UBIQUITIN AND PROTEOSOME

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UBIQUITIN
(originally, ubiquitous immunopoietic polypeptide)

- small regulatory protein, found in almost all tissues (ubiquitously/universally) of eukaryotic cells.
- It directs proteins to compartments in the cell, including the proteasome
- Ubiquitin can be attached to proteins and label them for destruction
- Ubiquitin tags can also direct proteins to other locations in the cell, where they control other protein and cell mechanisms.
- The ubiquitylation system was initially characterized as an ATP-dependent proteolytic system (Apaf-1) present in cellular extracts.

- A heat-stable polypeptide ATP-dependent proteolysis factor 1 (APF-1), was found to become covalently attached to the model protein substrate lysozyme in an ATP- and Mg^{2+}-dependent process.
- Multiple APF-1 molecules were linked to a single substrate molecule by an isopeptide linkage, and conjugates were found to be rapidly degraded with the release of free **APF-1**.
- Soon after APF-1-protein conjugation was characterized, **APF-1 was identified as ubiquitin**.

Ubiquitination (ubiquitylation)

- Is an enzymatic, protein post-translational modification (PTM) process in which an isopeptide bond between a lysine of the target protein and the C-terminal glycine of ubiquitin occur
- Following addition of a single ubiquitin moiety to a protein substrate (monoubiquitination), further ubiquitin molecules can be added forming a polyubiquitin chain.

- The ubiquitination system functions in a wide variety of cellular processes, including:
  - Antigen processing
  - Apoptosis
  - Biogenesis of organelles
  - Cell cycle and division
  - DNA transcription and repair
  - Differentiation and development
  - Immune response and inflammation
  - Neural and muscular degeneration
  - Morphogenesis of neural networks
  - Modulation of cell surface receptors, ion channels and the secretory pathway
  - Response to stress and extracellular modulators
  - Ribosome biogenesis
  - Viral infection

- Monoubiquitylation of proteins also targets the localization of proteins - **proteasome**.
- The **proteasome** is a barrel-shaped structure with two chambers, within which proteolysis occurs.
- Proteins are rapidly degraded into small peptides (usually 3-24 amino acid residues in length).
- Ubiquitin molecules are cleaved off from the protein immediately prior to destruction and are recycled for further use.
- Although the majority of proteasomal substrates are ubiquitinated, there are examples of **non-ubiquitinated proteins targeted to the proteasome**.

Diagnostic use:

- Immunohistochemistry using antibodies to ubiquitin can identify abnormal accumulations of this protein inside cells, indicating a disease process.
- These protein accumulations are referred to as inclusion bodies. Examples include:
  - Neurofibrillary tangles in Alzheimer's disease
  - Lewy body in Parkinson's disease
  - Mallory bodies in alcoholic liver disease etc.

Ubiquitin-like modifiers

- ubiquitin-like proteins (UBLs) that modify cellular targets include:
  - small ubiquitin-like modifier (SUMO),
  - ubiquitin cross-reactive protein (UCRP, also known as interferon-stimulated gene-15 : ISG15),
  - ubiquitin-related modifier-1 (URM1),
  - neuronal-precursor-cell-expressed developmentally downregulated protein-8 (NEDD8),
  - human leukocyte antigen F-associated (FAT10),
  - autophagy-8 (ATG8) and -12 (ATG12),
  - Fau ubiquitin-like protein (FUB1),
  - MUB (membrane-anchored UBL),
  - ubiquitin fold-modifier-1 (UFM1)
  - ubiquitin-like protein-5 (UBL5).

Ubiquitin-like modifiers

- **SUMO** modification often acts antagonistically to that of ubiquitination and serves to stabilize protein substrates.
- Proteins conjugated to UBLs are typically not targeted for degradation by the proteasome, but rather function in diverse regulatory activities.
- Attachment of UBLs might
  - alter substrate conformation,
  - affect the affinity for ligands or other interacting molecules,
  - alter substrate localization
  - influence protein stability.

Proteasome

- Proteasomes are protein complexes inside all eukaryotes and archaea, and in some bacteria.
- In eukaryotes, they are located in the nucleus and the cytoplasm.
- The main function of the proteasome is to degrade unneeded or damaged proteins by proteolysis through proteases
- Proteasomes are part of a major mechanism by which cells regulate the concentration of particular proteins and degrade misfolded proteins.
- Proteins are tagged for degradation with a small protein called ubiquitin.
- Once a protein is tagged with a single ubiquitin molecule, this result in polyubiquitin chain formation that is bound by the proteasome, allowing it to degrade the tagged protein.

- The proteasomal degradation pathway is essential for many cellular processes, including
  - the cell cycle,
  - the regulation of gene expression,
  - responses to oxidative stress.
- Before the discovery of the **ubiquitin proteasome system**, protein degradation in cells was thought to rely mainly on **lysosomes**

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