UBIQUITYLATION

Choosing to be single

How does the cellular machinery decide whether proteins should be monoubiquitylated rather than polyubiquitylated? This decision is crucial because being tagged with a single copy of ubiquitin determines whether proteins become involved in controlling endocytosis rather than being targeted for destruction by the proteasome.

Now, three reports shed light on this process by showing that ubiquitin recognition and monoubiquitylation depend on the ubiquitin-interacting motif (UIM), which is found in many groups of proteins, including endocytic proteins and ubiquitin ligases.

Polo and colleagues report in *Nature* that the determinants that are necessary for monoubiquitylation of the endocytosis-regulating proteins Eps15 and eps15R mapped to the carboxy-terminal regions of the proteins — a region that contains two UIM domains. Deletion analysis of this region showed that one of these domains was crucial for monoubiquitylation and binding to ubiquitin-containing proteins, whereas the other domain was crucial only for monoubiquitylation. The function of the UIM domain was further illustrated by the demonstration that the ubiquitin ligase Nedd4 — which catalyses the transfer of ubiquitin — could monoubiquitylate eps15 but not a UIM-deleted mutant.

Two reports in *Nature Cell Biology* show that other proteins that are involved in endocytosis — epsins, which interact directly with clathrin and are involved in internalization, and Hrs (hepatocyte growth-factor regulated tyrosine kinase substrate), which is involved in recruiting clathrin to endosomes — also function through the UIM domain. Hicke and colleagues and Stenmark and colleagues found that UIM domains in epsins and Hrs, respectively, were required for ubiquitin binding. Stenmark and colleagues also found that ubiquitylated proteins localize specifically to Hrs- and clathrin-coated microdomains, which indicates that the Hrs/clathrin coat concentrates ubiquitylated proteins before the formation of vesicles.

There is obviously more to be learnt about the UIM and its function, but as Riezman concludes in a News and Views article, this work "marks the beginning of an exciting quest".

Simon Frantz

References and links

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NEURODEGENERATIVE DISEASE

The early results of misfolding

It has long been known that many serious diseases are associated with problems in protein folding. Insoluble clusters of misfolded proteins are a common pathological feature of conditions such as Alzheimer's disease and encephalopathies, but the connection between these disease-linked aggregates and the detrimental effects on health is still unclear. Now, two studies published in *Nature* provide evidence that early, misfolded intermediates on the path to forming these aggregates might be responsible for at least part of the damage.

Newly synthesized proteins adopt characteristic three-dimensional structures on the basis of their individual amino-acid sequences, but a common feature of all protein folding is that hydrophobic residues tend to be hidden within the body of the folded molecule. When folding goes wrong, and hydrophobic residues are exposed on the surface of proteins, the misfolded proteins clump together to form aggregates. Once of a certain size, these are deposited within or outside cells as insoluble deposits, the most well known being the amyloid-β (Aβ) plaques seen in Alzheimer's disease.

Investigating the toxicity of intermediates in the path from simple Aβ monomers to insoluble Aβ plaques, Walsh *et al.* found that soluble Aβ oligomers, formed from two or three associated Aβ molecules, were able to interfere with synaptic plasticity. When injected into rat brains, Aβ oligomers derived from cultured cells expressing mutated amyloid-β precursor protein (APP) were able to inhibit hippocampal long-term potentiation. This ability to disrupt processes thought to be crucial to memory formation indicates that Aβ oligomers might be the leading culprits in inhibiting neuronal function in Alzheimer's disease. Furthermore, the extent of the dementia experienced by Alzheimer's patients has previously been found to correlate well with their levels of soluble Aβ, but not with the density of amyloid plaques.

The second paper, by Bucciantini *et al.*, similarly finds that a species formed early during the process of protein aggregation is damaging to cell function. In this case, however, the misfolded protein is the amino-terminal domain of the bacterial regulatory HypF protein (HypF-N), one not normally associated with any disease state. The authors have previously shown that under suitable conditions, amyloid fibrils can be induced to form from proteins not thought to be linked to any disease, and here, they study the cytotoxicity of intermediates on the route to forming the HypF-N fibrils. Their finding that aggregates and protofibrils of HypF-N are deadly to cultured mouse fibroblasts, but that mature HypF-N fibrils are not, leads them to conclude that minute amounts of early aggregates of proteins might impair cellular function, without insoluble deposits being present. The fact that a protein totally unassociated with neurological disease can cause these effects raises the possibility that the spontaneous development of early aggregates of proteins that are not under suspicion at present might underlie the development of various diseases.

Together, these demonstrations of aggregate pathogenicity indicate that an understanding of the mechanisms that underlie aggregate formation, and of why the cellular control mechanisms that normally prevent it break down, might be just as important for future therapeutic approaches as an understanding of the biology of individual disease-associated proteins.

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References and links

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