Characterization of TDP-43 NTD-RRM1-RRM2 Interacting with GU-Rich RNA Using Multi-Scale MD Simulations and SAXS Data

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TDP-43, a nuclear RNA-binding protein, is a hallmark of neurodegenerative diseases including ALS, FTLD, and Alzheimer's disease.[1] Despite its pathological significance, the structural basis of TDP-43 conformational regulation and pathological aggregation remains poorly understood, largely because of the disordered linker between the NTD and RRM1 domains and the intrinsically disordered C-terminal domain, both of which resist classical structural characterization.[2]

Experimental studies have demonstrated that the two conserved and ordered RNA recognition motifs (RRM-1 and RRM-2) of TDP-43 bind GU-rich RNA sequences with high affinity,[3] and that RNA binding can prevent pathological aggregation.[4] However, the molecular details of this RNA-mediated protective effect, including the molecular organization between ordered and disordered domains in TDP-43 assemblies, remain elusive. Molecular dynamics simulations provide a powerful approach to investigate conformational dynamics and assemblies that are difficult to characterize experimentally.

Here, a combined all-atom (AA) and coarse-grained (CG) molecular dynamics study of the TDP-43 monomer (NTD-RRM1-RRM2) is presented in the absence and presence of GU-rich RNA. AA simulations employed the AMBER03ws force field with χOL3 RNA parameters, while CG simulations used the MARTINI 3[5] force field in combination with an in-house developed MARTINI 3-compatible GU-rich RNA model to access longer-timescale dynamics. Conformational ensembles from both resolution levels were refined against experimental small-angle X-ray scattering (SAXS) data using the GAJOE genetic algorithm.[6]

Across resolutions, RNA binding consistently reshapes the conformational ensemble of TDP-43 by promoting closer packing of the RRM domains while subtly reorienting the N-terminal domain. Ensemble refinement improves agreement with SAXS data without perturbing the residue-level interaction network, indicating that the native RNA recognition interface is accurately captured by the simulations.

This work establishes a validated multiscale framework bridging atomistic accuracy, large-scale CG conformational sampling, and experimental data integration, laying the groundwork for future multimeric and condensate-level modeling of TDP-43 proteinopathy.

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