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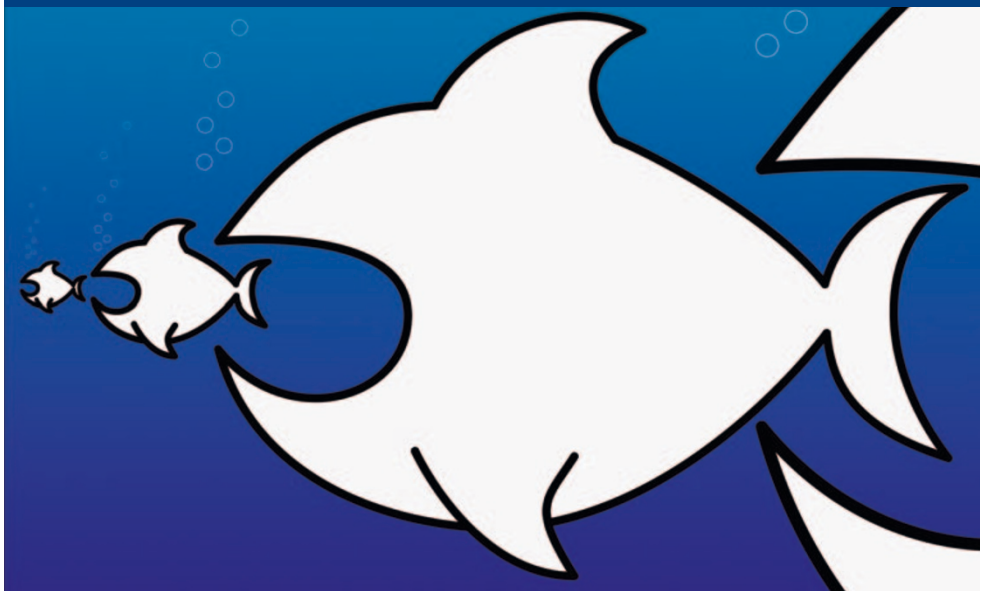
POST-TRANSLATIONAL MODIFICATION

Chain reaction



> …a poly-ubiquitin chain can be transferred *en bloc* from the E2 to a Lys residue of a target substrate.

Ubiquitylation is a versatile post-translational modification, whereby an E2 ubiquitin-conjugating enzyme, aided by an E3 ubiquitin ligase, links ubiquitin to a Lys residue of a target protein. In addition to being added individually, ubiquitin molecules can be linked together through their own Lys residues to form polyubiquitin chains. For example, Lys48-linked polyubiquitin chains form on substrates to mark them for degradation by the proteasome. Two recent papers provide insight into how these polyubiquitin chains might be synthesized.

It has been presumed that polyubiquitin chains are constructed at the site of the substrate's Lys residue by the sequential addition of ubiquitin monomers to the distal end of a growing chain. A study by Li *et al.* has challenged this view by showing that polyubiquitin chains rapidly form on an E2 when purified E2, E3 and ubiquitin are incubated together. The chains are formed on the catalytic Cys residue, from where ubiquitin is transferred to a substrate. Further, the authors showed that, at least *in vitro*, a polyubiquitin chain can be transferred *en bloc* from the E2 to a Lys residue of a target substrate. Whether this occurs *in vivo* is not known, but Li *et al.* were able to detect polyubiquitin chains attached to the catalytic Cys residue of the same E2 in proteasome-inhibitor-treated cells.

In a separate study, Ravid and Hochstrasser observed the *in vivo* formation of polyubiquitin chains on the catalytic Cys residue of yeast Ubc7, the orthologue of the E2 studied by Li *et al.* The stability of Ubc7 depends on a binding partner, Cue1, because Ubc7 is downregulated in the absence of Cue1. The signal for degradation of Ubc7 was shown to be the polyubiquitin chain formed on the catalytic Cys residue, but Cue1 does not rescue Ubc7 by preventing the formation of these chains. Instead, Cue1 appears to function as a protective chaperone that allows the assembly of polyubiquitin chains on Ubc7, but prevents them from being recognized as degradation signals by the proteasome.

So how are polyubiquitin chains generated on E2? Li *et al.* show that ubiquitin can be transferred from one E2 to another to form a di-ubiquitin chain. This probably requires the E2 enzymes to dimerize, which has been reported for several different E2s. Further investigation is required to determine how the di-ubiquitin chains are then elongated to the longer ubiquitin chains that are necessary for efficient protein degradation.

James Pickett

ORIGINAL RESEARCH PAPERS Li, W. *et al.* A ubiquitin ligase transfers preformed polyubiquitin chains from a conjugating enzyme to a substrate. *Nature* 18 Feb 2007 (doi: 10.1038/nature05542) | Ravid, T. & Hochstrasser, M. Autoregulation of an E2 enzyme by ubiquitin-chain assembly on its catalytic residue. *Nature Cell Biol.* 21 Feb 2007 (doi: 10.1038/ncb1558)
FURTHER READING Hochstrasser, M. Lingering mysteries of the ubiquitin-chain assembly. *Cell* **124**, 27–34 (2006) | Welchman, R. L. *et al.* Ubiquitin and ubiquitin-like proteins as multifunctional signals. *Nature Rev. Mol. Cell Biol.* **6**, 599–609 (2005)