Structural Insights into Liquid–Liquid Phase Separation and Aggregation of TDP-43 Low Complexity Domain

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The aggregation of TDP-43 is a significant pathological feature of neurodegenerative disorders, notably Amyotrophic Lateral Sclerosis (ALS) and Frontotemporal Lobar Degeneration (FTLD-TDP). In pathological states, TDP-43 relocates from the nucleus to the cytoplasm, leading to the formation of harmful inclusions, characterized by nuclear depletion and cytoplasmic aggregation. These aggregates can trigger additional protein misfolding, interfere with intracellular transport, and are frequently observed to co-aggregate with other proteins. While TDP-43 aggregates are recognized as pathological entities in the final stages of these conditions, the molecular alterations that trigger the start of aggregation remain largely unclear [1].
This study aimed to explore the impact of phase separation on both self-aggregation and aggregation initiated by pre-existing aggregates of TDP-43, either the low-complexity domain (LCD), its mutant A321V, or its short aggregation-promoting regions (APRs). First, we applied various experimental techniques, theoretical calculations, and modelling to determine the fundamental physicochemical parameters of the studied biomolecules, such as monomer size, cross-sectional area, dependence of the nominal charge on pH, and isoelectric points. Additionally, molecular dynamics modeling was applied to gain insights into the mechanism of pH-driven changes in protein molecule conformations and their influence on the oligomerization process. By systematically altering the physicochemical conditions, we found that liquid–liquid phase separation (LLPS) facilitated spontaneous aggregation. However, seeded aggregation was less efficient under phase-separation conditions. By examining a wide array of conditions using the Hofmeister series of buffers, we verified that stabilizing hydrophobic interactions dominate over destabilizing electrostatic forces. RNA influences the cooperation between LLPS and aggregation in a "reentrant" manner, exerting the most significant positive effect at moderate concentrations. In summary, we conclude that conditions favoring LLPS enhance the subsequent aggregation of the TDP-43 LCD with a complex dependency but also adversely impact seeding kinetics.

Keywords:
Amyotrophic Lateral Sclerosis; Phase Separation; Biocondensates; Aggregation; TDP-43

References:

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Acknowledgements:
This work was financially supported by the National Science Center, Poland, Opus Project, 2021/43/B/ST8/01900. We gratefully acknowledge Polish high-performance computing infrastructure PLGrid (HPC Center: ACK Cyfronet AGH) for providing computer facilities and support within computational grant no. PLG/2026/019457.