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Induction and control of the type I interferon pathway by Bluetongue virus

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ABSTRACT

The innate immune response is the first line of defence against viruses, involving the production of type I IFN (IFN-α/β) and other pro-inflammatory cytokines that control the infection. It also shapes the adaptive immune response generated by both T and B cells. Production of type I IFN occurs both *in vivo* and *in vitro* in response to Bluetongue virus (BTV), an arthropod-borne virus. However, the mechanisms responsible for the production of IFN-β in response to BTV remained unknown until recently and are still not completely understood. In this review, we describe the recent advances in the identification of cellular sensors and signalling pathways involved in this process. The RNA helicases retinoic acid-inducible gene-I (RIG-I) and melanoma differentiation-associated gene 5 (MDA5) were shown to be involved in the expression of IFN-β as well as in the control of BTV infection in non-haematopoietic cells. In contrast, induction of IFN-α/β synthesis in sheep primary plasmacytoid dendritic cells (pDCs) required the MyD88 adaptor independently of the Toll-like receptor 7 (TLR7), as well as the kinases dsRNA-activated protein kinase (PKR) and stress-activated protein kinase (SAPK)/Jun N-terminal protein kinase (JNK). As type I IFN is essential for the establishment of an antiviral cellular response, most of viruses have elaborated counteracting mechanisms to hinder its action. This review also addresses the ability of BTV to interfere with IFN-β synthesis and the recent findings describing the non-structural viral protein NS3 as a powerful antagonist of the host cellular response.

1. Introduction

The innate immunity is the first line of defence against viral infections. It allows infected and neighbour cells to establish an antiviral state in the host early after virus exposure and prepares the adaptive immune response. This antiviral response is initiated upon recognition of viral molecular signatures by specialized cellular receptors, which in turn activates signalling cascades that ultimately culminate in the production of antiviral factors.

1.1. Type I IFN: a key cytokine of the innate antiviral immunity

In 1957, Isaacs and Lindenmann observed that the culture medium of cells infected with an inactive influenza virus could protect naive cells against virulent infection (Isaacs and Lindenmann, 1957). This result indicated that the culture medium was containing a protective substance able to interfere with viral dissemination that was subsequently named "interferon" (IFN). Since its original discovery, many types of IFNs have been identified that are now classified into three distinct families: type I, type II and type III IFNs.

Type I IFNs are ubiquitously expressed cytokines that are conserved among vertebrates. They are divided into several classes including IFN-α, IFN-β, IFN-κ, IFN-ε and IFN-ω, IFN-α/β being the major key players. All these cytokines are structurally and genetically related as they derive from common ancestors (Roberts et al., 1998; Pestka et al., 2004; Bonjardim et al., 2009). In humans, there are 13 different IFN-α genes while all the other type I IFNs are encoded by only one gene. IFN-τ and IFN-δ are absent in humans. In ruminants however, several subtypes of IFN-β have been described (Pestka et al., 2004) and IFN-τ plays a crucial role during gestation (Hansen et al., 2010). Type I IFNs do not display antiviral properties *per se* but are mediators that propagate signals within and between cells to induce the expression of antiviral effectors (Pestka et al., 2004). They also exhibit antiproliferative and proapoptotic properties and are prone to shape the immune adaptive response. They contribute to the maturation of dendritic cells (DC), the cytotoxicity of natural killer (NK) cells, the differentiation of T cytotoxic lymphocytes and to the production of high antibody titres upon

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viral infections (Deal et al., 2013). Because of all these properties, IFNs are considered as crucial cytokines to circumvent viral propagation early during infection and to achieve the switch between the host innate and adaptive responses.

Besides type I IFNs, two other IFN families exist, type II (IFN-γ) and type III (IFN-λ) IFNs, that will not be discussed in details in this review. IFN-γ is expressed in cells of the immune system such as lymphocytes and NK cells. It participates to the induction and the regulation of the immune response by activating immune cells and favouring antigen presentation. It also contributes to cell proliferation and activation of apoptosis (Schroder et al., 2004). Its expression is induced by a variety of cytokines including IL-12 and IL-18 that are produced during viral infections. IFN-λ (IL-28A, IL-28B and IL-29) were discovered more recently. They share many common features with IFN-α/β and regulate similar clusters of genes although they use a distinct receptor (Ank et al., 2006; Kotenko, 2011). *In vivo*, the difference between these two types of IFNs relies mainly on the distinct tissue-specific expression of their dedicated receptors. Indeed, IFN-λ expression is restricted to immune and epithelial cells while type I IFNs are ubiquitously expressed (Sommereyns et al., 2008).

The IFN signalling pathway is divided into two phases: (1) an induction phase triggered by the detection of viral molecules by specialized cellular receptors leading to a first wave of IFN synthesis; (2) a response phase activated upon IFN binding to its cognate receptor and characterized by the expression of hundreds of genes involved in the establishment of an antiviral state. In a first part, we will review the different steps involved in the production and function of IFN-α/β and will give some examples of mechanisms evolved by viruses to counteract these pathways. Then, we will give insights into the mechanisms of IFN-α/β induction during BTV infection and will describe the recently uncovered role of NS3 of BTV as a type I IFN antagonist.

1.1.1. Induction of type I IFN synthesis in response to virus infection

The production of IFN in response to virus intrusion is triggered by the recognition of pathogen associated molecular patterns (PAMPs) by the infected cell. There are four main types of viral PAMPs: dsRNA, ssRNA, non-methylated CpG DNA and envelop glycoproteins. The detection of these viral molecules is accomplished by specialized receptors named pattern recognition receptors (PRRs). These receptors or sensors have two essential functions: (1) to discriminate between self and non-self molecules and (2) to promote an antimicrobial response towards multiple pathogens. The two best characterized families of PRRs involved in the detection of viruses are the Toll-like receptors (TLRs) and the RIG-I-like receptors (RLRs).

1.1.1.1. Extracellular receptors of viral PAMPs: the TLRs family. Thirteen members of the TLR family have been identified to date, each one being involved in the detection of distinct patterns (Takeda and Akira, 2005; Akira et al., 2006; Thompson and Locarnini, 2007). TLRs are expressed mainly by antigen presenting cells like DCs or macrophages. However, their expression is not restricted to these cells and most host cells can express at least some TLRs (Akira, 2006). Moreover, expression of TLRs can be induced by many pathogens, cytokines or cellular stresses. Although they recognize diverse molecular structures, TLRs share a common structural organization with a leucine-rich extracellular domain (LRR) which is required for PAMPs recognition, a transmembrane hydrophobic domain and a cytosolic signalling domain, called TIR (Toll/interleukin-1 receptor) domain which mediates homotypic interactions with adaptor proteins containing a TIR domain (Akira et al., 2006; Kaisho and Akira, 2006; Beutler, 2009). TLRs are key actors in the antiviral response, especially in plasmacytoid DCs (pDCs) that are among the main IFN producing cells of the organism. TLR3, TLR7/8 and TLR9 are the most important TLRs for the detection of viral infections and are all specialized in the recognition of nucleic acids (Kawai and Akira, 2007; Eisenacher et al., 2008). Interestingly, these TLRs are localized in endosomal compartments with the exception of TLR3 which is also expressed at the surface of human fibroblasts (Akira et al., 2006; Vercammen et al., 2008). The binding of ligand to its dedicated TLR provokes the dimerization and a conformational change of the receptor, and the subsequent recruitment of specific adaptive molecules: MyD88 for TLR7-9 and TRIF/TICAM1 for TLR3. These adaptors coordinate the activation of multiple signalling pathways that contribute to the synthesis of IFN-α/β and pro-inflammatory cytokines (O'Neill and Bowie, 2007).

TLR3. Until recently, dsRNA was considered as the main viral PAMP. It is contained in the genome of certain viruses such as members of the *Reoviridae* family, appears transiently as intermediates of replication or represents structured RNA generated during most viral infections. Interaction of TLR3 with dsRNA or poly(I:C), a synthetic analogue of dsRNA, allows the recruitment of a unique adaptor, TRIF, that activates the transcription factors IRF-3/7, NF-κB and AP-1 (Alexopoulou et al., 2001; Yamamoto et al., 2003; Vercammen et al., 2008). IRF-3 and IRF-7 essentially control the expression of IFN-α/β genes, while NF-κB and AP-1 regulate various genes encoding inflammatory cytokines. TRIF interacts with TRAF3, a member of the TRAF family, which in turn activates TBK1 and IKKε kinases (Hacker et al., 2006; Oganesyan et al., 2006). The activation of these kinases is also controlled by other proteins such as TANK, SINTBAD or NEMO within a large complex (Ryzhakov and Randow, 2007; Zhao et al., 2007). TBK1 and IKKε phosphorylate IRF-3 and IRF-7 transcription factors which consequently dimerize and translocate into the nucleus to activate the transcription of IFN-α/β genes (Panne et al., 2007). IRF-3 is constitutively expressed and can be engaged early upon viral infection. In contrast, IRF-7 is poorly expressed at basal level but is highly induced upon IFN release, thus playing a key role in the second wave of IFN synthesis (Honda et al., 2005; Hiscott, 2007). TRIF adaptor associates with TRAF6, an E3 ubiquitin ligase, and RIP1 which is involved in the activation of NF-κB mediated by the TNF receptor. These two proteins activate the MAPK (JNK, p38) pathway which governs the activation of members of the AP-1 family (ATF2/c-Jun) and the IKK complex (α/β/γ-NEMO). Activation of this complex leads to the phosphorylation and the subsequent degradation of the IκBα inhibitor, allowing NF-κB (p65/p50) to translocate into the nucleus to induce a pro-inflammatory response. The TLR3/TRIF signalling pathway appears essential in certain types of cells such as conventional DCs (cDCs) where TLR3 is highly expressed. Nevertheless, the role of this sensor appears negligible for the innate antiviral response in most cases although a physiological relevance of TLR3 has been suggested for some viruses like encephalomyocarditis virus (EMCV), herpes simplex virus type-1 (HSV-1) or respiratory syncytial virus (RSV) (Hardarson et al., 2007; Zhang et al., 2007; Vercammen et al., 2008).

TLR7/8. TLR7 and TLR8 (7/8) are highly homologous and both use ssRNA as ligand (Heil et al., 2004; Akira et al., 2006; Eisenacher et al., 2008; Unterholzner and Bowie, 2008). They are highly expressed in pDCs. The induction of IFN-α/β synthesis in response to the activation of these PRRs occurs through the specific activation and homodimerization of IRF-7 which induces the expression of most IFN-α genes. Interestingly, while IRF-7 is inducible in most cell types, it is constitutively expressed in pDCs, suggesting this sensor plays a major role in these cells. Upon ssRNA binding in endosomal compartments, TLR7/8 recruits the adaptive molecule MyD88 through its TIR domain. Then, MyD88 forms a complex with IRAK-4 and IRAK-1 kinases that trigger the activation of the IRF-7 and NF-κB pathways with the involvement of other factors

including TRAF3 and TRAF6 (Kawai et al., 2004; Honda et al., 2005; Hacker et al., 2006; Oganesyan et al., 2006).

TLR9. The TLR9 receptor detects unmethylated dinucleotides CpG DNA molecules from bacterial or viral origin (Hemmi et al., 2000; Eisenacher et al., 2008). Several genomes from DNA viruses like HSV-1/2, Epstein–Barr virus and murine cytomegalovirus (MCMV) have been described as TLR9 stimulators (Lund et al., 2003; Hochrein et al., 2004; Tabeta et al., 2004; Fiola et al., 2010). The TLR9 pathway is very similar to the one triggered by TLR7/8 and appears highly active in pDCs.

TLRs and viral proteins. Not only nucleic acids but also viral proteins are able to activate TLRs (Thompson and Locarnini, 2007; Unterholzner and Bowie, 2008). For example, the F protein of RSV (Kurt-Jones et al., 2000) and the G glycoprotein of vesicular stomatitis virus activate TLR4. In parallel, TLR2 can also induce cellular responses upon binding to viral proteins such as measles haemaglutinin (H), hepatitis C virus (HCV) core and NS3 proteins or gB and gH envelop glycoproteins of human CMV (Boehme et al., 2006).

1.1.1.2. Cytosolic receptors of viral PAMPs. As depicted above, TLRs constitute an efficient alert system in case of pathogen intrusion and allow the infected cells to initiate a rapid and strong antimicrobial response. However, the main limit of action of these cellular sentinels is linked to their distribution: TLRs are localized at the cell surface or within endosomal compartments and are thus unable to sense intracytoplasmic viruses. Moreover, TLRs are expressed mainly in immune cells and therefore seem to play a minor role in most cell types. Finally, the use of models in which the activation or expression of TLRs is impaired or absent has strengthened the hypothesis that an alternative system exists for the intracellular detection of viral replication.

The RIG-I-like receptor pathway. Two studies have identified RIG-I and MDA5 helicases as intracellular PRRs able to induce the production of IFN-α/β and inflammatory cytokines in response to the presence of dsRNA in the cytoplasm (Andrejeva et al., 2004; Yoneyama et al., 2004). These helicases share many similar features and were named RIG-I like receptors (RLRs). They contain a DExD/H type helicase domain, a C-terminal domain (CTD) which is involved in the detection of nucleic acids, a repressor domain (RD) – absent in MDA5 – and two CARD domains at the amino-terminus end that are essential for signalling (Cui et al., 2008; Takahasi et al., 2008). Expression of the CARD region alone is sufficient to trigger type I IFN production. RIG-I and MDA5 are expressed ubiquitously in most tissues and are induced by IFN, allowing an autocrine and paracrine amplification of this detection system. However, while RIG-I is crucial for the induction of IFN-α/β after infection with RNA viruses in fibroblasts and most subsets of conventional DCs (cDCs), plasmacytoid DCs (pDCs) preferentially use the TLR system (Kato et al., 2005; Luber et al., 2010). The activity and stability of the RLRs is well controlled by regulating proteins including TRIM25 and RNF125 (Arimoto et al., 2007; Gack et al., 2007). Although RIG-I and MDA5 share high sequence identity, they are activated by different RNA subsets (Saito and Gale, 2008). RIG-I displays higher affinity for ssRNA than dsRNA (Saito et al., 2008; Takahasi et al., 2008). Among ssRNA, those containing a 5′-triphosphate bind RIG-I with a very high affinity and strongly induce IFN-α/β synthesis (Hornung et al., 2006; Pichlmair et al., 2006; Plumet et al., 2007; Saito et al., 2007; Cui et al., 2008; Takahasi et al., 2008). These types of 5′ triphosphate RNAs are generally absent from non-infected cells making the 5′-triphosphate motif a major viral PAMP. RIG-I is also activated by transcripts generated by the T7 RNA polymerase as well as influenza genomic RNA (Rehwinkel et al., 2010). More recent data have indicated that RIG-I activation can be influenced by many characteristics including secondary structure, the presence of "panhandle" structure, nucleosidic modifications and sequences (Marques et al., 2006; Saito et al., 2008; Takahasi et al., 2008; Schmidt et al., 2009; Takahasi et al., 2009; Lu et al., 2010). RIG-I has been described as a RNA sensor for many viruses including Flaviviviruses, Orthomyxoviruses, Paramyxoviruses and Rhabdoviruses (Hornung et al., 2006; Kato et al., 2006; Plumet et al., 2007; Cui et al., 2008). Recent additional structural data have also given insights into the mode of binding and activation of RIG-I (Cui et al., 2008; Takahasi et al., 2008) (reviewed in (Kolakofsky et al., 2012)). The mode of recognition of non-self RNA by MDA5 is less clear. *In vivo*, MDA5 was described as a sensor for poly(I:C) and members of the *Picornaviridae* family (Gitlin et al., 2006; Kato et al., 2006). MDA5 do not bind 5′-triphosphate RNAs but has a high affinity for dsRNA. Interestingly, while RIG-I preferentially recognizes ssRNA, it can also associates and is activated by short dsRNA (<2 kb). In contrast, long dsRNA molecules (>2 kb) are necessary for MDA5 activation (Kato et al., 2008). Members of the *Reoviridae* family that possess dsRNA genomes segments ranging from 1.2 to 3.9 kb can then activate both helicases (Kato et al., 2008). Members of the *Flaviridae* family are also able to activate RIG-I and MDA5 (Fredericksen and Gale, 2006). A third RLR member, LGP2, has also been described. LGP2 is highly homologous to RIG-I and MDA5 but lacks the CARD signalling domain (Yoneyama et al., 2005). LGP2 was originally considered as a negative regulator of the RLR pathway but its role in the antiviral response seems more complex and virus-dependent (Rothenfusser et al., 2005; Yoneyama et al., 2005; Komuro and Horvath, 2006; Saito et al., 2007; Venkataraman et al., 2007; Vitour and Meurs, 2007; Satoh et al., 2010).

In the absence of stimulating RNA molecules, RIG-I is maintained in a closed inactive conformation by the RD domain (Saito et al., 2007; Vitour and Meurs, 2007). Binding of viral RNA onto the CTD domain of the protein induces a conformational change that allows CARDs exposure and activation of the helicase domain. RIG-I can then multimerize and interact with the mitochondrial adaptor protein MAVS (*Mitochondrial antiviral signalling*, also named IPS-1, VISA, CARDIF) (Kawai et al., 2005; Meylan et al., 2005; Seth et al., 2005; Xu et al., 2005). This interaction occurs through a homotypic interaction between the CARD region of RIG-I and a CARD domain present at the amino-terminus of MAVS. Upon dsRNA binding, MDA5 forms dimers and filaments, a step required for subsequent association with MAVS (Berke and Modis, 2012). Like TRIF, MAVS interacts with TRAF3 (Saha et al., 2006) to activate TBK1 and IKKε and subsequently IRF-3/7 transcription factors. In parallel, activation of the NF-κB pro-inflammatory pathway is ensured by other adaptors including FADD/RIP1 and/or members of the TRAF family (Kawai et al., 2005; Seth et al., 2005; Xu et al., 2005). Interaction of MAVS with STING, a protein anchored at the endoplasmic reticulum, has been described and may also play a role in IFN-α/β induction (Kawai et al., 2005; Ishikawa and Barber, 2008; Arnoult et al., 2009). Some other sensors may also play a role in RNA sensing but are less characterized. Among those, a complex made of the DDX1, DDX21 and DHX36 helicases recognizes dsRNA in mDCs and uses TRIF to induce the signalling cascade leading to IFN-α/β production (Zhang et al., 2011). Many cytoplasmic DNA receptors have also been described but will not be discussed here (Unterholzner, 2013).

1.1.2. Response to type I IFN

The best-characterized pathway triggered by the secretion of type I IFN is the Janus tyrosine kinase (JAK)/signal transducer and activator of transcription protein (STAT) signalling pathway that was first described in the 1990s and is functional in most cell types (Fu et al., 1992; Schindler et al., 1992; Silvennoinen et al., 1993; Platanias, 2005). Activation of this pathway leads to the expression of hundreds of proteins encoded by IFN-stimulated genes (ISGs) that act on cellular processes such as the innate and adaptive immune system, cellular proliferation and survival and protein synthesis to establish an antiviral state within the cell. Once

IFN-α/β is secreted into the extracellular compartment, it binds to its cell surface receptor called IFNAR. IFNAR is composed of two transmembrane sub-units, IFNAR1 and IFNAR2, that are expressed in most tissues (de Weerd et al., 2007). IFNAR1 is constitutively associated with tyrosine kinase 2 (TYK2) (Colamonici et al., 1994a,b; Gauzzi et al., 1996), a kinase belonging to the JAK family and IFNAR2 is pre-associated with JAK1 and STAT2 (Uddin et al., 1995). Interaction of type I IFN with one subunit of IFNAR leads to the recruitment of the other subunit (Lamken et al., 2004) and their subsequent dimerization. This initial step triggers the phosphorylation of TYK2 on tyrosine 1054 and/or 1055 *via* a mechanism dependent on JAK1 (Silvennoinen et al., 1993; Gauzzi et al., 1996; Krishnan et al., 1996). Tyk2 then phosphorylates JAK1 to activate it further (putative tyrosine 1022/1023). The activated kinases subsequently phosphorylate tyrosine residues located in the cytoplasmic domain of IFNAR1 (tyrosine 466) and IFNAR2 (tyrosine 510) (Platanias and Colamonici, 1992; Abramovich et al., 1994; Platanias et al., 1994; Krishnan et al., 1996; Zhao et al., 2008). STAT2 is then able to bind to the phosphorylated residue of IFNAR1 and is phosphorylated by the JAK kinases on tyrosine 690. STAT1 subsequently binds through its SH2 domain to the newly phosporylated tyrosine of STAT2 and becomes phosphorylated on tyrosine 701 (Krishnan et al., 1996; Yan et al., 1996). The activated STAT1/STAT2 dimer is then released from IFNAR2 and associates in the cytoplasm with IRF9 to form the transcription factor ISG factor 3 (ISGF3). This heterotrimeric complex migrates to the nucleus where it binds to specific promoter elements called IFN-stimulated response elements (ISRE) and initiates the transcription of ISGs.

Other STATs such as STAT3, STAT4, STAT5 and STAT6 can be activated by type I IFN but this activation is mainly specific to certain cell types such as endothelial cells, T lymphocytes or lymphoblasts (Beadling et al., 1994; Meinke et al., 1996; Fasler-Kan et al., 1998; Matikainen et al., 1999; Torpey et al., 2004). Different combinations of STAT homo- or hetero-dimers other than STAT1–STAT2 can then form. They bind to IFN-γ-activated site (GAS) elements present in the promoter of certain ISG and induce the transcription of distinct genes. These different STAT complexes have different functions such as the regulation of the type I IFN response, the production of IFN-γ or the promotion of cell growth.

Many mechanisms are present within a cell to regulate the JAK/STAT pathway. Some post-translational biochemical modifications of STAT and JAK proteins by phosphorylation, ubiquitination, acetylation, methylation, ISGylation and sumoylation are important for full transcriptional activation or regulation of the JAK/STAT pathway (Shuai and Liu, 2003). Several key regulatory proteins are also expressed to regulate STAT and JAK proteins. They include protein tyrosine phosphatases (PTP), suppressor of cytokine signalling (SOCS) and protein inhibitor of activated STAT1 (PIAS) proteins.

Type I IFN can trigger other signalling cascades such as the mitogen activated protein kinase (MAPK), the phosphoinositide 3-kinase (PI3K), the v-crk sarcoma virus CT10 oncogene homolog (avian)-like (CRKL) and the NF-κB pathways. Additionally, activation of JAK results in the activation of several MAPK such as p38 and extracellular signal regulated kinases (ERK) that display antiviral activity and control cell differentiation and growth.

1.1.3. Function of ISGs

Type I IFN induces the expression of hundreds of genes involved in multiple cellular processes. These genes encode proteins that possess an intrinsic antiviral activity, act directly on different signalling cascades involved in the type I IFN system to enhance its action and/or play a role in other cellular responses such as apoptosis. The best-characterized antiviral proteins induced by type I IFN are dsRNA-dependent protein kinase R (PKR), myxovirus resistance (Mx) proteins and 2′,5′-oligoadenylate synthetase (OAS) proteins. Other proteins thought to play an important role in this antiviral response include ISG15, ISG20, members of the tripartite motif (TRIM) family (PML) and proteins encoded by the *ISG56* (*IFIT1*) family of genes (Liu et al., 2011).

PKR is a serine/threonine kinase consisting of an amino-terminal regulatory domain containing two dsRNA binding site and a carboxy-terminal catalytic domain (Meurs et al., 1990). In response to viral or synthetic dsRNA or other stimuli, PKR undergoes a conformational change leading to the activation of its kinase domain. Activated PKR phosphorylates eukaryotic initiation factor 2α-subunit (eIF2α) and subsequently blocks the translation of cellular and viral mRNA (Williams, 1999). As PKR is able to detect dsRNA, the kinase was first considered as a sensor for viral infections capable to induce the production of type I IFN (Yang et al., 1995; Diebold et al., 2003; Gilfoy and Mason, 2007; McAllister and Samuel, 2009). Nevertheless, the importance of PKR as a sensor for the induction of type I IFN remains unclear and seems to be less crucial than RIG-I and MDA5 (Smith et al., 2001). Another antiviral mechanism triggered by the type I IFN system is the recognition of dsRNA by OAS. The enzyme directly binds to viral dsRNA and is activated to synthesize 2′,5′-linked oligoadenylates from ATP, leading to the dimerization and activation of ribonuclease L (RNase L) (Dong and Silverman, 1995; Malathi et al., 2007). RNase L cleaves cellular and viral ssRNA downstream UU and UA sequences and generates small, often duplex, RNA (Malathi et al., 2007). This action limits viral replication and packaging as well as host and viral protein synthesis. Mx proteins (MxA and MxB in humans, Mx1 and Mx2 in mice) are guanosine triphosphatases (GTPase) belonging to the dynamin family able to inhibit early stage of the replication cycle of a wide range of viruses. They are conserved among the majority of vertebrate species but all of them do not seem to have an antiviral activity. Their mode of action is still not completely understood but seems to involve the confinement of essential viral components necessary for viral replication to block their functions (Haller et al., 2007a,b).

The proteins described above are only examples as the expression of more than 380 ISGs are induced by type I IFN (Schoggins et al., 2011). Many of them play a role in the initiation of rapid, robust and efficient cellular responses interfering with the establishment of viral infections.

1.1.4. Immunomodulation properties of type I IFN

The role of type I IFN is not restricted to the establishment of an antiviral state within cells as the cytokine is also involved in the modulation of many responses of the innate and adaptive immune system. Type I IFN can act directly on immune cells *via* the activation of IFNAR or indirectly *via* the induction of other cytokines and chemokines and the stimulation of specific cells (Bonjardim, 2005; Stetson and Medzhitov, 2006; Hervas-Stubbs et al., 2011; Huber and Farrar, 2011). Type I IFN is able to regulate natural killer (NK) cells by enhancing their ability to kill target cells and by stimulating their proliferation *via* the induction of IL-15 (Nguyen et al., 2002). Type I IFN also modulates the activity of macrophages and antigen-presenting cells such as pDCs and enhances the expression on these cells of surface receptors and co-stimulatory molecules that interact with immune cells of the adaptive response. For example, type I IFN enhances the expression of major histocompatibility complex (MHC) class I and proteins involved in antigen processing and presentation to increase antigen recognition and CD8 T-cell responses. Type I IFN is also directly involved in the survival, proliferation and differentiation of T cells into memory and effector T cells (Tough, 2012). Additionally, type I IFN modulates antiviral responses mediated by B lymphocytes *via* direct and indirect mechanisms by affecting their ability to produce cytotoxic and neutralizing antibodies and promoting their survival and activation (Kiefer et al., 2012).

1.2. Modulation of the type I IFN pathway by viruses

As described above, viral infections can trigger rapidly an antiviral state in host cells to circumvent virus propagation. In order to limit this cellular response, viruses have evolved a great variety of escape mechanisms to counteract the IFN pathway at the synthesis as well as the response level, thus demonstrating the physiological importance of this pathway. Certain viruses and viral proteins have the ability to interfere at multiple levels of the IFN signalling to increase the efficiency of this evasion. Some viruses also act more globally by inducing a large transcriptional shut-off that blocks the expression of ISGs and genes involved in IFN signalling (Haller et al., 2006). The list of mechanisms described below is not exhaustive and many other strategies have been evolved by viruses to counteract the IFN system, illustrating the great capacity of viruses to fight the innate cellular response of the host. This topic has been thoroughly discussed in recent reviews (Haller et al., 2006; Bowie and Unterholzner, 2008; Lee et al., 2009; Taylor and Mossman, 2012) and only a few examples of counteracting measures are given below.

1.2.1. Inhibition of type I IFN synthesis

The initial step in the production of type I IFN requires the detection of viral PAMPs by PRRs. These cellular sensors are thus the first line of defence of the antiviral response and their activation constitutes relevant targets for viruses. One frequent strategy adopted by many RNA viruses resides in the protection of their nucleic acids inside protein structures or cellular compartments that render them inaccessible to cellular sensors (Randall and Goodbourn, 2008). Certain viruses can avoid detection of the 5′ moiety of their genomic or mRNA molecules by specialized PRRs *via* capping events or 5′ triphosphate cleavage. Vaccinia virus is a very powerful antagonist of TLRs through at least two proteins, A46 and A52. A46 contains a TIR domain which associates with the TIR domain of the adaptor proteins MyD88, Mal, TRIF and TRAM and prevents their recruitment to their dedicated TLR (Stack et al., 2005). A52 targets TRAF6 and IRAK-2 and limits the activation of the NF-κB pathway in response to several TLRs including TLR4 (Harte et al., 2003). Hepatitis B virus negatively regulates the expression of TLR2 (Visvanathan et al., 2007). In the cytoplasm of infected cells, the activity of RLRs is a common target for viral inhibitors. Several viral proteins like NS1 of influenza, VP35 of Ebola, σ3 of reovirus, σA of avian reovirus or E3L of vaccinia virus share the same ability to bind and sequester viral RNAs that are responsible for the activation of RIG-I and MDA5 (Imani and Jacobs, 1988; Jacobs and Langland, 1998; Martinez-Costas et al., 2000; Unterholzner and Bowie, 2008). Through the same mechanism, most of these proteins also inhibit the activation of PKR and the OAS system. The RLRs and the adaptor protein MAVS are also directly targeted by viral proteins. For example, NS1 of influenza associates with RIG-I and inhibits its transactivation function (Pichlmair et al., 2006; Mibayashi et al., 2007). The interaction of the V protein of paramyxoviruses with the helicase domain of MDA5 blocks the activation of NF-κB and IRF-3 signalling pathways (Andrejeva et al., 2004; Childs et al., 2007). The 3ABC precursor of 3C(pro) protease of hepatitis A virus and the NS3/4A proteases of HCV and GBV-B cleave MAVS at its carboxy-terminus to interfere with its mitochondrial localization which is crucial for its antiviral function (Meylan et al., 2005; Chen et al., 2007; Yang et al., 2007). IRF-3 and IRF-7 transcription factors, which are essential to ensure IFN-α/β gene expression, also constitute interesting targets for viruses. The phosphoproteins of several negative-strand RNA viruses prevent IRF-3 activation by TBK1. For example, the P protein of Borna virus is phosphorylated by TBK1 and serves as viral decoy for IRF-3 phosphorylation (Unterstab et al., 2005). Another intriguing strategy used by human herpes virus 8 resides in the synthesis of viral analogues of IRFs (vIRFs) which act as dominant negative on the action of cellular IRFs through a molecular mimetic mechanism (Lin et al., 2001; Lubyova et al., 2004). Alternatively, some viral proteins favour the degradation of IRFs (Haller et al., 2007a,b). For example, NSP1 of rotavirus interacts with IRF-3, IRF-5 and IRF-7 and provokes their degradation through the proteasome (Barro and Patton, 2005, 2007; Liu et al., 2009). More recently, it was shown that truncated forms of NSP1 from the OSU and SA11 strains are unable to trigger IRF degradation but can still interfere with type I IFN synthesis by interacting with RIG-I (Qin et al., 2011). The protein ICP0 of HSV interacts and sequesters IRF-3 inside nuclear bodies to prevent binding to its target DNA (Melroe et al., 2007).

1.2.2. Inhibition of type I IFN response

Many viral antagonists interfere with the JAK/STAT pathway to block the synthesis of antiviral proteins such as PKR, Mx and OAS and the induction of antiviral responses. Interestingly, by blocking this signalling pathway, viruses also inhibit the expression of ISGs such as RIG-I, MDA5 or IRF-7 and subsequently interfere with the positive feedback loop triggered by the IFN response to enhance its effect. Some viral proteins act upstream of the JAK/STAT pathway by interfering with the binding of type I IFN to its receptor. For example, the vaccinia virus protein B18R, a soluble homologue of IFNAR, binds type I IFN to block its antiviral effect (Colamonici et al., 1995; Symons et al., 1995). JAKs and STATs are more frequently targeted and many viruses can sequester them, induce their degradation or interfere with their phosphorylation or their transport. Human Metapneumovirus has been shown to decrease the cellular levels of JAK1 and TYK2 and the surface expression of IFNAR (Ren et al., 2011). Rotaviruses are also able to interfere with STATs. A study has shown that monkey rotavirus RRV and human rotavirus Wa were able to block the nuclear translocation of STAT1 and STAT2 in response to type I IFN but not their phosphorylation (Holloway et al., 2009). Some viruses act downstream of the JAK/STAT pathway by interfering with IRF9 and the formation of the ISG3 complex. For example, reovirus is able to inhibit type I IFN response in a strain-dependent manner. μ2 from a strain causing mild myocarditis is able to induce a nuclear accumulation of IRF9 but not μ2 from a non-myocarditic reovirus strain (Zurney et al., 2009). This effect suggests that the viral protein modulates the interaction between IRF9 and STAT and the function of ISG3. Some viruses are also able to target the expression of specific ISGs. For example, HCV infection up-regulates the expression of microRNA miR-130A that targets the expression of the antiviral protein interferon-induced transmembrane 1 (IFITM1) (Bhanja Chowdhury et al., 2012). Finally, some viruses are able to induce the expression of regulatory proteins such as PTP, SOCS and PIAS that negatively modulate the JAK/STAT pathway. For example, influenza A virus inhibits the JAK/STAT pathway *via* the induction of SOCS-3 (Pauli et al., 2008; Pothlichet et al., 2008).

1.2.3. Inhibition of IFN-induced antiviral factors

Viruses are also able to target the antiviral factors synthesized in response to type I IFN to promote their replication and survival. By interfering with ISG proteins such as PKR and OAS/RNase L, viruses are able to block their function as sensors of viral infection, to inhibit the production of IFN and consequently to down-regulate their expression. Viral antagonists can target the activity of PKR by acting as pseudo-substrates, by interacting directly with the cellular protein or by inducing its degradation. For example, E2 from HCV contains a homologue sequence to PKR and eIF2α phosphorylation sites and is able to bind PKR and inhibit its kinase activity (Taylor et al., 1999). Reovirus σ3 and σA inhibit the dsRNA-dependent kinase activity of PKR (Imani and Jacobs, 1988; Yue and Shatkin, 1997; Martinez-Costas et al., 2000; Gonzalez-Lopez et al., 2003). Influenza virus uses another mechanism and activates $p58^{IPK}$, a cellular inhibitor of PKR (Lee et al., 1990, 1992; Melville

et al., 1999; Goodman et al., 2007). Several viruses also interfere with the OAS/RNase L system using different mechanisms. Some viruses such as human immunodeficiency virus and EMCV induce the expression of the cellular RNase L inhibitor (RLI) (Martinand et al., 1998, 1999). Rotavirus VP3 contains a phosphodiesterase that cleaves 2-5A to prevent the activation of RNAse L (Zhang et al., 2013). Other ISGs can also be targeted by viruses. For example, it has been shown that the nsp3 papain-like protease (PLP) 2 domain of a human coronavirus is able to deconjugate ISG15 from cellular substrates (Clementz et al., 2010). It was also suggested that membranous structures induced by the Kunjin strain of West Nile virus act as protection for the virus to hide partially from MxA (Hoenen et al., 2007) and that the adenovirus E4 ORF3 protein induces a rearrangement of PML nuclear bodies and interferes with the induction of a PML-dependent antiviral response (Ullman et al., 2007; Ullman and Hearing, 2008).

2. Induction of type I IFN synthesis by BTV

Most viruses are able to induce IFN synthesis although this event can vary greatly depending on the virus and host features. It is known that for RNA viruses, dsRNA are synthesized as intermediates of replication that represent a major PAMP. For dsRNA viruses, such as members of the *Reoviridae* family, the genome itself is composed of dsRNA molecules that have the ability to activate cellular sensors. In 1967, Tytell et al. (1967) first demonstrated that the viral genome of a dsRNA virus, *i.e.* reovirus, was sufficient to induce IFN production. Bluetongue virus (BTV), a dsRNA virus from the *Reoviridae* family, has been described for many years as a strong inducer of IFN. Here we review the current knowledge regarding the ability of BTV to induce type I IFN and the mechanisms and cellular pathways involved.

2.1. BTV induces type I IFN production both in vitro and in vivo

In 1969, Hendrik Huismans from the Onderstepoort Veterinary Research Institute in South Africa infected primary embryonic murine cells with an attenuated American vaccine strain of BTV serotype 10 (BTV-10A) (Huismans, 1969). At different time post-infection (p.i.), cell supernatant was collected to measure the IFN titre in the medium of these infected cells. The presence of IFN was detectable as soon as 5 h p.i. and increased until 24 h p.i. This constituted the first evidence of *in vitro* IFN production in response to BTV infection. In the following years, BTV was reported to induce IFN in many cell types from various tissues and species including foetal and adult leukocytes and spleen cells from sheep (Rinaldo et al., 1975), endothelial cells from bovine and ovine origin (Coen et al., 1991; Russell et al., 1996; Chauveau et al., 2012) and kidney cells from rabbits, hamsters, monkeys, cats and pigs (Jameson and Grossberg, 1981; Fulton and Pearson, 1982; Taylor and O'Brien, 1985). Importantly, BTV was also shown to induce the production of IFN in multiple human cells with a high efficiency particularly in tumour-derived cell lines (Jameson and Grossberg, 1978, 1979, 1981; Joklik, 1981; Chauveau et al., 2012).

The initial demonstration of IFN production upon BTV infection was obtained with an attenuated strain (BTV-10A) (Huismans, 1969). The potent IFN induction ability of this vaccine strain was latter confirmed by other groups (Jameson et al., 1978; Joklik, 1981) but Lyons et al. (1982) later found that this strain induced low IFN level in comparison with field strains of BTV-2, 4 and 6. Huismans used other attenuated (A) or virulent (V) serotypes (BTV-8A, BTV-1A and BTV-1V) and found that BTV-8A was the best IFN inducer while BTV-1A/V poorly induced IFN synthesis. However, this lack of production could be due to an impaired infectivity of BTV-1A/V in mouse embryonic cells (Huismans, 1969). In another study, Fulton and Pearson observed that strains from BTV-10, 11, 13 and 17 could also efficiently induce IFN synthesis. Recently, we found that European field strains of BTV-4 and BTV-8 are strong IFN-β inducers in bovine and human cells (Chauveau et al., 2012). Overall, it seems that both attenuated and virulent strains induce IFN and that many if not all serotypes can induce IFN, suggesting that a conserved mechanism of induction exists among the different BTV serotypes and strains.

Using different amounts of virus, H. Huismans initially defined a multiplicity of infection (MOI) of 1 PFU/cell as the minimum input required to obtain detectable levels of IFN and he observed a linear increase between IFN level and virus titre until an MOI of 10 (Huismans, 1969). However, this linear correlation between IFN yields and viral titre is not systematically observed and might depend on BTV strains, cell types and/or experimental conditions (Lyons et al., 1982). Upon infection of A549 cells with different amounts of BTV-8 and BTV-4, we observed a strong correlation between the level of IFN-β mRNA and the quantity of virus used (Chauveau et al., 2012).

In vivo IFN synthesis during BTV infection was initially demonstrated after intravenous injection of 4×10^8 PFU of partially purified BTV-10V in mice (Huismans, 1969). IFN synthesis was assessed in blood samples collected at different time p.i. and was detected as early as 4 h p.i. Maximum titre was reached between 8 and 12 h p.i. and then decreased to undetectable level at 24 h p.i. The same result was found with highly purified BTV-10A (Huismans, 1969; Eksteen and Huismans, 1972). Jameson et al. (1978) performed a similar experiment few years later and found that injection of $10^{6.8}$ PFU of BTV in mice induced a rapid production of IFN that reached a peak at 8 h post-injection. Although BTV can infect newborn mice, an adult mouse model was required to study the acquired immune response to BTV and prophylactic approaches. Blocking IFN-α/β activity in mice increases dramatically the sensitivity to many viruses. Indeed, genetically targeted (knockout) mice lacking the β subunit of the IFN-α/β receptor ($IFNAR^{(-/-)}$ mice) are unable to establish an antiviral state and, as a consequence, are highly susceptible to many viral infections (Muller et al., 1994; Fiette et al., 1995). Recently, Calvo-Pinilla et al. showed that adult $IFNAR^{(-/-)}$ mice support the growth of BTV *in vivo*, reproducing many aspects of its natural host infection, and were prone to respond to vaccine strategies (Calvo-Pinilla et al., 2009a,b, 2010, 2012). Thus, $IFNAR^{(-/-)}$ mice represent a good small animal surrogate model to study BTV pathogenicity and to evaluate vaccine strategies against this virus.

Domestic and wild ruminants are natural targets of BTV infection. To address whether BTV induces IFN in its hosts, several experimental infections were conducted in sheep and cattle. It has been shown that bovine foetuses experimentally infected as early as 125 days of gestation with BTV-10 can produce IFN both in serum and tissues and that this production might limit virus spread within infected tissues (MacLachlan et al., 1984). In calves infected experimentally with a BTV-10 strain, production of circulating IFN persisted between 2 and 4 days post-infection with a peak between 1 and 3 days p.i. (MacLachlan and Thompson, 1985). Titres of BTV measured in the blood were higher when IFN was no longer detectable in serum collected at the same time. This early transient synthesis of IFN suggests a preponderant role in the initial antiviral response rather than an involvement in the subsequent elimination of the infection. In sheep, IFN was detected in the serum of animals infected with BTV-10, -11, -13 and -17 (American strains) 5 days p.i. and peaked at day 6 p.i., which correlated with a first peak of viraemia (Foster et al., 1991). In comparison to the IFN antiviral response detected in cattle, the IFN index in sheep was higher and lasted longer. The IFN peak correlated with a 90% reduction in the viral titre. The decrease in IFN level was concomitant with a second peak of viraemia at day 10 p.i. Diphasic viraemia could be attributed

to the induction of high levels of IFN concomitantly to a first peak of viral titre, followed by the production of specific antibodies and a subsequent reduction of the viraemia during the second peak.

At the cellular level, it has been shown that BTV targets cDCs and pDCs early after infection in sheep (Hemati et al., 2009; Ruscanu et al., 2012). Interestingly, inoculation of an attenuated strain of BTV-2 (a-BTV-2) and a wild-type strain of BTV-8 (BTV-8) in the skin of sheep induced IFN-α/β production in afferent lymph, peaking at day 6 p.i. with a-BTV-2 (16 IU/ml) and at day 5 p.i. with BTV-8 (12 IU/ml), which corresponds to the peak of cDC-dependent viral dissemination from the skin (Hemati et al., 2009). Circulating IFN-α/β was also found in the blood of sheep inoculated with BTV-8 at days 2 and 6 p.i. These results suggest that lymph and blood pDCs and/or cDCs can be activated by BTV to produce IFN-α/β as both pDCs and cDCs are found in blood and lymph in sheep (Pascale et al., 2008). Enrichment of lymph and blood cells in cDCs and pDCs by low-density (LD) gradient centrifugation lead to IFN-α/β synthesis upon BTV stimulation independently of BTV strain and attenuation. In order to identify the cell type that was responsible for the IFN-α/β produced by the LD lymph and blood cells after BTV stimulation, cDCs ($CD11c^+$ cells) and pDCs (B^- $CD11c^-$ $CD45RB^+$ cells) were isolated using immunomagnetic beads (Pascale et al., 2008) and were subsequently subjected to BTV-2 stimulation. No IFN-α/β was detected in the lymph $CD11c^+$ cell fraction in response to BTV-2, while high levels were measured in the $CD11c^-$ cell fraction. Sheep lymph pDCs were then isolated and were found to produce IFN-α/β upon exposure to a-BTV-2. In blood, only $CD45RB^+$ cells were also able to synthesize IFN-α/β following BTV exposure. Altogether, these findings show that pDCs are the main producers of IFN-α/β upon BTV infection among immune cells. However, in other viral infections, pDCs are responsible for a burst of IFN-α/β within the first 24 h, a time frame that remains to be investigated in BTV infection *in vivo* (Zucchini et al., 2008).

Besides IFN production, BTV induces an inflammatory response which results in the synthesis of many inflammatory cytokines and co-stimulatory molecules which have an impact on the immune response triggered by the virus. In addition, several studies have indicated that a strong correlation exists between BTV-induced inflammatory cytokines and the pathogenesis of the disease (DeMaula et al., 2001, 2002). This is further discussed in the review from James MacLachlan published in this issue. In A549 cells, we found that the expression of IL-8 mRNA, a pro-inflammatory cytokine whose induction is predominantly dependent on NF-κB, is induced upon BTV infection and is concomitant with the expression of IFN-β mRNA (Chauveau et al., 2012). Ruscanu et al. (2012) have found that BTV can induce the expression of multiple cytokine genes like TNF-α, IL-6 and IL-12, as well as surface CD80/86, in purified primary sheep blood pDCs *in vitro*. Furthermore, upon *in vivo* infection, sheep blood pDCs displayed a strong inflammatory gene expression profile, with up-regulation of CCL2, CCL4, IL-8, IFN-γ and TNF transcripts (Ruscanu et al., 2013).

2.2. Mechanisms involved in the production of type I IFN production by BTV

As described above, IFN induction by BTV has been greatly documented *in vivo* and *in vitro* but the underlying mechanisms of this induction were unknown for a long time. Recent data have shed light on the cellular events required for activation of this antiviral response during BTV infection.

2.2.1. BTV PAMPs

One important question regarding IFN induction is the type of PAMPs that triggers this induction. As early as 1967, it was shown that the dsRNA genome of *Reoviridae* is a potent inducer of IFN synthesis (Tytell et al., 1967). Soon after, it was shown that injection of 8 μg of dsRNA isolated from BTV-10A in mice induced IFN synthesis after a 2 h latency phase and reached a maximum 6 h post-injection. This peak preceded of about 4 h the one obtained after infection with the corresponding virus (Huismans, 1969; Eksteen and Huismans, 1972). This delay might correspond to the time needed for decapsidation, release of viral genome or synthesis of new dsRNA. In preliminary experiments, we found that transfection of cells with purified BTV dsRNA activated IFN-β promoter (not shown). These data suggest that viral dsRNA of BTV genome is sufficient to trigger IFN synthesis both *in vivo* and *in vitro*.

The need for replication to induce IFN synthesis upon BTV infection has been extensively addressed *in vitro* and *in vivo* but remains unclear. Jameson et al. (1978) first suggested that, in mice, IFN synthesis occurs independently of the production of viral progeny as a UV-inactivated BTV induced similar levels of IFN as an infectious virus. *In vitro*, Taylor and O'Brien (1985) observed that IFN production was effective in rabbit cells infected with a UV-inactivated vaccine strain from serotype 10. This was confirmed in the human cell line HT-1376 which produced high amount of IFN after BTV infection. In these cells, infectious and UV-inactivated BTV (MOI of 10 PFU/cell) induced comparable level of IFN (Jameson and Grossberg, 1979). In contrast, they also observed that a UV-inactivated virus used at an MOI of 50 induced low level of IFN in the A375 human cell line. H. Huismans previously suggested that IFN production is not linked to replication efficiency as BTV-8A, which replicates at lower level than BTV-10A, induced more IFN (Huismans, 1969). Similarly, we found that while BTV-8 seemed to replicate more efficiently than BTV-4 in A549 cells at 48–72 h p.i., the BTV-4 strain induced more strongly the synthesis of IFN than BTV-8 at 36–48 h p.i. (Chauveau et al., 2012). In A549 cells infected with BTV-8, we also observed that the level of IFN-β mRNA followed the kinetic of BTV RNA synthesis, suggesting that viral replication is required to trigger IFN-β synthesis. Unlike live BTV-8, we found that the levels of IFN-β mRNA and protein quantified in cells infected with the UV-inactivated virus were close to background level, indicating that virus replication is essential for IFN-β induction in A549 cells (Chauveau et al., 2012). Similar results were obtained in the bovine cell line MDBK.

In pDCs, IFN-α/β production usually occurs independently of viral replication but exceptions do exist (Hornung et al., 2004; Guzylack-Piriou et al., 2006). In a recent work, Ruscanu et al. (2012) found that active BTV infection occurs in sheep pDCs (see above). When UV-inactivated BTV-8 was used, IFN-α/β was still produced in the supernatant of LD lymph cells and highly purified blood pDCs but at a lower level than with live virus (Ruscanu et al., 2012). The same results were also found with purified BTV-1 and UV-BTV-1 (Ruscanu et al., 2012).

2.2.2. RIG-I and MDA5 are involved in IFN-β production in BTV-infected epithelial cells

As mentioned previously in this review, the innate cellular responses activated during viral infections require recognition of PAMPs by PRRs. Several PRRs have been identified for the *Reoviridae* family, including TLR3 (Alexopoulou et al., 2001), RIG-I and MDA5 (Broquet et al., 2011; Sen et al., 2011; Yoneyama et al., 2004), PKR (Sen et al., 2011; Garcia et al., 2007) and the newly described TRIF-dependent DexD/H-box helicases (Zhang et al., 2011). Although BTV has been recognized for many years as a potent inducer of type I interferon (see above), the PRRs and signalling pathways involved in this cellular response remained uncharacterized.

Using A549 cells, we aimed to identify which signalling pathway(s) is triggered by BTV for IFN-α/β synthesis. We used a siRNA approach to silence the expression of MAVS, TLR3, MyD88, TRIF and DDX1 which are critical components of major pathways involved in viral sensing. Upon BTV infection, the expression of IFN-β mRNA expression was severely impaired only after MAVS silencing,

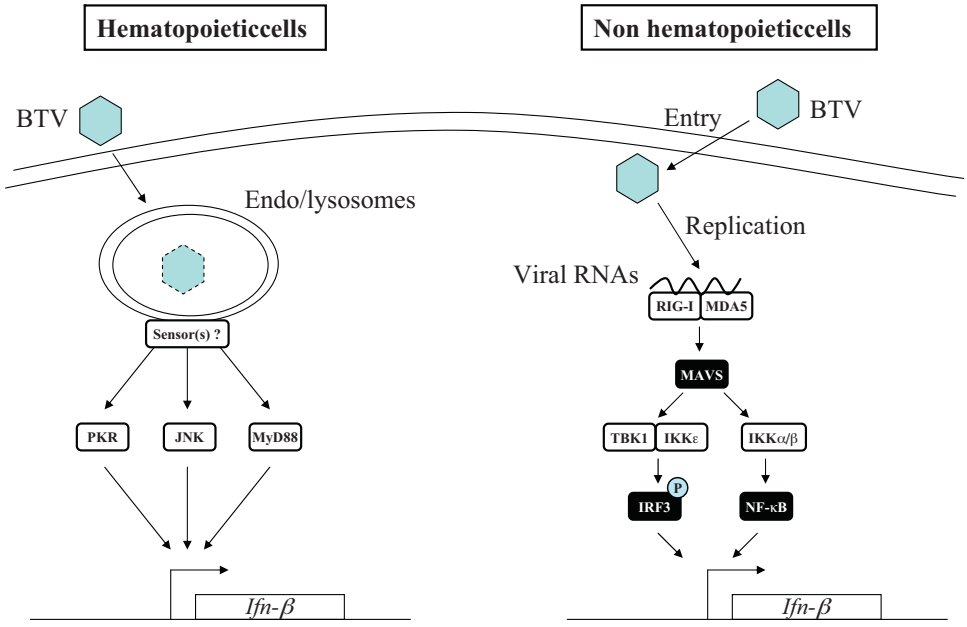

Fig. 1. Mechanisms of IFN-α/β induction upon BTV infection in haematopoietic and non-haematopoietic cells. In pDCs, IFN-α/β production involves endo/lysosomal acidification after BTV infection. This synthesis occurs through a TLR7-independent pathway implicating the MyD88 adaptor, PKR and JNK kinases, *via* a new sensor(s) whose identity and subcellular location remain to be determined. In contrast, in non-haematopoietic cells, BTV can activate the RNA helicases RIG-I and MDA5 that trigger a signalling cascade involving the mitochondrial protein MAVS and the subsequent activation of the IRF3 and NF-κB pathways that are both required for IFN-β synthesis.

indicating that MAVS is involved in IFN-β production following BTV infection (Chauveau et al., 2012). As MAVS is an adaptor protein for RIG-I and MDA5 helicases, we assessed the role of these viral RNA sensors in the production of IFN-β during BTV infection. When the expression of RIG-I or MDA5 was silenced with specific siRNA prior to BTV infection, IFN-β mRNA level was significantly reduced in infected cells. These results were confirmed at the protein level using an IFN-β ELISA assay. Interestingly, overexpression of RIG-I, MDA5 or both proteins in A549 cells prior to BTV infection resulted in a decrease in viral RNA, VP5 protein expression and virus titre, highlighting the importance of these sensors to mount an antiviral response towards BTV (Fig. 1).

2.2.3. *Induction of type I IFN in ovine plasmacytoid dendritic cells occurs via a Myd88-dependent pathway*

As depicted above, BTV targets cDCs and pDCs early after infection in sheep and induces IFN-α/β synthesis in pDCs (Hemati et al., 2009; Ruscanu et al., 2012). In order to investigate the mechanism of IFN-α/β induction in pDCs, Ruscanu et al. first addressed whether IFN-α/β induction by UV-BTV required intracellular processing *via* endo-/lysosomal vesicle maturation. Inhibitors of endolysosomal maturation blocked IFN-α/β synthesis, suggesting that a TLR-mediated signalling is involved in IFN-α/β induction after UV-BTV stimulation, as usually observed with pDCs during viral infections. The use of A151, an oligonucleotide described as a TLR antagonist inhibiting TLR7 and, to a lesser extent, TLR9 did not inhibit IFN-α/β induction by UV-BTV, indicating that UV-BTV activates IFN-α/β production *via* a TLR7/8-independent mechanism.

The role of MyD88 adaptor was also investigated as it mediates TLR7 and TLR9 signalling but also TLR-independent pathways (Kim et al., 2010). LD PBMCs from five different sheep were cultured overnight with a MyD88 inhibitory peptide (Loiarro et al., 2005) or a control peptide and then activated with CpG-A or with UV-BTV for 12 h. The MyD88 inhibitory peptide significantly inhibited IFN-α/β synthesis induced by both CpG and BTV, indicating that UV-BTV-dependent IFN-α/β synthesis in pDCs involves, at least partly, the MyD88 adaptor independently of TLR7/8 activation.

Ruscanu et al. also assessed the contribution of the cytosolic serine/threonine kinase PKR in IFN-α/β production induced by UV-BTV as its role in IFN-α/β induction in response to viral infections was not yet known in pDCs (Diebold et al., 2003; Hornung et al., 2004; Schulz et al., 2010; Sen et al., 2011). After treatment with PKR inhibitors, IFN-α/β production was impaired in LD PBMCs as well as in purified blood pDCs. PKR is known to activate mitogen-activated protein kinases (MAPK) which have also been implicated in the induction of IFN-α/β in response to virus infection (Goh et al., 2000; Fejer et al., 2008). To assess the contribution of this family of kinases, Ruscanu et al. cultured LD PBMCs and purified pDCs with UV-BTV in the presence of specific drugs that target the activity of stress-activated protein kinase (SAPK)/JNK or ERK1/2, two members of the MAPK family. The authors found that UV-BTV IFN-α/β induction was inhibited by the SAPK/JNK inhibitor by over 60%, whereas the ERK1/2 inhibitor had no effect.

Altogether, these data show that the induction of IFN-α/β by UV-BTV in pDCs involves a TLR7/8-independent and MyD88-dependent mechanism. Based on the use of pharmacological inhibitors, they also implicate PKR- and SAPK/JNK-dependent mechanisms (Fig. 1).

3. Modulation of the IFN pathway by NS3 of BTV

Production of type I IFN induces a cellular response that is detrimental for viral propagation. In order to dampen this host antiviral response, most of viruses have evolved counteracting strategies. This includes members of the *Reoviridae* family such as rotavirus and reovirus. The ability of BTV to control the IFN pathway has been poorly studied in the past and only recently, a study from our group has shown that BTV is also able to inhibit this antiviral response (Chauveau et al., 2013).

The effect of BTV infection on the activation of IFN-β promoter was first assessed using a luciferase reporter assay. 293T cells were infected with a BTV-8 strain isolated in France and transfected 8 h p.i. with a IFN-β reporter plasmid, a control vector and an expression vector coding for a constitutively active form of RIG-I (NRIG-I). IFN-β promoter activation was greatly impaired in BTV-infected cells in comparison to mock-infected cells, showing that BTV-8 is

able to inhibit IFN-β promoter activity after stimulation of the RIG-I-like pathway. The impact of the overexpression of the different BTV ORFs on the activity of IFN-β promoter was assessed using the same assay and the Polo-like kinase 1 Polo-box domain (Plk1-PBD), an antagonist of the RLR pathway, was used as control (Vitour et al., 2009). The strongest inhibitory effect on IFN-β promoter activity was achieved in cells expressing the viral protein NS3. This effect was conserved between field and attenuated strains from different BTV serotypes. Indeed, live South African vaccine strains of BTV-2 and -4 were also able to block this activation. Furthermore, we showed that NS3 reduced the level of IFN-β mRNA transcripts upon activation with NRIG-I, confirming that NS3 affects the transcription of *IFN-β* gene at the endogenous level. NS3 was also able to block the activation of IFN-β promoter induced by other stimuli of the RLR pathway, including Sendai virus (RNA virus), poly(I:C) (synthetic dsRNA) or poly(dA/dT) (transcribed to dsRNA by RNA polymerase III). Interestingly, NS3 had no effect on the activity of an *in vitro*-transcribed mRNA encoding the luciferase protein, suggesting that NS3 does not interfere with translation. NS3 had no effect on the activity of a CMV promoter and a promoter dependent on c-jun, a transcription factor involved in the expression of multiple genes, showing that NS3 did not induce a complete host transcriptional shutoff. However, it is still possible that NS3 affect a broad range of promoters.

In order to identify at which level of the RLR pathway NS3 acts, the effect of NS3 on the ability of overexpressed MAVS, TBK1, IKKε and IRF-3 5D (a constitutively active form of IRF-3) to activate IFN-β promoter was assessed in the same study. Results from this experiment showed that NS3 interferes with the IFN synthesis pathway downstream RIG-I and upstream TBK1/IKKε activation.

4. Conclusions

BTV is a major animal health concern but remains poorly characterized especially when it comes to the understanding of how the virus interacts with the immune innate response. BTV is a strong inducer of IFN-α/β, a key player of the innate antiviral response, in many cells types including endothelial and epithelial cells of bovine and human origin. In non-haematopoietic cells, the viral RNA sensors RIG-I and MDA5 are upregulated by BTV infection and these helicases control both the sensing and the pro-inflammatory and anti-viral responses to BTV. In contrast, BTV activates the IFN-α/β signalling pathway in pDCs *via* the MyD88 adaptor, independently of TLR7/8, and *via* a mechanism implicating PKR and JNK kinases. These data suggest for the first time that a dsRNA virus can trigger IFN-α/β in primary host pDCs *via* a mechanism independent on TLR7/8 but dependent on MyD88. Deeper mechanistic investigations should be undertaken to discover additional viral sensors in pDCs that would be pertinent for dsRNA viruses and possibly other pathogens.

The discrepancy between the sensors and signalling cascades activated by BTV in immune *versus* non-immune cells could be explained by the fact that different cellular responses exist in these cells. It is possible that, during the first stage of infection, a strong systemic production of type I IFN and other cytokines is induced in specialized antigen presenting cells (mainly pDC) through a replication- and RLR-independent mechanism and later, in the main target cells of BTV infection, such as endothelial cells, distinct PRRs respond to products of BTV replication to trigger a tissue-localized antiviral response (Fig. 1). These findings help in the understanding of the pathogenesis associated with BTV infection and, importantly, facilitate the development of better vaccines against dsRNA viruses. For example, they suggest that industrial processes of viral inactivation must maintain the ability of BTV and/or other members of the *Reoviridae* family to trigger type I IFN production by pDCs in order to induce optimal adaptive responses after vaccination. Indeed, type I IFN production by pDCs has recently been shown to be essential for the induction of high antibody titres upon rotavirus infection (Deal et al., 2013).

In this review, we also discussed evidences given on the ability of NS3 of BTV to interfere with the induction of the innate immune response in non-haematopoietic cells by inhibiting the RLR-dependent signalling pathway. Thus, as most viruses, BTV has evolved at least one strategy to interfere with the production of type I IFN. NS3 was shown to modulate the induction of type I IFN but it is possible that other genes/pathways are affected by this viral protein. The mode of action of NS3 remains poorly characterized and further studies need to be undertaken to fully understand the different functions of this viral protein. Some other viral proteins might also interfere with host antiviral responses and the IFN system. In a reporter assay, NS3 was shown to be the strongest inhibitor of IFN-β promoter activation but other proteins also displayed antagonistic activity although to a lesser extent (Chauveau et al., 2013). Moreover, an additional protein encoded by BTV, NS4, has been recently discovered and is thought to interact with the IFN pathway (Ratinier et al., 2011).

Interestingly, in another study, several BTV proteins including VP1, VP2 and NS2 were suggested to be determinants of virulence as the genome encoding these proteins consistently showed non-synonymous changes between virulent and attenuated BTV strain pairs (Caporale et al., 2011). It would be of great interest to investigate further whether these virulent factors contributing to viral pathogenesis are able to modulate the strength of type I IFN signalling and to precise the putative role of some other viral proteins in the control of this response (Stewart and Roy, 2010; Ratinier et al., 2011). The understanding of the interplay between BTV and the IFN pathway is only at its beginning and many unknowns remain to be addressed.

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References

Abramovich, C., Shulman, L.M., et al., 1994. Differential tyrosine phosphorylation of the IFNAR chain of the type I interferon receptor and of an associated surface protein in response to IFN-alpha and IFN-beta. EMBO J. 13 (24), 5871–5877.
Akira, S., 2006. TLR signaling. Curr. Top. Microbiol. Immunol. 311, 1–16.
Akira, S., Uematsu, S., et al., 2006. Pathogen recognition and innate immunity. Cell 124 (4), 783–801.
Alexopoulou, L., Holt, A.C., et al., 2001. Recognition of double-stranded RNA and activation of NF-kappaB by Toll-like receptor 3. Nature 413 (6857), 732–738.
Andrejeva, J., Childs, K.S., et al., 2004. The V proteins of paramyxoviruses bind the IFN-inducible RNA helicase, mda-5, and inhibit its activation of the IFN-beta promoter. Proc. Natl. Acad. Sci. U. S. A. 101 (49), 17264–17269.
Ank, N., West, H., et al., 2006. IFN-lambda: novel antiviral cytokines. J. Interferon Cytokine Res. 26 (6), 373–379.
Arimoto, K., Takahashi, H., et al., 2007. Negative regulation of the RIG-I signaling by the ubiquitin ligase RNF125. Proc. Natl. Acad. Sci. U. S. A. 104 (18), 7500–7505.
Arnoult, D., Carneiro, L., et al., 2009. The role of mitochondria in cellular defense against microbial infection. Semin. Immunol. 21 (4), 223–232.
Barro, M., Patton, J.T., 2005. Rotavirus nonstructural protein 1 subverts innate immune response by inducing degradation of IFN regulatory factor 3. Proc. Natl. Acad. Sci. U. S. A. 102 (11), 4114–4119.
Barro, M., Patton, J.T., 2007. Rotavirus NSP1 inhibits expression of type I interferon by antagonizing the function of interferon regulatory factors IRF3, IRF5, and IRF7. J. Virol. 81 (9), 4473–4481.
Beadling, C., Guschin, D., et al., 1994. Activation of JAK kinases and STAT proteins by interleukin-2 and interferon alpha, but not the T cell antigen receptor, in human T lymphocytes. EMBO J. 13 (23), 5605–5615.
Berke, I.C., Modis, Y., 2012. MDA5 cooperatively forms dimers and ATP-sensitive filaments upon binding double-stranded RNA. EMBO J. 31 (7), 1714–1726.
Beutler, B.A., 2009. TLRs and innate immunity. Blood 113 (7), 1399–1407.

Bhanja Chowdhury, J., Shrivastava, S., et al., 2012. Hepatitis C virus infection modulates expression of interferon stimulatory gene IFITM1 by upregulating miR-130A. J. Virol. 86 (18), 10221–10225.
Boehme, K.W., Guerrero, M., et al., 2006. Human cytomegalovirus envelope glycoproteins B and H are necessary for TLR2 activation in permissive cells. J. Immunol. 177 (10), 7094–7102.
Bonjardim, C.A., 2005. Interferons (IFNs) are key cytokines in both innate and adaptive antiviral immune responses – and viruses counteract IFN action. Microbes Infect. 7 (3), 569–578.
Bonjardim, C.A., Ferreira, P.C., et al., 2009. Interferons: signaling, antiviral and viral evasion. Immunol. Lett. 122 (1), 1–11.
Bowie, A.G., Unterholzner, L., 2008. Viral evasion and subversion of pattern-recognition receptor signalling. Nat. Rev. Immunol. 8 (12), 911–922.
Broquet, A.H., Hirata, Y., et al., 2011. RIG-I/MDA5/MAVS are required to signal a protective IFN response in rotavirus-infected intestinal epithelium. J. Immunol. 186 (3), 1618–1626.
Calvo-Pinilla, E., Navasa, N., et al., 2012. Multiserotype protection elicited by a combinatorial prime-boost vaccination strategy against Bluetongue virus. PLoS ONE 7 (4), e34735.
Calvo-Pinilla, E., Nieto, J.M., et al., 2010. Experimental oral infection of Bluetongue virus serotype 8 in type I interferon receptor-deficient mice. J. Gen. Virol. 91 (Pt 11), 2821–2825.
Calvo-Pinilla, E., Rodriguez-Calvo, T., et al., 2009a. Establishment of a Bluetongue virus infection model in mice that are deficient in the alpha/beta interferon receptor. PLoS ONE 4 (4), e5171.
Calvo-Pinilla, E., Rodriguez-Calvo, T., et al., 2009b. Heterologous prime boost vaccination with DNA and recombinant modified vaccinia virus Ankara protects IFNAR(−/−) mice against lethal Bluetongue infection. Vaccine 28 (2), 437–445.
Caporale, M., Wash, R., et al., 2011. Determinants of Bluetongue virus virulence in murine models of disease. J. Virol. 85 (21), 11479–11489.
Chauveau, E., Doceul, V., et al., 2012. Sensing and control of Bluetongue virus infection in epithelial cells via RIG-I and MDA5 helicases. J. Virol. 86 (21), 11789–11799.
Chauveau, E., Doceul, V., et al., 2013. NS3 of Bluetongue virus interferes with the induction of type I interferon. J. Virol. 87 (14), 8241–8246.
Chen, Z., Benureau, Y., et al., 2007. GB virus B disrupts RIG-I signaling by NS3/4A-mediated cleavage of the adaptor protein MAVS. J. Virol. 81 (2), 964–976.
Childs, K., Stock, N., et al., 2007. mda-5, but not RIG-I, is a common target for paramyxovirus V proteins. Virology 359 (1), 190–200.
Clementz, M.A., Chen, Z., et al., 2010. Deubiquitinating and interferon antagonism activities of coronavirus papain-like proteases. J. Virol. 84 (9), 4619–4629.
Coen, M.L., Ellis, J.A., et al., 1991. Cytokine modulation of the interaction between Bluetongue virus and endothelial cells in vitro. Vet. Pathol. 28 (6), 524–532.
Colamonici, O., Yan, H., et al., 1994a. Direct binding to and tyrosine phosphorylation of the alpha subunit of the type I interferon receptor by p135tyk2 tyrosine kinase. Mol. Cell. Biol. 14 (12), 8133–8142.
Colamonici, O.R., Domanski, P., et al., 1995. Vaccinia virus B18R gene encodes a type I interferon-binding protein that blocks interferon alpha transmembrane signaling. J. Biol. Chem. 270 (27), 15974–15978.
Colamonici, O.R., Uyttendaele, H., et al., 1994b. p135tyk2, an interferon-alpha-activated tyrosine kinase, is physically associated with an interferon-alpha receptor. J. Biol. Chem. 269 (5), 3518–3522.
Cui, S., Eisenacher, K., et al., 2008. The C-terminal regulatory domain is the RNA 5′-triphosphate sensor of RIG-I. Mol. Cell 29 (2), 169–179.
de Weerd, N.A., Samarajiwa, S.A., et al., 2007. Type I interferon receptors: biochemistry and biological functions. J. Biol. Chem. 282 (28), 20053–20057.
Deal, E.M., Lahl, K., et al., 2013. Plasmacytoid dendritic cells promote rotavirus-induced human and murine B cell responses. J. Clin. Invest. 123 (6), 2464–2474.
DeMaula, C.D., Jutila, M.A., et al., 2001. Infection kinetics, prostacyclin release and cytokine-mediated modulation of the mechanism of cell death during Bluetongue virus infection of cultured ovine and bovine pulmonary artery and lung microvascular endothelial cells. J. Gen. Virol. 82 (Pt 4), 787–794.
DeMaula, C.D., Leutenegger, C.M., et al., 2002. The role of endothelial cell-derived inflammatory and vasoactive mediators in the pathogenesis of Bluetongue. Virology 296 (2), 330–337.
Diebold, S.S., Montoya, M., et al., 2003. Viral infection switches non-plasmacytoid dendritic cells into high interferon producers. Nature 424 (6946), 324–328.
Dong, B., Silverman, R.H., 1995. 2-5A-dependent RNase molecules dimerize during activation by 2-5A. J. Biol. Chem. 270 (8), 4133–4137.
Eisenacher, K., Steinberg, C., et al., 2008. The role of viral nucleic acid recognition in dendritic cells for innate and adaptive antiviral immunity. Immunobiology 212 (9–10), 701–714.
Eksteen, P.A., Huismans, H., 1972. Interferon induction by Bluetongue virus and Bluetongue virus ribonucleic acid. Onderstepoort J. Vet. Res. 39 (3), 125–131.
Fasler-Kan, E., Pansky, A., et al., 1998. Interferon-alpha activates signal transducers and activators of transcription 5 and 6 in Daudi cells. Eur. J. Biochem. 254 (3), 514–519.
Fejer, G., Drechsel, L., et al., 2008. Key role of splenic myeloid DCs in the IFN-alphabeta response to adenoviruses in vivo. PLoS Pathog. 4 (11), e1000208.
Fiette, L., Aubert, C., et al., 1995. Theiler's virus infection of 129 Sv mice that lack the interferon alpha/beta or interferon gamma receptors. J. Exp. Med. 181 (6), 2069–2076.
Fiola, S., Gosselin, D., et al., 2010. TLR9 contributes to the recognition of EBV by primary monocytes and plasmacytoid dendritic cells. J. Immunol. 185 (6), 3620–3631.
Foster, N.M., Luedke, A.J., et al., 1991. Temporal relationships of viremia, interferon activity, and antibody responses of sheep infected with several Bluetongue virus strains. Am. J. Vet. Res. 52 (2), 192–196.
Fredericksen, B.L., Gale Jr., M., 2006. West Nile virus evades activation of interferon regulatory factor 3 through RIG-I-dependent and -independent pathways without antagonizing host defense signaling. J. Virol. 80 (6), 2913–2923.
Fu, X.Y., Schindler, C., et al., 1992. The proteins of ISGF-3, the interferon alpha-induced transcriptional activator, define a gene family involved in signal transduction. Proc. Natl. Acad. Sci. U. S. A. 89 (16), 7840–7843.
Fulton, R.W., Pearson, N.J., 1982. Interferon induction in bovine and feline monolayer cultures by four Bluetongue virus serotypes. Can. J. Comp. Med. 46 (1), 100–102.
Gack, M.U., Shin, Y.C., et al., 2007. TRIM25 RING-finger E3 ubiquitin ligase is essential for RIG-I-mediated antiviral activity. Nature 446 (7138), 916–920.
Garcia, M.A., Meurs, E.F., et al., 2007. The dsRNA protein kinase PKR: virus and cell control. Biochimie 89 (6–7), 799–811.
Gauzzi, M.C., Velazquez, L., et al., 1996. Interferon-alpha-dependent activation of Tyk2 requires phosphorylation of positive regulatory tyrosines by another kinase. J. Biol. Chem. 271 (34), 20494–20500.
Gilfoy, F.D., Mason, P.W., 2007. West Nile virus-induced interferon production is mediated by the double-stranded RNA-dependent protein kinase PKR. J. Virol. 81 (20), 11148–11158.
Gitlin, L., Barchet, W., et al., 2006. Essential role of mda-5 in type I IFN responses to polyriboinosinic:polyribocytidylic acid and encephalomyocarditis picornavirus. Proc. Natl. Acad. Sci. U. S. A. 103 (22), 8459–8464.
Goh, K.C., deVeer, M.J., et al., 2000. The protein kinase PKR is required for p38 MAPK activation and the innate immune response to bacterial endotoxin. EMBO J. 19 (16), 4292–4297.
Gonzalez-Lopez, C., Martinez-Costas, J., et al., 2003. Evidence that avian reovirus sigmaA protein is an inhibitor of the double-stranded RNA-dependent protein kinase. J. Gen. Virol. 84 (Pt 6), 1629–1639.
Goodman, A.G., Smith, J.A., et al., 2007. The cellular protein P58IPK regulates influenza virus mRNA translation and replication through a PKR-mediated mechanism. J. Virol. 81 (5), 2221–2230.
Guzylack-Piriou, L., Bergamin, F., et al., 2006. Plasmacytoid dendritic cell activation by foot-and-mouth disease virus requires immune complexes. Eur. J. Immunol. 36 (7), 1674–1683.
Hacker, H., Redecke, V., et al., 2006. Specificity in Toll-like receptor signalling through distinct effector functions of TRAF3 and TRAF6. Nature 439 (7073), 204–207.
Haller, O., Kochs, G., et al., 2006. The interferon response circuit: induction and suppression by pathogenic viruses. Virology 344 (1), 119–130.
Haller, O., Kochs, G., et al., 2007a. Interferon, Mx, and viral countermeasures. Cytokine Growth Factor Rev. 18 (5–6), 425–433.
Haller, O., Stertz, S., et al., 2007b. The Mx GTPase family of interferon-induced antiviral proteins. Microbes Infect. 9 (14–15), 1636–1643.
Hansen, T.R., Henkes, L.K., et al., 2010. Endocrine actions of interferon-tau in ruminants. Soc. Reprod. Fertil. Suppl. 67, 325–340.
Hardarson, H.S., Baker, J.S., et al., 2007. Toll-like receptor 3 is an essential component of the innate stress response in virus-induced cardiac injury. Am. J. Physiol. Heart Circ. Physiol. 292 (1), H251–H258.
Harte, M.T., Haga, I.R., et al., 2003. The poxvirus protein A52R targets Toll-like receptor signaling complexes to suppress host defense. J. Exp. Med. 197 (3), 343–351.
Heil, F., Hemmi, H., et al., 2004. Species-specific recognition of single-stranded RNA via toll-like receptor 7 and 8. Science 303 (5663), 1526–1529.
Hemati, B., Contreras, V., et al., 2009. Bluetongue virus targets conventional dendritic cells in skin lymph. J. Virol. 83 (17), 8789–8799.
Hemmi, H., Takeuchi, O., et al., 2000. A Toll-like receptor recognizes bacterial DNA. Nature 408 (6813), 740–745.
Hervas-Stubbs, S., Perez-Gracia, J.L., et al., 2011. Direct effects of type I interferons on cells of the immune system. Clin. Cancer Res. 17 (9), 2619–2627.
Hiscott, J., 2007. Triggering the innate antiviral response through IRF-3 activation. J. Biol. Chem. 282 (21), 15325–15329.
Hochrein, H., Schlatter, B., et al., 2004. Herpes simplex virus type-1 induces IFN-alpha production via Toll-like receptor 9-dependent and -independent pathways. Proc. Natl. Acad. Sci. U. S. A. 101 (31), 11416–11421.
Hoenen, A., Liu, W., et al., 2007. West Nile virus-induced cytoplasmic membrane structures provide partial protection against the interferon-induced antiviral MxA protein. J. Gen. Virol. 88 (Pt 11), 3013–3017.
Holloway, G., Truong, T.T., et al., 2009. Rotavirus antagonizes cellular antiviral responses by inhibiting the nuclear accumulation of STAT1, STAT2, and NF-kappaB. J. Virol. 83 (10), 4942–4951.
Honda, K., Yanai, H., et al., 2005. IRF-7 is the master regulator of type-I interferon-dependent immune responses. Nature 434 (7034), 772–777.
Hornung, V., Ellegast, J., et al., 2006. 5′-Triphosphate RNA is the ligand for RIG-I. Science 314 (5801), 994–997.
Hornung, V., Schlender, J., et al., 2004. Replication-dependent potent IFN-alpha induction in human plasmacytoid dendritic cells by a single-stranded RNA virus. J. Immunol. 173 (10), 5935–5943.
Huber, J.P., Farrar, J.D., 2011. Regulation of effector and memory T-cell functions by type I interferon. Immunology 132 (4), 466–474.
Huismans, H., 1969. Bluetongue virus-induced interferon synthesis. Onderstepoort J. Vet. Res. 36 (2), 181–185.
Imani, F., Jacobs, B.L., 1988. Inhibitory activity for the interferon-induced protein kinase is associated with the reovirus serotype 1 sigma 3 protein. Proc. Natl. Acad. Sci. U. S. A. 85 (21), 7887–7891.
Isaacs, A., Lindenmann, J., 1957. Virus interference. I. The interferon. Proc. R. Soc. Lond. B: Biol. Sci. 147 (927), 258–267.

Ishikawa, H., Barber, G.N., 2008. STING is an endoplasmic reticulum adaptor that facilitates innate immune signalling. Nature 455 (7213), 674–678.
Jacobs, B.L., Langland, J.O., 1998. Reovirus sigma 3 protein: dsRNA binding and inhibition of RNA-activated protein kinase. Curr. Top. Microbiol. Immunol. 233 (Pt 1), 185–196.
Jameson, P., Grossberg, S.E., 1978. Production of interferon in human cell cultures by a new, potent viral inducer. Adv. Exp. Med. Biol. 110, 37–53.
Jameson, P., Grossberg, S.E., 1979. Production of interferon by human tumor cell lines. Arch. Virol 62 (3), 209–219.
Jameson, P., Grossberg, S.E., 1981. Induction of interferon with Bluetongue virus in various cells. Methods Enzymol. 78 (Pt A), 254–258.
Jameson, P., Schoenherr, C.K., et al., 1978. Bluetongue virus, an exceptionally potent interferon inducer in mice. Infect. Immun. 20 (1), 321–323.
Joklik, W.K., 1981. Induction of interferon by nonmultiplying virus. Methods Enzymol. 79 (Pt B), 330–336.
Kaisho, T., Akira, S., 2006. Toll-like receptor function and signaling. J. Allergy Clin. Immunol. 117 (5), 979–987 (quiz 988).
Kato, H., Sato, S., et al., 2005. Cell type-specific involvement of RIG-I in antiviral response. Immunity 23 (1), 19–28.
Kato, H., Takeuchi, O., et al., 2008. Length-dependent recognition of double-stranded ribonucleic acids by retinoic acid-inducible gene-I and melanoma differentiation-associated gene 5. J. Exp. Med. 205 (7), 1601–1610.
Kato, H., Takeuchi, O., et al., 2006. Differential roles of MDA5 and RIG-I helicases in the recognition of RNA viruses. Nature 441 (7089), 101–105.
Kawai, T., Akira, S., 2007. Antiviral signaling through pattern recognition receptors. J. Biochem. (Tokyo) 141 (2), 137–145.
Kawai, T., Sato, S., et al., 2004. Interferon-alpha induction through Toll-like receptors involves a direct interaction of IRF7 with MyD88 and TRAF6. Nat. Immunol. 5 (10), 1061–1068.
Kawai, T., Takahashi, K., et al., 2005. IPS-1, an adaptor triggering RIG-I- and Mda5-mediated type I interferon induction. Nat. Immunol. 6 (10), 981–988.
Kiefer, K., Oropallo, M.A., et al., 2012. Role of type I interferons in the activation of autoreactive B cells. Immunol. Cell Biol. 90 (5), 498–504.
Kim, T., Pazhoor, S., et al., 2010. Aspartate-glutamate-alanine-histidine box motif (DEAH)/RNA helicase A helicases sense microbial DNA in human plasmacytoid dendritic cells. Proc. Natl. Acad. Sci. U. S. A. 107 (34), 15181–15186.
Kolakofsky, D., Kowalinski, E., et al., 2012. A structure-based model of RIG-I activation. RNA 18 (12), 2118–2127.
Komuro, A., Horvath, C.M., 2006. RNA- and virus-independent inhibition of antiviral signaling by RNA helicase LGP2. J. Virol. 80 (24), 12332–12342.
Kotenko, S.V., 2011. IFN-lambdas. Curr. Opin. Immunol. 23 (5), 583–590.
Krishnan, K., Yan, H., et al., 1996. Dimerization of a chimeric CD4-interferon-alpha receptor reconstitutes the signaling events preceding STAT phosphorylation. Oncogene 13 (1), 125–133.
Kurt-Jones, E.A., Popova, L., et al., 2000. Pattern recognition receptors TLR4 and CD14 mediate response to respiratory syncytial virus. Nat. Immunol. 1 (5), 398–401.
Lamken, P., Lata, S., et al., 2004. Ligand-induced assembling of the type I interferon receptor on supported lipid bilayers. J. Mol. Biol. 341 (1), 303–318.
Lee, H.R., Kim, M.H., et al., 2009. Viral interferon regulatory factors. J. Interferon Cytokine Res. 29 (9), 621–627.
Lee, T.G., Tomita, J., et al., 1990. Purification and partial characterization of a cellular inhibitor of the interferon-induced protein kinase of Mr 68,000 from influenza virus-infected cells. Proc. Natl. Acad. Sci. U. S. A. 87 (16), 6208–6212.
Lee, T.G., Tomita, J., et al., 1992. Characterization and regulation of the 58,000-dalton cellular inhibitor of the interferon-induced, dsRNA-activated protein kinase. J. Biol. Chem. 267 (20), 14238–14243.
Lin, R., Genin, P., et al., 2001. HHV-8 encoded vIRF-1 represses the interferon antiviral response by blocking IRF-3 recruitment of the CBP/p300 coactivators. Oncogene 20 (7), 800–811.
Liu, K., Yang, X., et al., 2009. Rotavirus strategies to evade host antiviral innate immunity. Immunol. Lett. 127 (1), 13–18.
Liu, S.Y., Sanchez, D.J., et al., 2011. New developments in the induction and antiviral effectors of type I interferon. Curr. Opin. Immunol. 23 (1), 57–64.
Loiarro, M., Sette, C., et al., 2005. Peptide-mediated interference of TIR domain dimerization in MyD88 inhibits interleukin-1-dependent activation of NF-{kappa}B. J. Biol. Chem. 280 (16), 15809–15814.
Lu, C., Xu, H., et al., 2010. The structural basis of 5′ triphosphate double-stranded RNA recognition by RIG-I C-terminal domain. Structure 18 (8), 1032–1043.
Luber, C.A., Cox, J., et al., 2010. Quantitative proteomics reveals subset-specific viral recognition in dendritic cells. Immunity 32 (2), 279–289.
Lubyova, B., Kellum, M.J., et al., 2004. Kaposi's sarcoma-associated herpesvirus-encoded vIRF-3 stimulates the transcriptional activity of cellular IRF-3 and IRF-7. J. Biol. Chem. 279 (9), 7643–7654.
Lund, J., Sato, A., et al., 2003. Toll-like receptor 9-mediated recognition of Herpes simplex virus-2 by plasmacytoid dendritic cells. J. Exp. Med. 198 (3), 513–520.
Lyons, S.F., Schoub, B.D., et al., 1982. Induction of interferon by Sendai virus versus blue tongue virus. J. Interferon Res. 2 (4), 543–547.
MacLachlan, N.J., Schore, C.E., et al., 1984. Antiviral responses of Bluetongue virus-inoculated bovine fetuses and their dams. Am. J. Vet. Res. 45 (7), 1469–1473.
MacLachlan, N.J., Thompson, J., 1985. Bluetongue virus-induced interferon in cattle. Am. J. Vet. Res. 46 (6), 1238–1241.
Malathi, K., Dong, B., et al., 2007. Small self-RNA generated by RNase L amplifies antiviral innate immunity. Nature 448 (7155), 816–819.
Marques, J.T., Devosse, T., et al., 2006. A structural basis for discriminating between self and nonself double-stranded RNAs in mammalian cells. Nat. Biotechnol. 24 (5), 559–565.
Martinand, C., Montavon, C., et al., 1999. RNase L inhibitor is induced during human immunodeficiency virus type 1 infection and down regulates the 2-5A/RNase L pathway in human T cells. J. Virol. 73 (1), 290–296.
Martinand, C., Salehzada, T., et al., 1998. RNase L inhibitor (RLI) antisense constructions block partially the down regulation of the 2-5A/RNase L pathway in encephalomyocarditis-virus-(EMCV)-infected cells. Eur. J. Biochem. 254 (2), 248–255.
Martinez-Costas, J., Gonzalez-Lopez, C., et al., 2000. Possible involvement of the double-stranded RNA-binding core protein sigmaA in the resistance of avian reovirus to interferon. J. Virol. 74 (3), 1124–1131.
Matikainen, S., Sareneva, T., et al., 1999. Interferon-alpha activates multiple STAT proteins and upregulates proliferation-associated IL-2Ralpha, c-myc, and pim-1 genes in human T cells. Blood 93 (6), 1980–1991.
McAllister, C.S., Samuel, C.E., 2009. The RNA-activated protein kinase enhances the induction of interferon-beta and apoptosis mediated by cytoplasmic RNA sensors. J. Biol. Chem. 284 (3), 1644–1651.
Meinke, A., Barahmand-Pour, F., et al., 1996. Activation of different Stat5 isoforms contributes to cell-type-restricted signaling in response to interferons. Mol. Cell. Biol. 16 (12), 6937–6944.
Melroe, G.T., Silva, L., et al., 2007. Recruitment of activated IRF-3 and CBP/p300 to herpes simplex virus ICP0 nuclear foci: potential role in blocking IFN-beta induction. Virology 360 (2), 305–321.
Melville, M.W., Tan, S.L., et al., 1999. The cellular inhibitor of the PKR protein kinase, P58(IPK), is an influenza virus-activated co-chaperone that modulates heat shock protein 70 activity. J. Biol. Chem. 274 (6), 3797–3803.
Meurs, E., Chong, K., et al., 1990. Molecular cloning and characterization of the human double-stranded RNA-activated protein kinase induced by interferon. Cell 62 (2), 379–390.
Meylan, E., Curran, J., et al., 2005. Cardif is an adaptor protein in the RIG-I antiviral pathway and is targeted by hepatitis C virus. Nature 437 (7062), 1167–1172.
Mibayashi, M., Martinez-Sobrido, L., et al., 2007. Inhibition of retinoic acid-inducible gene I-mediated induction of beta interferon by the NS1 protein of influenza A virus. J. Virol. 81 (2), 514–524.
Muller, U., Steinhoff, U., et al., 1994. Functional role of type I and type II interferons in antiviral defense. Science 264 (5167), 1918–1921.
Nguyen, K.B., Salazar-Mather, T.P., et al., 2002. Coordinated and distinct roles for IFN-alpha beta, IL-12, and IL-15 regulation of NK cell responses to viral infection. J. Immunol. 169 (8), 4279–4287.
O'Neill, L.A., Bowie, A.G., 2007. The family of five: TIR-domain-containing adaptors in Toll-like receptor signalling. Nat. Rev. Immunol. 7 (5), 353–364.
Oganesyan, G., Saha, S.K., et al., 2006. Critical role of TRAF3 in the Toll-like receptor-dependent and -independent antiviral response. Nature 439 (7073), 208–211.
Panne, D., Maniatis, T., et al., 2007. An atomic model of the interferon-beta enhanceosome. Cell 129 (6), 1111–1123.
Pascale, F., Contreras, V., et al., 2008. Plasmacytoid dendritic cells migrate in afferent skin lymph. J. Immunol. 180 (9), 5963–5972.
Pauli, E.K., Schmolke, M., et al., 2008. Influenza A virus inhibits type I IFN signaling via NF-kappaB-dependent induction of SOCS-3 expression. PLoS Pathog. 4 (11), e1000196.
Pestka, S., Krause, C.D., et al., 2004. Interferons, interferon-like cytokines, and their receptors. Immunol. Rev. 202, 8–32.
Pichlmair, A., Schulz, O., et al., 2006. RIG-I-mediated antiviral responses to single-stranded RNA bearing 5′-phosphates. Science 314 (5801), 997–1001.
Platanias, L.C., 2005. Mechanisms of type-I- and type-II-interferon-mediated signalling. Nat. Rev. Immunol. 5 (5), 375–386.
Platanias, L.C., Colamonici, O.R., 1992. Interferon alpha induces rapid tyrosine phosphorylation of the alpha subunit of its receptor. J. Biol. Chem. 267 (33), 24053–24057.
Platanias, L.C., Uddin, S., et al., 1994. Tyrosine phosphorylation of the alpha and beta subunits of the type I interferon receptor. Interferon-beta selectively induces tyrosine phosphorylation of an alpha subunit-associated protein. J. Biol. Chem. 269 (27), 17761–17764.
Plumet, S., Herschke, F., et al., 2007. Cytosolic 5′-triphosphate ended viral leader transcript of measles virus as activator of the RIG I-mediated interferon response. PLoS ONE 2 (3), e279.
Pothlichet, J., Chignard, M., et al., 2008. Cutting edge: innate immune response triggered by influenza A virus is negatively regulated by SOCS1 and SOCS3 through a RIG-I/IFNAR1-dependent pathway. J. Immunol. 180 (4), 2034–2038.
Qin, L., Ren, L., et al., 2011. Rotavirus nonstructural protein 1 antagonizes innate immune response by interacting with retinoic acid inducible gene I. Virol. J. 8, 526.
Randall, R.E., Goodbourn, S., 2008. Interferons and viruses: an interplay between induction, signalling, antiviral responses and virus countermeasures. J. Gen. Virol. 89 (Pt 1), 1–47.
Ratinier, M., Caporale, M., et al., 2011. Identification and characterization of a novel non-structural protein of Bluetongue virus. PLoS Pathog. 7 (12), e1002477.
Rehwinkel, J., Tan, C.P., et al., 2010. RIG-I detects viral genomic RNA during negative-strand RNA virus infection. Cell 140 (3), 397–408.
Ren, J., Kolli, D., et al., 2011. Human metapneumovirus inhibits IFN-beta signaling by downregulating Jak1 and Tyk2 cellular levels. PLoS ONE 6 (9), e24496.
Rinaldo Jr., C.R., Overall Jr., J.C., et al., 1975. Viral replication and interferon production in fetal and adult ovine leukocytes and spleen cells. Infect. Immun. 12 (5), 1070–1077.
Roberts, R.M., Liu, L., et al., 1998. The evolution of the type I interferons. J. Interferon Cytokine Res. 18 (10), 805–816.

Rothenfusser, S., Goutagny, N., et al., 2005. The RNA helicase Lgp2 inhibits TLR-independent sensing of viral replication by retinoic acid-inducible gene-I. J. Immunol. 175 (8), 5260–5268.
Ruscanu, S., Jouneau, L., et al., 2013. Dendritic cell subtypes from lymph nodes and blood show contrasted gene expression programs upon Bluetongue virus infection. J. Virol. 87 (16), 9333–9343.
Ruscanu, S., Pascale, F., et al., 2012. The double-stranded RNA Bluetongue virus induces type I interferon in plasmacytoid dendritic cells via a MYD88-dependent TLR7/8-independent signaling pathway. J. Virol. 86 (10), 5817–5828.
Russell, H., O'Toole, D.T., et al., 1996. Comparative effects of Bluetongue virus infection of ovine and bovine endothelial cells. Vet. Pathol. 33 (3), 319–331.
Ryzhakov, G., Randow, F., 2007. SINTBAD, a novel component of innate antiviral immunity, shares a TBK1-binding domain with NAP1 and TANK. EMBO J. 26 (13), 3180–3190.
Saha, S.K., Pietras, E.M., et al., 2006. Regulation of antiviral responses by a direct and specific interaction between TRAF3 and Cardif. EMBO J. 25 (14), 3257–3263.
Saito, T., Gale Jr., M., 2008. Differential recognition of double-stranded RNA by RIG-I-like receptors in antiviral immunity. J. Exp. Med. 205 (7), 1523–1527.
Saito, T., Hirai, R., et al., 2007. Regulation of innate antiviral defenses through a shared repressor domain in RIG-I and LGP2. Proc. Natl. Acad. Sci. U. S. A. 104 (2), 582–587.
Saito, T., Owen, D.M., et al., 2008. Innate immunity induced by composition-dependent RIG-I recognition of hepatitis C virus RNA. Nature 454 (7203), 523–527.
Satoh, T., Kato, H., et al., 2010. LGP2 is a positive regulator of RIG-I- and MDA5-mediated antiviral responses. Proc. Natl. Acad. Sci. U. S. A. 107 (4), 1512–1517.
Schindler, C., Shuai, K., et al., 1992. Interferon-dependent tyrosine phosphorylation of a latent cytoplasmic transcription factor. Science 257 (5071), 809–813.
Schmidt, A., Schwerd, T., et al., 2009. 5′-Triphosphate RNA requires base-paired structures to activate antiviral signaling via RIG-I. Proc. Natl. Acad. Sci. U. S. A. 106 (29), 12067–12072.
Schoggins, J.W., Wilson, S.J., et al., 2011. A diverse range of gene products are effectors of the type I interferon antiviral response. Nature 472 (7344), 481–485.
Schroder, K., Hertzog, P.J., et al., 2004. Interferon-gamma: an overview of signals, mechanisms and functions. J. Leukoc. Biol. 75 (2), 163–189.
Schulz, O., Pichlmair, A., et al., 2010. Protein kinase R contributes to immunity against specific viruses by regulating interferon mRNA integrity. Cell Host Microbe 7 (5), 354–361.
Sen, A., Pruijssers, A.J., et al., 2011. The early interferon response to rotavirus is regulated by PKR and depends on MAVS/IPS-1, RIG-I, MDA-5, and IRF3. J. Virol. 85 (8), 3717–3732.
Seth, R.B., Sun, L., et al., 2005. Identification and characterization of MAVS, a mitochondrial antiviral signaling protein that activates NF-kappaB and IRF 3. Cell 122 (5), 669–682.
Shuai, K., Liu, B., 2003. Regulation of JAK-STAT signalling in the immune system. Nat. Rev. Immunol. 3 (11), 900–911.
Silvennoinen, O., Ihle, J.N., et al., 1993. Interferon-induced nuclear signalling by Jak protein tyrosine kinases. Nature 366 (6455), 583–585.
Smith, E.J., Marie, I., et al., 2001. IRF3 and IRF7 phosphorylation in virus-infected cells does not require double-stranded RNA-dependent protein kinase R or Ikappa B kinase but is blocked by Vaccinia virus E3L protein. J. Biol. Chem. 276 (12), 8951–8957.
Sommereyns, C., Paul, S., et al., 2008. IFN-lambda (IFN-lambda) is expressed in a tissue-dependent fashion and primarily acts on epithelial cells in vivo. PLoS Pathog. 4 (3), e1000017.
Stack, J., Haga, I.R., et al., 2005. Vaccinia virus protein A46R targets multiple Toll-like-interleukin-1 receptor adaptors and contributes to virulence. J. Exp. Med. 201 (6), 1007–1018.
Stetson, D.B., Medzhitov, R., 2006. Type I interferons in host defense. Immunity 25 (3), 373–381.
Stewart, M.E., Roy, P., 2010. Role of cellular caspases, nuclear factor-kappa B and interferon regulatory factors in Bluetongue virus infection and cell fate. Virol. J. 7, 362.
Symons, J.A., Alcami, A., et al., 1995. Vaccinia virus encodes a soluble type I interferon receptor of novel structure and broad species specificity. Cell 81 (4), 551–560.
Tabeta, K., Georgel, P., et al., 2004. Toll-like receptors 9 and 3 as essential components of innate immune defense against mouse cytomegalovirus infection. Proc. Natl. Acad. Sci. U. S. A. 101 (10), 3516–3521.
Takahasi, K., Kumeta, H., et al., 2009. Solution structures of cytosolic RNA sensor MDA5 and LGP2 C-terminal domains: identification of the RNA recognition loop in RIG-I-like receptors. J. Biol. Chem. 284 (26), 17465–17474.
Takahasi, K., Yoneyama, M., et al., 2008. Nonself RNA-sensing mechanism of RIG-I helicase and activation of antiviral immune responses. Mol. Cell 29 (4), 428–440.
Takeda, K., Akira, S., 2005. Toll-like receptors in innate immunity. Int. Immunol. 17 (1), 1–14.
Taylor, D.R., Shi, S.T., et al., 1999. Inhibition of the interferon-inducible protein kinase PKR by HCV E2 protein. Science 285 (5424), 107–110.
Taylor, J.L., O'Brien, W.J., 1985. Interferon production and sensitivity of rabbit corneal epithelial and stromal cells. Invest. Ophthalmol. Vis. Sci. 26 (11), 1502–1508.
Taylor, K.E., Mossman, K.L., 2012. Recent advances in understanding viral evasion of type I interferon. Immunology.
Thompson, A.J., Locarnini, S.A., 2007. Toll-like receptors, RIG-I-like RNA helicases and the antiviral innate immune response. Immunol. Cell Biol. 85 (6), 435–445.
Torpey, N., Maher, S.E., et al., 2004. Interferon alpha but not interleukin 12 activates STAT4 signaling in human vascular endothelial cells. J. Biol. Chem. 279 (25), 26789–26796.
Tough, D.F., 2012. Modulation of T-cell function by type I interferon. Immunol. Cell Biol. 90 (5), 492–497.
Tytell, A.A., Lampson, G.P., et al., 1967. Inducers of interferon and host resistance. 3. Double-stranded RNA from reovirus type 3 virions (reo 3-RNA). Proc. Natl. Acad. Sci. U. S. A. 58 (4), 1719–1722.
Uddin, S., Chamdin, A., et al., 1995. Interaction of the transcriptional activator Stat-2 with the type I interferon receptor. J. Biol. Chem. 270 (42), 24627–24630.
Ullman, A.J., Hearing, P., 2008. Cellular proteins PML and Daxx mediate an innate antiviral defense antagonized by the adenovirus E4 ORF3 protein. J. Virol. 82 (15), 7325–7335.
Ullman, A.J., Reich, N.C., et al., 2007. Adenovirus E4 ORF3 protein inhibits the interferon-mediated antiviral response. J. Virol. 81 (9), 4744–4752.
Unterholzner, L., 2013. The interferon response to intracellular DNA: why so many receptors? Immunobiology 218 (11), 1312–1321.
Unterholzner, L., Bowie, A.G., 2008. The interplay between viruses and innate immune signaling: recent insights and therapeutic opportunities. Biochem. Pharmacol. 75 (3), 589–602.
Unterstab, G., Ludwig, S., et al., 2005. Viral targeting of the interferon-{beta}-inducing Traf family member-associated NF-{kappa}B activator (TANK)-binding kinase-1. Proc. Natl. Acad. Sci. U. S. A. 102 (38), 13640–13645.
Venkataraman, T., Valdes, M., et al., 2007. Loss of DExD/H box RNA helicase LGP2 manifests disparate antiviral responses. J. Immunol. 178 (10), 6444–6455.
Vercammen, E., Staal, J., et al., 2008. Sensing of viral infection and activation of innate immunity by toll-like receptor 3. Clin. Microbiol. Rev. 21 (1), 13–25.
Visvanathan, K., Skinner, N.A., et al., 2007. Regulation of Toll-like receptor-2 expression in chronic hepatitis B by the precore protein. Hepatology 45 (1), 102–110.
Vitour, D., Dabo, S., et al., 2009. Polo-like kinase 1 (PLK1) regulates interferon (IFN) induction by MAVS. J. Biol. Chem. 284 (33), 21797–21809.
Vitour, D., Meurs, E.F., 2007. Regulation of interferon production by RIG-I and LGP2: a lesson in self-control. Sci. STKE 2007 (384), pe20.
Williams, B.R., 1999. PKR; a sentinel kinase for cellular stress. Oncogene 18 (45), 6112–6120.
Xu, L.G., Wang, Y.Y., et al., 2005. VISA is an adapter protein required for virus-triggered IFN-beta signaling. Mol. Cell 19 (6), 727–740.
Yamamoto, M., Sato, S., et al., 2003. Role of adaptor TRIF in the MyD88-independent toll-like receptor signaling pathway. Science 301 (5633), 640–643.
Yan, H., Krishnan, K., et al., 1996. Phosphorylated interferon-alpha receptor 1 subunit (IFNaR1) acts as a docking site for the latent form of the 113 kDa STAT2 protein. EMBO J. 15 (5), 1064–1074.
Yang, Y., Liang, Y., et al., 2007. Disruption of innate immunity due to mitochondrial targeting of a picornaviral protease precursor. Proc. Natl. Acad. Sci. U. S. A. 104 (17), 7253–7258.
Yang, Y.L., Reis, L.F., et al., 1995. Deficient signaling in mice devoid of double-stranded RNA-dependent protein kinase. EMBO J. 14 (24), 6095–6106.
Yoneyama, M., Kikuchi, M., et al., 2005. Shared and unique functions of the DExD/H-box helicases RIG-I, MDA5, and LGP2 in antiviral innate immunity. J. Immunol. 175 (5), 2851–2858.
Yoneyama, M., Kikuchi, M., et al., 2004. The RNA helicase RIG-I has an essential function in double-stranded RNA-induced innate antiviral responses. Nat. Immunol. 5 (7), 730–737.
Yue, Z., Shatkin, A.J., 1997. Double-stranded RNA-dependent protein kinase (PKR) is regulated by reovirus structural proteins. Virology 234 (2), 364–371.
Zhang, R., Jha, B.K., et al., 2013. Homologous 2′,5′-phosphodiesterases from disparate RNA viruses antagonize antiviral innate immunity. Proc. Natl. Acad. Sci. U. S. A. 110 (32), 13114–13119.
Zhang, S.Y., Jouanguy, E., et al., 2007. TLR3 deficiency in patients with herpes simplex encephalitis. Science 317 (5844), 1522–1527.
Zhang, Z., Kim, T., et al., 2011. DDX1, DDX21, and DHX36 helicases form a complex with the adaptor molecule TRIF to sense dsRNA in dendritic cells. Immunity 34 (6), 866–878.
Zhao, T., Yang, L., et al., 2007. The NEMO adaptor bridges the nuclear factor-kappaB and interferon regulatory factor signaling pathways. Nat. Immunol. 8 (6), 592–600.
Zhao, W., Lee, C., et al., 2008. A conserved IFN-alpha receptor tyrosine motif directs the biological response to type I IFNs. J. Immunol. 180 (8), 5483–5489.
Zucchini, N., Bessou, G., et al., 2008. Individual plasmacytoid dendritic cells are major contributors to the production of multiple innate cytokines in an organ-specific manner during viral infection. Int. Immunol. 20 (1), 45–56.
Zurney, J., Kobayashi, T., et al., 2009. Reovirus mu2 protein inhibits interferon signaling through a novel mechanism involving nuclear accumulation of interferon regulatory factor 9. J. Virol. 83 (5), 2178–2187.