Contribution ID: **26** Type: **not specified**

Structural basis for the recognition of type-2 N-degron substrate by PRT1 E3 ubiquitin ligase

Thursday 26 June 2025 13:00 (40 minutes)

PROTEOLYSIS1 (PRT1), an N-recognin of Arabidopsis thaliana, has a specificity for recognizing the N-terminal aromatic hydrophobic residue (Tyr/Phe/Trp) of its substrates, subsequently degrading them through ubiquitylation. Here, I represent the complex structures of the ZZ domain of A. thaliana PRT1 (PRT1ZZ) with bulky hydrophobic N-degron peptides. Unlike other ZZ domains, the binding site of PRT1ZZ has a novel structure organized into two hydrophobic regions. The N-terminal aromatic residues of N-degron interact hydrophobically with Ile333 and Phe352 in the flexible loops, which undergo dramatic conformational change. A third N-degron residue participating in the hydrophobic network with N-recognin was also identified. Moreover, the ubiquitylation assay of PRT1 using the N-terminal tyrosine-exposed substrate BIG BROTHER showed that the tandem RING organization in PRT1 is critical for its robust activity. Therefore, the current study expands our knowledge of the structural repertoire in the N-degron pathway and provides insights into the regulation of E3 ubiquitin ligases containing tandem RING domains.

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Session Classification: Session 4: X-ray Crystallography for Bio-Science (Chair : Sun-Shin Cha)