Ubiquitin Binding Mechanism of Hrs-UIM

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Mono-ubiquitination plays an important role in degradation of growth factor receptors. Monoubiquitinated receptors are sorted into multivesicular bodies, which then fuse with lysosomes. Hepatocyte growth factor-regulated trypsine kinase substrate (Hrs) is one of the essential proteins for the sorting mechanism. Hrs can interact with ubiquitin by its ubiquitin interaction motif (UIM). The ability to bind ubiquitin is essential for the function of Hrs in sorting of ubiquitinated proteins. We present a crystal structure of an Hrs-UIM/ubiquitin complex. Data sets were collected to 1.7Å resolution with good statistics (R_{merge} = 5.1%) using synchrotron radiation (1.0 Å wavelength) at beamline PF-AR NW12 of Photon Factory, Tsukuba, Japan. Using the molecular replacement method, we have determined and refined the complex structure. It consists of two ubiquitin molecules and one UIM peptide, suggesting that Hrs-UIM can interact with two ubiquitin molecules simultaneously. Together with a binding assay using surface plasmon resonance, the crystal structure sheds light on the molecular mechanism of double-side ubiquitin recognition by Hrs-UIM, which facilitates efficient binding of multi-mono-ubiquitinated protein complexes. We propose the double-sided UIM as a new sub-class of UIM based on a sequence search which yielded a number of putative double-sided UIMs.

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