Molecular Mechanism of Ubiquitin Recognition by GGA3 GAT Domain

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GGA (Golgi-localizing, γ-adaptin ear domain homology, ARF-binding) proteins, which constitute a family of clathrin coat adaptor proteins, have recently been shown to be involved in the ubiquitin-dependent sorting of receptors, through the interaction between the C-terminal three-helix-bundle of the GAT (GGA and Tom1) domain (C-GAT) and ubiquitin. We report the crystal structure of human GGA3 C-GAT in complex with ubiquitin. At the center of the interface, three pockets on the hydrophobic Ile44 surface of ubiquitin accommodate three hydrophobic residues from helices α1 and α2 of C-GAT. Two distinct orientations of ubiquitin Arg42 determine the shape and the charge distribution of the third pocket of the ubiquitin Ile44 surface, leading to tight and loose binding modes of C-GAT. The flexibility of the third pocket explains why ubiquitin Ile44 surface can interact with structurally divergent ubiquitin binding modules. In addition, biochemical and NMR data suggest another hydrophobic binding site on C-GAT helices α2 and α3, opposite to the first binding site, also binds ubiquitin although weakly. The double-sided ubiquitin binding provides the GAT domain with higher efficiency in recognizing ubiquitinated receptors for lysosomal receptor degradation.

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