

**GRADUATE SCHOOL OF BIOMEDICAL SCIENCES**

**BIOCHEMISTRY AND MOLECULAR PHARMACOLOGY**

**Ph.D. THESIS DEFENSE**

**DAVIDE TAVELLA**

MENTOR: Francesca Massi, PhD

Friday, November 16, 2018 10:00 a.m.

LRB 816

Investigating the contribution of disordered domains to the biological activity of RNA-binding proteins

Many proteins contain disordered domains under physiological conditions. These disordered regions may be functional, although under pathological conditions they may lead to protein aggregation and degradation. For my dissertation defense, I will focus my presentation on my work elucidating how the equilibrium between structural order and disorder in two proteins, MEX-5 in *Caenorhabditis elegans* and TDP-43 in human, affects their function in the cell.

Using NMR spectroscopy and MD simulations, I found that the RNA-binding domain (RBD) of MEX-5 is partially unfolded, but folds upon RNA-binding. To assess if this disorder-to-order transition contributes to MEX-5 function, I designed a variant MEX-5 where the RBD is fully folded in the absence of RNA. I found that the RNA-binding affinity and specificity of this variant are unchanged compared to the wild type. However, worms homozygous for this MEX-5 variant form massive uterine tumors. These results show that the unfolded state of MEX-5 is critical to its function *in vivo*.

The RRM2 of the ALS protein TDP-43 contains peptide regions that are particularly prone to fibril formation. In addition, RRM2 populates, to a small degree, one or more partially folded states under native conditions. I characterized the structures of these states using MD simulations including enhanced sampling methods and restraints from experimental chemical shifts. I found that in these states the protein exposes to the solvent aggregation-prone regions that are instead buried in the protein core in the native state. These results suggest a role in fibrogenesis for the transient partially folded states of TDP-43 RRM2.

|  |  |
| --- | --- |
| Mentor(s)  Francesca Massi, PhD | Dissertation Exam Committee  Lawrence Stern, PhD (Chair)  Celia Schiffer, PhD  Osman Bilsel, PhD  Brian Kelch, PhD  Vincenzo Venditti, PhD |