Antiviral Signalling Pathways
IPS-1/VISA/MAVS/Cardif – New Mediator of the RIG-I/MDA5 Pathway

Viral infection of mammalian cells lead to an innate immune response by which type I interferons (IFNs) are activated to limit viral replication. Antiviral signalling pathways are initiated upon detection of viral dsRNA, which is generated during the life cycle of most viruses. Recognition of the dsRNA is mediated by either the two RNA helicases retinoic acid inducible gene-1 (RIG-I [1]; Ddx58) and melanoma differentiation associated gene 5 (MDA5 [2]; Helicard [3]; Ifih1), or by toll-like receptor 3 (TLR3) located to an endosomal-like compartment. Recently, four independent studies identified a new mediator of the RIG-I/MDA5 (Helicard) pathway and named it IPS-1 (interferon-β promoter stimulator 1) [4, 5], VISA (virus-induced signalling adaptor) [6], MAVS (mitochondrial antiviral signalling) [7, 8] and Cardif [9] (Figure 1).

IPS-1/VISA/MAVS/Cardif contains a C-terminal transmembrane (TM) domain that targets it to the mitochondrial membrane. By its caspase activation and recruitment domain (CARD) IPS-1/VISA/MAVS/Cardif interacts with the corresponding CARD domain of MDA5 (Helicard) (not shown) or RIG-I. CARD-CARD association leads to the activation of transcription factor NF-κB, as well as to the activation of transcription factor IRF3 in a TBK1 and IKKe dependent way.

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Antiviral Signalling Pathways continued

(Figure 2). The same transcription factors are activated during the TLR3 pathway to force expression of type I IFNs, such as IFN-α and IFN-β. Between the different studies there are discrepancies including the role and/or recruitment of signalling molecules such as TRAF6, RIP-1 and FADD (see also Table 1). Beside these studies, TRAF3 has been identified as a new interacting partner of IPS-1/VISA/MAVS/Cardif [10]. First in vivo studies with IPS-1/VISA/MAVS/Cardif-deficient mice support an essential role for the newly recognized mediator in antiviral signalling [11, 12].

Hepatitis C virus protease NS3-4A colocalizes with IPS-1/VISA/MAVS/Cardif in the mitochondrial membrane and has been shown to cleave the TLR3 adapter TRIF [13] and Cardif [9, 14-16] to eliminate the antiviral signalling pathways (Figure 3).

**FIGURE 3:** Viral Replication

**Product Highlights**

**PAb to MDA5 [Helicard] (mouse) (AL180)**

ALX-210-352-C100  100 µg

From rabbit. IMMUNOGEN: Recombinant mouse MDA5 (Helicard) (aa 2-208). SPECIFICITY: Recognizes mouse MDA5. APPLICATION: WB.

**PAb to MDA5 [Helicard] (human) (AT113)**

ALX-210-935-C100  100 µg

From rabbit. IMMUNOGEN: Recombinant human MDA5 (Helicard). SPECIFICITY: Recognizes human MDA5. APPLICATION: IP, WB.

**PAb to RIG-I (human) (AT111)**

ALX-210-932-C100  100 µg

From rabbit. IMMUNOGEN: Recombinant human RIG-I (aa 201-713) fused at the N-terminus to a tag. SPECIFICITY: Recognizes human RIG-I. APPLICATION: IP, WB.
**Antiviral Signalling Pathways**


**Related Products**

**Interferon Regulatory Factor 3 (IRF3)**

**PAb to IRF3 (100-150) (human)** (BL1820) BET-A30-426A 0.1 mg


**IPS-1/VISA/MAVS/Cardiﬁd**

**PAb to IPS-1/VISA/MAVS/Cardiﬁd (BL3172) BET-A30-782A 0.02 mg


**PAb to IPS-1/VISA/MAVS/Cardiﬁd (BL3173) BET-A30-783A 0.1 mg


**TANK-binding Kinase 1 (TBK1)**

**PAb to TANK-binding Kinase 1 (675-729) (human)** (BL703) BET-A30-093A 0.1 mg

From rabbit. IMMUNOGEN: Synthetic peptide corresponding to aa 675-729 (C-termius) of human TBK1 (TANK-binding kinase 1). SPECIFICITY: Recognizes human TBK1. APPLICATION: IP, WB. BP: BET-BP300-093.

**MAb to TANK-binding Kinase 1 (1084A29)**

**ALX-804-372-C100 100 µg

CLONE: 1084A29. ISO TYPE: Mouse IgG1. IMMUNOGEN: Recombinant human TBK1 (TANK-binding kinase 1). SPECIFICITY: Recognizes human and mouse TBK1. Detects a band of ~80kDa by Western blot. APPLICATION: WB.


**Toll-like Receptor 3 [TLR3]**

**MAb to Toll-like Receptor 3 (human)** (TOL385.6) ALX-804-362-C100 100 µg

ALX-804-362B-C100  Biotin 100 µg

ALX-804-362-F-C100  FITC 100 µg

ALX-804-362-C100  R-PE 100 µg

**CLONE:** 40C1285.6. ISO TYPE: Mouse IgG1. IMMUNOGEN: Synthetic peptides derived from the cytoplasmic portion of human TLR3 (Toll-like receptor 3). SPECIFICITY: Recognizes human TLR3. APPLICATION: FC, IHC (PS), ICC, IP, WB.


**MAb to Toll-like Receptor 3 (human)** (TLR3.7) ALX-804-474-C050 50 µg

**CLONE:** TLR3.7. ISO TYPE: Mouse IgG1. IMMUNOGEN: Recombinant human TLR3 (Toll-like receptor 3). SPECIFICITY: Recognizes human TLR3. APPLICATION: FC, IHC, IP, PB. **TIONS:** Blocks double-stranded RNA-mediated signalling.


**TRAF6**

**PAb to TRAF6 (Bur 30) ALX-210-028-R050 50 µl

From rabbit. IMMUNOGEN: Synthetic peptide corresponding to aa 451-469 (DYKDELQLTQ) of human TRAF6 (tumor necrosis factor receptor-associated factor 6). This sequence is completely conserved in mouse TRAF6. SPECIFICITY: Recognizes human and mouse TRAF6. Detects a band of ~65kDa by Western blot. Does not cross-react with other members of the TRAF family. APPLICATION: IHC (PS), WB.

**TRIF**

**PAb to TRIF (human)** (AL227) ALX-210-908-C050 50 µg

From rabbit. IMMUNOGEN: Synthetic peptide corresponding to aa 4-31 (TGFSPLSFFDDILAAGQDLLLYKHLKTL) of human TRIF. SPECIFICITY: Recognizes human TRIF. APPLICATION: IP, WB.

**TGF-β Activated Kinase 1 [TAK1]**

**PAb to TAK1 (NT) PSC-3385-C100 100 µg

From rabbit. IMMUNOGEN: Synthetic peptide corresponding to aa 44-156 of human TAK1. SPECIFICITY: Recognizes human, mouse and rat TAK1. APPLICATION: WB.

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**Selected Latest Review Articles**

NS3, NS3-NS4A & NS5B

Hepatitis C virus (HCV) was first characterized in 1989 as the major cause of non-A and non-B hepatitis infections [1]. HCV is a single-stranded, positive-sense RNA virus of the Flaviviridae family. Its 9,600 nucleotide genome encodes for a single polypeptide of approx. 3,000 amino acids, which is processed by host cell and viral proteases into three structural proteins (C, E1 and E2) and six non-structural proteins (NS2, NS3, NS4A, NS4B, NS5A, and NS5B) [2].

The NS3 protein is a 631 aa residue bi-functional enzyme with a serine protease localized to the N-terminal 180 residues and an RNA helicase located in the C-terminal 451 residues. The helicase domain of NS3 (NS3h) is essential for viral RNA replication, while the protease is responsible for proteolytic cleavage of the polyprotein. NS3 assembles as a noncovalent complex with cofactor NS4A to form the mature protease. NS3-NS4A blocks the phosphorylation and effector action of interferon regulatory factor-3 (IRF-3), a key cellular antiviral signalling molecule [3]. It seems to cleave the TLR3 adapter TRIF [4] and CARD15 [5, 6], as it has been shown recently.

Another of the non-structural proteins, NS5B, catalyzes the RNA-dependent RNA polymerization of a negative strand intermediate and the subsequent generation of multiple copies of the plus strand viral genome.

NS3, NS3-NS4A and NS5B represent important targets for antiviral drugs [7, 8].

Product Highlight

**Mab to NS5 (HCV) (1B6)**

**APPLICATION:** WB


**Mab to NS5B (HCV) (SB-3B1)**

**APPLICATION:** WB


**Mab to NS5B (HCV) (SB-1287)**

**APPLICATION:** WB


**Product Highlight**

**Nitric Oxide & Viral Signalling**

Hepatitis C virus (HCV) infection has been shown to cause double-stranded DNA breaks and to enhance the mutation frequency of immunoglobulin genes and protooncogenes [1]. Such damages are thought to be mediated by nitric oxide (NO). Accordingly, it has been shown that HCV infections stimulate the production of NO. The stimulation is dependent on the activation of the gene for iNOS (NOSII) by the viral core and NS3 proteins [2].

**Latest Insight**

T. Uemehara, et al. show that the serine palmitoyltransferase inhibitor myriocin suppresses hepatitis C virus (HCV) replication (in a mouse model).

**Myriocin**

**APPLICATION:** WB

**UT:** Produced in E. coli. Recombinant hepatitis C virus (HCV) NS3-NS4A serine protease complex is fused to a His-tag, MW: 68.7kDa (NS3 subunit), 6.2kDa (NS4A subunit).

**Product Highlight**

**NS3-NS4A (HCV) (rec.) (His)**

**APPLICATION:** WB

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**Antiviral Signalling**

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