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REVIEW

NLRP7: From inflammasome regulation to human disease

Jessica Carriere[1] | **Andrea Dorfleutner**[1,2] | **Christian Stehlik**[1,2,3]

[1]Department of Academic Pathology, Cedars Sinai Medical Center, Los Angeles, CA, USA

[2]Department of Biomedical Sciences, Cedars Sinai Medical Center, Los Angeles, CA, USA

[3]Samuel Oschin Comprehensive Cancer Institute, Cedars Sinai Medical Center, Los Angeles, CA, USA

Correspondence
Andrea Dorfleutner or Christian Stehlik, Department of Academic Pathology, Cedars Sinai Medical Center, Advanced Health Sciences Pavilion, 127 S. San Vicente Blvd, Los Angeles CA 90048, USA.
Emails: andrea.dorfleutner@cshs.org; christian.stehlik@cshs.org

Senior author: Andrea Dorfleutner and Christian Stehlik

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Abstract

Nucleotide-binding oligomerization domain (NOD) and leucine-rich repeat (LRR)-containing receptors or NOD-like receptors (NLRs) are cytosolic pattern recognition receptors, which sense conserved microbial patterns and host-derived danger signals to elicit innate immune responses. The activation of several prototypic NLRs, including NLR and pyrin domain (PYD) containing (NLRP) 1, NLRP3 and NLR and caspase recruitment domain (CARD) containing (NLRC) 4, results in the assembly of inflammasomes, which are large, cytoplasmic multiprotein signalling platforms responsible for the maturation and release of the pro-inflammatory cytokines IL-1β and IL-18, and for the induction of a specialized form of inflammatory cell death called pyroptosis. However, the function of other members of the NLR family, including NLRP7, are less well understood. NLRP7 has been linked to innate immune signalling, but its precise role is still controversial as it has been shown to positively and negatively affect inflammasome responses. Inflammasomes are essential for homeostasis and host defence, but inappropriate inflammasome responses due to hereditary mutations and somatic mosaicism in inflammasome components and defective regulation have been linked to a broad spectrum of human diseases. A compelling connection between *NLRP7* mutations and reproductive diseases, and in particular molar pregnancy, has been established. However, the molecular mechanisms by which NLRP7 mutations contribute to reproductive diseases are largely unknown. In this review, we focus on NLRP7 and discuss the current evidence of its role in inflammasome regulation and its implication in human reproductive diseases.

KEYWORDS

inflammasome, inflammation, innate receptors, macrophage, reproductive immunology

Abbreviations: AIM2, absent in melanoma 2; ALRs, AIM2-like receptors; ASC, apoptosis-associated speck-like protein containing a CARD; BBox, B-box-type zinc finger domain; B30·2, B30·2 and SPRY (SPla and the RYanodine Receptor); BIR, baculovirus inhibitor of apoptosis repeat; CAPS, cryopyrin-associated periodic syndrome; CARD, caspase recruitment domain; CC, coiled-coil; CHM, complete HM; COP, CARD-only protein; DAMPs, danger-associated molecular patterns; FGR, fetal growth restriction; FIIND, function to find domain; FRHM, familial recurrent HM; gDMR, germline differentially methylated region; GSDMD, gasdermin D; HM, hydatidiform mole; IFI16, interferon gamma-inducible protein 16; IL, interleukin; KSHV, Kaposi sarcoma-associated herpesvirus; LPS, lipopolysaccharide; LRR, leucine-rich repeat; MAMPs, microbial-associated molecular patterns; MDP, muramyl dipeptide; mtDNA, mitochondrial DNA; NAD, NACHT-associated domain; NBD, nucleotide-binding domain; NF-κB, nuclear factor kappa B; NLR, NOD-like, LRR-containing receptor; NLRA, NLR family acidic domain containing; NLRB, NLR family BIR domain containing; NLRC, NLR family CARD containing; NLRP, NLR family PYD containing; NOD, nucleotide-binding oligomerization domain; PAMPs, pathogen-associated molecular patterns; PHM, partial HM; POP, PYD-only protein; PRR, pattern recognition receptors; PYD, pyrin domain; ROS, reactive oxygen species; STAM, signal-transducing adaptor molecule; STAMBP, STAM-binding protein; STAND, signal-transducing ATPase with numerous domains; TLR, Toll-like receptor; TNF, tumour necrosis factor; TTSS, type III secretion system; UC, ulcerative colitis; YY1, ying yang 1; ZBTB16, zinc finger and BTB domain containing 16.

INTRODUCTION

The innate immune system maintains homeostasis and responds to tissue damage and pathogen infections [1]. The presence of microbes and their microbial-associated molecular patterns (MAMPs) and pathogen-associated molecular patterns (PAMPs) or host-derived danger signals or danger-associated molecular patterns (DAMPs) is sensed by a wide variety of specialized pattern recognition receptors (PRRs) expressed on the cell surface or in the cytoplasm of phagocytes and other cells [2]. Nucleotide-binding oligomerization domain (NOD)- and leucine-rich repeat (LRR)-containing receptors (NLRs) represent the largest cytosolic PRR family, which together with absent in melanoma 2 (AIM2)-like receptors (ALRs), and Pyrin, are involved in inflammasome assembly and activation [3]. The NLR gene superfamily encodes 22 human and 34 mouse proteins, which are characterized by an evolutionarily conserved tripartite domain architecture with (i) an N-terminal effector domain, (ii) a central nucleotide-binding domain (NBD) – comprised of the NACHT domain (NAIP, CIITA, HET-E and TP1) and NACHT-associated domain (NAD) – and (iii) a C-terminal autoregulatory LRR domain [4]. The five subfamilies are distinguished by the nature of the N-terminal effector domain, containing either an acidic domain (NLRA), a baculovirus inhibitor of apoptosis repeat (BIR) domain (NLRB), a CARD domain (NLRC), a pyrin domain (PYD; NLRP) or an N-terminal domain with no homology with other NLR members (NLRX) [4]. NLR activation triggers multiple innate immune signalling pathways to eradicate infections, promote wound healing and maintain tissue homeostasis, including MHC class I and II gene transcription, regulation of nuclear factor kappa B (NF-κB), mitogen-activated protein kinase, Akt, type I interferons, autophagy and developmental processes [5]. While most NLRs trigger a transcriptional response upon ligand sensing, several are capable of promoting the assembly of large multiprotein signalling platforms called inflammasomes (Figure 1).

Inflammasomes are responsible for the recruitment and proximity-mediated activation of the protease caspase-1, which is required for proteolytic maturation of the pro-inflammatory cytokines interleukin (IL)-1β and IL-18 [3,6]. Upon recognition of their respective ligands and/or activating signals, NLR sensors oligomerize and subsequently recruit the adaptor protein apoptosis-associated speck-like protein containing a CARD (ASC) through homotypic PYD-PYD or CARD-CARD interactions, depending on the effector domain of the NLR sensor. Inflammasome assembly progresses with the self-perpetuating polymerization of ASC prion-like filaments, and the exposed CARD of ASC then recruits caspase-1 through CARD-CARD interactions, resulting in clustering and proximity-mediated activation of caspase-1 [7]. In macrophages, inflammasome activation also induces an inflammatory form of cell death called pyroptosis through the caspase-1-mediated proteolytic cleavage of gasdermin D (GSDMD), which removes its inhibitory C-terminal domain and allows the N-terminal fragment to bind to phosphoinositides and cardiolipin, insert into membranes, oligomerize and form a lytic pore [8-10]. Noticeably, inflammasome assembly can be fine-tuned by small endogenous PYD-only proteins (POPs) and CARD-only proteins (COPs), which are incorporated into these structures by interfering with the PYD-PYD and CARD-CARD interactions [11-22].

To date, thirteen inflammasomes have been identified (Table 1) [23]. NLRP1 was the first NLR sensor identified to form an inflammasome and together with NLRC4, Pyrin, AIM2 and NLRP3 is among the best studied inflammasomes [24-29]. *NLRP3* has initially been identified as the causative gene for a group of autoinflammatory diseases referred to as Cryopyrin-associated periodic syndrome (CAPS) and has a unique position by sensing not only infections, but primarily sterile tissue damage, and its activation is now well defined and requires a two-step mechanism (Figure 1) [28,30]. The first signal ('priming') induces the transcription and various post-translational modifications of NLRP3 to prepare the sensor for activation while remaining inactive. The second and activating signal is unique for NLRP3, as it senses a diverse spectrum of signals culminating in an altered intracellular milieu caused by (i) changes in the ionic balance (potassium efflux, calcium influx), (ii) lysosomal and mitochondrial damage, (iii) pore forming toxins, cell death-induced pores and other cellular stress signals, and (iv) changes in the subcellular localization of NLRP3 inflammasome components [28]. In addition to these established inflammasome sensors, several other proteins have been recognized as inflammasome sensors, but their mechanisms are less well understood, including interferon gamma-inducible protein 16 (IFI16), NLRP2, NLRP6, NLRP9b, NLRP12, NLRC5, CARD8 and NLRP7(Table 1).

Advances have been made to better understand the role of inflammasomes during pathogen infection and sterile injury, and there is compelling evidence for their central role in maintaining tissue homeostasis and human health. Hence, a tight control and proper regulation of inflammasome activation is crucial [11,31]. The malfunction, dysregulation or the presence of mutations or allelic variations in genes encoding inflammasome sensors, including NLRP1, NLRP3, NLRP7, NLRP12, NLRC4, AIM2, CARD8 and Pyrin, results in either gain or loss of function, and is associated with various pathologies [31-35]. In this review, we focus on one of the less well-characterized NLRPs, NLRP7. We will discuss the controversial findings regarding its role in inflammasome signalling, and we will give an update on the current knowledge on its implication in human reproductive diseases.

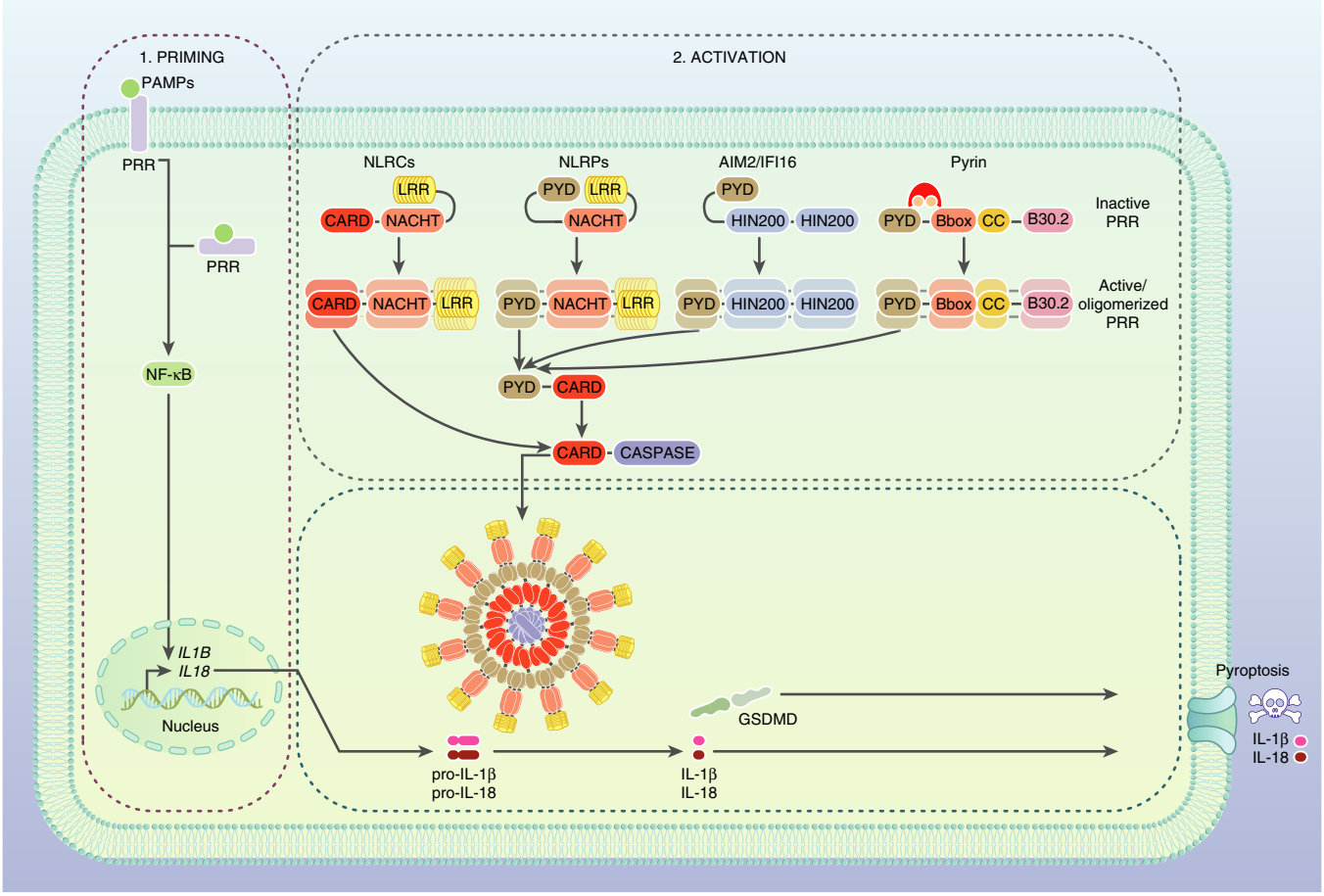
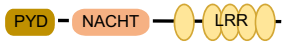
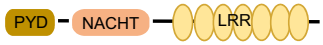
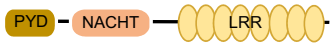
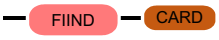
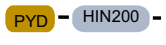


FIGURE 1 Inflammasome assembly. The activation of inflammasomes requires a 'priming' step resulting in activation of the transcription factor NF-κB and, consequently, the expression of pro-IL-1β and pro-IL-18 and inflammasome components. In addition, inflammasome-activating PRRs undergo various post-translational modifications to confer a ready state, as well as the subcellular redistribution of inflammasome components. Inflammasome activation is initiated upon ligand sensing, resulting in the release of the inactive conformation of the inflammasome sensors and sensor oligomerization. The active, oligomerized PRR recruits ASC usually through PYD-PYD interactions, which nucleates ASC polymerization and in turn caspase-1 polymerization through CARD-CARD interactions resulting in proximity-induced caspase-1 activation, while NLRC4 can also directly recruit caspase-1. Active caspase-1 proteolytically matures pro-IL-1β and pro-IL-18 into their bioactive form, which are then released. Caspase-1 also proteolytically cleaves GSDMD, which polymerizes in membranes to generate pores resulting in pyroptosis

IDENTIFICATION AND ORIGIN OF NLRP7

NLRP7 is located on the long arm of chromosome 19q13·4, and its first link to disease was recognized in 1999 when an *NLRP7* variant was identified in a woman with a history of recurrent molar pregnancy [36]. Taxonomic analyses suggest that the human *NLRP7* gene may have originally evolved in Simiiformes (*i.e.* Simians) nearly 43 million years ago and then evolved in Catarrhini, Hominoidea, Hominidae, Homininae and finally Homo sapiens. Further, phylogenetic analyses revealed that NLRP genes are conserved and that all NLRPs are monophyletic, except for *NLRP2*, which encompasses the primate-specific *NLRP7*, suggesting a gene duplication of the *NLRP2/7* ancestor in primates [37]. Significantly, this study also revealed a clade of NLRPs that are related to the reproductive system, including NLRP2, NLRP4, NLRP5, NLRP7, NLRP8, NLRP9, NLRP11, NLRP13 and NLRP14, which are all expressed in human oocytes and embryos [38]. Like all NLRP family members, NLRP7 is composed of an N-terminal PYD, a central NACHT domain followed by a NAD and several C-terminal LRRs (Table 1; Figure 2). Multiple isoforms have been described and predicted with varying LRR numbers due to alternative splicing (Figure 2). Whether the number of LRRs or the amino acid composition of these additional or lacking domains have consequences on NLRP7 function/pathway is a question largely remaining to be answered. However, the analysis of two NLRP7 isoforms with 5 and 7 LRRs revealed a higher pro-inflammatory activity of

TABLE 1 Inflammasomes and their activators. The protein structure and the ligands, NCBI Accession numbers and activating signals for known inflammasomes are shown

Sensor	Protein domains	Accession #	Activators/PAMPs/DAMPs
NLRP1		(NP_127497.1)	*Bacillus anthracis* lethal toxin [117-120] K^+ efflux [121] MDP [118, 119] dsRNA [122]
NLRP2		(NP_001167552.1)	ATP [111, 112]
NLRP3		(NP_004886.3)	Pathogen mRNA [123] Extracellular ATP [124-126] Crystals [b-amyloid [127], asbestos [128], silica [128, 129], MSU [130], cholesterol [131] mtDNA [132, 133] cAMP [134] K^+ efflux/Ca^{2+} influx [121, 135, 136] Lysosomal proteases [129] ROS [137] Ceramide [138] Cardiolipin [139] Aluminium salts [140]
NLRP6		(NP_612202.2)	Lipoteichoic acid [65]
NLRP7		(NP_631915.2)	Gram-positive bacteria [39] *Mycoplasma* spp. [39] Bacterial acylated lipopeptides [39] *Mycobacterium bovis* [63] K^+ efflux? Lysosomal proteases?
Nlrp9b		(NP_918947.2)	short double-stranded RNA [141]
NLRP12		(NP_001264055.1)	Acylated lipid A [142]
NLRC4		(NP_067032.3)	Flagellin [143-147] Bacterial TTSS [148] Gram-negative bacteria [147-150] K^+ efflux/Ca^{2+} influx [151] Lysosomal proteases [152]
NLRC5		(NP_001371879.1)	Unknown
CARD8		(NP_001171829.1)	Inhibition of dipeptidyl-peptidases [153, 154]
AIM2		(NP_004824.1)	cytosolic dsDNA [155-158]
IFI16		(NP_001193496.1)	KSHV DNA [159]
Pyrin		(NP_000234.1)	Loss of Rho GTPase-mediated PKN1/2 phosphorylation [160]

the longer isoform, suggesting a potential functional consequence based on the number of LRRs [39]. Functional consequences of alternative splicing of LRR-encoding exons have also been demonstrated for human NLRP3, where exons 5 and 7 are frequently skipped in human macrophages. The variant lacking exon 5 is inactive due to the absence of specific surface amino acids present in the missing LRRs, which impairs the essential interaction with NEK7 [40,41].

NLRP7 AND INFLAMMASOME SIGNALLING

NLRP7: a negative regulator of inflammasome signalling

NLRP7, also known as NALP7, PAN7, NOD12, CLR19·4, PYPAF3 and HYDM, has the necessary domain architecture

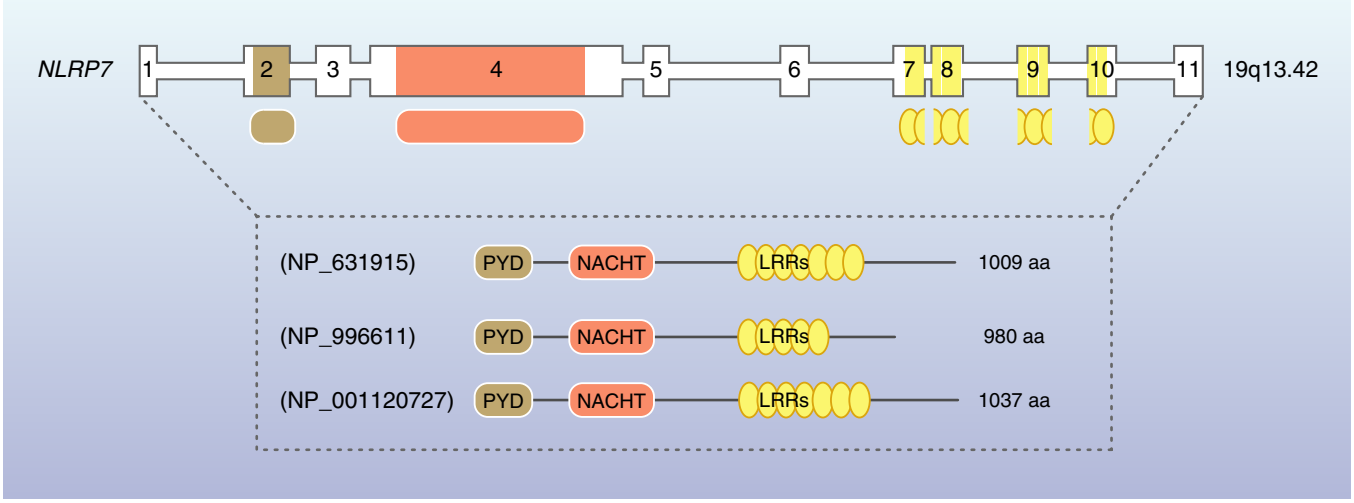


FIGURE 2 NLRP7 structure and splice forms. *NLRP7* exon organization of the 3 main domains: PYD, NACHT and LRRs (predicted by InterPro, https://www.ebi.ac.uk/interpro/) [113] and the main 3 annotated isoforms present in the NCBI protein database are indicated with their Accession numbers

to function as an inflammasome sensor. However, in 2002, a functional analysis based on the overexpression of several PYPAF family members, including NLRP7 (PYPAF3) in non-immune HEK293 cells, demonstrated the lack of NF-κB and caspase-1 activation [42]. Another overexpression study in HEK293 cells observed that NLRP7 inhibited NLRP3-mediated IL-1β release without impacting NF-κB activation and the authors hypothesized that this inhibition is a consequence of the interaction between NLRP7, pro-caspase-1 and pro-IL-1β, which could block pro-IL-1β maturation [43]. Reduced IL-1β release was also observed in some LPS-treated peripheral blood mononuclear cells isolated from hydatidiform mole (HM) patients carrying disease-associated NLRP7 variants (as discussed further below), compared with cells from healthy individuals. However, in most HM patients analysed, the presence of NLRP7 mutations was not associated with IL-1β levels [44]. Transient overexpression of wild-type or HM-associated mutant NLRP7 in HEK293 cells decreased the protein levels of both IL-1β precursor and mature form, suggesting effects on transcription/translation rather than maturation of IL-1β [45]. Moreover, this phenotype of reduced pro-IL-1β and IL-1β was abolished in HM-associated *NLRP7* mutants that result in a premature stop codon after the PYD and therefore generate a truncated protein, reminiscent of POPs [11,12,45]. A similar phenotype was observed with mutants harbouring deletions of each of the main functional domains, except the PYD [45]. Consequently, NLRP7 was viewed to have anti-inflammatory activity, reminiscent of other NLRs that can dampen pro-inflammatory signalling under certain conditions when expressed in HEK293 cells, including NLRP2, NLRP4, NLRP6, NLRP10, NLRP11, NLRP12, NLRX, NLRC3 and NLRC5 [46-60]. However, this anti-inflammatory activity requires further validation, as contradictory results have been reported in fibroblasts, THP-1 macrophages and primary macrophages, where NLRP7 did not inhibit NF-κB, NF-κB-mediated cytokine production or inflammatory responses [39, 42, 61-63].

NLRP7: a sensor for the inflammasome

Contrary to the above-described anti-inflammatory responses, evidence also supports the formation and activation of an NLRP7 inflammasome upon bacterial infection in human macrophages [39]. An RNAi screen identified NLRP7 as a component required for IL-1β release in response to *Mycoplasma* spp. infection in human macrophages. The NLRP7 inflammasome was also activated in response to infection with Gram-positive bacteria *Staphylococcus aureus* and *Listeria monocytogenes*, as silencing of NLRP7 prevented IL-1β and IL-18 release and phenocopied NLRP3 silencing. Furthermore, both NLRs restricted bacterial replication, although only silencing of NLRP3, but not NLRP7, reduced pyroptosis. In addition, even though both NLRs sensed Gram-positive bacteria, only NLRP7 specifically sensed bacterial di- and triacylated lipopeptides, reminiscent of Toll-like receptor (TLR) 2, and accordingly, TLR2 and NLRP7 were required for IL-1β release and restriction of replication of *L. monocytogenes* [39]. In contrast to earlier studies in HEK293 cells, silencing of NLRP7 did not affect NF-κB activation and therefore did not alter the transcription of pro-IL-1β, pro-IL-18 and other inflammatory cytokines, but supported an inflammasome role by binding to ASC and assembling with caspase-1 into a high molecular weight complex exhibiting caspase-1 activity [39]. The NLRP7 inflammasome was recently confirmed upon *Mycobacterium bovis* infection in THP-1 cells, as RNAi-mediated silencing of NLRP7 reduced pro-IL-1β processing and IL-1β release in response to

M. bovis infection. However, the authors also observed decreased *IL1B*, but upregulated *IL18* mRNA levels that were not observed in the earlier study, which will require further investigation. TNF and CCL3 mRNA levels were also reduced, which may be a feedback from autocrine IL-1β signalling [63]. NLRP7 belongs to the signal-transducing ATPase with numerous domain (STAND) family, and ATP binding and hydrolysis by the NLRP7 NACHT domain is required for assembly of the NLRP7 inflammasome in response to *S. aureus* infection [62]. Complex post-translational modifications regulate inflammasome sensors, and accordingly, ubiquitination of NLRP7 regulates inflammasome activity [61]. Lysine residues 288 and 290 are K-63-polyubiquitinated in resting macrophages, and the STAM-binding protein (STAMBP) deubiquitinates NLRP7 in response to TLR activation to prevent trafficking of NLRP7 to lysosomes and degradation. Accordingly, STAMBP silencing or inhibition impairs TLR-mediated increase in NLRP7 expression levels and dampens IL-1β release, providing evidence for ubiquitin-mediated NLRP7 inflammasome regulation [61]. The direct interaction between NLRP7 and STAM or STAMBP remains to be determined, as is the identity of the NLRP7 ubiquitin-conjugating enzyme. Further studies will also be necessary to elucidate additional factors controlling NLRP7 inflammasome activity, based on the complex interplay between transcriptional and post-translational modifications regulating NLRP3 and other NLRs [64]. Finally, while lipoteichoic acid (LTA) from Gram-positive bacteria binds to the LRR of NLRP6 and activates the NLRP6 inflammasome [65], it is still unclear whether there is any direct interaction between NLRP7 and acylated lipopeptides.

Taken together, these studies provided evidence for a role of NLRP7 in regulating inflammation, particularly in macrophages. While NLRP7 inflammasome activation has been demonstrated in response to bacterial infection in macrophages, its anti-inflammatory role has been largely observed by overexpression in non-immune cells. Therefore, elucidating the precise mechanism will require additional studies due to the conflicting activities reported for inflammasome activation and/or NF-κB regulation.

NLRP7 AND REPRODUCTIVE DISEASES

NLRP7 expression in placental development and early pregnancy

NLRP7 belongs to a reproduction-associated human gene cluster whose members are highly expressed in reproductive organs [66]. Interestingly, NLRP7 mutations in males do not have any adverse effects on reproductive outcomes, indicating that NLRP7 specifically regulates female reproduction [67,68]. While NLRP2 and NLRP7 expression decreases during oocyte maturation but then increases 3-5 days after fertilization in the blastocyst stage of embryonic development, the expression of other reproduction-associated NLRs decreases irreversibly. Hence, NLRP2 and NLRP7 seem to have a unique function [38]. NLRP7 expression is also very critical in the early stages of placenta development, and particularly in trophoblasts, which provide critical nutrients for the embryo and develop into a large part of the placenta, and in fibroblast-like endometrial stromal cells, which differentiate into epithelioid-like decidual cells during decidualization. Trophoblasts undergo a hypoxic period during placental development and express NLRP7 in a hypoxia-inducible manner. Furthermore, NLRP7 expression increases trophoblast proliferation but negatively regulates trophoblast differentiation [69]. Yet, another study confirms that impaired NLRP7 expression is associated with excessive trophoblast differentiation, which was dependent on bone morphogenetic protein 4 signalling [70,71]. The importance of controlled NLRP7 expression during placental development is further underscored by the fact that NLRP7 expression is elevated in the placenta of pregnancies suffering from fetal growth restriction (FGR). Interestingly, FGR pregnancies are often characterized by increased inflammation, hinting that the NLRP7 inflammasome could be involved. Indeed, expression of other NLRP7 inflammasome components, including ASC, cleaved caspase-1 and mature IL-1β, is also increased in FGR placentas, and circulating IL-1β, but not IL-18 levels, is significantly increased in the sera from FGR patients [69]. NLRP7 is also important during decidualization, a process that involves the crucial remodelling of endometrial tissue and cellular differentiation at the beginning of a pregnancy, which is necessary for embryo implantation and placental development. Here, NLRP7 is mainly expressed in the cytosol and nucleus of swollen decidualized human endometrial stromal cells, and in an *in vitro* decidualization model, NLRP7 is upregulated and translocated to the nucleus after decidualization. Accordingly, NLRP7 silencing impairs, but exogenous NLRP7 expression enhances, decidualization [72]. NLRP7 is also abundantly expressed in M2-polarized decidual macrophages from first-trimester pregnancies. While NLRP7 overexpression suppresses M1, it increases M2 macrophage marker expression, which may contribute to endometrial haemostasis and reproductive success [73]. Overall, there is growing evidence that NLRP7 plays an important role during early pregnancy by affecting oocyte maturation, endometrial remodelling and placental development.

NLRP7 and hydatidiform mole

Molar pregnancy or HM is an abnormal pregnancy characterized by trophoblast neoplasia, which leads to defective

embryonic development, reproductive wastage and/or spontaneous abortion. Complete HM (CHMs) and partial HM (PHMs) are the two subtypes distinguished by histopathological features and the genetic origin of the molar tissue [74]. The risk of recurrence is relatively low, except for women suffering from rare autosomal-recessive familial recurrent HM (FRHM) [75,76]. 5-25% of women with CHM and 2-3% of women with PHM may develop an aggressively spreading gestational choriocarcinoma [77]. In the last 15 years, ample evidence has been collected to convincingly link HM to NLRP7, as numerous *NLRP7* gene variants are clearly associated with reproduction and imprinting defects [44,66,78-83]. In particular, non-synonymous variants of *NLRP7* are linked to HM. The first variants of *NLRP7* located on the long arm of chromosome 19q13·4 were observed in women with a history of recurrent HM, and more than 200 sequence variants (https://infevers.umai-montpellier.fr) have now been reported in 48-80% of recurrent HM patients [36,67,68,84-88]. Mutations include substitutions, insertions, deletions and duplications, which affect most *NLRP7* exons and directly alter NLRP7 (Figure 3). A contributing factor may be the high frequency of Alu repeats in *NLRP7* introns, which are susceptible to recombination and are known to promote genomic instability.

While the link between *NLRP7* genomic variants and HM is compelling, the molecular mechanism and the functional consequences of these mutations are still poorly understood. An emerging hypothesis is that inflammation may contribute

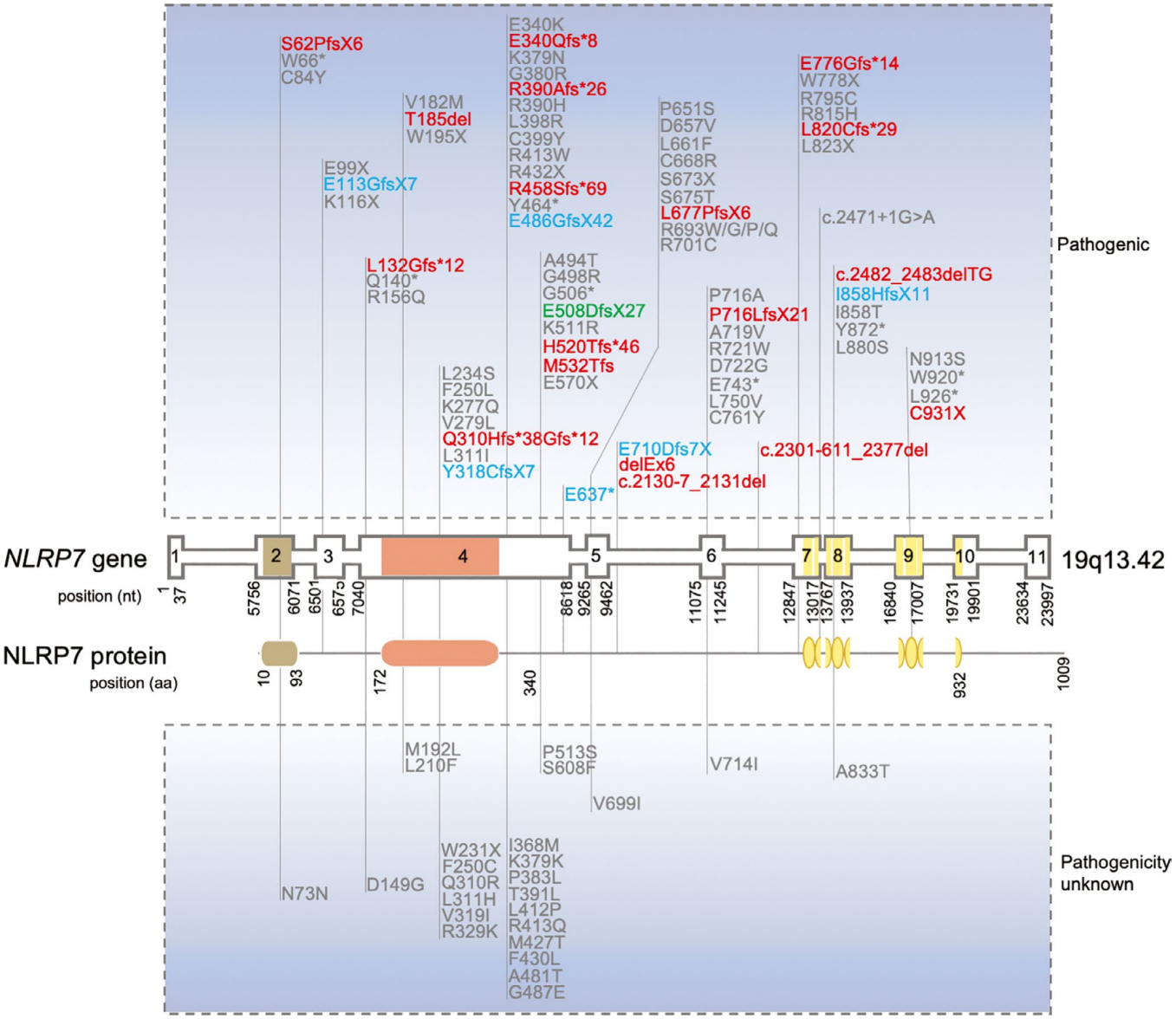


FIGURE 3 Mapping and pathogenicity of NLRP7 variants. Usual human genome variation society (HGVS) names of the variants impacting the NLRP7 protein sequence and their pathogenicity are shown and mapped along the *NLRP7* gene and NLRP7 protein. Grey, substitutions; red, deletions; blue, duplication; green, insertions. Database: Infevers: an online database for autoinflammatory mutations. https://infevers.umai-montpellier.fr/ Accessed June 2020 [88,114-116]

to HM pathogenesis, as placental tissues from HM patients exert histopathological features of chorioamnionitis (inflammation of the fetal membranes) and chorangiosis (tissue hypoxaemia leading to an excessive chorion neoangiogenesis and necrosis) [89,90]. The impact of *NLRP7* mutations on inflammatory signalling, the aetiology of HM, and whether the mutations confer gain or loss of function are still elusive. Attenuated NLRP7 may render the host more susceptible to infections by commensal or pathogenic microorganisms, and increasing infections in the lower reproductive tract and/or the loss of immune surveillance could lead to the development of moles. In contrast, hyperactive NLRP7 may exacerbate infection-triggered inflammation or promote sterile inflammation [39,62,63]. Supporting a gain-of-function phenotype, three independent HM-associated mutations displayed elevated basal activity and were hyperactive in promoting inflammasome responses, even in the absence of microbial triggers [39]. These mutations are reminiscent of the NACHT and LRR mutations present in other NLRs in patients with autoinflammatory diseases [31,33,34].

NLRP7 and genomic imprinting

Genomic imprinting is an epigenetic mechanism governing imprinted gene expression from only one of the parental alleles [91]. At the chromosomal level, both PHMs and CHMs have an androgenetic origin, which means that two copies of the paternal genome are transmitted, and this results in the overexpression of genes from the paternally inherited allele [92]. Indeed, 75% of HMs are CHMs and have a diploid androgenetic origin, while the 25% of PHMs have a triploid origin [89,90,93]. The importance of the androgenetic origin of HMs suggests genomic imprinting defects. Studies carried out on embryonic tissues from women carrying *NLRP7* variants revealed abnormal DNA methylation of imprinted germline differentially methylated regions (gDMRs), and abortion is linked to defective placental imprinting [85,86,94-96]. Accordingly, NLRP7 has been implicated in proper DNA methylation and interacts with DMRs in a methylation-dependent manner [97]. However, *NLRP7* variants were absent from women suffering from the imprinting disorder Beckwith–Wiedemann syndrome [98,99]. A relation between NLRP7, HM and the establishment and/or maintenance of maternal genomic imprinting is emerging, but not every patient suffering from FRHM harbours *NLRP7* variants, which is consistent with a multifactorial aetiology [100]. Truncating K homology domain containing 3 like mutations were observed in patients negative for *NLRP7* mutations in FRHM, and both proteins colocalized in lymphoblastoid cells [101,102], suggesting an intrinsic link between these two proteins, but any biological significance of this interaction is unknown. Studying NLRP7 binding partners involved in chromatin remodelling, and DNA methylation identified the transcription and chromatin-binding factor ying yang 1 (YY1) [97], which binds to imprinted gDMRs in a methylation-dependent manner to regulate imprinted gene expression. Reduced NLRP7 expression affected DNA methylation and accelerated trophoblast lineage differentiation, but the significance and impact of its interaction with YY1 on genomic imprinting is still unclear, as NLRP7 binding did not affect YY1 expression nor its DNA binding affinity in gDMRs [97]. NLRP7 also binds to the transcriptional repressor zinc finger and BTB domain containing 16 (ZBTB16), which is involved in chromatin remodelling and transcriptional silencing [103,104]. NLRP7 overexpression increased the cytoplasmic presence of ZBTB16, and NLRP7 may retain ZBTB16 in the cytoplasm, disrupt its chromatin remodelling function and promote the proliferation and differentiation of myeloid progenitors in HM.

Taken together, support exists for a role of NLRP7 in HM, decidualization and regulation of genomic imprinting, and one could envision distinct, cell type-specific NLRP7 roles in the nucleus and the cytosol, such as imprinting and inflammasome responses, respectively. Deciphering the NLRP7 interaction network and the biological significance of these interactions will help to understand the roles of NLRP7 in inflammasome regulation, genomic imprinting and molar pregnancy. Appropriate NLRP7 expression and NLRP7 inflammasome activity seem to be essential during the early stages of pregnancy. However, further investigation is required to establish how HM-associated *NLRP7* variants might affect NLRP7 inflammasome activity and lead to reproductive wastage.

CONCLUDING REMARKS

A role of *NLRP7* in the reproductive system and establishment of pregnancy is strongly supported, but mechanistic details remain elusive. However, NLRP7 expression requires a precise regulation during oocyte maturation and preimplantation of the embryo. NLRP7 is also crucial in decidualization, that is the preparation of the endometrium for and during pregnancy, and is involved in placental development by regulating trophoblast differentiation and proliferation. Excessive trophoblast proliferation is a hallmark of HM and an oncogenic role of NLRP7 in the pathological proliferation of germline cells in testicular seminomas [105], and NLRP7 expression in endometrial cancer tissues [106] has been proposed. Furthermore, the impact of *NLRP7* mutations observed in HM patients suffering from autoimmune disorders, such as Hashimoto disease, lupus, vitiligo and Crohn's disease, remains to be explored [89,107]. More recently, whole-exome sequencing identified a significant association of the low-frequency NLRP7 S361L variant with an increased risk of developing

ulcerative colitis (UC) [107]. However, any functional insights on how *NLRP7* variants contribute to immune system pathologies are still elusive and will require further investigations, but inflammasomes and NF-κB are crucial for immune homeostasis. While the link of *NLRP7* and *NLRP7* mutations to reproductive wastage is now established, the precise function of NLRP7 in pre-embryo and placental development and the causative link between HM-associated *NLRP7* variants and reproductive disorders remain to be discovered. Furthermore, the involvement of the NLRP7 inflammasome in these processes is still elusive. While the first studies proposed an anti-inflammatory role of NLRP7 in non-immune cells, further studies showed the assembly of a functional NLRP7 inflammasome particularly in response to pathogenic bacteria in macrophages. Growing evidence suggests a connection between inflammasome regulation in normal pregnancy and supports the possibility of inflammation-triggered reproductive wastage [108]. The NLRP7 inflammasome could also be involved in the inflammation of fetal membranes and in inflammation-mediated placental defects leading to fetal growth restriction, but further studies are required. However, the rarity of human oocytes and embryos available for research, as well as the absence of *NLRP7* in mice, complicates these studies. In addition to *in vitro* fertilization and maturation of oocytes, leveraging humanized mouse models could advance this field. Interestingly, *NLRP7* likely originated from a gene duplication of *NLRP2*, which also has a role in embryonic development. Like NLRP7, *NLRP2* mutations are also associated with recurrent miscarriage and *NLRP2* deficiency is associated with maternal age-associated fertility loss in mice [109,110]. Similar to NLRP7, NLRP2 has also been implicated in inflammasome activation and inhibition of NF-κB [54,111,112], but the precise mechanisms are elusive. Exciting opportunities therefore exist to further precisely define the role of NLRP7 and NLRP2, not only in inflammasome signalling during inflammatory responses but also during reproduction, and to determine the consequences for physiology and pathology.

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CONFLICT OF INTEREST

None.

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ORCID

Andrea Dorfleutner https://orcid.org/0000-0003-4406-2986
Christian Stehlik https://orcid.org/0000-0002-3782-2484

REFERENCES

1. Beutler B. Innate immunity: an overview. Mol Immunol. 2004;40:845–59.
2. Takeuchi O, Akira S. Pattern recognition receptors and inflammation. Cell. 2010;140:805–20.
3. Khare S, Luc N, Dorfleutner A, Stehlik C. Inflammasomes and their activation. Crit Rev Immunol. 2010;30:463–87.
4. Ting JP, Lovering RC, Alnemri ES, Bertin J, Boss JM, Davis BK, et al. The NLR gene family: a standard nomenclature. Immunity. 2008;28:285–7.
5. Motta V, Soares F, Sun T, Philpott DJ. NOD-like receptors: versatile cytosolic sentinels. Physiol Rev. 2015;95:149–78.
6. Schroder K, Tschopp J. The inflammasomes. Cell. 2010;140:821–32.
7. Kesavardhana S, Malireddi RKS, Kanneganti TD. Caspases in cell death, inflammation, and pyroptosis. Annu Rev Immunol. 2020;38:567–95.
8. Kayagaki N, Stowe IB, Lee BL, O'Rourke K, Anderson K, Warming S, et al. Caspase-11 cleaves gasdermin D for non-canonical inflammasome signalling. Nature. 2015;526:666–71.
9. Shi J, Gao W, Shao F. Pyroptosis: gasdermin-mediated programmed necrotic cell death. Trends Biochem Sci. 2017;42:245–54.
10. Shi J, Zhao Y, Wang K, Shi X, Wang Y, Huang H, et al. Cleavage of GSDMD by inflammatory caspases determines pyroptotic cell death. Nature. 2015;526:660–5.
11. Dorfleutner A, Chu L, Stehlik C. Inhibiting the inflammasome: one domain at a time. Immunol Rev. 2015;265:205–16.
12. Stehlik C, Dorfleutner A. COPs and POPs: modulators of inflammasome activity. J Immunol. 2007;179:7993–8.
13. Bedoya F, Sandler LL, Harton JA. Pyrin-only protein 2 modulates NF-kappaB and disrupts ASC:CLR interactions. J Immunol. 2007;178:3837–45.
14. de Almeida L, Khare S, Misharin AV, Patel R, Ratsimandresey RA, Wallin MC, et al. The PYRIN domain-only protein POP1 inhibits inflammasome assembly and ameliorates inflammatory disease. Immunity. 2015;43:264–76.
15. Dorfleutner A, Bryan NB, Talbott SJ, Funya KN, Rellick SL, Reed JC, et al. Cellular pyrin domain-only protein 2 is a candidate regulator of inflammasome activation. Infect Immun. 2007;75:1484–92.
16. Humke EW, Shriver SK, Starovasnik MA, Fairbrother WJ, Dixit VM. ICEBERG: a novel inhibitor of interleukin-1beta generation. Cell. 2000;103:99–111.
17. Khare S, Ratsimandresey RA, de Almeida L, Cuda CM, Rellick SL, Misharin AV, et al. The PYRIN domain-only protein POP3 inhibits ALR inflammasomes and regulates responses to infection with DNA viruses. Nat Immunol. 2014;15:343–53.
18. Lamkanfi M, Denecker G, Kalai M, D'Hondt K, Meeus A, Declercq W, et al. INCA, a novel human caspase recruitment domain protein that inhibits interleukin-1beta generation. J Biol Chem. 2001;279:51729–38.
19. Lee SH, Stehlik C, Reed JC. Cop, a caspase recruitment domain-containing protein and inhibitor of caspase-1 activation processing. J Biol Chem. 2001;276:34495–500.

20. Periasamy S, Porter KA, Atianand MK, Le H, Earley S, Duffy EB, et al. Pyrin-only protein 2 limits inflammation but improves protection against bacteria. Nat Commun. 2017;8:15564.
21. Ratsimandresey RA, Chu LH, Khare S, de Almeida L, Gangopadhyay A, Indramohan M, et al. The PYRIN domain-only protein POP2 inhibits inflammasome priming and activation. Nat Commun. 2017;8:15556.
22. Stehlik C, Krajewska M, Welsh K, Krajewski S, Godzik A, Reed JC. The PAAD/PYRIN-only protein POP1/ASC2 is a modulator of ASC-mediated NF-kB and pro-Caspase-1 regulation. Biochem J. 2003;373:101–13.
23. Malik A, Kanneganti TD. Inflammasome activation and assembly at a glance. J Cell Sci. 2017;130:3955–63.
24. Kumari P, Russo AJ, Shivcharan S, Rathinam VA. AIM2 in health and disease: Inflammasome and beyond. Immunol Rev. 2020;297:83–95.
25. Martinon F, Burns K, Tschopp J. The inflammasome: a molecular platform triggering activation of inflammatory caspases and processing of proIL-beta. Mol Cell. 2002;10:417–26.
26. Mitchell PS, Sandstrom A, Vance RE. The NLRP1 inflammasome: new mechanistic insights and unresolved mysteries. Curr Opin Immunol. 2019;60:37–45.
27. Schnappauf O, Chae JJ, Kastner DL, Aksentijevich I. The pyrin inflammasome in health and disease. Front Immunol. 2019;10:1745.
28. Swanson KV, Deng M, Ting JP. The NLRP3 inflammasome: molecular activation and regulation to therapeutics. Nat Rev Immunol. 2019;19:477–89.
29. Vance RE. The NAIP/NLRC4 inflammasomes. Curr Opin Immunol. 2015;32:84–9.
30. Hoffman HM, Mueller JL, Broide DH, Wanderer AA, Kolodner RD. Mutation of a new gene encoding a putative pyrin-like protein causes familial cold autoinflammatory syndrome and Muckle-Wells syndrome. Nat Genet. 2001;29:301–5.
31. Broderick L, De Nardo D, Franklin BS, Hoffman HM, Latz E. The inflammasomes and autoinflammatory syndromes. Annu Rev Pathol. 2015;10:395–424.
32. Guo H, Callaway JB, Ting JP. Inflammasomes: mechanism of action, role in disease, and therapeutics. Nat Med. 2015;21:677–87.
33. Harapas CR, Steiner A, Davidson S, Masters SL. An update on autoinflammatory diseases: inflammasomopathies. Curr Rheumatol Rep. 2018;20:40.
34. Manthiram K, Zhou Q, Aksentijevich I, Kastner DL. The monogenic autoinflammatory diseases define new pathways in human innate immunity and inflammation. Nat Immunol. 2017;18:832–42.
35. Taabazuing CY, Griswold AR, Bachovchin DA. The NLRP1 and CARD8 inflammasomes. Immunol Rev. 2020;297:13–25.
36. Moglabey YB, Kircheisen R, Seoud M, El Mogharbel N, Van den Veyver I, Slim R. Genetic mapping of a maternal locus responsible for familial hydatidiform moles. Hum Mol Genet. 1999;8:667–71.
37. Tian X, Pascal G, Monget P. Evolution and functional divergence of NLRP genes in mammalian reproductive systems. BMC Evol Biol. 2009;9:202.
38. Zhang P, Dixon M, Zucchelli M, Hambiliki F, Levkov L, Hovatta O, et al. Expression analysis of the NLRP gene family suggests a role in human preimplantation development. PLoS One. 2008;3:e2755.
39. Khare S, Dorfleutner A, Bryan NB, Yun C, Radian AD, de Almeida L, et al. An NLRP7-containing inflammasome mediates recognition of microbial lipopeptides in human macrophages. Immunity. 2012;36:464–76.
40. Shi H, Wang Y, Li X, Zhan X, Tang M, Fina M, et al. NLRP3 activation and mitosis are mutually exclusive events coordinated by NEK7, a new inflammasome component. Nat Immunol. 2016;17:250–8.
41. Hoss F, Mueller JL, Rojas Ringeling F, Rodriguez-Alcazar JF, Brinkschulte R, Seifert G, et al. Alternative splicing regulates stochastic NLRP3 activity. Nat Commun. 2019;10:3238.
42. Grenier JM, Wang L, Manji GA, Huang WJ, Al-Garawi A, Kelly R, et al. Functional screening of five PYPAF family members identifies PYPAF5 as a novel regulator of NF-kappaB and caspase-1. FEBS Lett. 2002;530(1–3):73–8.
43. Kinoshita T, Wang Y, Hasegawa M, Imamura R, Suda T. PYPAF3, a PYRIN-containing APAF-1-like protein, is a feedback regulator of caspase-1-dependent interleukin-1beta secretion. J Biol Chem. 2005;280:21720–5.
44. Messaed C, Chebaro W, Di Roberto RB, Rittore C, Cheung A, Arseneau J, et al. NLRP7 in the spectrum of reproductive wastage: rare non-synonymous variants confer genetic susceptibility to recurrent reproductive wastage. J Med Genet. 2011;48:540–8.
45. Messaed C, Akoury E, Djuric U, Zeng J, Saleh M, Gilbert L, et al. NLRP7, a nucleotide oligomerization domain-like receptor protein, is required for normal cytokine secretion and Co-localizes with Golgi and the Microtubule-organizing Center*. J Biol Chem. 2011;286:43313–23.
46. Allen IC, Moore CB, Schneider M, Lei Y, Davis BK, Scull MA, et al. NLRX1 protein attenuates inflammatory responses to infection by interfering with the RIG-I-MAVS and TRAF6-NF-κB signaling pathways. Immunity. 2011;34:854–65.
47. Anand PK, Kanneganti TD. NLRP6 in Infection and Inflammation. Microbes Infect. 2013;15(10-11):661–8.
48. Benko S, Magalhaes JG, Philpott DJ, Girardin SE. NLRC5 limits the activation of inflammatory pathways. J Immunol. 2010;185:1681–91.
49. Cui J, Li Y, Zhu L, Liu D, Songyang Z, Wang HY, et al. NLRP4 negatively regulates type I interferon signaling by targeting the kinase TBK1 for degradation via the ubiquitin ligase DTX4. Nat Immunol. 2012;13:387–95.
50. Cui J, Zhu L, Xia X, Wang HY, Legras X, Hong J, et al. NLRC5 negatively regulates the NF-kappaB and type I interferon signaling pathways. Cell. 2010;141:483–96.
51. Ellwanger K, Becker E, Kienes I, Sowa A, Postma Y, Cardona Gloria Y, et al. The NLR family pyrin domain-containing 11 protein contributes to the regulation of inflammatory signaling. J Biol Chem. 2018;293:2701–10.
52. Eren E, Berber M, Ozoren N. NLRC3 protein inhibits inflammation by disrupting NALP3 inflammasome assembly via competition with the adaptor protein ASC for pro-caspase-1 binding. J Biol Chem. 2017;292:12691–701.
53. Fiorentino L, Stehlik C, Oliveira V, Ariza ME, Godzik A, Reed JC. A novel PAAD-containing protein that modulates NF-kappa B induction by cytokines tumor necrosis factor-alpha and interleukin-1beta. J Biol Chem. 2002;277:35333–40.
54. Fontalba A, Gutierrez O, Fernandez-Luna JL. NLRP2, an inhibitor of the NF-kappaB pathway, is transcriptionally activated by NF-kappaB and exhibits a nonfunctional allelic variant. J Immunol. 2007;179:8519–24.
55. Gharagozloo M, Mahvelati TM, Imbeault E, Gris P, Zerif E, Bobbala D, et al. The nod-like receptor, Nlrp12, plays an anti-inflammatory role in experimental autoimmune encephalomyelitis. J Neuroinflammation. 2015;12:198.

56. Imamura R, Wang Y, Kinoshita T, Suzuki M, Noda T, Sagara J, et al. Anti-inflammatory activity of PYNOD and its mechanism in humans and mice. J Immunol. 2010;184:5874–84.
57. Qin Y, Su Z, Wu Y, Wu C, Jin S, Xie W, et al. NLRP11 disrupts MAVS signalosome to inhibit type I interferon signaling and virus-induced apoptosis. EMBO Rep. 2017;18:2160–71.
58. Schneider M, Zimmermann AG, Roberts RA, Zhang L, Swanson KV, Wen H, et al. The innate immune sensor NLRC3 attenuates Toll-like receptor signaling via modification of the signaling adaptor TRAF6 and transcription factor NF-κB. Nat Immunol. 2012;13:823–31.
59. Wu C, Su Z, Lin M, Ou J, Zhao W, Cui J, et al. NLRP11 attenuates Toll-like receptor signalling by targeting TRAF6 for degradation via the ubiquitin ligase RNF19A. Nat Commun. 2017;8:1977.
60. Zhang L, Mo J, Swanson KV, Wen H, Petrucelli A, Gregory SM, et al. NLRC3, a member of the NLR family of proteins, is a negative regulator of innate immune signaling induced by the DNA sensor STING. Immunity. 2014;40:329–41.
61. Bednash JS, Weathington N, Londino J, Rojas M, Gulick DL, Fort R, et al. Targeting the deubiquitinase STAMBP inhibits NALP7 inflammasome activity. Nat Commun. 2017;8:15203.
62. Radian AD, Khare S, Chu LH, Dorfleutner A, Stehlik C. ATP binding by NLRP7 is required for inflammasome activation in response to bacterial lipopeptides. Mol Immunol. 2015;67:294–302.
63. Zhou Y, Shah SZ, Yang L, Zhang Z, Zhou X, Zhao D. Virulent mycobacterium bovis beijing strain activates the NLRP7 inflammasome in THP-1 macrophages. PLoS One. 2016;11:e0152853.
64. Christgen S, Place DE, Kanneganti TD. Toward targeting inflammasomes: insights into their regulation and activation. Cell Res. 2020;30:315–27.
65. Hara H, Seregin SS, Yang D, Fukase K, Chamaillard M, Alnemri ES, et al. The NLRP6 inflammasome recognizes lipoteichoic acid and regulates gram-positive pathogen infection. Cell. 2018;175:1651–64 e14.
66. Van Gorp H, Kuchmiy A, Van Hauwermeiren F, Lamkanfi M. NOD-like receptors interfacing the immune and reproductive systems. Febs j. 2014;281:4568–82.
67. Qian J, Cheng Q, Murdoch S, Xu C, Jin F, Chebaro W, et al. The genetics of recurrent hydatidiform moles in China: correlations between NLRP7 mutations, molar genotypes and reproductive outcomes. Mol Hum Reprod. 2011;17:612–9.
68. Wang CM, Dixon PH, Decordova S, Hodges MD, Sebire NJ, Ozalp S, et al. Identification of 13 novel NLRP7 mutations in 20 families with recurrent hydatidiform mole; missense mutations cluster in the leucine-rich region. J Med Genet. 2009;46:569–75.
69. Abi Nahed R, Reynaud D, Borg AJ, Traboulsi W, Wetzel A, Sapin V, et al. NLRP7 is increased in human idiopathic fetal growth restriction and plays a critical role in trophoblast differentiation. J Mol Med (Berl). 2019;97:355–67.
70. Alici-Garipcan A, Ozcimen B, Suder I, Ulker V, Onder TT, Ozoren N. NLRP7 plays a functional role in regulating BMP4 signaling during differentiation of patient-derived trophoblasts. Cell Death Dis. 2020;11:658.
71. Li Yingchun, Parast MM. BMP4 regulation of human trophoblast development. Int J Dev Biol. 2014;58:239–246. http://dx.doi.org/10.1387/ijdb.130341mp.
72. Huang JY, Yu PH, Li YC, Kuo PL. NLRP7 contributes to *in vitro* decidualization of endometrial stromal cells. Reprod Biol Endocrinol. 2017;15:66.
73. Tsai P-Y, Chen K-R, Li Y-C, Kuo P-L. NLRP7 is involved in the differentiation of the decidual macrophages. Int J Mol Sci. 2019;20:5994.
74. Candelier JJ. The hydatidiform mole. Cell Adh Migr. 2016;10(1–2):226–35.
75. Sebire NJ, Fisher RA, Foskett M, Rees H, Seckl MJ, Newlands ES. Risk of recurrent hydatidiform mole and subsequent pregnancy outcome following complete or partial hydatidiform molar pregnancy. BJOG. 2003;110:22–6.
76. Sebire NJ, Savage PM, Seckl MJ, Fisher RA. Histopathological features of biparental complete hydatidiform moles in women with NLRP7 mutations. Placenta. 2013;34:50–6.
77. Slim R, Mehio A. The genetics of hydatidiform moles: new lights on an ancient disease. Clin Genet. 2007;71:25–34.
78. Caliebe A, Richter J, Ammerpohl O, Kanber D, Beygo J, Bens S, et al. A familial disorder of altered DNA-methylation. J Med Genet. 2014;51:407–12.
79. Docherty LE, Rezwan FI, Poole RL, Turner CL, Kivuva E, Maher ER, et al. Mutations in NLRP5 are associated with reproductive wastage and multilocus imprinting disorders in humans. Nat Commun. 2015;6:8086.
80. Landolsi H, Rittore C, Philibert L, Hmissa S, Gribaa M, Touitou I, et al. NLRP7 mutation analysis in sporadic hydatidiform moles in Tunisian patients: NLRP7 and sporadic mole. Arch Pathol Lab Med. 2012;136:646–51.
81. Landolsi H, Rittore C, Philibert L, Missaoui N, Hmissa S, Touitou I, et al. Screening for NLRP7 mutations in familial and sporadic recurrent hydatidiform moles: report of 2 Tunisian families. Int J Gynecol Pathol. 2011;30:348–53.
82. Meyer E, Lim D, Pasha S, Tee LJ, Rahman F, Yates JRW, et al. Germline mutation in NLRP2 (NALP2) in a familial imprinting disorder (Beckwith-Wiedemann Syndrome). PLoS Genet. 2009;5:e1000423.
83. Murdoch S, Djuric U, Mazhar B, Seoud M, Khan R, Kuick R, et al. Mutations in NALP7 cause recurrent hydatidiform moles and reproductive wastage in humans. Nat Genet. 2006;38:300–2.
84. Estrada H, Buentello B, Zenteno JC, Fiszman R, Aguinaga M. The p. L750V mutation in the NLRP7 gene is frequent in Mexican patients with recurrent molar pregnancies and is not associated with recurrent pregnancy loss. Prenat Diagn. 2013;33:205–8.
85. Hayward BE, De Vos M, Talati N, Abdollahi MR, Taylor GR, Meyer E, et al. Genetic and epigenetic analysis of recurrent hydatidiform mole. Hum Mutat. 2009;30:E629–E639.
86. Kou YC, Shao L, Peng HH, Rosetta R, del Gaudio D, Wagner AF, et al. A recurrent intragenic genomic duplication, other novel mutations in NLRP7 and imprinting defects in recurrent biparental hydatidiform moles. Mol Hum Reprod. 2008;14:33–40.
87. Slim R, Bagga R, Chebaro W, Srinivasan R, Agarwal N. A strong founder effect for two NLRP7 mutations in the Indian population: an intriguing observation. Clin Genet. 2009;76:292–5.
88. Touitou I, Lesage S, McDermott M, Cuisset L, Hoffman H, Dode C, et al. Infevers: an evolving mutation database for auto-inflammatory syndromes. Hum Mutat. 2004;24:194–8.
89. Deveault C, Qian JH, Chebaro W, Ao A, Gilbert L, Mehio A, et al. NLRP7 mutations in women with diploid androgenetic and triploid moles: a proposed mechanism for mole formation. Hum Mol Genet. 2009;18:888–97.
90. Williams D, Hodgetts V, Gupta J. Recurrent hydatidiform moles. Eur J Obstet Gynecol Reprod Biol. 2010;150:3–7.

91. Tucci V, Isles AR, Kelsey G, Ferguson-Smith AC. Genomic imprinting and physiological processes in mammals. Cell. 2019;176:952–65.
92. Fallahian M, Sebire NJ, Savage PM, Seckl MJ, Fisher RA. Mutations in NLRP7 and KHDC3L confer a complete hydatidiform mole phenotype on digynic triploid conceptions. Hum Mutat. 2013;34:301–8.
93. Kajii T, Ohama K. Androgenetic origin of hydatidiform mole. Nature. 1977;268:633–4.
94. Djuric U, El-Maarri O, Lamb B, Kuick R, Seoud M, Coullin P, et al. Familial molar tissues due to mutations in the inflammatory gene, NALP7, have normal postzygotic DNA methylation. Hum Genet. 2006;120:390–5.
95. Ito Y, Maehara K, Kaneki E, Matsuoka K, Sugahara N, Miyata T, et al. Novel nonsense mutation in the NLRP7 gene associated with recurrent hydatidiform mole. Gynecol Obstet Invest. 2016;81:353–8.
96. Sanchez-Delgado M, Martin-Trujillo A, Tayama C, Vidal E, Esteller M, Iglesias-Platas I, et al. Absence of maternal methylation in biparental hydatidiform moles from women with NLRP7 maternal-effect mutations reveals widespread placenta-specific imprinting. PLoS Genet. 2015;11:e1005644.
97. Mahadevan S, Wen S, Wan YW, Peng HH, Otta S, Liu Z, et al. NLRP7 affects trophoblast lineage differentiation, binds to overexpressed YY1 and alters CpG methylation. Hum Mol Genet. 2014;23:706–16.
98. Boonen SE, Porksen S, Mackay DJ, Oestergaard E, Olsen B, Brondum-Nielsen K, et al. Clinical characterisation of the multiple maternal hypomethylation syndrome in siblings. Eur J Hum Genet. 2008;16:453–61.
99. Soejima H, Higashimoto K. Epigenetic and genetic alterations of the imprinting disorder Beckwith-Wiedemann syndrome and related disorders. J Hum Genet. 2013;58:402–9.
100. Dixon PH, Trongwongsa P, Abu-Hayyah S, Ng SH, Akbar SA, Khawaja NP, et al. Mutations in NLRP7 are associated with diploid biparental hydatidiform moles, but not androgenetic complete moles. J Med Genet. 2012;49:206–11.
101. Parry DA, Logan CV, Hayward BE, Shires M, Landolsi H, Diggle C, et al. Mutations causing familial biparental hydatidiform mole implicate c6orf221 as a possible regulator of genomic imprinting in the human oocyte. Am J Hum Genet. 2011;89:451–8.
102. Reddy R, Akoury E, Phuong Nguyen NM, Abdul-Rahman OA, Dery C, Gupta N, et al. Report of four new patients with protein-truncating mutations in C6orf221/KHDC3L and colocalization with NLRP7. Eur J Hum Genet. 2013;21:957–64.
103. Singer H, Biswas A, Nuesgen N, Oldenburg J, El-Maarri O. NLRP7, involved in hydatidiform molar pregnancy (HYDM1), interacts with the transcriptional repressor ZBTB16. PLoS One. 2015;10:e0130416.
104. Barna M, Merghoub T, Costoya JA, Ruggero D, Branford M, Bergia A, et al. Plzf mediates transcriptional repression of HoxD gene expression through chromatin remodeling. Dev Cell. 2002;3:499–510.
105. Okada K, Hirota E, Mizutani Y, Fujioka T, Shuin T, Miki T, et al. Oncogenic role of NALP7 in testicular seminomas. Cancer Sci. 2004;95:949–54.
106. Ohno S, Kinoshita T, Ohno Y, Minamoto T, Suzuki N, Inoue M, et al. Expression of NLRP7 (PYPAF3, NALP7) protein in endometrial cancer tissues. Anticancer Res. 2008;28(4C):2493–7.
107. Onoufriadis A, Stone K, Katsiamides A, Amar A, Omar Y, de Lange KM, et al. Exome Sequencing and Genotyping Identify a Rare Variant in NLRP7 Gene Associated With Ulcerative Colitis. J Crohns Colitis. 2018;12:321–6.
108. Gomez-Lopez N, Motomura K, Miller D, Garcia-Flores V, Galaz J, Romero R. Inflammasomes: Their Role in Normal and Complicated Pregnancies. J Immunol. 2019;203:2757–69.
109. Huang JY, Su M, Lin SH, Kuo PL. A genetic association study of NLRP2 and NLRP7 genes in idiopathic recurrent miscarriage. Hum Reprod. 2013;28:1127–34.
110. Kuchmiy AA, D'Hont J, Hochepied T, Lamkanfi M. NLRP2 controls age-associated maternal fertility. J Exp Med. 2016;213:2851–60.
111. Matsuoka Y, Yamashita A, Matsuda M, Kawai K, Sawa T, Amaya F. NLRP2 inflammasome in dorsal root ganglion as a novel molecular platform that produces inflammatory pain hypersensitivity. Pain. 2019;160:2149–60.
112. Minkiewicz J, de Rivero Vaccari JP, Keane RW. Human astrocytes express a novel NLRP2 inflammasome. Glia. 2013;61:1113–21.
113. Blum M, Chang HY, Chuguransky S, Grego T, Kandasaamy S, Mitchell A, et al. The InterPro protein families and domains database: 20 years on. Nucleic Acids Res. 2021;49(D1):D344–D354.
114. Milhavet F, Cuisset L, Hoffman HM, Slim R, El-Shanti H, Aksentijevich I, et al. The infevers autoinflammatory mutation online registry: update with new genes and functions. Hum Mutat. 2008;29:803–8.
115. Sarrauste de Menthiere C, Terriere S, Pugnere D, Ruiz M, Demaille J, Touitou I. INFEVERS: the Registry for FMF and hereditary inflammatory disorders mutations. Nucleic Acids Res. 2003;31:282–5.
116. Van Gijn ME, Ceccherini I, Shinar Y, Carbo EC, Slofstra M, Arostegui JI, et al. New workflow for classification of genetic variants' pathogenicity applied to hereditary recurrent fevers by the International Study Group for Systemic Autoinflammatory Diseases (INSAID). J Med Genet. 2018;55:530–7.
117. Boyden ED, Dietrich WF. Nalp1b controls mouse macrophage susceptibility to anthrax lethal toxin. Nat Genet. 2006;38:240–4.
118. Bruey JM, Bruey-Sedano N, Luciano F, Zhai D, Balpai R, Xu C, et al. Bcl-2 and Bcl-XL regulate proinflammatory caspase-1 activation by interaction with NALP1. Cell. 2007;129:45–56.
119. Hsu LC, Ali SR, McGillivray S, Tseng PH, Mariathasan S, Humke EW, et al. A NOD2-NALP1 complex mediates caspase-1-dependent IL-1beta secretion in response to Bacillus anthracis infection and muramyl dipeptide. Proc Natl Acad Sci U S A. 2008;105:7803–8.
120. Wickliffe KE, Leppla SH, Moayeri M. Anthrax lethal toxin-induced inflammasome formation and caspase-1 activation are late events dependent on ion fluxes and the proteasome. Cell Microbiol. 2008;10:332–43.
121. Petrilli V, Papin S, Dostert C, Mayor A, Martinon F, Tschopp J. Activation of the NALP3 inflammasome is triggered by low intracellular potassium concentration. Cell Death Differ. 2007;14:1583–9.
122. Bauernfried S, Scherr MJ, Pichlmair A, Duderstadt KE, Hornung V. Human NLRP1 is a sensor for double-stranded RNA. Science. 2021;371:eabd0811.
123. Sha W, Mitoma H, Hanabuchi S, Bao M, Weng L, Sugimoto N, et al. Human NLRP3 inflammasome senses multiple types of bacterial RNAs. Proc Natl Acad Sci U S A. 2014;111:16059–64.

124. Di Virgilio F, Dal Ben D, Sarti AC, Giuliani AL, Falzoni S. The P2X7 Receptor in Infection and Inflammation. Immunity. 2017;47:15–31.
125. Ferrari D, Pizzirani C, Adinolfi E, Lemoli RM, Curti A, Idzko M, et al. The P2X7 receptor: a key player in IL-1 processing and release. J Immunol. 2006;176:3877–83.
126. Idzko M, Ferrari D, Eltzschig HK. Nucleotide signalling during inflammation. Nature. 2014;509:310–7.
127. Halle A, Hornung V, Petzold GC, Stewart CR, Monks BG, Reinheckel T, et al. The NALP3 inflammasome is involved in the innate immune response to amyloid-beta. Nat Immunol. 2008;9:857–65.
128. Dostert C, Petrilli V, Van Bruggen R, Steele C, Mossman BT, Tschopp J. Innate immune activation through Nalp3 inflammasome sensing of asbestos and silica. Science. 2008;320:674–7.
129. Hornung V, Bauernfeind F, Halle A, Samstad EO, Kono H, Rock KL, et al. Silica crystals and aluminum salts activate the NALP3 inflammasome through phagosomal destabilization. Nat Immunol. 2008;9:847–56.
130. Martinon F, Petrilli V, Mayor A, Tardivel A, Tschopp J. Gout-associated uric acid crystals activate the NALP3 inflammasome. Nature. 2006;440:237–41.
131. Duewell P, Kono H, Rayner KJ, Sirois CM, Vladimer G, Bauernfeind FG, et al. NLRP3 inflammasomes are required for atherogenesis and activated by cholesterol crystals. Nature. 2010;464:1357–61.
132. Shimada K, Crother TR, Karlin J, Dagvadorj J, Chiba N, Chen S, et al. Oxidized mitochondrial DNA activates the NLRP3 inflammasome during apoptosis. Immunity. 2012;36:401–14.
133. Zhong Z, Liang S, Sanchez-Lopez E, He F, Shalapour S, Lin XJ, et al. New mitochondrial DNA synthesis enables NLRP3 inflammasome activation. Nature. 2018;560:198–203.
134. Lee GS, Subramanian N, Kim AI, Aksentijevich I, Goldbach-Mansky R, Sacks DB, et al. The calcium-sensing receptor regulates the NLRP3 inflammasome through Ca2+ and cAMP. Nature. 2012;492:123–7.
135. Muñoz-Planillo R, Kuffa P, Martínez-Colón G, Smith BL, Rajendiran TM, Núñez G. K^+ efflux is the common trigger of NLRP3 inflammasome activation by bacterial toxins and particulate matter. Immunity. 2013;38:1142–53.
136. Murakami T, Ockinger J, Yu J, Byles V, McColl A, Hofer AM, et al. Critical role for calcium mobilization in activation of the NLRP3 inflammasome. Proc Natl Acad Sci U S A. 2012;109:11282–7.
137. Zhou R, Yazdi AS, Menu P, Tschopp J. A role for mitochondria in NLRP3 inflammasome activation. Nature. 2011;469:221–5.
138. Scheiblich H, Schlutter A, Golenbock DT, Latz E, Martinez-Martinez P, Heneka MT. Activation of the NLRP3 inflammasome in microglia: the role of ceramide. J Neurochem. 2017;143:534–50.
139. Iyer SS, He Q, Janczy JR, Elliott EI, Zhong Z, Olivier AK, et al. Mitochondrial cardiolipin is required for Nlrp3 inflammasome activation. Immunity. 2013;39:311–23.
140. Eisenbarth SC, Colegio OR, O'Connor W, Sutterwala FS, Flavell RA. Crucial role for the Nalp3 inflammasome in the immunostimulatory properties of aluminium adjuvants. Nature. 2008;453:1122–6.
141. Zhu S, Ding S, Wang P, Wei Z, Pan W, Palm NW, et al. Nlrp9b inflammasome restricts rotavirus infection in intestinal epithelial cells. Nature. 2017;546:667–70.
142. Vladimer GI, Weng D, Paquette SW, Vanaja SK, Rathinam VA, Aune MH, et al. The NLRP12 inflammasome recognizes Yersinia pestis. Immunity. 2012;37:96–107.
143. Amer A, Franchi L, Kanneganti TD, Body-Malapel M, Ozoren N, Brady G, et al. Regulation of Legionella phagosome maturation and infection through flagellin and host Ipaf. J Biol Chem. 2006;281:35217–23.
144. Franchi L, Amer A, Body-Malapel M, Kanneganti TD, Ozoren N, Jagirdar R, et al. Cytosolic flagellin requires Ipaf for activation of caspase-1 and interleukin 1beta in salmonella-infected macrophages. Nat Immunol. 2006;7:576–82.
145. Miao EA, Alpuche-Aranda CM, Dors M, Clark AE, Bader MW, Miller SI, et al. Cytoplasmic flagellin activates caspase-1 and secretion of interleukin 1beta via Ipaf. Nat Immunol. 2006;7:569–75.
146. Ren T, Zamboni DS, Roy CR, Dietrich WF, Vance RE. Flagellin-deficient Legionella mutants evade caspase-1- and Naip5-mediated macrophage immunity. PLoS Pathog. 2006;2:e18.
147. Sutterwala FS, Mijares LA, Li L, Ogura Y, Kazmierczak BI, Flavell RA. Immune recognition of Pseudomonas aeruginosa mediated by the IPAF/NLRC4 inflammasome. J Exp Med. 2007;204:3235–45.
148. Miao EA, Mao DP, Yudkovsky N, Bonneau R, Lorang CG, Warren SE, et al. Innate immune detection of the type III secretion apparatus through the NLRC4 inflammasome. Proc Natl Acad Sci U S A. 2010;107:3076–80.
149. Mariathasan S, Newton K, Monack DM, Vucic D, French DM, Lee WP, et al. Differential activation of the inflammasome by caspase-1 adaptors ASC and Ipaf. Nature. 2004;430:213–8.
150. Vinzing M, Eitel J, Lippmann J, Hocke AC, Zahlten J, Slevogt H, et al. NAIP and Ipaf control Legionella pneumophila replication in human cells. J Immunol. 2008;180:6808–15.
151. Freeman L, Guo H, David CN, Brickey WJ, Jha S, Ting JP. NLR members NLRC4 and NLRP3 mediate sterile inflammasome activation in microglia and astrocytes. J Exp Med. 2017;214:1351–70.
152. Lage SL, Buzzo CL, Amaral EP, Matteucci KC, Massis LM, Icimoto MY, et al. Cytosolic flagellin-induced lysosomal pathway regulates inflammasome-dependent and -independent macrophage responses. Proc Natl Acad Sci U S A. 2013;110:E3321–E3330.
153. Johnson DC, Taabazuing CY, Okondo MC, Chui AJ, Rao SD, Brown FC, et al. DPP8/DPP9 inhibitor-induced pyroptosis for treatment of acute myeloid leukemia. Nat Med. 2018;24:1151–6.
154. Linder A, Bauernfried S, Cheng Y, Albanese M, Jung C, Keppler OT, et al. CARD8 inflammasome activation triggers pyroptosis in human T cells. Embo J. 2020;39:e105071.
155. Burckstummer T, Baumann C, Bluml S, Dixit E, Durnberger G, Jahn H, et al. An orthogonal proteomic-genomic screen identifies AIM2 as a cytoplasmic DNA sensor for the inflammasome. Nat Immunol. 2009;10:266–72.
156. Fernandes-Alnemri T, Yu JW, Datta P, Wu J, Alnemri ES. AIM2 activates the inflammasome and cell death in response to cytoplasmic DNA. Nature. 2009;458:509–13.
157. Hornung V, Ablasser A, Charrel-Dennis M, Bauernfeind F, Horvath G, Caffrey DR, et al. AIM2 recognizes cytosolic dsDNA and forms a caspase-1-activating inflammasome with ASC. Nature. 2009;458:514–8.
158. Roberts TL, Idris A, Dunn JA, Kelly GM, Burnton CM, Hodgson S, et al. HIN-200 proteins regulate caspase activation in response to foreign cytoplasmic DNA. Science. 2009;323:1057–60.

159. Kerur N, Veettil MV, Sharma-Walia N, Bottero V, Sadagopan S, Otageri P, et al. IFI16 acts as a nuclear pathogen sensor to induce the inflammasome in response to Kaposi Sarcoma-associated herpesvirus infection. Cell Host Microbe. 2011;9:363–75.
160. Xu H, Yang J, Gao W, Li L, Li P, Zhang L, et al. Innate immune sensing of bacterial modifications of Rho GTPases by the Pyrin inflammasome. Nature. 2014;513:237–41.

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