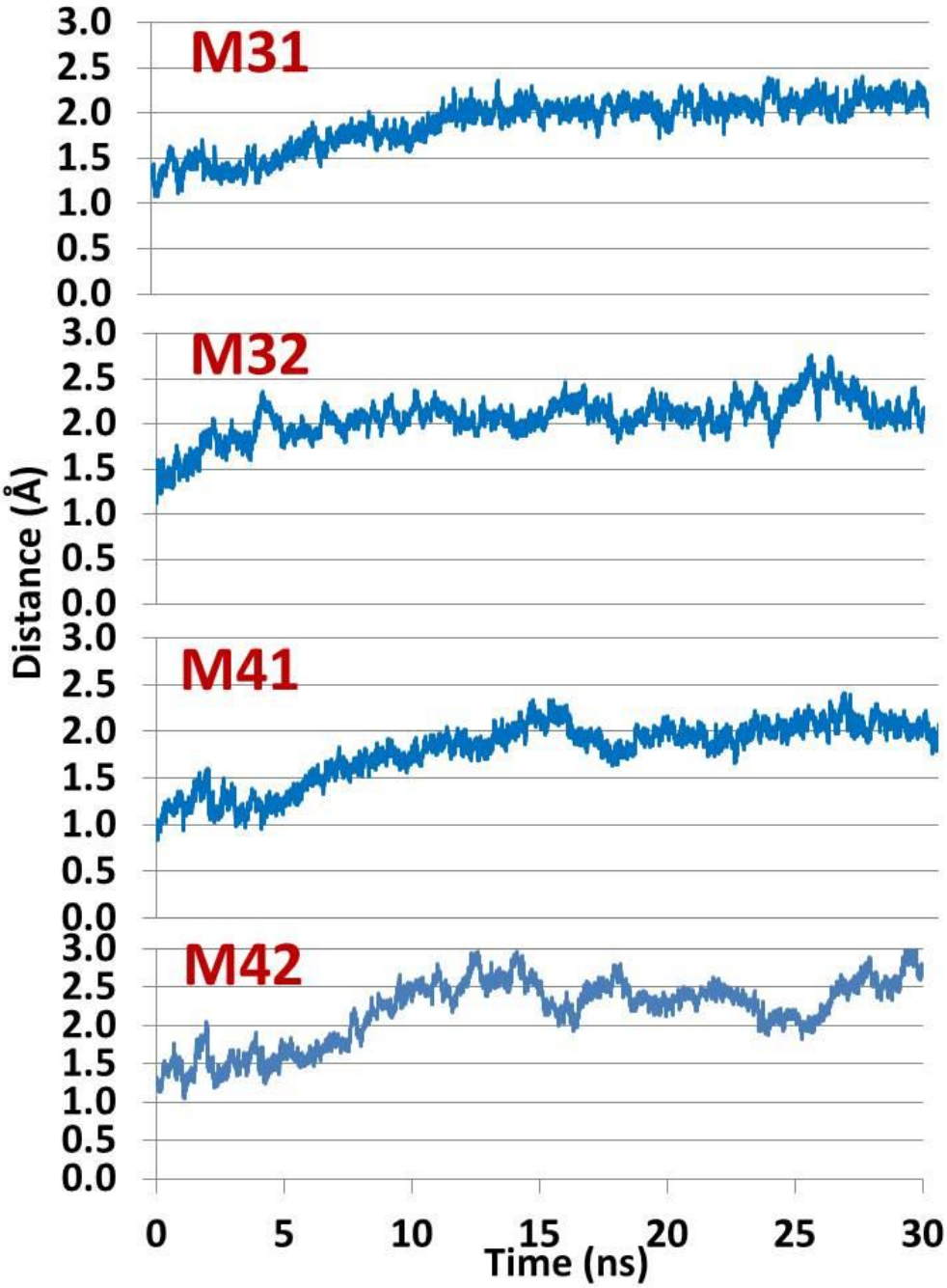


Figure S14: RMSDs of M31, M32, M41 and M42 models.

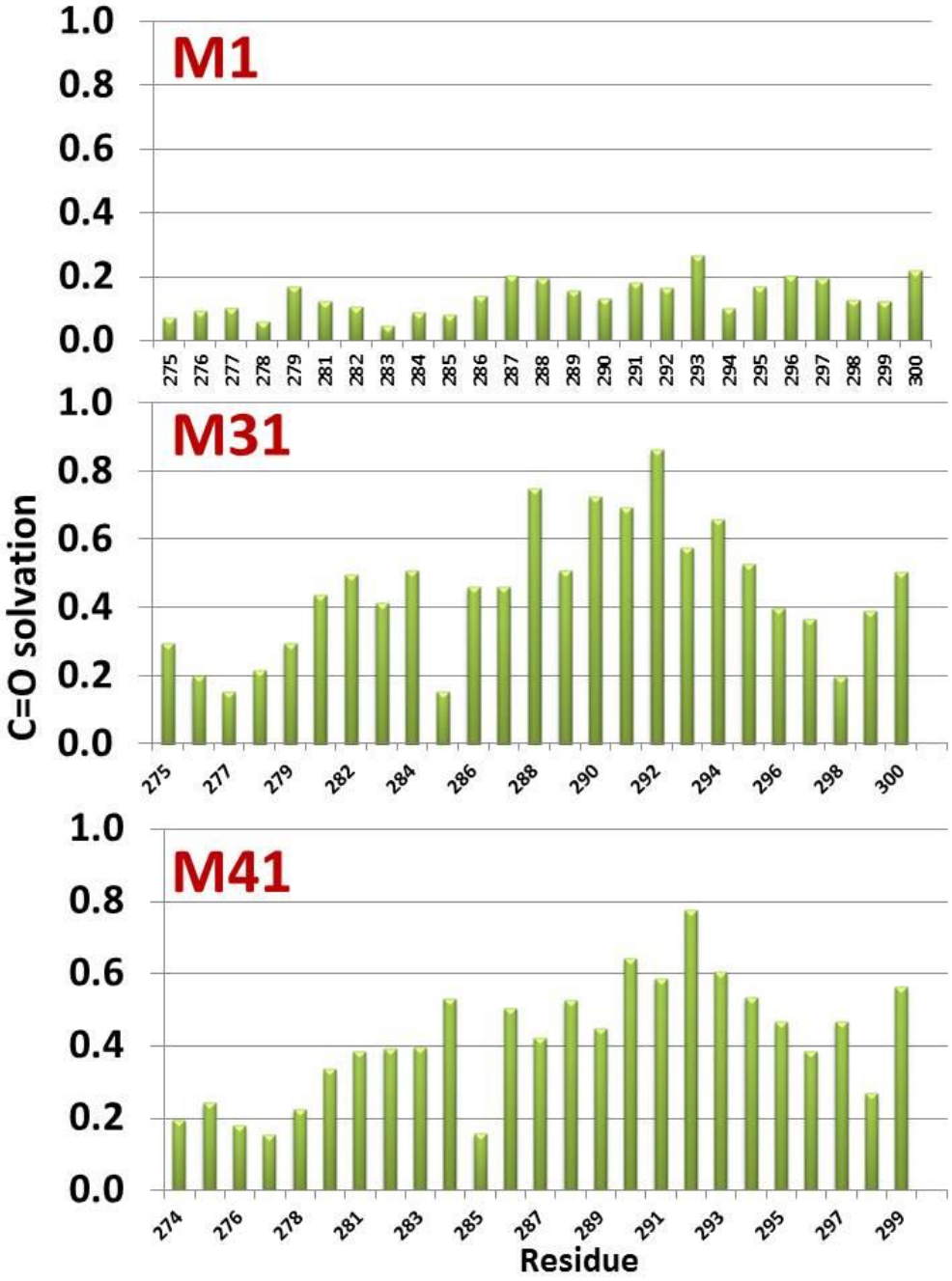


Figure S15: The average number of water molecules around each side chain Cβ carbon (within 4 Å) for M1, M31 and M41 models.

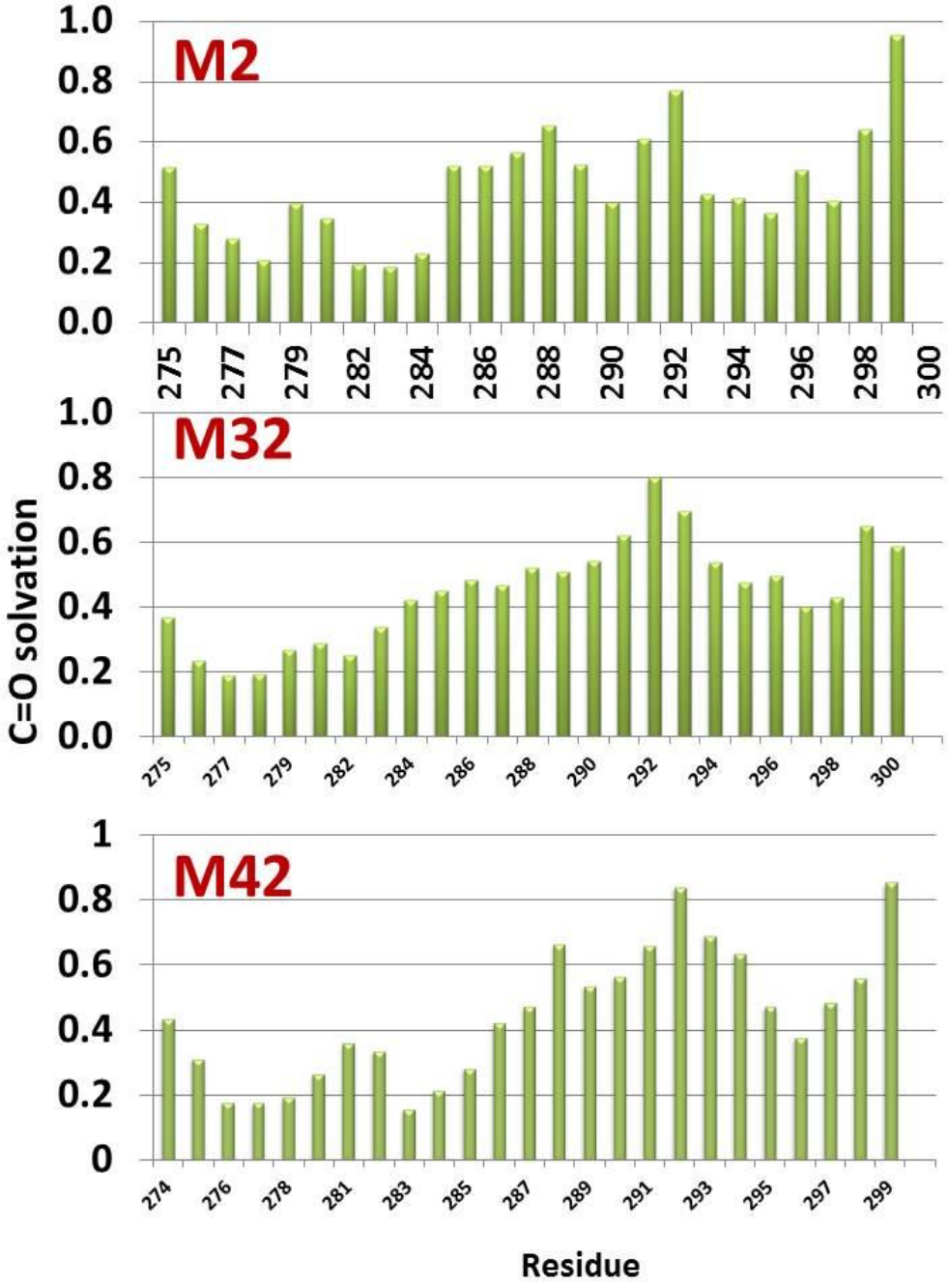


Figure S16: The average number of water molecules around each side chain Cβ carbon (within 4 Å) for M2, M32 and M42 models.

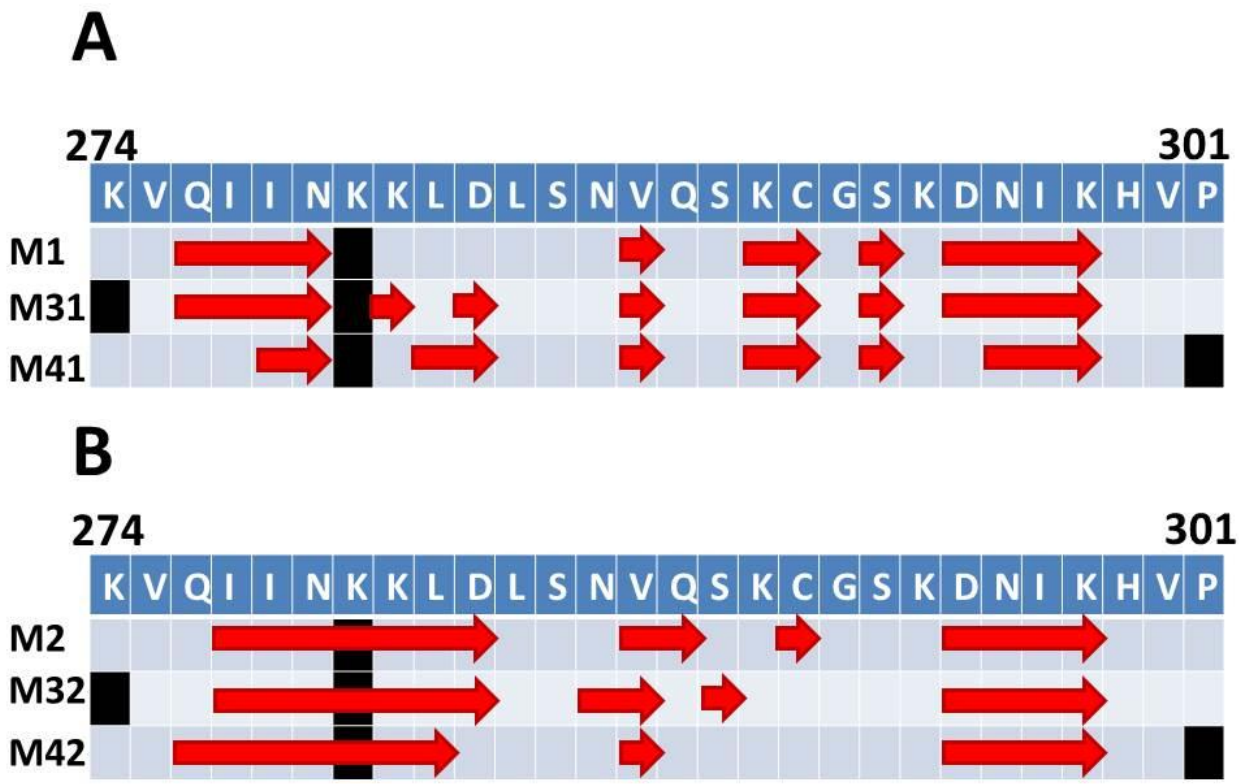


Figure S17: Schematic representations of the secondary structure (β-sheet indicated as arrow) of the M1, M31, M41, M2, M41 and M42 models proposed by our simulated constructed models. The secondary structure proposed by simulations had been estimated by computing the ψ and Φ dihedral angles from the last 5 ns for each residue. Angles with the range of ψ = 90-180° and Φ = (-60)-(-120)° were considered as β-sheet structure.[2]

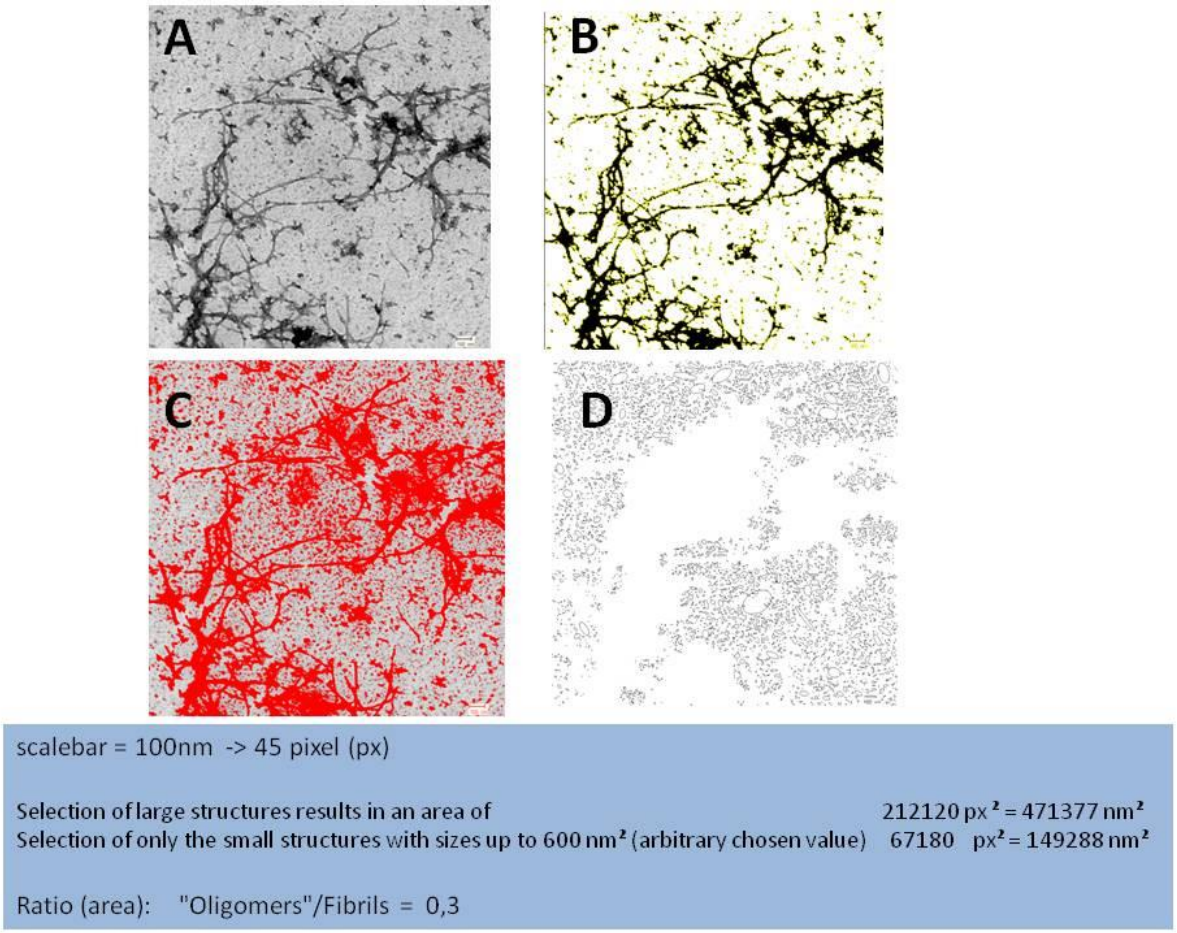


Figure S18: TEM images for the ΔK280 mutated tau R2 repeat with extension of the C-terminal by one residue. (A) Original image, (B) selection of the large structures (fibrils), (C) selection of large structures (fibrils) and small aggregates (oligomers), (D) selection of only small aggregates (oligomers).

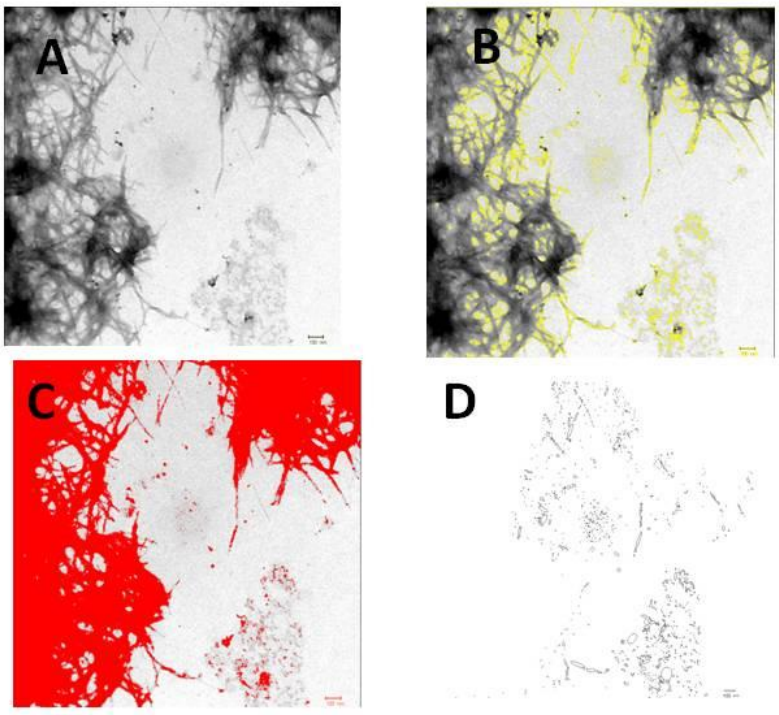


Figure S19: TEM images for the ΔK280 mutated tau R2 repeat with extension of the N-terminal by one residue. (A) Original image, (B) selection of the large structures (fibrils), (C) selection of large structures (fibrils) and small aggregates (oligomers), (D) selection of only small aggregates (oligomers).

Table S1: ^{13}C Chemical shift values of the labeled residues (in ppm) and assignment to secondary structure according to their isotropic chemical shift

Residue	Carbon atom	Wild-Type Chemical shift (ppm)	Wild-Type Secondary structure	M1/M2 Chemical shift (ppm)	M1/M2 Secondary structure
D283	Cα	51.3		51.4	
	Cβ	41.3		41.2	
	Cγ	178.5		178.8	
	Cα- Cβ	10	β-sheet	10.2	β-sheet
V287	Cα	59 / 57.9		59.1	
	Cβ	34.3 / 32.9		34.3	
	Cγ	19.1 / 20.6		19	
	Cα- Cβ	24.7	β-sheet	24.8	β-sheet
K290	Cα	52.8		53	
	Cβ	33.7		32	
	Cγ	23.3		23.2	
	Cδ	27.8		27.7	
	Cε	40.4		40.4	
	Cα- Cβ	19.4	β-sheet	21	β-sheet
G292	Cα	42.4		43.3	

Table S2: The averaged structural values of WT, M1 and M2 models from the last 5 ns.

	WT	M1	M2
Hydrogen bond (%)	60	80	70
Cα backbone-backbone distance (Å)	22.5	13.7	22.0
RMSD (Å)	2.6	1.5	2.3

Table S3: The conformational energies (computed using the GBMV calculations[3,4]) and the populations of models M1 and M2.

	Conformational energy (kcal /mol)	Standard deviation (kcal/mol)	Population (%)
M1	-4618.0	94.1	74
M2	-4349.1	87.2	26

Table S4: The experimental and computational order parameters values for the WT, M1 and M2 models.

Residue	Carbon atom	Wild-Type			M1			M2		
		Exp.	Com.	Dev. (%)	Exp.	Com.	Dev. (%)	Exp.	Com.	Dev. (%)
D283	Cα	0.837	0.957	14.3	0.872	0.959	10.0	0.872	0.956	9.6
	Cβ	0.371	0.920	148.0	0.417	0.850	103.8	0.417	0.913	118.9
V287	Cα	0.859	0.950	10.6	0.896	0.948	5.8	0.896	0.878	2.0
	Cβ	0.743	0.771	3.8	0.790	0.913	15.6	0.790	0.699	15.3
	Cγ	0.243	0.316	30.2	0.257	0.369	43.6	0.257	0.286	11.3
K290	Cα	0.776	0.862	11.1	0.822	0.877	6.7	0.822	0.891	8.4
	Cβ	0.624	0.768	23.0	0.664	0.582	12.4	0.664	0.867	30.5
	Cγ	0.339	0.642	89.4	0.408	0.458	12.3	0.408	0.747	83.1
	Cδ	0.325	0.509	56.6	0.360	0.360	0.1	0.360	0.626	73.9
	Cε	0.371	0.486	31.0	0.417	0.278	33.5	0.417	0.509	22.1
G292	Cα	0.545	0.751	37.7	0.568	0.858	5.3	0.568	0.781	37.9

Table S5: The conformational energies (computed using the GBMV calculations[3,4]) and the populations of models M31, M32, M41 and M42.

	Conformational energy (kcal /mol)	Standard deviation (kcal/mol)	Population (%)
M31	-4374.1	97.0	51
M32	-4368.3	95.0	49
M41	-4995.0	101.4	58
M42	-4941.3	92.9	42

Table S6: The averaged structural values of M31, M32, M41 and M42 models from the last 5 ns.

	M31	M32	M41	M42
Hydrogen bond (%)	80	70	85	65
Cα backbone-backbone distance (Å)	16.9	26.5	15.8	26.3
RMSD (Å)	2.1	2.3	2.1	2.4

Table S7: The experimental and computational order parameters values for M31 and M32 models.

Residue	Carbon atom	M31			M32		
		Exp.	Com.	Dev. (%)	Exp.	Com.	Dev. (%)
D283	Cα	0.896	0.961	7.3	0.896	0.966	7.8
	Cβ	0.446	0.897	101.1	0.446	0.941	111.0
V287	Cα	0.922	0.951	3.2	0.922	0.936	1.5
	Cβ	0.819	0.923	12.7	0.819	0.818	0.1
	Cγ	0.254	0.350	37.8	0.254	0.348	36.9
K290	Cα	0.858	0.892	4.0	0.858	0.911	6.1
	Cβ	0.689	0.713	3.5	0.689	0.744	7.9
	Cγ	0.689	0.435	0.5	0.689	0.681	55.8
	Cδ	0.385	0.299	22.3	0.385	0.650	68.7
	Cε	0.446	0.202	54.8	0.446	0.494	10.8
G292	Cα	0.667	0.830	24.5	0.667	0.771	15.5

Table S8: The experimental and computational order parameters values for M41 and M42 models.

Residue	Carbon atom	M41			M42		
		Exp.	Com.	Dev. (%)	Exp.	Com.	Dev. (%)
D283	Cα	0.885	0.906	2.3	0.885	0.958	8.2
	Cβ	0.885	0.892	114.4	0.885	0.940	125.8
V287	Cα	0.903	0.900	0.3	0.903	0.952	5.4
	Cβ	0.787	0.790	0.4	0.787	0.884	12.4
	Cγ	0.258	0.429	66.3	0.258	0.290	12.3
K290	Cα	0.819	0.895	9.2	0.819	0.936	14.3
	Cβ	0.661	0.683	3.4	0.661	0.738	11.6
	Cγ	0.391	0.495	26.5	0.391	0.630	61.2
	Cδ	0.351	0.386	9.9	0.351	0.475	35.5
	Cε	0.416	0.288	30.9	0.416	0.400	3.9
G292	Cα	0.558	0.837	50.0	0.558	0.838	50.2

References:

(1) Mukrasch, M. D.; Bibow, S.; Korukottu, J.; Jeganathan, S.; Biernat, J.; Griesinger, C.; Mandelkow, E.; Zweckstetter, M. *PLoS Biol* **2009**, *7*, e34.

(2) Voet, D. V., J. *Biochemistry*; 3rd Ed. ed.; John Wiley & Sons, 2004.

(3) Lee, M. S.; Feig, M.; Salsbury, F. R.; Brooks, C. L. *J Comp Chem* **2003**, *24*, 1348.

(4) Lee, M. S.; Salsbury, F. R.; Brooks, C. L. *J Chem Phys* **2002**, *116*, 10606.