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.

The development of Reactome is supported by grants from the US National Institutes of Health (P41 HG003751), University of Toronto (CFREF Medicine by Design), European Union (EU STRP, EMI-CD), and the European Molecular Biology Laboratory (EBI Industry program).

Literature references


Reactome database release: 71

This document contains 4 pathways and 9 reactions (see Table of Contents)
TNFR2 non-canonical NF-kB pathway

Stable identifier: R-HSA-5668541

Compartments: plasma membrane, nucleoplasm

Tumor necrosis factor-alpha (TNFA) exerts a wide range of biological effects through TNF receptor 1 (TNFR1) and TNF receptor 2 (TNFR2). Under normal physiological conditions TNFR2 exhibits more restricted expression, being found on certain subpopulation of immune cells and few other cell types (Grell et al. 1995). TNFR1 mediated signalling pathways have been very well characterized but, TNFR2 has been much less well studied. TNFR1 upon activation by TNFA activates apoptosis through two pathways, involving the adaptor proteins TNFR1-associated death domain (TRADD) and fas-associated death domain (FADD). In contrast, TNFR2 signalling especially in highly activated T cells, induces cell survival pathways that can result in cell proliferation by activating transcription factor NF-kB (nuclear factor-kB) via the alternative non-canonical route. TNFR2 signalling seems to play an important role, in particular for the function of regulatory T cells. It offers protective roles in several disorders, including autoimmune diseases, heart diseases, demyelinating and neurodegenerative disorders and infectious diseases (Faustman & Davis 2010).

Activation of the non-canonical pathway by TNFR2 is mediated through a signalling complex that includes TNF receptor-associated factor (TRAF2 and TRAF3), cellular inhibitor of apoptosis (cIAP1 and cIAP2), and NF-kB-inducing kinase (NIK). In this complex TRAF3 functions as a bridging factor between the cIAP1/2:TRAF2 complex and NIK. In resting cells cIAP1/2 in the signalling complex mediates K48-linked polyubiquitination of NIK and subsequent proteasomal degradation making NIK levels invisible. Upon TNFR2 stimulation, TRAF2 is recruited to the intracellular TRAF binding motif and this also indirectly recruits TRAF1 and cIAP1/2, as well as TRAF3 and NIK which are already bound to TRAF2 in unstimulated cells. TRAF2 mediates K63-linked ubiquitination of cIAP1/2 and this in turn mediates cIAP dependent K48-linked ubiquitination of TRAF3 leading to the proteasome-dependent degradation of the latter. As TRAF3 is degraded, NIK can no longer interact with TRAF1/2:cIAP complex. As a result NIK concentration in the cytosol increases and NIK gets stabilised and activated. Activated NIK phosphorylates IKKalpha, which in turn phosphorylates p100 (NFkB2) subunit. Phosphorylated p100 is also ubiquitinated by the SCF-beta-TRCP ubiquitin ligase complex and is subsequently processed by the proteasome to p52, which is a transcriptionally competent NF-kB subunit in conjunction with RelB (Petrus et al. 2011, Sun 2011, Vallabhapurapu & Karin 2009).
**Literature references**


**Editions**

<table>
<thead>
<tr>
<th>Date</th>
<th>Action</th>
<th>Author(s)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2015-01-26</td>
<td>Authored, Edited</td>
<td>Garapati, P V.</td>
</tr>
<tr>
<td>2015-05-12</td>
<td>Reviewed</td>
<td>Ware, CF., Rajput, A., Virgen-Slane, R.</td>
</tr>
</tbody>
</table>
Mitogen-activated protein kinase kinase kinase 14 (MAP3K14 also named as NIK) is a central signalling component of the non-canonical pathway which integrates signals from TNFR2 and activates IkB kinase-alpha (IkBA) for triggering p100 phosphorylation and processing (Sun 2011). A tight control of NIK stability is essential to achieve controlled activation of the noncanonical NF-kB signalling upon TNFR2 activation. In unstimulated cells the level of NIK protein is extremely low, which is due to constant degradation by a ubiquitination-dependent mechanism (Liao et al. 2004). Proteasomal degradation of NIK occurs on assembly of a regulatory complex through TRAF3 complexed with NIK and TRAF2 which exists in a pre-assembled complex with cellular Inhibitor of apoptosis 1 (cIAP1) and cIAP2 (cIAP1,2:TRAF2::TRAF3:NIK). The c-IAPs do not directly contact TNFR2, but rather associate with TRAF2 through their N-terminal BIR motif-comprising domain (Rothe et al. 1995, Shu et al. 1996). TRAF3 functions as a bridging factor between cIAP1/2-TRAF2 E3 complex and NIK enabling cIAP to mediate K48 linked ubiquitination of NIK (Zarnegar et al. 2008, Vallabhapurapu et al. 2008, Li et al. 2004).

Followed by: cIAP1,2 ubiquitinates NIK in cIAP1,2:TRAF2::TRAF3:NIK

Literature references


Editions

<table>
<thead>
<tr>
<th>Date</th>
<th>Action</th>
<th>Author</th>
</tr>
</thead>
<tbody>
<tr>
<td>2015-01-26</td>
<td>Authored, Edited</td>
<td>Garapati, P V.</td>
</tr>
<tr>
<td>2015-05-12</td>
<td>Reviewed</td>
<td>Ware, CF., Rajput, A., Virgen-Slane, R.</td>
</tr>
</tbody>
</table>
cIAP1,2 ubiquitinates NIK in cIAP1,2:TRAF2::TRAF3:NIK

Location: TNFR2 non-canonical NF-κB pathway

Stable identifier: R-HSA-5668534

Type: omitted

Compartments: cytosol

TNF receptor-associated factor 2 (TRAF2) and cellular inhibitors of apoptosis 1 and 2 (cIAP1,2) are both RING-containing ubiquitin protein ligase (E3), and the interplay between these two protein families and other molecules in the receptor complex is critical in the propagation of downstream signals (Rothe et al. 1995, Yang et al., 2000). cIAP1,2 initiates proteasomal degradation of NIK by mediating its K48-linked ubiquitination (K48-(Ub)n).

Preceded by: TRAF3:NIK binds TRAF2:cIAP1/2

Followed by: 26Sproteasome degrades K48polyUb-NIK

Literature references


Editions

<table>
<thead>
<tr>
<th>Date</th>
<th>Action</th>
<th>Author/Editor</th>
</tr>
</thead>
<tbody>
<tr>
<td>2015-01-26</td>
<td>Authored, Edited</td>
<td>Garapati, P V.</td>
</tr>
<tr>
<td>2015-05-12</td>
<td>Reviewed</td>
<td>Ware, CF., Rajput, A., Virgen-Slane, R.</td>
</tr>
</tbody>
</table>
**26S proteasome degrades K48polyUb-NIK**

**Location:** TNFR2 non-canonical NF-kB pathway

**Stable identifier:** R-HSA-5668520

**Type:** omitted

**Compartments:** cytosol

Polyubiquitinated NIK is targeted for proteasomal degradation and as a consequence the NIK levels are invisible and thus prevent non-canonical NF-kB activation.

**Preceded by:** cIAP1,2 ubiquitinates NIK in cIAP1,2:TRAF2::TRAF3:NIK

**Literature references**


**Editions**

<table>
<thead>
<tr>
<th>Date</th>
<th>Action</th>
<th>Author(s)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2015-01-26</td>
<td>Authored, Edited</td>
<td>Garapati, P V.</td>
</tr>
<tr>
<td>2015-05-12</td>
<td>Reviewed</td>
<td>Ware, CF., Rajput, A., Virgen-Slane, R.</td>
</tr>
</tbody>
</table>
Tumor necrosis factor alpha (TNFA) exists in two biologically active forms, a transmembrane TNF-alpha (tmTNFA) and secretory TNF-alpha (sTNFA). tmTNFA is expressed as a functional 26kDa homotrimer transmembrane protein, which can be cleaved by a metalloproteinase TNFα-converting enzyme (TACE or ADAM17) to release the extracellular C-terminal portion with 17kDa, sTNFA. Tumor necrosis factor receptor superfamily member 1B (TNFRSF1B or hereafter referred as TNFR2) preferentially binds with membrane integrated TNFA (Grell et al. 1995) and can activate the noncanonical NF-kB pathway (Rauert et al. 2010) as well as canonical NF-kB pathway (Marchetti et al. 2004).

Followed by: 2xTNFR2 binds TNFA trimer:TNFR2

Literature references


**2xTNFR2 binds TNFA trimer:TNFR2**

**Location:** TNFR2 non-canonical NF-κB pathway

**Stable identifier:** R-HSA-5668467

**Type:** binding

**Compartments:** plasma membrane

TNF receptor-associated factor 2 (TNFR2) undergoes trimerization upon binding of transmembrane bound TNFA (tmTNFA) trimers, which leads to recruitment of TRAF2 to the intracellular TRAF-binding motif of TNFR2.

**Preceded by:** TNFA binds TNFR2

**Followed by:** cIAP1,2:TRAF2:TRAF3:NIK complex binds TNFR2 trimer

**Literature references**


**Editions**

<table>
<thead>
<tr>
<th>Date</th>
<th>Action</th>
<th>Author/Editor</th>
</tr>
</thead>
<tbody>
<tr>
<td>2015-01-26</td>
<td>Authored, Edited</td>
<td>Garapati, P V.</td>
</tr>
<tr>
<td>2015-05-12</td>
<td>Reviewed</td>
<td>Ware, CF., Rajput, A., Virgen-Slane, R.</td>
</tr>
</tbody>
</table>
cIAP1,2:TRAF2:TRAF3:NIK complex binds TNFR2 trimer

**Location:** TNFR2 non-canonical NF-kB pathway

**Stable identifier:** R-HSA-5668417

**Type:** binding

**Compartments:** plasma membrane, cytosol

Tumor necrosis factor receptor 2 (TNFR2) signalling starts with recruitment of the adaptor protein TNF receptor-associated factor 2 (TRAF2) to the intracellular TRAF-binding motif and consequently to indirect recruitment of the TRAF2 associated proteins cellular inhibitors of apoptosis 1 and 2 (cIAP1 and cIAP2), along with a complex of TRAF3 and NF-kB-inducing kinase (NIK) which already interact with TRAF2 in unstimulated cells (Song & Donner. 1994, Rothe et al. 1995, Rothe et al. 1994). In resting cells usually NIK is targeted for ubiquitination and proteasomal degradation however in stimulated cells NIK escapes the ubiquitination by the cIAP1,2:TRAF2::TRAF3 complex due to rapid degradation of TRAF3.

**Preceded by:** 2xTNFR2 binds TNFA trimer:TNFR2

**Followed by:** TRAF2 ubiquitinates cIAP1,2 in cIAP1,2:TRAF1:TRAF2:TRAF3:NIK

**Literature references**


**Editions**

<table>
<thead>
<tr>
<th>Date</th>
<th>Action</th>
<th>User</th>
</tr>
</thead>
<tbody>
<tr>
<td>2015-01-26</td>
<td>Authored, Edited</td>
<td>Garapati, P V.</td>
</tr>
<tr>
<td>2015-05-12</td>
<td>Reviewed</td>
<td>Ware, CF., Rajput, A., Virgen-Slane, R.</td>
</tr>
</tbody>
</table>
TNF receptor superfamily (TNFSF) members mediating non-canonical NF-kB pathway

Location: TNFR2 non-canonical NF-kB pathway

Stable identifier: R-HSA-5676594

Compartments: plasma membrane, cytosol, extracellular region

Activation of NF-kB is fundamental to signal transduction by members of the TNFRSF. Expression of NF-kB target genes is essential for mounting innate immune responses to infectious microorganisms but is also important for the proper development and cellular compartmentalization of secondary lymphoid organs necessary to orchestrate an adaptive immune response.

NF-kB transcription factor family is activated by two distinct pathways: the canonical pathway involving NF-kB1 and the non-canonical pathway involving NF-kB2. Unlike NF-kB1 signalling, which can be activated by a wide variety of receptors, the NF-kB2 pathway is typically activated by a subset of receptor and ligand pairs belonging to the tumor necrosis factor receptor (TNF) super family (TNFRSF) members. These members include TNFR2 (Rauert et al. 2010), B cell activating factor of the TNF family receptor (BAFFR also known as TNFRSF13C) (Kayagaki et al. 2002, CD40 (also known as TNFRSF5) (Coope et al. 2002, lymphotoxin beta-receptor (LTBR also known as TNFRSF3) (Dejardin et al. 2002), receptor activator for nuclear factor kB (RANK also known as TNFRSF11A) (Novack et al. 2003), CD27 and Fibroblast growth factor-inducible immediate-early response protein 14 (FN14 also known as TNFRSF12A) etc. These receptors each mediate specific biological roles of the non-canonical NF-kB. These non-canonical NF-kB-stimulating receptors have one thing in common and is the presence of a TRAF-binding motif, which recruits different TNF receptor-associated factor (TRAF) members, particularly TRAF2 and TRAF3, to the receptor complex during ligand ligation (Grech et al. 2004, Bishop & Xie 2007). Receptor recruitment of these TRAF members leads to their degradation which is a critical step leading to the activation of NIK and induction of p100 processing (Sun 2011, 2012).

Literature references


**Editions**

<table>
<thead>
<tr>
<th>Date</th>
<th>Action</th>
<th>Author</th>
</tr>
</thead>
<tbody>
<tr>
<td>2015-01-26</td>
<td>Author, Edited</td>
<td>Garapati, P V.</td>
</tr>
<tr>
<td>2015-05-12</td>
<td>Reviewed</td>
<td>Ware, CF., Rajput, A., Virgen-Slane, R.</td>
</tr>
</tbody>
</table>
Following recruitment to receptor, TRAF2 mediates K63-linked polyubiquitination of cIAP1 and cIAP2. In addition to being an adaptor that recruits cIAP1/2 and TRAF3 to the receptor TRAF2 is also an E3 that activates cIAP1/2, through their K63-linked ubiquitination. This K63-linked ubiquitination stimulates the K48-ubiquitin ligase function of cIAP1/2 and may impose a change in the substrate specificity of cIAP1/2. Thus, rather than ubiquitinating NIK, cIAP1/2 ubiquitinates TRAF3 leading to its degradation (Vallabhapurapu et al. 2008, Wallach & Kovalenko 2008).

**Preceded by:** cIAP1,2:TRAF2:TRAF3:NIK complex binds TNFR2 trimer

**Followed by:** K63polyUb-cIAP1,2 ubiquitinates TRAF3

**Literature references**


**Editions**

<table>
<thead>
<tr>
<th>Date</th>
<th>Event</th>
<th>Author(s)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2015-01-26</td>
<td>Authored, Edited</td>
<td>Garapati, P V.</td>
</tr>
<tr>
<td>2015-05-12</td>
<td>Reviewed</td>
<td>Ware, CF., Rajput, A., Virgen-Slane, R.</td>
</tr>
</tbody>
</table>
K63polyUb-cIAP1,2 ubiquitinates TRAF3

Location: TNFR2 non-canonical NF-κB pathway

Stable identifier: R-HSA-5668454

Type: omitted

Compartments: plasma membrane, cytosol

cIAP1,2 targets NIK for ubiquitination and degradation under unstimulated conditions but redirects its destructive action towards TRAF3 in response to receptor signals. K63polyUb-cIAP1,2 catalyses K48-linked polyubiquitination of TRAF3 leading to signal-induced TARF3 degradation.

Preceded by: TRAF2 ubiquitinates cIAP1,2 in cIAP1,2:TRAF1:TRAF2:TRAF3:NIK

Followed by: Proteasomal degradation of K48polyUb-TRAF3

Literature references


Editions

<table>
<thead>
<tr>
<th>Date</th>
<th>Action</th>
<th>Author/Editor</th>
</tr>
</thead>
<tbody>
<tr>
<td>2015-01-26</td>
<td>Authored, Edited</td>
<td>Garapati, P V.</td>
</tr>
<tr>
<td>2015-05-12</td>
<td>Reviewed</td>
<td>Ware, CF., Rajput, A., Virgen-Slane, R.</td>
</tr>
</tbody>
</table>
Proteosomal degradation of K48polyUb-TRAF3

**Location:** TNFR2 non-canonical NF-κB pathway

**Stable identifier:** R-HSA-5668481

**Type:** omitted

**Compartments:** plasma membrane, cytosol

K48-linked ubiquitination of TRAF3 further leads to its proteasome-dependent degradation. The resultant degradation of TRAF3 releases NIK from the cIAPs ubiquitin-ligase complex. NIK can escape from ubiquitination and subsequent degradation resulting in accumulation in the cytosol (Sas et al. 2012).

**Preceded by:** K63polyUb-cIAP1,2 ubiquitinates TRAF3

**Literature references**


**Editions**

<table>
<thead>
<tr>
<th>Date</th>
<th>Action</th>
<th>Author(s)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2015-01-26</td>
<td>Authored, Edited</td>
<td>Garapati, P V.</td>
</tr>
<tr>
<td>2015-05-12</td>
<td>Reviewed</td>
<td>Ware, CF., Rajput, A., Virgen-Slane, R.</td>
</tr>
</tbody>
</table>
NIK→noncanonical NF-κB signaling

Location: TNFR2 non-canonical NF-κB pathway

Stable identifier: R-HSA-5676590

Compartments: cytosol, nucleoplasm

In addition to the activation of canonical NF-κB subunits, activation of SYK pathway by Dectin-1 leads to the induction of the non-canonical NF-κB pathway, which mediates the nuclear translocation of RELB-p52 dimers through the successive activation of NF-κB-inducing kinase (NIK) and IκB kinase-alpha (IKKa) (Geijtenbeek & Gringhuis 2009, Gringhuis et al. 2009). Noncanonical activity tends to build more slowly and remain sustained several hours longer than does the activation of canonical NF-κB. The noncanonical NF-κB pathway is characterized by the post-translational processing of NFKB2 (Nuclear factor NF-kappa-B) p100 subunit to the mature p52 subunit. This subsequently leads to nuclear translocation of p52:RELB (Transcription factor RelB) complexes to induce cytokine expression of some genes (C-C motif chemokine 17 (CCL17) and CCL22) and transcriptional repression of others (IL12B) (Gringhuis et al. 2009, Geijtenbeek & Gringhuis 2009, Plato et al. 2013).

Literature references


<table>
<thead>
<tr>
<th>Editions</th>
<th>Date</th>
<th>Action</th>
<th>Author(s)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>2015-01-26</td>
<td>Authored, Edited</td>
<td>Garapati, P V.</td>
</tr>
<tr>
<td></td>
<td>2015-05-12</td>
<td>Reviewed</td>
<td>Ware, CF., Rajput, A., Virgen-Slane, R.</td>
</tr>
</tbody>
</table>
TNFs bind their physiological receptors ➔

**Location:** TNFR2 non-canonical NF-κB pathway

**Stable identifier:** R-HSA-5669034

**Compartments:** plasma membrane

Members of the tumour necrosis factor superfamily (TNFSF) and TNF receptor superfamily (TNFRSF) have crucial roles in both innate and adaptive immunity. These members are implicated in various acquired or genetic human diseases, ranging from septic shock to autoimmune disorders, allograft rejection and cancer (So et al. 2006).

**Literature references**


**Editions**

<table>
<thead>
<tr>
<th>Date</th>
<th>Action</th>
<th>Author(s)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2015-01-30</td>
<td>Authored, Edited</td>
<td>Jassal, B.</td>
</tr>
<tr>
<td>2015-05-12</td>
<td>Reviewed</td>
<td>Ware, CF., Rajput, A., Virgen-Slane, R.</td>
</tr>
</tbody>
</table>
# Table of Contents

Introduction

- TNFR2 non-canonical NF-kB pathway
  - TRAF3:NIK binds TRAF2:cIAP1/2
  - cIAP1,2 ubiquitinates NIK in cIAP1,2:TRAF2::TRAF3:NIK
  - 26Sproteasome degrades K48polyUb-NIK
- TNFA binds TNFR2
  - 2xTNFR2 binds TNFA trimer:TNFR2
  - cIAP1,2:TRAF2:TRAF3:NIK complex binds TNFR2 trimer
- TNF receptor superfamily (TNFSF) members mediating non-canonical NF-kB pathway
  - TRAF2 ubiquitinates cIAP1,2 in cIAP1,2:TRAF1:TRAF2:TRAF3:NIK
  - K63polyUb-cIAP1,2 ubiquitinates TRAF3
  - Protesomal degradation of K48polyUb-TRAF3
- NIK-->noncanonical NF-kB signaling
- TNFs bind their physiological receptors

Table of Contents