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Supplemental Information

Structural Model of the Proline-Rich Domain of Huntingtin Exon-1 Fibrils

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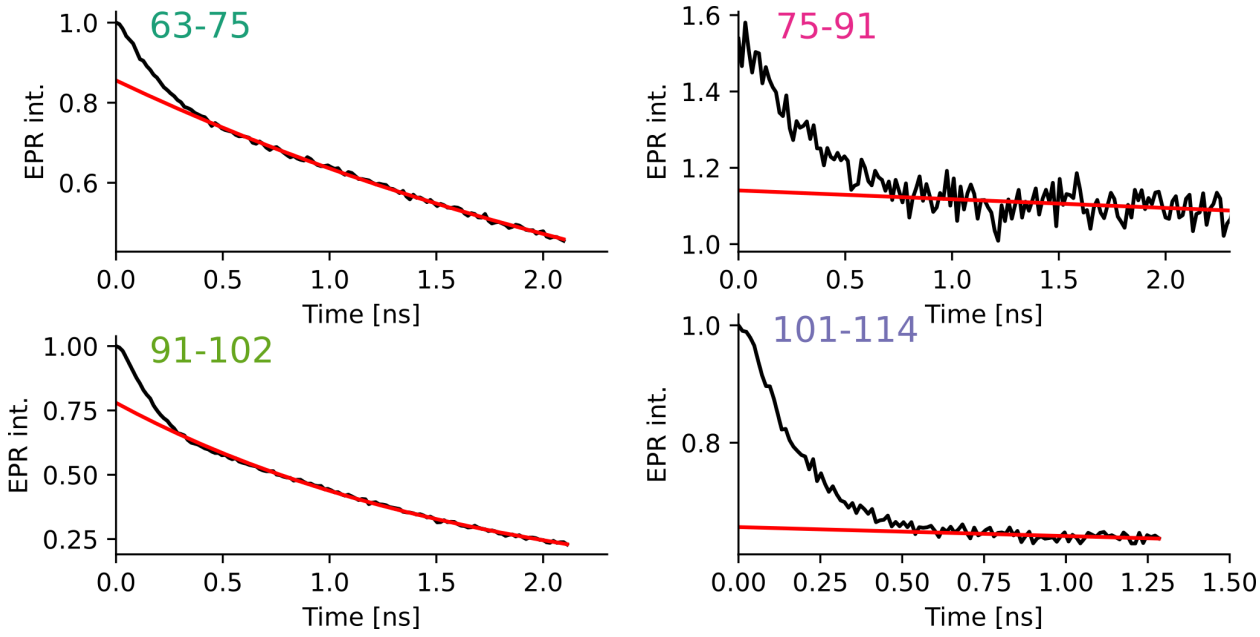

Figure S1: *Original DEER traces of distances indicated are shown in black. Backgrounds that were subtracted from these curves before fitting them to distance distributions using Tikhonov regularization are shown in red.*

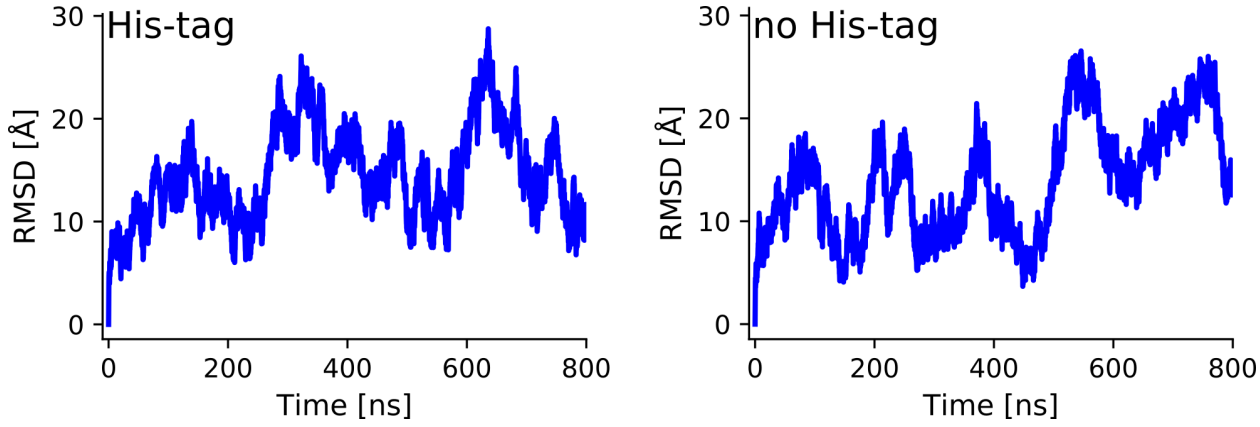

Figure S2*: Root mean square deviation (RMSD) timeline of structures in the simulation trajectory compared to the starting structure (i.e. extended PPII helix). Based on these data, we chose 200 ns of equilibration because the at this time both simulations had finished their initial increase in RMSD.*

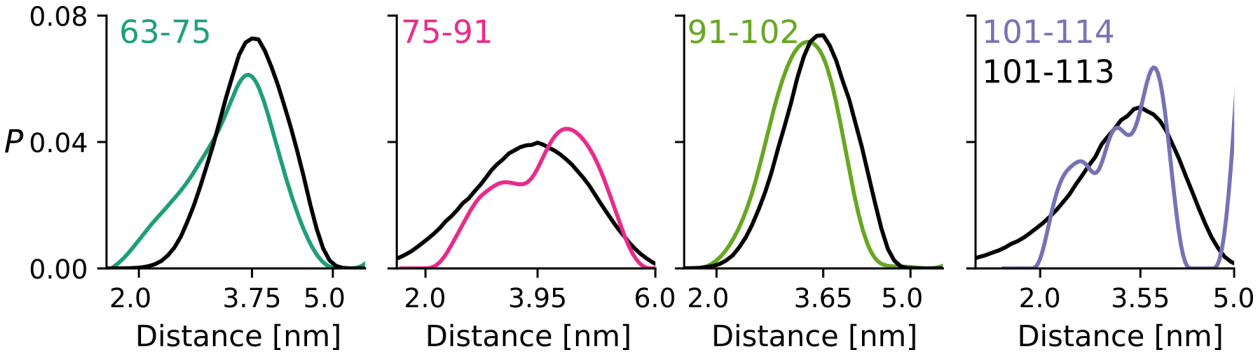

Figure S3: *DEER distance distributions (P_{DEER}) and MTSL spin label distance distributions calculated from the MD trajectory (P_{MD}) similar to Figure 3 but using the simulation without HIS-tag as basis for calculating P_{MD}. DEER distance distributions between residue pairs as indicated are shown in colors. Distance distributions derived from MD without HIS-tag using the RotamerConvolveMD algorithm (P_{MD}) are shown in black. The mode of P_{MD} is indicated on the x-axis. Note that due to the absence of the HIS-tag P_{MD} of 101-113 is shown instead of 101-114.*

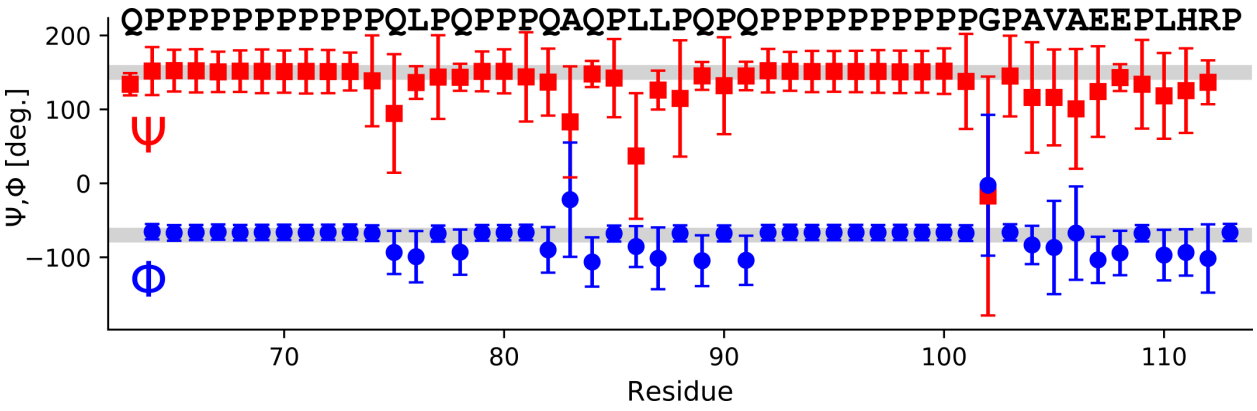

Figure S4: *Similar to the simulation without His-tag, the simulation with His-tag has relatively stable PPII helices for the polyP stretches and an increase in disorder for the L17 and C12 region. Average per residue φ and ψ angles with their standard deviations are shown in red and blue respectively. The φ and ψ angles for an idealized PPII helix is indicated with gray bars.*

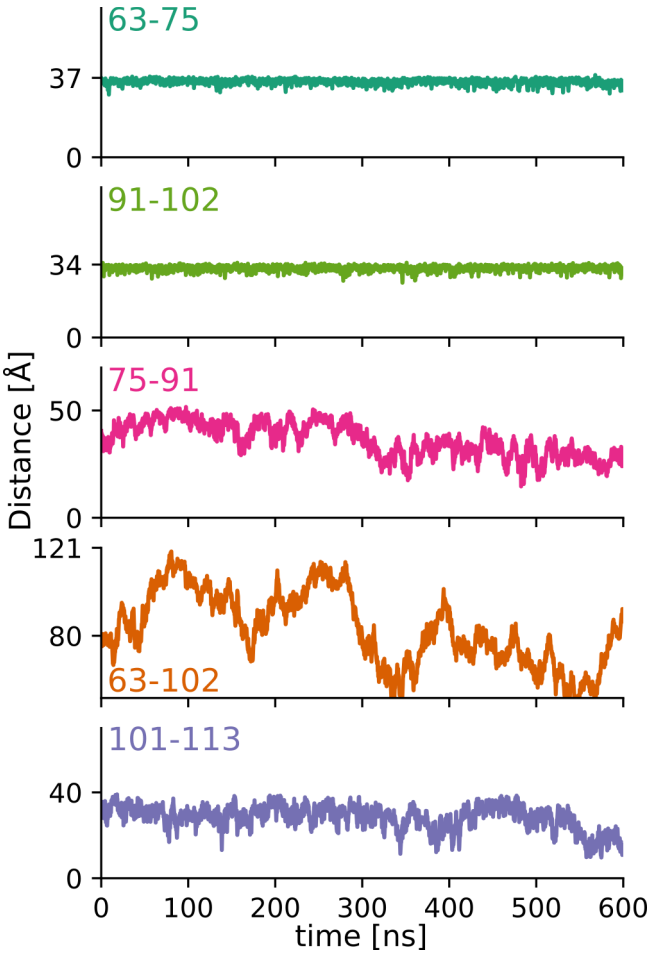

Figure S5: *Change in Cα-Cα distances (as defined in Figure 1) over time for the simulation without HIS-tag. The upper y-axis tick indicates an ideal polyproline II helical distance (see Figure 2f). All distance variations are plotted with comparable y-axis scaling.*

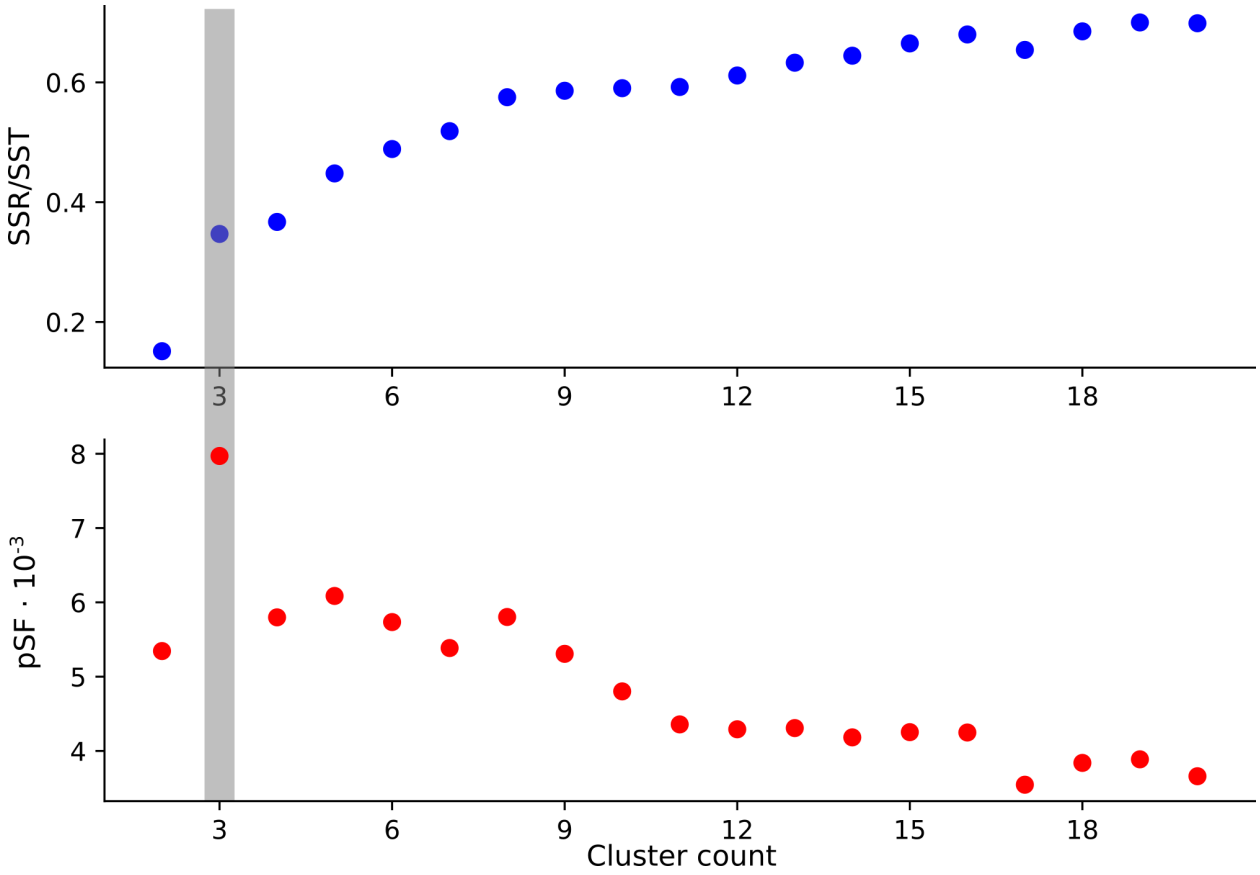

Figure S6: *SSR/SST ratios and pSF values indicate that 3 K-mean clusters are a suitable division to represent the structural ensemble of the PRD. K-mean clusters were calculated of the trajectory with His-tag using the MDAnalysis package and custom python scripts. The maximum of pSF and the beginning plateau of the SSR/SST ratio at 3 clusters is a measure of suitable clustering.*

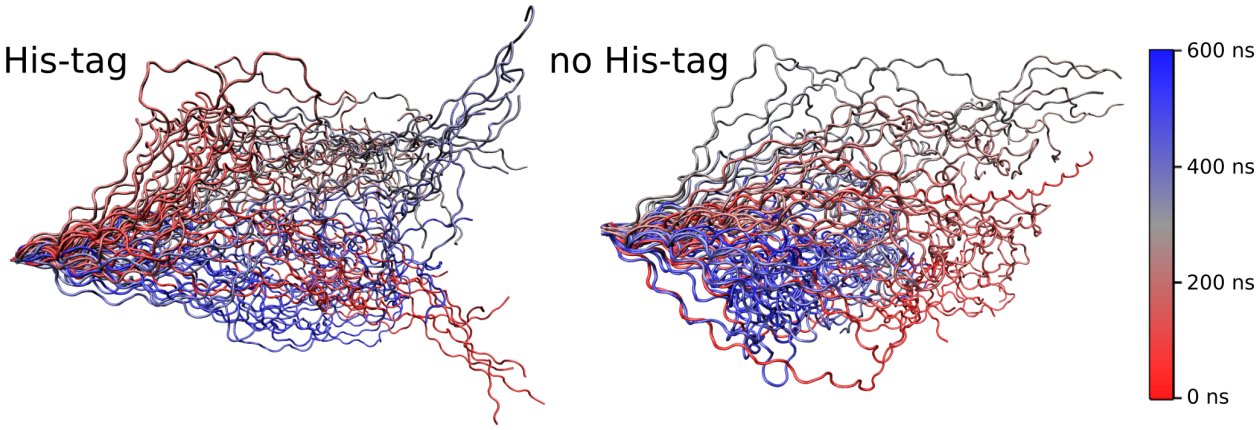

Figure S7: *Trajectory backbone representation of simulation with and without His-tag. Trajectory was plotted in 1 ns steps and colored according to the color scale bar on the right. The fixed residue Q63 is oriented to the left and the plot shows how the PRD generally extends to the right.*

Table S1: *Initial simulation attempts using different force fields, water models, and starting structures. The resulting average CA-CA distances corresponding to the DEER distance measurements are given. The simulation with implicit water (Generalized Born Implicit Solvent, GBIS) resulted in compact structures that were incompatible with the experimental data. Rosetta generated starting structures using EPR distances as restraints or not resulted in CA-CA distances that were generally shorter than experimentally determined. Using an extended polyproline II helical starting structure in combination with an explicit solvent model gave distances most compatible with the EPR data.*

Program	**Force Field**	**H_2O Model**	**Initial Structure**	**63-75**	**75-91**	**91-102**	**101-114**	**63-102**
NAMD	CHARMM36	TIP3P	Extended PPII	34.0	41.8	31.7	31.0	99.3
NAMD	CHARMM36	GBIS	Extended PPII	33.4	7.7	31.6	10.5	20.2
NAMD	CHARMM36*	TIP3P	RosettaEPR	34.3	35.2	31.0	14.8	52.8
OpenMM	AMBER ff99SB	TIP4P-D	Rosetta	28.8	27.2	31.8	21.4	79.4
OpenMM	AMBER ff99SB	TIP4P-D	Extended PPII	35.5	47.5	31.0	36.7	102.0

* DEER distances were used to generate the initial starting structure and as constraints in the following simulation. The CA-CA distances still turned out to be too short because the resulting structures were compact with the MTSL labels pointing in opposite directions to fulfill the distance constraints.