Structural Basis of the Vinculin–F-actin Interaction

Awardee: Gregory M. Alushin
Award: Early Independence Award
Awardee Institution: National Heart, Lung, and Blood Institute
Co-authors: Kim, L.Y., Thompson, P.M., Campbell, S.L.
Co-authors institutions: National Heart, Lung, and Blood Institute, University of North Carolina at Chapel Hill

The essential protein vinculin is a key component of cell–matrix and cell–cell adhesions, acting as a linker between these connection points and the actin cytoskeleton. Accumulation of vinculin is necessary for adhesions to sustain high traction forces, and its direct interaction with F-actin is required for mechanotransduction. Due to its physiological significance, the molecular determinants of this interface have been under scrutiny for some time, yet conflicting structural models have been reported, largely because of the absence of high-resolution analysis. To address this issue, we have obtained a subnanometer-resolution (8.5 Å, gold-standard FSC 0.143) cryo-EM reconstruction of vinculin's C-terminal "tail" domain (Vt) bound to F-actin, sufficient for constructing an unambiguous pseudo-atomic model of the interface by flexible docking of crystallographic structures of the components. Our findings support a recent experimentally derived model of the interaction (Thompson et al., Structure 2014). Additionally, we find that Vt undergoes a substantial conformational change upon actin binding, characterized by a twisting of helices 4 and 5 and unfolding of helix 1 from the bundle, a segment of which contacts the surface of the actin filament. We postulate that this structural transition enables additional interactions between vinculin and binding partners upon actin engagement. Additionally, we observe a similar conformational transition in the actin-bound structure of the cardiomyopathy-associated vinculin splice variant metavinculin. This suggests that disease mutations harbored in metavinculin helix 1 (H1') may disrupt function by modulating the H1' – actin interface.