Antigen processing: Ubiquitination & Proteasome degradation

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Introduction

Reactome is open-source, open access, manually curated and peer-reviewed pathway database. Pathway annotations are authored by expert biologists, in collaboration with Reactome editorial staff and cross-referenced to many bioinformatics databases. A system of evidence tracking ensures that all assertions are backed up by the primary literature. Reactome is used by clinicians, geneticists, genomics researchers, and molecular biologists to interpret the results of high-throughput experimental studies, by bioinformaticians seeking to develop novel algorithms for mining knowledge from genomic studies, and by systems biologists building predictive models of normal and disease variant pathways.

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Literature references


Reactome database release: 73

This document contains 1 pathway and 9 reactions (see Table of Contents)
Antigen processing: Ubiquitination & Proteasome degradation

Stable identifier: R-HSA-983168

Compartments: cytosol

Intracellular foreign or aberrant host proteins are cleaved into peptide fragments of a precise size, such that they can be loaded on to class I MHC molecules and presented externally to cytotoxic T cells. The ubiquitin-26S proteasome system plays a central role in the generation of these class I MHC antigens.

Ubiquitination is the mechanism of adding ubiquitin to lysine residues on substrate protein leading to the formation of a polyubiquitinated substrate. This process involves three classes of enzyme, an E1 ubiquitin-activating enzyme, an E2 ubiquitin-conjugating enzyme, and an E3 ubiquitin ligase. Polyubiquitination through lysine-48 (K48) generally targets the substrate protein for proteasomal destruction. The protease responsible for the degradation of K48-polyubiquitinated proteins is the 26S proteasome. This proteasome is a two subunit protein complex composed of the 20S (catalytic core) and 19S (regulatory) proteasome complexes. The proteasome eliminates most of the foreign and non-functional proteins from the cell by degrading them into short peptides; only a small fraction of the peptides generated are of the correct length to be presented by the MHC class I system. It has been calculated that between 994 and 3122 protein molecules have to be degraded for the formation of a single, stable MHC class I complex at the cell surface, with an average efficiency of 1 in 2000 (Kloetzel et al. 2004, Princiotta et al. 2003).

Literature references


Editions

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**E1 mediated ubiquitin activation**

**Location:** Antigen processing: Ubiquitination & Proteasome degradation

**Stable identifier:** R-HSA-983153

**Type:** transition

**Compartments:** cytosol

Ubiquitin is activated in an ATP-dependent manner, catalyzed by E1 ubiquitin-activating enzymes. In the first step of ubiquitin activation, the E1 enzyme binds ATP, Mg\(^{2+}\) and ubiquitin, and catalyses ubiquitin C-terminal acyl-adenylation (Ubiquitin-AMP). In the second step, the catalytic cystine in the E1 attacks the ubiquitin-adenylate (Ub-AMP) to form the activated ubiquitin-E1 thioester-bonded complex and an AMP leaving group. The intermediate reaction involving the formation of the ubiquitin-AMP complex is not represented here.

**Followed by:** Transfer of ubiquitin from E1 to E2

**Literature references**


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Activated ubiquitin is transferred from E1 to the active site cystine of ubiquitin conjugating enzymes (E2s) via a trans-esterification reaction. E2s catalyze covalent attachment of ubiquitin to target proteins. They all share an active-site ubiquitin-binding cysteine residue and are distinguished by the presence of a ubiquitin-conjugating catalytic (UBC) fold required for binding of distinct ubiquitin ligases or E3s. Once conjugated to ubiquitin, the E2 molecule binds one of several E3s (Glickman et al. 2002).

**Preceded by:** E1 mediated ubiquitin activation

**Followed by:** Interaction of E3 with substrate and E2-Ub complex

**Literature references**


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Interaction of E3 with substrate and E2-Ub complex

Location: Antigen processing: Ubiquitination & Proteasome degradation

Stable identifier: R-HSA-983157

Type: binding

Compartments: cytosol

Ubiquitin E3 ligases confer specificity to ubiquitination by recognizing target substrates and mediating transfer of ubiquitin from an E2 ubiquitin-conjugating enzyme to substrate (Raymond et al. 2009). E3 ligases includes a large, diverse set of proteins characterized by several defining motifs which include a HECT (homologous to E6-associated C-terminus), RING (Really Interesting New Gene) and U-box domains. The E3 ligases can be multisubunit complexes rather than a single polypeptide. Presently three different kinds of E3 complexes have been described called SCF, APC, and VHL. E3 ligases binds to both substrate and an E2 thioesterified with ubiquitin (E2-Ub).

Preceded by: Transfer of ubiquitin from E1 to E2

Followed by: Transfer of Ub from E2 to substrate and release of E2

Literature references


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Transfer of Ub from E2 to substrate and release of E2

**Location:** Antigen processing: Ubiquitination & Proteasome degradation

**Stable identifier:** R-HSA-983140

**Type:** transition

**Compartments:** cytosol

Interaction of E3 with both substrate and E2-Ub, brings them into proximity so that ubiquitin is transferred from E2 to the substrate. In most cases the transfer of ubiquitin is direct from E2 to substrate, but in a small subset of E3s, it occurs via a covalent E3-Ub thioester intermediate (Raymond et al. 2009).

**Preceded by:** Interaction of E3 with substrate and E2-Ub complex

**Followed by:** Polyubiquitination of substrate

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Polyubiquitination of substrate

**Location:** Antigen processing: Ubiquitination & Proteasome degradation

**Stable identifier:** R-HSA-983156

**Type:** omitted

**Compartments:** cytosol

Monoubiquitinated substrate acquires additional ubiquitin modifications in the form of multiple single attachments or a ubiquitin chain. Polyubiquitin chains added through K48 residue of ubiquitin typically targets the substrate for degradation (Raymond et al. 2009).

**Preceded by:** Transfer of Ub from E2 to substrate and release of E2

**Followed by:** Release of E3 from polyubiquitinated substrate

**Literature references**


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Release of E3 from polyubiquitinated substrate

**Location:** Antigen processing: Ubiquitination & Proteasome degradation

**Stable identifier:** R-HSA-983147

**Type:** dissociation

**Compartments:** cytosol

K48 polyubiquitinated substrate dissociates from E3 to become a substrate for a multicatalytic complex called the 26S proteasome.

**Preceded by:** Polyubiquitination of substrate

**Followed by:** Proteasomal cleavage of substrate

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Proteasomal cleavage of substrate

**Location:** Antigen processing: Ubiquitination & Proteasome degradation

**Stable identifier:** R-HSA-983150

**Type:** omitted

**Compartments:** cytosol

The 26S proteasome complex consists of the 20S catalytic core particle which harbours the proteolytically active sites and the regulatory 19S particle which is responsible for substrate interaction. This process generates a vast number (perhaps hundreds) of different peptides, depending on the length and sequence of the substrate protein. Only a small fraction of these peptides (nearly 10%) form the exact length to be presented by class I MHC; most (approximately 70%) are too short to bind. The remaining proteasome products (10-20%) are N-terminally extended precursors that require additional cleavage by cytosolic aminopeptidases for presentation by MHC class I molecules.

**Preceded by:** Release of E3 from polyubiquitinated substrate

**Followed by:** THOP1 cleaves oligopeptide fragment (8-16aa), Trimming of N-ter extended precursor fragments by cytosolic aminopeptidases

**Literature references**


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Trimming of N-ter extended precursor fragments by cytosolic aminopeptidases

Location: Antigen processing: Ubiquitination & Proteasome degradation

Stable identifier: R-HSA-983162

Type: omitted

Compartments: cytosol

Some peptides generated by the 26S proteasome are too long to bind to MHC class I molecules. These N-terminal extended precursor peptides may be trimmed by cytosolic aminopeptidases, such as Tripeptidyl peptidase II (TPP2), puromycin-sensitive aminopeptidase (PSA), bleomycin hydrolase (BH), and leucine aminopeptidase (LAP).

Preceded by: Proteasomal cleavage of substrate

Followed by: THOP1 cleaves oligopeptide fragment (8-16aa)

Literature references


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2010-10-29 Authored, Edited Garapati, P V.
2011-02-11 Reviewed Elliott, T.
THOP1 cleaves oligopeptide fragment (8-16aa)

**Location:** Antigen processing: Ubiquitination & Proteasome degradation

**Stable identifier:** R-HSA-8940641

**Type:** omitted

**Compartments:** cytosol

Thimet oligopeptidase (THOP1, EP24.15) is a zinc-dependent peptidase of the metallopeptidase M3 family (Pierotti et al. 1990). It was first described as a neuropeptide-degrading enzyme present in the soluble fraction of brain homogenates (Orlowski et al. 1983). However, its predominant location in the cytosol and nucleus suggests that extracellular degradation of neuropeptides and hormones is not its primary function (Fontenele-Neto et al. 2001). THOP1 can metabolize peptides within cells, thereby affecting antigen presentation and G protein-coupled receptor signal transduction. It was shown in vivo to participate in antigen presentation through MHC-I (Silva et al. 1999, Kim et al. 2003, Yorl et al. 2003) and in vitro to bind (Portaro et al. 1999) or degrade (Saric et al. 2001) some MHC-I associated peptides. THOP1 can degrade a broad range of intracellular peptides containing 5–17 amino acids (Oliveira et al. 2001, Berti et al. 2009). Substrate size is restricted because its catalytic center is located in a deep channel (Ray et al. 2004). THOP1 can both degrade existing peptides and generate new peptides, making it a versatile enzyme for regulating intracellular peptide function including antigen presentation (Berti et al. 2009, Russo et al. 2012).

Cytotoxic T lymphocytes (CTLs) recognize peptides presented by HLA class I molecules on the cell surface. The C terminus of these CTL epitopes is considered to be produced by the proteasome, but is complemented by THOP1 and other cytosolic endopeptidases such as Nardilysin (Kessler et al. 2011, Oliveira & van Hall 2015).

**Preceded by:** Proteasomal cleavage of substrate, Trimming of N-ter extended precursor fragments by cytosolic aminopeptidases

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