Applying electron microscopy to better understanding targeted protein degradation

Gabriel C. Lander
Department of Integrative Structural and Computational Biology,
Scripps Research, La Jolla, CA, USA

Abstract:

The attainable resolution of structures determined by cryo-EM continues to increase, making this technique an attractive replacement for crystallography. Here, we will explore the utility of combining electron microscopy with crystallographic approaches in exploring a protein of high biomedical importance: the cereblon (CRBN) is a ubiquitin ligase (E3) substrate receptor. This protein is co-opted by CRBN E3 ligase modulatory drug (CELMoD) agents that target therapeutically relevant proteins for degradation. Prior crystallographic studies defined the drug-binding site within CRBN's thalidomide-binding domain (TBD), but the allostery of drug-induced neosubstrate binding remained unclear. Cryo-electron microscopy analyses of the DNA damage-binding protein 1 (DDB1)-CRBN in the absence and presence of CELMoD compounds showed that association of CELMoD compounds to the TBD is necessary and sufficient for triggering an allosteric rearrangement from an open conformation to the canonical closed conformation. The neosubstrate Ikaros only stably associates with the closed CRBN conformation, illustrating the importance of allostery for CELMoD compound efficacy and informing structure-guided design strategies to improve therapeutic efficacy.