Journal of Autoimmunity xxx (xxxx) xxxx

Contents lists available at ScienceDirect

Journal of Autoimmunity

journal homepage: www.elsevier.com/locate/jautimm

# Inflammasomes and autoimmune and rheumatic diseases: A comprehensive review☆

Jae Il Shin[a,b,*], Keum Hwa Lee[a,b], Yo Han Joo[c], Jiwon M. Lee[d], Jaewook Jeon[c], Hee Jae Jung[c], Minkyue Shin[c], Seobum Cho[c], Tae Hwan Kim[c], Seonghyuk Park[c], Bong Yeol Jeon[c], Hyunwoo Jeong[c], Kangto Lee[c], Kyutae Kang[c], Myungsuk Oh[c], Hansang Lee[c], Seungchul Lee[c], Yeji Kwon[c], Geun ho Oh[c], Andreas Kronbichler[e]

[a] Department of Pediatrics, Yonsei University College of Medicine, Seoul, South Korea
[b] Division of Pediatric Nephrology, Severance Children's Hospital, Seoul, South Korea
[c] Yonsei University College of Medicine, Seoul, South Korea
[d] Department of Pediatrics, Chungnam National University Hospital, Daejeon, South Korea
[e] Department of Internal Medicine IV, Medical University Innsbruck, Innsbruck, Austria

ARTICLE INFO

*Keywords:*
Inflammasome
Autoimmunity
Caspase-1
Interleukin-1β

ABSTRACT

Inflammasomes are a multi-protein platform forming a part of the innate immune system. Inflammasomes are at standby status and can be activated when needed. Inflammasome activation is an important mechanism for the production of active interleukin (IL)-1β and IL-18, which have important roles to instruct adaptive immunity. Active forms of inflammasomes trigger a series of inflammatory cascades and lead to the differentiation and polarization of naïve T cells and secretion of various cytokines, which can induce various kinds of autoimmune and rheumatic diseases such as systemic lupus erythematosus (SLE), rheumatoid arthritis (RA), gout, Sjögren's syndrome, Behçet's disease, anti-neutrophil cytoplasmic antibody (ANCA)-associated vasculitis and IgA vasculitis (former Henoch-Schönlein purpura ). In this review, we summarize studies published on inflammasomes and review their roles in various autoimmune diseases. Understanding of the role of inflammasomes may facilitate the diagnosis of autoimmune diseases and the development of tailored therapies in the future.

## 1. Introduction

An inflammasome, first reported by Tschopp and his team in the year 2002, is a multiprotein platform which is formed in response to various physiologic and pathogenic stimuli [1]. Activation of inflammasome is an essential component of the innate immune response and is important for the clearance of pathogens or damaged cells. It should be tightly regulated to appropriately defend against the pathogens and to prevent the pathological host damage [2]. However, excess activation of inflammasome can also drive autoimmune diseases ranging from rheumatoid arthritis (RA), juvenile idiopathic arthritis (JIA), systemic lupus erythematosus (SLE) and metabolic disorders such as type 1 and 2 diabetes, and therefore, it is important to understand this process in physiological and pathological contexts [2–4].

Inflammasomes have critical roles in both innate and adaptive immunity. An immune response begins with sensing of conserved molecular patterns derived from pathogens and host tissue damage, which are called pathogen associated molecular patterns (PAMPs) and damage (danger) associated molecular patterns (DAMPs), respectively, and the innate immune system activates the adaptive immune system primarily through the actions of antigen presenting cells (APCs). Inflammasomes and interleukin (IL)-1 family cytokines have been implicated in the generation of adaptive immunity via T and B lymphocytes [2,4].

PAMPs, such as cell wall components and nucleic acids of microbial pathogens, are recognized by pattern recognition receptors (PRRs) which can be divided into membrane-bound PRRs (eg. toll like receptors (TLRs) and c-type lectin receptors (CLRs)) and cytoplasmic PRRs (eg. NOD-like receptors (NLRs)). Interaction of TLRs with their specific PAMPs is mediated through either MyD88-dependent pathway and triggers the signaling through nuclear factor-kappaB (NF-κB) and the mitogen-activated protein (MAP) kinase pathway and therefore the

☆ The authors disclose no financial or non-financial conflicts of interest.

* Corresponding author. 50 Yonsei-ro, Seodaemun-gu, C.P.O. Box 8044, Department of Pediatrics, Yonsei University College of Medicine, Seoul, 03722, South Korea.
*E-mail address:* shinji@yuhs.ac (J.I. Shin).

https://doi.org/10.1016/j.jaut.2019.06.010
Received 12 April 2019; Received in revised form 27 June 2019; Accepted 29 June 2019
0896-8411/ © 2019 Elsevier Ltd. All rights reserved.

secretion of pro-inflammatory cytokines and co-stimulatory molecules or TIR-domain-containing adapter-inducing interferon-β (TRIF)-dependent signaling pathway [2,4,5]. PRRs can also detect structurally related chemical moieties from damaged host cells and DAMPs act as endogenous PRR ligands. Recognition of PAMPs and DAMPs can not only lead to the induction of pro-inflammatory and anti-microbial activity but also result in the adaptive immune response [4]. In addition to these processes, APCs, such as dendritic cells (DCs), become mature in response to recognition of microbial products and participate on the several activities which are important for the initiation of T cell-mediated adaptive immune responses such as (1) presentation of processed microbial antigen to T cells with major histocompatibility complexes (MHCs) on their surfaces, (2) increase the expression of co-stimulatory molecules (CD80 and CD86) on the cell surface which are required for appropriate T cell activation and (3) release of cytokines (e.g. IL-12), which adaptive immune response are necessary to differentiate naïve T cells into their effector and memory cells. Inflammasome-dependent cytokines (IL-1β and IL-18) have important roles to instruct these adaptive immunity [2,4].

Inflammasomes typically comprise caspase-1, the nucleotide-binding and oligomerization (NACHT), leucine rich repeat (LRR), and pyrin domain (PYD)-containing proteins and the apoptosis-associated speck-like protein containing a carboxy-terminal caspase activation and recruitment domain (ASC) [2,4,5]. Assembled inflammasomes can include a nucleotide-binding oligomerization domain and leucine-rich repeat-containing receptors and may be absent in melanoma 2 (AIM 2)-like receptors (ALRs). The exact composition depends on the type of initiating molecules that trigger their assembly [5,6]. Serving as a platform for recruiting and activating caspase-1, inflammasomes lead to maturation of the pro-inflammatory cytokines IL-1β and IL-18 [7]. The cytoplasmic complex eventually results in the secretion of these cytokines, thus modulating various inflammatory responses [8]. Inflammasome activation can also lead to pyroptosis, a type of pro-inflammatory cell death programmed to end the replication of pathogens within a cell [9]. Detailed process of NLRP3 and AIM2 inflammasome activation are presented in Fig. 1.

A variety of inflammasome subsets are reported to date: the NLR family subset including NLRP1, NLRP3, NLRP6, NLRP12, and caspase activation and recruitment domains (CARD)-containing protein 4 (NLRC4), and the non-NLR family subset including AIM 2 [10]. Among these, the NLRP3 inflammasome is the most studied subtype and its assembly can be triggered by TLR agonists. TLR agonists such as lipopolysaccharides (LPS) and lipid A can regulate inflammatory responses by inducing assembly of the NLRP3 inflammasome via pro-inflammatory cytokines including tumor necrosis factor (TNF)-α [11]. A more recently reported scaffold of a different kind is the AIM 2 inflammasome and its activation depends upon the recognition of host- and pathogen-associated double-stranded deoxyribonucleic acid (dsDNA) [12].

Once activated, the inflammasome can trigger an inflammatory cascade. A fully assembled inflammasome induces the recruitment and processing of pro-caspase-1 [1]. Recruited pro-caspase-1 proteins cleave themselves via oligomerization to form active caspase-1 [13]. Active

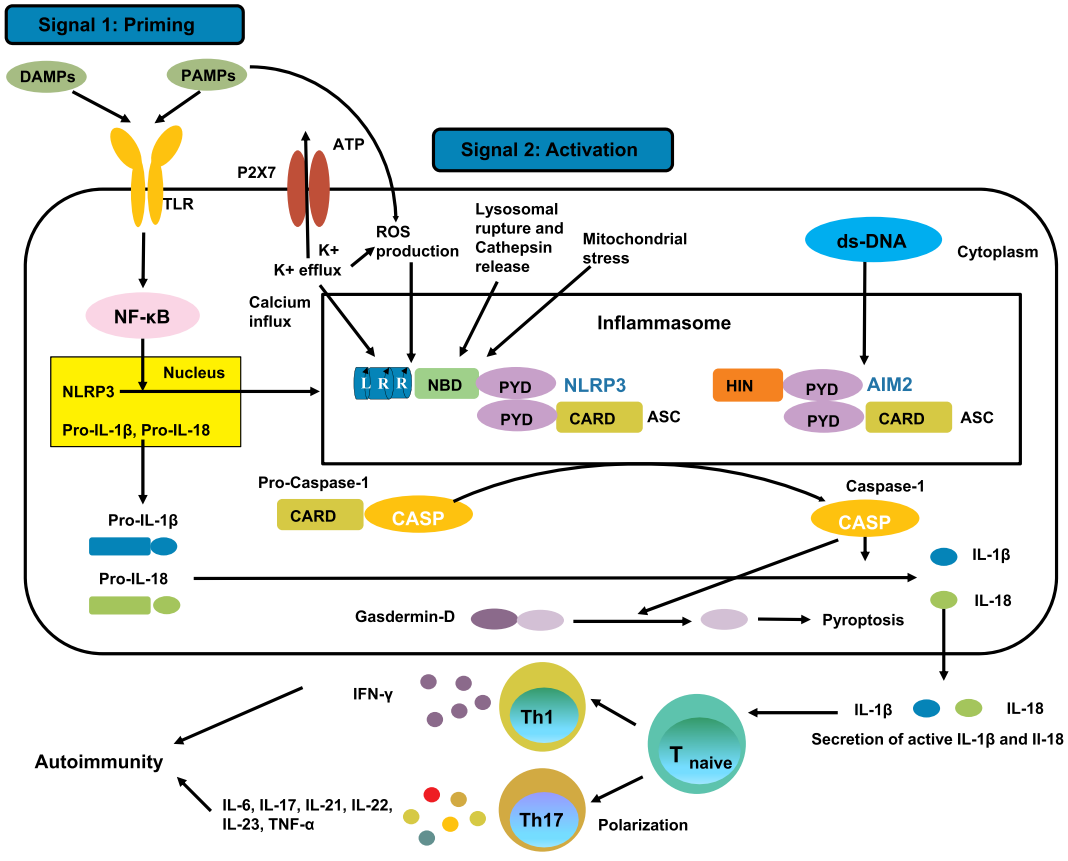


**Fig. 1.** Mechanisms for assembly and activation of NLRP3 and AIM2 inflammasomes. Inflammasomes are formed by two signals of priming and activation. Pathogen-associated molecular patterns (PAMPs) or endogenous damage-associated molecular patterns (DAMPs) are recognized by toll-like receptors (TLRs) and NF-κB (nuclear factor κB) is activated, which mediate priming of NLRP3 (nucleotide-binding domain, leucine-rich–containing family, pyrin domain–containing-3) inflammasome. The functional NLRP3 inflammasome is formed by various secondary signals such as potassium efflux, ROS (reactive oxygen species) production, lysosomal rupture and mitochondrial stress. AIM2 (absent in melanoma-2) inflammasome is sensed by dsDNA (double stranded DNA). Finally, caspase-1, IL (interleukin)-1β, and IL-18 are produced on the platform of NLRP3 or AIM2 inflammasomes. Pyroptosis (gasdermin-mediated necrotic cell death) acts as an immune defense against infection. Active forms of inflammasomes lead to the differentiation and polarization of naïve T cells and secretion of various cytokines, which can induce various kinds of autoimmunity.

caspase-1 generates matureinterleukin (IL)-1β and IL-18 via cleavage of their respective precursors, pro-IL-1β and pro-IL-18 [14–17].

Inflammasomes are reported as important molecules in understanding the pathogenesis of inflammation [18–20], which can be closely associated with the development and progression of various kinds of autoimmune diseases. In this article, we comprehensively reviewed the role of various inflammatory molecules in relation to inflammasomes, and their associated molecules in autoimmune diseases.

## 2. Inflammasome and systemic lupus erythematosus (Table 1 and Supplemental Table 1S)

### 2.1. Animal studies on inflammasomes using "lupus prone mice"

Several animal studies have demonstrated the relevance of inflammasomes in SLE. Two studies revealed that activation of the AIM2 inflammasome plays a key role in SLE pathogenesis via apoptotic DNA (apopDNA)-induced macrophage activation and IL-1β production [21,22]. In contrast, one study suggested that decreased activity of the AIM2 inflammasome contributes to lupus by producing interferon (IFN) [23]. Three studies have shown that NLRP family inflammasomes are important in SLE pathogenesis [24–26]. In lupus prone mice, the NLRP inflammasome, which is activated by disinhibition of Deoxyribonuclease 1 Like 3 (Dnase1L3) [24] and deficiency of adenosine triphosphate binding cassette transporter A1/G1 (ABCA1/G1) gene [25], activates the cascades including caspase-1 [26].

Regarding the NLRP3 inflammasome, four studies have demonstrated its relationship with lupus nephritis or cardiopulmonary manifestations [27–30]. Fu et al. reported that NLRP3 activation has an impact on podocyte injuries and the pathogenesis of proteinuria [27]. Lu et al. reported that NLRP3-R258W mutation of myeloid cells induces proteinuria and mesangial cell destruction in pristine challenged lupus-prone mice [28]. Zhao et al. showed that activation of the NLRP3 inflammasome induces lupus nephritis through activation of glycogen synthase kinase 3-β (GSK3-β) [29]. On the other hand, Lech et al. demonstrated that inactivation of the NLRP3/(NLRP3/ASC) inflammasome induces exacerbation of lupus nephritis by inhibiting transforming growth factor (TGF)-β signaling [30] and reported a link between lung infiltration and the inactivation of NLRP3/ASC inflammasomes [30]. However, one study reported that inflammasomes are not associated with the induction of diffuse alveolar hemorrhage [31]. Two other studies explained that the risk of cardiovascular disease increases in SLE patients via inflammasome activation [26,32]. IFN-α causes activation of inflammasomes, which activates caspase-1 and IL-18, resulting in a reduced number of endothelial progenitor cells and circulating angiogenic cells leading to premature atherosclerosis [26,32].

**Table 1**
List of studies related to the role of inflammasomes in lupus.

| Main study findings |
|---|
| **Genetic variant** |
| SNP in IL1B gene [33] |
| SNP in NLRP1 gene [34] |
| SNP in CARD8 gene [45] |
| **NLRP3 inflammasome** |
| **Pathogenic role of NLRP3 inflammasome** |
| Activated by *anti*-dsDNA Abs mediated by TLR4 and mitochondrial ROS [36] or potassium efflux [37] |
| Activated by endogenous RNA-containing U1-snRNP [38] |
| Activated by IRF-1 through long term type I IFN exposure [39] |
| Activated by NETs [52] |
| Activated by P2X7 signal [46] |
| Associated with podocyte injuries and proteinuria in lupus nephritis [27] |
| A role of GSK3-β in pathogenesis of lupus nephritis by activating NLRP3 inflammasome [29] |
| Serum IL-18 represents the activity of SLE, including renal flares [41] |
| **Protective role of NLRP3 inflammasome** |
| Inactivation of NLRP3 inflammasome leads to progression of lupus [30] |
| Downregulation of NLRP3/NLRP1 inflammasome is associated with lupus [40] |
| **AIM2 inflammasome** |
| **Pathogenic role of AIM2 inflammasome** |
| Activated in MRL/lpr mice [21]. |
| Activated in SLE patients with high disease activity [21]. |
| Important role in apopDNA-induced macrophage activation [22] |
| **Protective role of AIM2 inflammasome** |
| Decreased activity of AIM2 inflammasome in some mouse strains by p202 [23]. |
| AIM2 deficiency plays a role in female lupus pathogenesis by activating Ifi202gene [42] |
| **Gender-dependent regulation of inflammasomes** |
| AIM2 and p202 differently regulated according to the cell type and gender [43] |
| Mechanism of NLRP3 inflammasome activation is different in male and female SLE patients [45] |
| **Therapeutic targets for NLRP3 inflammasomes** |
| Bay11-7082: NF-κB in MRL/lpr mice [47] |
| TLR 7, 8 and 9 antagonists: in lupus-prone NZBW/F1 mice [50] |
| EGCG: in lupus-prone mice [51] |

Abbreviations: NLRP: Nucleotide-binding oligomerization domain, Leucine rich Repeat and Pyrin domain containing, SNP: single nucleotide polymorphism, IL: interleukin, CARD8: caspase recruitment domain family, member 8; dsDNA: double strand DNA, TLR: Toll-like receptor, ROS: reactive oxygen species, snRNP: small nuclear ribonucleoproteins, IRF1: Interferon Regulatory Factor, IFN: interferon, NET: neutrophil extracellular trap, P2RX7: P2X purinoceptor 7, GSK3: glycogen synthase kinase 3, SLE: systemic lupus erythematosus, AIM: absent in melanoma, apopDNA: apoptotic DNA, Ifi: interferon inducible, NF-κB: Nuclear factor-kappa B, TLR: Toll-like receptor, EGCG: epogallocatechin-3-gallate.

### 2.2. Human studies on inflammasomes and lupus using human immune cells

Several human studies have demonstrated a strong correlation of inflammasomes with human SLE using several methods including genetic, cellular, and molecular analyses. Among these, two studies analyzed single nucleotide polymorphisms (SNPs) in inflammasome genes [33,34]. Pontillo et al. reported that polymorphisms of the inflammasome receptor gene NLRP1 play a significant role in the pathogenesis of SLE [33]. Furthermore, they revealed that among NLRP1, NLRP3, CARD8, IL-1β, NLRC4, AIM2, and NLRPX genes, the IL-1β gene plays a crucial role in juvenile SLE pathogenesis [34]. Although not tested in lupus, genetic variations of inflammasome genes were associated with inflammasome mRNA levels in peripheral blood mononuclear cells [35].

Human studies on inflammasomes so far have mostly used monocytes or macrophages from SLE patients. Three studies revealed that the autoantibodies that are commonly present in SLE patients lead to activation of inflammasomes [36–38]. Zhang et al. and Shin et al. demonstrated that *anti*-dsDNA antibodies could affect inflammasome activation via mitochondrial reactive oxygen species (ROS) and the potassium efflux signaling pathway [36,37]. Further, Shin et al. reported that the presence of *anti*-U1-SnRNP could influence the clinical progression of SLE patients [38].

Three other studies have investigated the correlation between the type 1 IFN response and the inflammasome pathway [32,39,40]. Liu et al. showed that long term type I IFN exposure leads to marked inflammasome activation related to IRF-1 in SLE patients [39]. In contrast, Yang et al. reported that NLRP1/3 protein and mRNA levels in peripheral blood mononuclear cells (PBMCs) are inversely proportional to the occurrence and severity of SLE [40]. Interestingly, Kahlenberg et al. discovered that IFN-α activates inflammasomes and maturates lL-18, but suppresses IL-1β [32].

In addition, two studies showed that AIM2 inflammasomes contribute to SLE progression by using human myeloid-derived suppressor cells with monocytic morphology (M-MDSCs) [21] and apopDNA-induced macrophages, respectively [22]. In addition, Wu et al. reported that serum IL-18 levels could be an indicator of clinical outcomes including lupus renal flares in pediatric-onset SLE [41].

### 2.3. Gender-dependent regulation of inflammasomes in SLE

Panchanathan et al. reported that female sex hormone increases the susceptibility of lupus by inducing p202 protein, which further

stimulates IFN-β, signal transducer and activator of transcription 1 (STAT1), and other molecules in immune cells [42]. On the other hand, the same group demonstrated that male sex hormone prevents lupus by inducing AIM2, which is in inverse correlation with p202 [43]. Furthermore, they also demonstrated that exposure to bisphenol A, an environmental estrogen, increases the risk of SLE by activating IFN signaling, p202 expression, and NLRP3 inflammasomes [44]. Yang et al. reported that the mechanisms of NLRP3 inflammasome hyperactivation in male and female SLE patients are similarly inducing IL-1β production, but differences exist between them [45]. NLRP3 mRNA is overexpressed in females, whereas AIM2 mRNA is overexpressed and the CARD8 variant allele is associated with the male sex [45].

### 2.4. Inflammasome modulation for the treatment of SLE

The idea that SLE patients could be treated by blocking the NLRP3 inflammasome pathway has triggered the search for promising therapeutic agents. There have been three studies using MRL/lpr mice treated with isoflurane, P2X7 antagonist brilliant blue G (BBG), and I-kappaB kinase-β inhibitor (bay 11–7082), respectively [18,46,47]. These agents restricted the formation of NLRP3 inflammasomes and sequentially downregulated the release of IL- 1β [18,46,47].

Furthermore, four studies examined the influence of related factors such as A20, citral, TLR antagonists, epigallocatechin gallate (EGCG) on inflammasomes [48–51]. Li et al. reported that overexpression of A20 suppresses lupus nephritis via inhibiting NF-κB -mediated NLRP3 inflammasomes activation [48]. Ka et al. showed that treatment with citral inhibited the activation signal of NLRP3 inflammasomes and stimulated activation of Nrf2 antioxidant signaling [49]. In addition, NZBW/FL lupus-prone mice treated with TLR 7, 8, and 9 receptor antagonists also demonstrated restriction in forming NLRP3 inflammasomes [50]. Further, Tsai et al. showed that EGCG has a preventive effect on lupus nephritis via anti-oxidation of Nrf2 and inactivation of NLRP3 inflammasomes [51]. The potential therapeutic targets for inflammasome pathways are presented in Fig. 2.

In patients with SLE, targeting IL-1β receptor using anakinra led to clinical and serological improvement in 4 patients with lupus arthritis, but one patient had an arthritic flare after 6 weeks, suggesting that anakinra could be an interesting alternative in individual patients with lupus arthritis not responding to conventional treatments [35].

### 2.5. Interaction between NETs and inflammasomes in SLE

Neutrophil extracellular traps (NETs) are known to be significant defenders against microorganisms. However, unregulated NETs could be toxic to the endothelium and lead to organ damage. Kahlenberg et al. suggested that NETs are hyper-stimulated especially in SLE patients through NLRP3 inflammasomes [52]. They showed that macrophages of SLE patients stimulated by NETs through P2X7 receptors, sequentially activate the NLRP3 inflammasomes pathway and release IL-18 [52]. Furthermore, IL-18 could reactivate NETs, causing NETosis by a 'feed-forward' reaction [52]. This study provided a new insight into NETs, which could potentially contribute to SLE pathogenesis [53,54].

In summary, NLRP3 inflammasome may have an important role in the pathogenesis of both lupus prone mice and human lupus through various inflammasome-driven pathways, but there have also been some reports on the protective role of inflammasomes in SLE models, requiring cautious interpretation and further additional studies will be necessary in the future. There were some reports on different gender-dependent regulation of inflammasomes in SLE and some variants in inflammasome genes (e.g. NLRP1 and IL-1β) were found to be associated the susceptibility of SLE. Though several studies have suggested inflammasomes as potential therapeutic targets in experimental models of SLE, the results may be partial and preliminary, requiring further validations in the future. The studies on the use of IL-1β receptor antagonist were also limited in human SLE, requiring prospective randomized controlled trials (RCTs) in the future.

## 3. Inflammasomes and rheumatoid arthritis (Table 2 and Supplemental Table 2S)

### 3.1. Animal studies on inflammasomes and RA

Several studies have suggested a role of inflammasomes in RA using

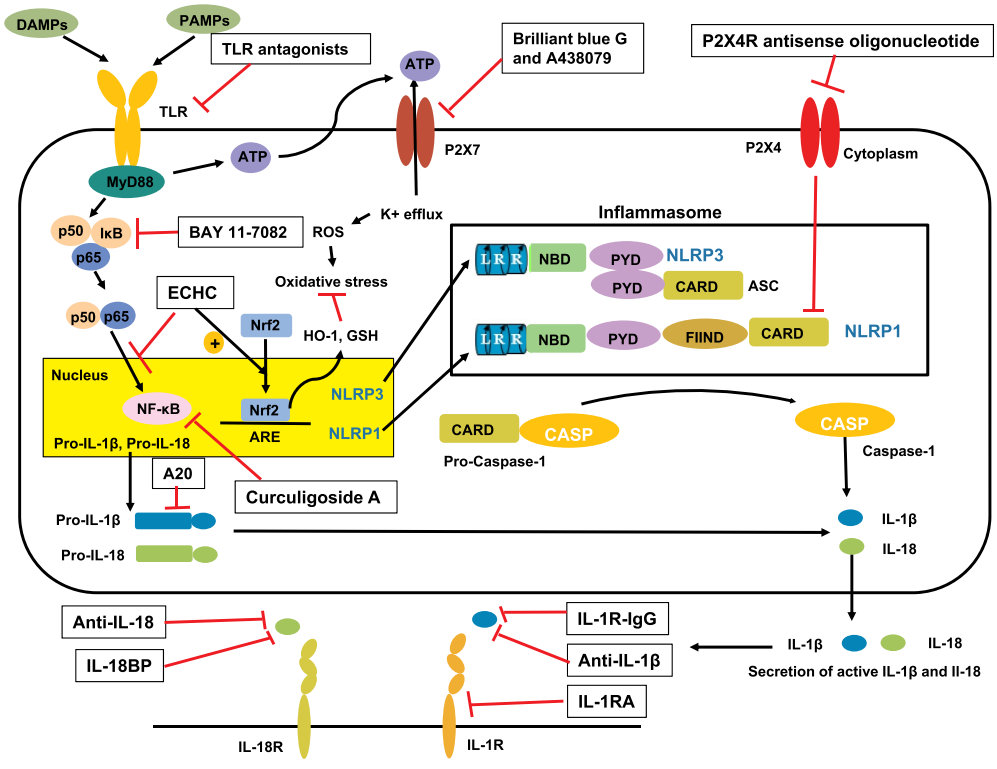


**Fig. 2.** Potential therapeutic targets for various activation pathways of NLRP1 and NLRP3 inflammasomes. Various cascades for inflammasome activation have been targeted through toll-like receptors (TLRs), P2X7 (P2X purinoceptor 7), P2X4 (P2X purinoceptor 4), ROS (reactive oxygen species), NF-κB (nuclear factor κB) pathways, IL (interleukin)-1β, and IL-18 and IL-1β receptor. Abbreviations: HO-1: heme oxygenase-1, GHS: glutathione, Nrf2: the nuclear factor erythroid 2–related factor 2, ARE: antioxidant response element

animal models or gene knock-outs [55–64]. One study showed a positive correlation between NLRP3 expression and clinical/radiographic severity in the mouse model of collagen induced arthritis (CIA) [55]. In another study, the NLRP3 signaling pathway was upregulated in a mouse model of adjuvant-induced arthritis (AIA) [59]. Although little is known about the role of NLRP3 in synovial tissue, one study that used IL-10 knock out AIA mice suggested that the degree of bone erosion by synovial regulation of the NLRP3 inflammasome was associated with IL-10 [60]. In another study using mice with a myeloid-cell-specific deletion of the A20/Tnfaip3 gene, deletion of NLRP3, caspase-1, and IL-1 receptor resulted in decreased RA-associated inflammation and cartilage destruction [63].

On the other hand, two studies suggested that caspase-1 inhibition had no therapeutic effect in RA [62,64]. One study revealed that NLRP3 and caspase-1 knock-out mice were prone to CIA whereas ASC knockout mice were not, as ASC plays an independent role in T cell priming of CIA [62]. One study revealed no therapeutic difference in joint inflammation in mice deficient for NALP-3, IPAF, and caspase-1, which are inflammasome components, whereas ASC knockout mice showed decreased inflammation in AIA, indicating that ASC is independent of caspase-1, NALP-3, and IPAF [64]. Because IL-1β is activated by caspase-1 as well as by serine proteinases, especially PR3, dual inhibition of caspase-1 and serine proteinase may have a therapeutic effect on inflammatory diseases such as RA [61].

**Table 2**
List of studies related to the role of inflammasomes in rheumatoid arthritis.

| Main study findings |
|---|
| **Genetic mutation** |
| NLRP1 mutation: NLRP1-associated autoinflammation with arthritis and dyskeratosis [73] |
| **Genetic variant** |
| **Susceptibility of RA** |
| SNPs in NLRP1 gene: Han Chinese [76] |
| SNPs in NLRP3 gene: Sweden [74], Brazil [75], Caucasian [77] |
| SNPs in CARD8 (TUCAN) gene: Sweden [74], Brazil [75], Caucasian [77] |
| **Disease course or activity (severity)** |
| SNPs in NLRP3 gene: Slovenia [78], Sweden [80] |
| SNPs in CARD8 gene: Slovenia [78] early RA from northern Sweden [81] |
| **Response to treatment (e.g. *anti*-TNF-α)** |
| SNPs in NLRP3 gene: Caucasian [77], naïve RA from Denmark [79] |
| SNPs in CARD8 gene: Caucasian [77] |
| **Comorbidities** |
| SNPs in NLRP3 gene: atherosclerosis in Sweden [80] |
| **No effect of genetic variant on RA** |
| SNPs in NLRP3 gene: RA susceptibility in France and Tunisia [82] |
| SNPs in CARD8 gene: RA susceptibility in France and Tunisia [82] and Spain [83], cardiovascular events in Spain [83] |
| **NLRP3 inflammasome** |
| **Pathogenic role of NLRP3 inflammasome** |
| **Animal models** |
| Correlated with clinical/radiographic severity in CIA mouse model [55] |
| Upregulated in AIA mice model [59] |
| ASC (−/−) mice were protected from CIA, while Nlrp3(−/−) and caspase-1(−/−) mice were not [62] |
| ASC (−/−) mice were protected from AIA, while mice deficient in NALP-3, IPAF, or caspase-1 were not [64] |
| Overexpressed in IL-10KO synovium [60] |
| RA inflammation was reduced by deletion of NLRP3, caspase-1, and IL-1 receptor in A20 (myel-KO) mice [63] |
| **RA patients** |
| IL-1β secretion and NLRP3 expression were significantly enhanced in RA [66] |
| MiR-33 activated NLRP3-caspase1-IL-1 pathway in RA patients [67] |
| IL-1β and IL-18 were markedly increased in patients with rheumatoid lung [68] |
| NLRP3 expression was increased via TLR 3 and 4 but not TLR 2 in active RA patients [70] |
| Hyperactivated caspase-1 in neutrophils of RA and high level of IL-18 but not IL-1β in RA patients [72] |
| **Protective role of NLRP3 inflammasome** |
| NLRP1 and caspase-1 mRNA in PBMC of RA group were lower in RA [71] |
| No difference in NLRP3 and ASC mRNA between RA and controls [71] |
| NLRP3, caspase-1 and ASC mRNA in granulocyte were lower in RA [71] |
| A negative correlation of NLRP1 mRNA with anti-rheumatoid factor antibody [71] |
| **Therapeutic targets for NLRP3 inflammasomes** |
| 11β-HSD 1 inhibitor: inhibition of NF-kB activation and NLRP1 assembly [85]. |
| P2X4 inhibition: suppression of NLRP 1 activation, serum IL-1β and disease activity in CIA mice [86] |
| Curculigoside A: reduction of NF-κB/NLRP3 pathway activity and serum IL-1β [87] |
| **Prognostic indicator** |
| Gene signature score of inflammasome gene predicted the treatment outcome of biologics [69] |

AbbreviationsNLRP: nucleotide binding domain and leucine rich repeat containing proteins, CIAS1: cold-induced autoinflammatory syndrome 1 gene encoding NLRP3, TUCAN (CARD8): caspase recruitment domain family, member 8, SNP: single nucleotide polymorphism; RA: rheumatoid arthritis, CIA: Collagen-induced arthritis, AIA: Adjuvant-Induced Arthritis, ASC: Apoptosis-Associated Speck-Like Protein Containing CARD, NALP: The NACHT, LRR and PYD domains containing protein, IPAF (NLRC4): NLR family CARD domain-containing protein 4, IL: interleukin; KO: knock-out, A20 (TNFAIP3): TNF α-induced protein 3, MiR: microRNA, TLR: Toll-like receptor, mRNA: messenger RNA, PBMC: peripheral blood mononuclear cell, HSD: Hydroxysteroid dehydrogenase, P2X4: P2X purinoceptor 4, NF-κB: Nuclear factor-kappa B, SDH: succinyl dehydrogenase.

### 3.2. Human studies on inflammasomes and RA

The human studies on inflammasomes suggest their important roles in RA pathogenesis [65–72]. Some studies have revealed that stimulation of the NLRP3 pathway induces IL-1β production and caspase-1 activation in RA patients [66–68,72]. In one study with RA patients, IL-1β and TNF production from peripheral blood monocytes was increased compared to that in healthy controls, and IL-1β secretion and NLRP3 expression in monocytes was elevated [66]. Another study found significant upregulation of IL-1β in bronchoalveolar lavage fluid macrophages of patients with RA-usual interstitial pneumonia (RA-UIP) [68]. Further, levels of microRNA (miR)-33, which is supposed to enhance the expression of NLRP3 and activate caspase-1 as well as NLRP3 inflammasomes, were enhanced in peripheral blood monocytes of RA patients [67]. In neutrophils, unlike in monocytes or macrophages, the expression of NLRP3, ASC, and pro-caspase-1 was decreased, whereas the expression of activated caspase-1 was increased [72]. This study showed no correlation between RA severity and serum IL-1β, but showed a positive correlation with serum IL-18 [72].

### 3.3. Genetic studies on inflammasomes and RA

Several studies have investigated various kinds of inflammasome-associated genes in relation to disease susceptibility, severity, or comorbidity of RA [73–83]. Grandemange et al. identified 3 patients from two unrelated families with diffuse skin dyskeratosis, autoimmunity, autoinflammation, arthritis, and high transitional B-cell levels, and called this type of disease as NAIAD (NLRP1-associated autoinflammation with arthritis and dyskeratosis), using a next generation sequencing approach and by analyzing NLRP1 mutations [73].

One study suggested that polymorphisms in genes encoding TUCAN (CARD8) and cryopyrin (CIAS1) are associated with RA susceptibility and severity [74]. Addobbati et al. analyzed six inflammasome genes (AIM2, CARD8, CASP1, NLRP1, NLRP3, NLRC4) along with IL-1β, IL-18, IL-1R genes and their gene expression levels in monocytes of an Eastern Brazilian population, and suggested that CARD8 and NLRP3 polymorphisms are related to RA development, and that RA patients showed high activity of IL-1β, IL-1R, and CASP1 genes [75]. One study showed that NLRP1 gene polymorphism enhances NLRP1 expression and risk of RA in Han Chinese [76]. Several other studies showed that NLRP and CARD8 are associated with RA susceptibility, high *anti*-TNF treatment response rate, or high disease activity [77–81]. However, variations in CARD8 and NLRP3 genes showed no effect on RA susceptibility in the French and Tunisian populations [82]. The association with genetic variants between NLRP3 inflammasomes and the risk of developing stroke/transient ischemic attacks (TIA), but not myocardial

infarction (MI)/angina, was reported in Swedish patients with established RA [80]. One study from northern Sweden concluded that progression in early RA is associated with the presence of CARD8-X [81]. However, it is still controversial whether inflammasomes are associated with comorbidities of RA such as cardiovascular disease (CVD). According to a study from Spain that assessed the CARD8 polymorphism in 1530 RA patients with cardiovascular disease or cardiovascular risk factors, the genotype for CARD8 gene variants between patients with RA and controls was not significantly different [83]. Therefore, they suggested that the CARD8 polymorphism did not influence the development of CVD nor did it increase the risk of cardiovascular events during the entire lifetime [83].

### 3.4. Inflammasome targeted treatment in RA

Animal studies for inflammasome-targeted RA treatment have been reported [56,84–88]. Human umbilical cord blood-derived mesenchymal stem cells (hUCB-MSCs) have the potential as a novel therapeutic strategy, inactivating NLRP3 inflammasomes and differentiating naïve macrophages to the M2 phenotype [56]. One study suggested NALP1 inflammasomes, which are activated in synovial tissues from AIA rats, as a therapeutic target [84]. BVT-2733, a selective 11β-hydroxysteroid dehydrogenase type 1 (11β-HSD1) inhibitor, reduced the severity of arthritis and the levels of serum IL-1, IL-6, IL-1β, and TNF-α in CIA mice by inhibiting NF-κB and NLRP1 inflammasome signaling pathways [85]. In another study, purinergic receptor P2X4 (P2X4R) antisense oligonucleotide, which suppresses NLRP1 inflammasome activation, was injected into CIA mice resulting in a decrease in both the disease severity and serum levels of IL-6, IL-17, IL-1β, and TNF-α [86]. Another study demonstrated the effect of curculigoside A on AIA rats by inhibiting NF-κB/NLRP3 activation [87]. The potential therapeutic targets for inflammasome pathways are presented in Fig. 2.

In summary, there have been great advances in the genetic studies of inflammasome-associated genes in RA. Mutations in the inflammasome genes such as NLRP1 and NLRP3 were gain of function and caused several kinds of autoimmunity in humans. It has been reported that SNPs in NLRP3 and CARD8 genes were associated with the susceptibility, disease course or activity, predicted response to treatment (e.g. *anti*-TNF-α) or comorbidities of RA, but some discrepant results were shown possibly due to ethnic differences. Although animal models of RA have shown the pathogenic role of NLRP3 inflammasome in its pathogenesis, there have been controversial results in patients with RA. There have been several studies showing inflammasomes as potential therapeutic targets in experimental models of RA, but the results are still preliminary, requiring further validations in the future. Targeting IL-1β receptor using anakinra was shown to be relatively safe and modestly efficacious biologic therapy for RA, but the degree of improvement is less pronounced compared to studies using other biologic therapies [88].

## 4. Inflammasomes and gout (Table 3 and Supplemental Table 3S)

Deposition of monosodium urate (MSU) crystals in human tissues, especially in the joints is the main cause of gout [89]. It induces inflammatory reactions in the body, causing severe pain and fever [89]. In this section, we summarize the previous published studies on gout, particularly focusing on the relationship between inflammasomes and gout.

### 4.1. In vitro studies on inflammasomes and gout using human cells

To understand the relationship between inflammasomes and gout, *in vitro* studies using human cells have been performed [90–99] by activating or inhibiting inflammasome-mediated immune response pathways. Several investigators have demonstrated inflammasome-mediated immune response pathway activation by serum amyloid A, TLR-2 ligands, various nucleotides including Adenosine triphosphate (ATP), soluble uric acid, and TNF-α [90–95]. NLRP3 inflammasomes were activated by these molecules and showed increased secretion of IL-1β [90–95]. In another study, NLRP3 inflammasome-mediated immune response pathway was inhibited by knockdown of the Nrf2 gene, using a heme oxygenase (HO)-1 inhibitor and knockdown of the Nek7 gene, resulting in decreased secretion of IL-1β [96]. One study demonstrated that neither MSU crystals nor soluble UA had any effect on activation of NF-κB and precipitation of UA into MSU crystals was essential for activation of the NLRP3 inflammasome and subsequent release of IL-1β in human monocytes [97].

In another *in vitro* study, phagocytosis of MSU by NLRP3 expressing osteoblasts was found [98]. MSU crystals were internalized in human osteoblasts via phagocytosis and NLRP3-mediated autophagy in this study, which can explain the presence of MSU in the bone tissue of chronic gout patients [98]. Moreover, accumulation of p62 protein by MSU and the resulting increased activity of inflammasome-mediated immune response pathway was found in another *in vitro* study [99].

### 4.2. In vivo studies on inflammasomes and gout using animal models

Several studies have investigated inflammasomes in gout using *in vivo* models [100–114]. Some genes thought to play a key role in the inflammasome-mediated immune response pathway were investigated by constructing genetically modified mice with defects in the pathway by removing the function of these genes including macrophage migration inhibitory factor (MIF), cryopyrin (Cryo), CD-14, small heterodimer partner (SHP), and miR-146a [100–105]. The results of these *in vivo* studies suggest that MIF, Cryo, and CD-14 genes play a positive role in the inflammasome-mediated immune response pathway because knockout mice for these genes produced low IL-1β [100,101,103]; in contrast, SHP and miR-146a play a negative role in the inflammasome-mediated immune response pathway because knockout mice for these genes produced high IL-1β levels [104,105].

In other *in vivo* studies, researchers have analyzed the inflammasome-mediated immune response pathway in mice using some well-known molecules like MSU and TNF-α [106,107]. They found that the MSU-induced inflammasome-mediated immune response pathway is induced by activating leukotriene B4 (LTB4) and C5a [106,107], and that the pathway is thought to be activated by low intracellular potassium concentration [108]. It was also found that the transmembrane form of TNF-α and MSU stimulation combined with free fatty acids (FFAs) or granulocyte-macrophage colony-stimulating factor (GM-CSF) are effective to activate the inflammasome-mediated immune response pathway [109–111], whereas exogenous or endogenous hydrogen sulfide (H2S), PMN-ecto (neutrophil-derived microvesicles), and antibiotics have an inhibitory effect on this immune response pathway [112–114].

### 4.3. Characteristics of genetic variants and gene expression levels in patients with gout

There have been some studies on genetic variants and gene expression levels in gout patients. Researchers have tried to find correlations between molecules – some genes and proteins - which are components of the inflammasome-mediated immune response pathway and gout [115–124].

One study showed functional variants in IL-1B and CARD8 genes, providing evidence suggesting that inflammasome activity could be related to gout severity by interaction between IL-1β and CARD8 genes [119]. In this study, the level of IL-1β was dependent on the gout-associated allele of IL1B and the multiplicative interaction between CARD8 and IL-1B was consistent with the synergy of greater inflammasome activity combined with higher levels of pre-IL-1β, causing overproduction of IL-1β in gout [119]. In another study, CARD 8 gene and P2X purinoceptor 7 (P2X7R) gene polymorphisms were partly

ARTICLE IN PRESS



**Table 3**
List of studies related to the role of inflammasomes in gout.

| Main study findings |
|---|
| **In vitro experiments** |
| **MSU stimulation alone** |
| MSU stimulation alone cannot result in IL-1β secretions in synovial fibroblasts [90]. |
| MSU crystals alone failed to induce IL-1β in PBMCs of both primary gouty patients and control groups [91] |
| MSU crystals or soluble uric acid alone cannot activate NF-κB in primary human monocytes [97] |
| Soluble uric acids induced NALP3, caspase-1 and IL-1β in proximal tubule epithelial cells [94] |
| **MSU + priming with others** |
| SAA: activation of caspase-1 and production of active IL-1β in synovial fibroblasts [90] |
| TLR-2 ligand: production of IL-1β in PBMCs of gout and healthy controls [91] |
| TNF-α: secretion of IL-1β in human neutrophils [95] |
| Soluble UA: activation of NLRP3 inflammasome in PBMCs [97] |
| **Potential mechanisms** |
| K (+)- and Ca (2+)- mediated dysfunction of mitochondria leading to iATP loss in macrophages [92] |
| Purinergic receptor signaling in macrophages [93] |
| TLR4-dependent ways in proximal tubule epithelial cells [94] |
| Nrf2 was found to be crucial in MSU crystal-induced inflammations [96] |
| Phagocytosis, autophagy and interrupted functional regulations of OBs [98]. |
| p62 accumulation through disruption of proteasomal degradation in autophagy [99] |
| **In vivo experiments** |
| **Knock-out animals showing decreased MSU-induced inflammation** |
| Inflammasome components-deficient mice: impaired neutrophil influx [102] |
| Inflammasome components-deficient mice: MSU/FFA-induced release of IL-1β was dependent on activation of caspase 1 and ASC, but not NLRP3 [110] |
| Inflammasome components-deficient mice: a role of LTB4 for the activation of NLRP3 inflammasome [106] |
| Cryo(-Z/-Z) and Cryo (DeltaLRR Z/DeltaLRR Z) mice: an important role of cryopyrin LRR domain [101] |
| CD14(−/−) mouse s.c. air pouches [103] |
| Complements (−/−) mice: an important role of C5a for the activation of NLRP3 inflammasome [107] |
| GPR43(−/−) mice: the role of microbiota to respond to an inflammasome-dependent stimulus [114] |
| **Knock-out animals showing increased MSU-induced inflammation** |
| SHP (−/−) mice: a negative regulatory role for SHP in NLRP3 inflammasome pathway [104] |
| miR-146a (−/−) mice: a negative regulatory role for miR-146a in NLRP3 inflammasome pathway [105] |
| **Activation of NLRP3 inflammasome** |
| The decrease in K (+) in air pouch MSU-injected mouse model [108] |
| Transmembrane form of TNF-α in murine gout model using C57BL/6J with joint injection of MSU crystals [109] |
| GM-CSF is important in MSU-recruited monocyte differentiation [111] |
| **Inhibition of NLRP3 inflammasome** |
| Exogenous or endogenous H2S: inhibitory effects on NLRP3 inflammasome [112] |
| Neutrophil microvesicles: inhibiting C5a-mediated priming of the inflammasome [113] |
| **Genetic studies for gout** |
| SNPs in PPARGC1B gene in gout [115] |
| MEFV gene mutations carriage: higher frequency [117], no association [121] |
| SNPs in P2X7R [118] and CARD8 gene [118,119] |
| SNPs in IL1B gene [119] |
| SNPs in TLR4 gene: increased risk in European and decreased the risk in Polynesians [120] |
| **Human studies for gout** |
| IL-18 levels were elevated in gout [122] |
| Negative association of NLRP3 levels serum uric acid levels [123] |
| No differences in synovial caspase-1 levels between gout and other arthritides [124] |
| **Targeted treatment for gout with IL-1 receptor blocker** |
| Anakinra: Improvement of renal function [125], pain and CRP [126], sign and symptoms of arthritis [127] |
| Rilonacept: Improvement of pain visual analogue scale, CRP and joint scores [128] |

Abbreviations: MSU: monosodium urate, IL: interleukin, PBMC: peripheral blood mononuclear cell, NF-κB: Nuclear factor-kappa B, NALP: The NACHT, LRR and PYD domains containing protein, SAA: Serum Amyloid A, TLR: toll like receptor, TNF: tumor necrosis factor, UA: uric acid, NLRP: nucleotide binding domain and leucine rich repeat containing proteins, iATP: intracellular ATP, Nrf2: nuclear factor E2-related factor 2, OB: osteoblast, free fatty acids: FFAs, ASC: apoptosis-associated speck-like protein containing a CARD, LTB4: Leukotriene B4, LRR: leucine-rich repeat, CD14: cluster of differentiation 14, GPR43: G-protein coupled receptor 43, SHP: small heterodimer partner, MiR: microRNA, GM-CSF: granulocyte-macrophage colony-stimulating factor, H2S: hydrogen sulfide, SNP: single nucleotide polymorphism, PPARGC1B: Peroxisome proliferator-activated receptor gamma coactivator 1-beta, MEFV: Mediterranean fever, P2X7R: P2X ligand-gated ion channel 7, CARD8: caspase recruitment domain 8, CRP: C-reactive protein.

associated with gout susceptibility [118]. Similarly, polymorphisms of PPARGC1B, MEFV, and TLR4 genes, which are involved in the inflammasome-mediated immune response pathway, were found to be associated with gout pathophysiology [115,117,120]. There was one study showing the effect of gene variants of mitochondrial DNA on inflammasome activity [116].

In addition to genetic variants, increased gene expression levels may affect the pathophysiology of gout. One study compared the cytokine levels produced by the inflammasome-mediated immune response pathway [122]. This study found increased IL-6 and IL-18 in gouty patients and showed that IL-6 is more related with the clinical features of gout (articular deformity, presence of tophi etc.), whereas IL-18 is more related with inflammatory activity in gout [122].

However, there are some studies providing contradictory evidence about the correlations between these genetic variants or increased gene expression levels and the pathophysiology of gout. The study by Rasheed et al. [120] showed a discrepant result on the association of TLR4 polymorphism according to race and Sari et al. [121] suggested that correlations between variants of the MEFV gene and the pathophysiology of gout are uncertain, because the clinical features of gouty

arthritis do not depend on the presence of MEFV variants. Besides, the caspase-1 levels in synovial fluid of gouty arthritis were not as high as those in other arthritis forms [124].

### 4.4. Effective inflammasome targeted treatment and potential treatment in gout

Some effective inflammasome targeted treatment options have been investigated in gout [125–128]. Inflammasome-targeted treatment is aimed to relieve and alleviate the severity of gout by blocking specific stages of the inflammasome-mediated immune response pathway [126–128]. Anakinra, an IL-1 receptor antagonist, is one of the effective inflammasome targeted treatments that acts by blocking IL-1 (product of inflammasome mediated immune response pathway) binding to the IL-1 receptor [126]. In this manner, it suppresses the immune response and greatly relieves the symptoms of patients with gout within 48 h after injection [127]. Anakinra was also effective in improving the renal function of diabetic patients suffering from gout [125]. Another drug, rilonacept, also known as an IL-1 trap, was developed to treat gout and was proven to be effective [128], but it has not been approved by United States Food and Drug Administration for the treatment of gout, because the benefits did not outweigh the risks associated with the drug.

There are some studies on the treatments of gout patients, including butyrate, ferulic acid, beta-hydroxybutyrate, sulforaphane, tranilast, EGCG, CY-09 (a NLRP3 inhibitor that blocks NLRP3 inflammasome assembly and activation), chondroitin sulphate, and doliroside A [129–138]. Each of these agents has their own inflammasome-targeting mechanisms similar to anakinra and rilonacept, which have been investigated using *in vivo* studies or in clinical trials. The potential therapeutic targets for inflammasome pathways are presented in Fig. 2.

In summary, MSU stimulation alone could not result in IL-1β secretions or inflammasome activation in various cells, but MSU primed with other factors could activate production of IL-1β. NLRP3 could be activated by dysfunction of mitochondria, purinergic receptor, TLR and autophagy. Studies on various kinds of knock-out animal models have found several molecules involved in the pathogenesis of MSU-induced inflammation. There have been several reports on the effect of several SNPs in IL1B, P2X7R, TLR4 and CARD8 genes on the susceptibility of gout. The studies on the levels of inflammasome-driven cytokines are limited, but several clinical trials have demonstrated that blocking IL-1 receptor (e.g. anakinra) lead to the improvement of symptoms and inflammatory markers such as C-reactive protein (CRP) in patients with gouty arthritis. Further studies are also necessary to evaluate whether targeting various pathways of inflammasome activation could be beneficial in the treatment of gout.

## 5. Inflammasomes and Behçet's disease (Supplemental Table 4S)

### 5.1. In vitro studies on inflammasomes and Behçet's disease using human cells

It has been controversial whether inflammasomes play a significant role in the pathogenesis of Behçet's disease (BD). Several *in vitro* studies using human cells support their role in the pathogenesis of BD [139–141]. Monocyte-derived macrophages (MDMs) obtained from BD patients showed increased TLR2/4 expression and IL-1β/ROS production, and showed inhibition of IL-1β production when the NLRP3 inflammasome was suppressed using RNA interference [140]. PBMCs from BD patients, when stimulated with LPS, expressed larger amounts of NLRP3 inflammasome components and produced more IL-1β compared to cells from healthy controls [139]. These results imply a possible link between the ROS-NLRP3-dependent pathway and IL-1β production in BD. On the other hand, dendritic cells and neutrophils from BD patients, which were stimulated with two types of PRRs revealed an insignificant difference in caspase-1 activity compared to healthy controls, suggesting a lack of association between the caspase-1-dependent pathway and BD pathogenesis [141].

### 5.2. Inflammasome-related genetic variants in patients with Behçet's disease

The mutational profile of inflammasome-related genes in BD has been partially elucidated so far [142–144]. One study showed that the NLRP3/cryopyrin V200 M mutant, found in some BD patients, induces IL-1β production [143]. SNPs in gamma-interferon-inducible protein (IFI16) and AIM2 genes were found to influence the risk of BD [142]. In this study, risk haplotype was associated with lower IFI16 expression compared to the protective counterpart, suggesting that IFI16 variant types may influence BD susceptibility [142]. A recent study screened seven candidate genes related to autoinflammatory diseases (AIDs) in BD patients, of which mevalonate kinase (MVK), nucleotide-binding oligomerization domain-containing protein 2 (NOD2), and proline-serine-threonine phosphatase-interacting protein 1 (PSTPIP1) were proven to be associated with AIDs by multiple different statistical methods [144].

In summary, PBMCs from BD patients show higher expression of NLRP3 inflammasome components and larger IL-1β secretion compared to healthy controls and BD patients also showed increased levels of IL-1β, suggesting an important role of inflammasome activation in BD. There have been several reports on the effect of the SNP in AIM2 and NLRP3/cryopyrin mutations in the susceptibility of BD. Although prospective RCTs for the treatment of Behçet's disease are still limited, the data from retrospective studies showed that there have also been acceptable results with IL-1 inhibitors for the management of refractory ocular disease, and with apremilast, anakinra, and ustekinumab for refractory mucocutaneous involvement [145].

## 6. Inflammasome and Sjögren's syndrome

### 6.1. In vivo study of inflammasome and Sjögren's syndrome using animal models

Researchers have suggested that the NLRP3 inflammasome pathway plays a role in the pathogenesis of Sjögren's syndrome (SS) [146–150]. Khalafalla et al. demonstrated the role of inflammasomes in SS in mouse models, and tested a potential therapeutic strategy by *in vivo* targeting of the inflammasome pathway [150]. P2X7R activation induced assembly of the NLRP3 inflammasome and IL-1β secretion in primary submandibular gland cells from wild-type mice, but not in the P2X7R-deficient strain [150]. *In vivo* injection of A438079, a P2X7R antagonist, was tested in an autoimmune salivary gland exocrinopathy mouse model, which resulted in reduced salivary gland inflammation, and enhanced saliva secretion, suggesting that P2X7R antagonism could be an effective therapeutic strategy for SS patients [150].

### 6.2. Human studies on inflammasomes and Sjögren's syndrome

In a study conducted by Baldini et al. [146], the gene expression levels of inflammasome components (NLRP3, ASC, and caspase-1) were significantly higher in primary SS gland specimens, and this was paralleled by the increased expression of mature IL-18 in primary SS saliva samples. The same group also showed that the expression levels of inflammasome components (NLRP3, ASC, and caspase-1) were even higher in patients later developing mucosa-associated lymphoid tissue non-Hodgkin's lymphoma (MALT-NHL) due to SS progression [147]. Another group demonstrated an increase in ASC, caspase-1, and IL-1β expression in SS patients, with IL-1β mRNA/protein levels being correlated with NLRP3 mRNA levels [148]. These evidences support the

hypothesis that the P2X7R-NLRP3 inflammasome-caspase-1 axis is involved in the pathogenesis of SS exocrinopathy and lymphomagenesis.

### 6.3. Mechanism underlying NLRP3 inflammasome activation in Sjögren's syndrome

A recent study proposed a mechanism for NLRP3 inflammasome activation in severe SS, suggesting that extranuclear accumulation of inflammagenic DNA and impaired DNA degradation may be the culprit [149]. In SS patients at risk of lymphoma and in those with established lymphoma, extranuclear DNA accumulation was found in the serum, PBMCs, and salivary gland tissues, with diminished DNaseI activity and DNaseII expression [149]. Evidences of NLRP3 inflammasome pathway activation were found in the monocytes and macrophages of these patients [149]. Addition of cell-free nucleic acids obtained from patients or DNaseII gene-silencing both effectively activated inflammasomes in healthy monocytes *in vitro* [149].

Although recent studies regarding inflammasomes have shed a light on their pathogenic roles and potentially new therapeutic options in SS, one randomized, double-blind, placebo-controlled trial of anakinra did not find a significant reduction of fatigue in pSS its primary endpoint [151]. The reported inefficacy of anakinra in SS could be due to the reason that other immunologic pathways such as B cells which are considered to be a central actor of primary SS pathogenicity, germinal center-like structures which are likely to support auto-Ab (*anti*-SSA/Ro) production, and T cell co-stimulation might be more important in the pathogenesis of SS [152].

## 7. Inflammasome and ANCA-associated glomerulonephritis

### 7.1. Inflammasome suppression by the effect of ROS in ANCA-associated glomerulonephritis

Experiments with the anti-neutrophil cytoplasmic antibody (ANCA)-associated glomerulonephritis (AAGN) mouse model revealed that ROS generated by ANCA-induced phagocyte NADPH oxidase (phox) suppresses caspase-1 activity, thus inhibiting inflammasome action and reducing ANCA-induced inflammation [153]. In an *anti*-myeloperoxidase (*anti*-MPO) antibody-mediated disease model, mice transplanted with either gp91(phox)-deficient or p47(phox)-deficient bone marrow showed accelerated disease with increased crescents, necrosis, glomerular monocytes, and renal IL-1β levels compared to mice transplanted with wild-type bone marrow, suggesting that ROS could limit the ANCA-induced inflammation [153]. In this study, IL-1β receptor blockade ameliorated the disease in gp91(phox)-deficient mice, implicating IL-1 receptor blockade as a promising strategy in AAGN [153].

### 7.2. NLRP3 inflammasome induced tubulointerstitial injury in ANCA-associated glomerulonephritis

Another study [154] in which kidney biopsy was performed in AAGN patients, showed that the NLRP3 inflammasome pathway plays a key role in tubulointerstitial injury (TII) [154]. IL-1β levels and TLR4/NLRP3 expression levels correlated with TII severity. NLRP3 protein and TLR4 protein were exclusively detected in tubulointerstitial infiltrating cells, and almost none were detected in the glomerular area [154]. These results imply that NLRP3 inflammasome-dependent processing in tubulointerstitial macrophages produces IL-1β, resulting in TII [154]. Since the NLRP3 inflammasome pathway seems to act only in the tubulointerstitial area, inhibiting the action of IL-1β may be a strategy for treating AAGN [154]. However, the role of inflammasomes in the pathogenesis of AAGN is currently limited due to the small number of studies in this field.

## 8. Inflammasomes and IgA vasculitis (Henoch-Schönlein purpura)

### 8.1. An inflammasome-related genetic variant in IgA vasculitis (Henoch-Schönlein purpura) patients

He et al. [155] reported a variant in the pyrin-encoding MEFV gene associated with IgA vasculitis (Henoch-Schönlein purpura) susceptibility. MEFV E148Q polymorphism (G > C) increased the risk of IgA vasculitis by an odds ratio of 2.76, and the allele was correlated with joint involvement in the disease [155].

## 9. Conclusion

In this study, we attempted to expand our understanding of the role of inflammasomes on the links between inflammasomes and autoimmune diseases though a comprehensive review of the literature. We also reviewed the central function of inflammasomes between innate and adaptive immunity. Inflammasome activation is an important mechanism for the production of active IL-1β and IL-18, which have important roles to instruct adaptive immunity. Activated cells of the innate immune system upregulate inflammatory mediators and induce increased expression of costimulatory and adhesion molecules which can also lead to the activation of the adaptive immune system. During this process, APCs, such as DCs have important roles for the initiation of T cell-mediated adaptive immune responses which are necessary to differentiate naïve T cells into their effector and memory cells. Therefore, the complex interplay between innate and adaptive immunity is critical in the development of various autoimmune and rheumatic diseases and therefore, they could be a potential therapeutic target for drug development.

Recently, there have been great advances in the genetic studies of inflammasome-associated genes in various kinds of autoimmune and rheumatic diseases, but some discrepant results were shown possibly due to ethnic differences. Some studies have revealed an association between inflammasomes and disease severity, complications, and treatment response. Nevertheless, many of these studies have been conducted in animal studies, and therefore, more studies in humans are needed. Despite these limitations, the current evidence implies a crucial role of inflammasomes in some autoimmune and rheumatic diseases.

Further studies that focus on effective inflammasome-targeted treatment are needed. Although some drugs like anakinra are in use and some are in clinical trials, further studies are necessary to evaluate their efficacy and safety. If targeting IL-1β is not effective in some autoimmune diseases such as SS, the pathogenesis may involve other crucial immunological pathways, such as B cells, germinal center-like structures which are likely to support auto-Ab (*anti*-SSA/Ro) production, T cells and co-stimulatory molecules which could be more important pathogenesis of SS. In addition, the pleiotropic actions of inflammasomes and potential harms after targeted treatments should also be considered. Although targeting inflammasomes and IL-1 has been shown to be highly effective in the treatment of rheumatic diseases, such as gout, the long-term safety profile has not been determined. Also, if the inflammasome is targeted, other adverse events, such as infection or development of other autoimmune diseases by imbalance of cytokines, should also be closely monitored because it is an important component of the innate immune system [156].

## Conflicts of interest

All authors state that they have no actual or potential conflict of interest including any financial, personal or other relationships with other people or organizations.

## Authorship

All authors made substantial contributions to all of the following:

ARTICLE IN PRESS



(1) the conception and design of the study, or acquisition of data, or analysis and interpretation of data, (2) drafting the article or revising it critically for important intellectual content, (3) final approval of the version to be submitted.

## Funding

None provided financial support for the conduct of the research and/or preparation of the article.

## Acknowledgements

We express our sincere gratitude to the students at Yonsei University College of Medicine who assisted in the preparation of the manuscript.

## Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.jaut.2019.06.010.

## References

[1] F. Martinon, K. Burns, J. Tschopp, The inflammasome: a molecular platform triggering activation of inflammatory caspases and processing of proIL-beta, Mol. Cell 10 (2002) 417–426.
[2] D. Sharma, T.D. Kanneganti, The cell biology of inflammasomes: mechanisms of inflammasome activation and regulation, J. Cell Biol. 213 (2016) 617–629.
[3] M.D. Cordero, E. Alcocer-Gómez, B. Ryffel, Gain of function mutation and inflammasome driven diseases in human and mouse models, J. Autoimmun. 91 (2018) 13–22.
[4] C.L. Evavold, J.C. Kagan, How inflammasomes inform adaptive immunity, J. Mol. Biol. 430 (2018) 217–237.
[5] H. Kumar, T. Kawai, S. Akira, Pathogen recognition by the innate immune system, Int. Rev. Immunol. 30 (2011) 16–34.
[6] D.A. Muruve, V. Petrilli, A.K. Zaiss, L.R. White, S.A. Clark, P.J. Ross, et al., The inflammasome recognizes cytosolic microbial and host DNA and triggers an innate immune response, Nature 452 (2008) 103–107.
[7] J. Chavarria-Smith, R.E. Vance, The NLRP1 inflammasomes, Immunol. Rev. 265 (2015) 22–34.
[8] J.G. Walsh, D.A. Muruve, C. Power, Inflammasomes in the CNS, Nat. Rev. Neurosci. 15 (2014) (2014) 84–97.
[9] E.A. Miao, I.A. Leaf, P.M. Treuting, D.P. Mao, M. Dors, A. Sarkar, et al., Caspase-1-induced pyroptosis is an innate immune effector mechanism against intracellular bacteria, Nat. Immunol. 11 (2010) 1136–1142.
[10] M.R. de Zoete, N.W. Palm, S. Zhu, R.A. Flavell, Inflammasomes. Cold Spring Harb. Perspect. Biol. 6 (2014) a016287.
[11] F.G. Bauernfeind, G. Horvath, A. Stutz, E.S. Alnemri, K. MacDonald, D. Speert, et al., Cutting edge: NF-kappaB activating pattern recognition and cytokine receptors license NLRP3 inflammasome activation by regulating NLRP3 expression, J. Immunol. 183 (2009) 787–791.
[12] T. Fernandes-Alnemri, J.W. Yu, P. Datta, J. Wu, E.S. Alnemri, AIM2 activates the inflammasome and cell death in response to cytoplasmic DNA, Nature 458 (2009) 509–513.
[13] X. Yang, H.Y. Chang, D. Baltimore, Autoproteolytic activation of pro-caspases by oligomerization, Mol. Cell 1 (1998) 319–325.
[14] Y. Gu, K. Kuida, H. Tsutsui, G. Ku, K. Hsiao, M.A. Fleming, et al., Activation of interferon-gamma inducing factor mediated by interleukin-1beta converting enzyme, Science 275 (1997) 206–209.
[15] T. Ghayur, S. Banerjee, M. Hugunin, D. Butler, L. Herzog, A. Carter, et al., Caspase-1 processes IFN-gamma-inducing factor and regulates LPS-induced IFN-gamma production, Nature 386 (1997) 619–623.
[16] C. Cayrol, J.P. Girard, The IL-1-like cytokine IL-33 is inactivated after maturation by caspase-1, Proc. Natl. Acad. Sci. U.S.A. 106 (2009) 9021–9026.
[17] B. Ostendorf, C. Iking-Konert, K. Kurz, G. Jung, O. Sander, M. Schneider, Preliminary results of safety and efficacy of the interleukin 1 receptor antagonist anakinra in patients with severe lupus arthritis, Ann. Rheum. Dis. 64 (2005) 630–633.
[18] Y. Yuan, Z. Liu, Isoflurane attenuates murine lupus nephritis by inhibiting NLRP3 inflammasome activation, Int. J. Clin. Exp. Med. 8 (2015) 17730–17738.
[19] E.A. Miao, D.P. Mao, N. Yudkovsky, R. Bonneau, C.G. Lorang, S.E. Warren, et al., Innate immune detection of the type III secretion apparatus through the NLRC4 inflammasome, Proc. Natl. Acad. Sci. U.S.A. 107 (2010) 3076–3080.
[20] M. Lamkanfi, V.M. Dixit, Inflammasomes and their roles in health and disease, Annu. Rev. Cell Dev. Biol. 28 (2012) 137–161.
[21] J. Ji, J. Xu, S. Zhao, F. Liu, J. Qi, Y. Song, et al., Myeloid-derived suppressor cells contribute to systemic lupus erythaematosus by regulating differentiation of Th17 cells and Tregs, Clin. Sci. 130 (2016) 1453–1467.
[22] W. Zhang, Y. Cai, W. Xu, Z. Yin, X. Gao, S. Xiong, AIM2 facilitates the apoptotic DNA-induced systemic lupus erythematosus via arbitrating macrophage functional maturation, J. Clin. Immunol. 33 (2013) 925–937.
[23] Q. Yin, D.P. Sester, Y. Tian, Y.S. Hsiao, A. Lu, J.A. Cridland, et al., Molecular mechanism for p202-mediated specific inhibition of AIM2 inflammasome activation, Cell Rep. 4 (2013) 327–339.
[24] G. Shi, K.N. Abbott, W. Wu, R.D. Salter, P.A. Keyel, Dnase1L3 regulates inflammasome-dependent cytokine secretion, Front. Immunol. 8 (2017) 522.
[25] M. Westerterp, E.L. Gautier, A. Ganda, M.M. Molusky, W. Wang, P. Fotakis, et al., Cholesterol accumulation in dendritic cells links the inflammasome to acquired immunity, Cell Metabol. 25 (2017) 1294–1304.e6.
[26] J.M. Kahlenberg, S. Yalavarthi, W. Zhao, J.B. Hodgin, T.J. Reed, N.M. Tsuji, et al., An essential role of caspase 1 in the induction of murine lupus and its associated vascular damage, Arthritis Rheum. 66 (2014) 152–162.
[27] R. Fu, C. Guo, S. Wang, Y. Huang, O. Jin, H. Hu, et al., Podocyte activation of NLRP3 inflammasomes contributes to the development of proteinuria in lupus nephritis, Arthritis Rheum. 69 (2017) 1636–1646.
[28] A. Lu, H. Li, J. Niu, S. Wu, G. Xue, X. Yao, et al., Hyperactivation of the NLRP3 inflammasome in myeloid cells leads to severe organ damage in experimental lupus, J. Immunol. 198 (2017) 1119–1129.
[29] J. Zhao, H. Wang, Y. Huang, H. Zhang, S. Wang, F. Gaskin, et al., Lupus nephritis: glycogen synthase kinase 3beta promotion of renal damage through activation of the NLRP3 inflammasome in lupus-prone mice, Arthritis Rheum. 67 (2015) 1036–1044.
[30] M. Lech, G. Lorenz, O.P. Kulkarni, M.O. Grosser, N. Stigrot, M.N. Darisipudi, et al., NLRP3 and ASC suppress lupus-like autoimmunity by driving the immunosuppressive effects of TGF-beta receptor signalling, Ann. Rheum. Dis. 74 (2015) 2224–2235.
[31] H. Zhuang, S. Han, P.Y. Lee, R. Khaybullin, S. Shumyak, L. Lu, et al., Pathogenesis of diffuse alveolar hemorrhage in murine lupus, Arthritis Rheum. 69 (2017) 1280–1293.
[32] J.M. Kahlenberg, S.G. Thacker, C.C. Berthier, C.D. Cohen, M. Kretzler, M.J. Kaplan, Inflammasome activation of IL-18 results in endothelial progenitor cell dysfunction in systemic lupus erythematosus, J. Immunol. 187 (2011) 6143–6156.
[33] A. Pontillo, E.C. Reis, B.L. Liphaus, C.A. Silva, M. Carneiro-Sampaio, Inflammasome polymorphisms in juvenile systemic lupus erythematosus, Autoimmunity 48 (2015) 434–437.
[34] A. Pontillo, M. Girardelli, A.J. Kamada, J.A. Pancotto, E.A. Donadi, S. Crovella, et al., Polimorphisms in inflammasome genes are involved in the predisposition to systemic lupus erythematosus, Autoimmunity 45 (2012) 271–278.
[35] G. Paramel Varghese, L. Folkersen, R.J. Strawbridge, B. Halvorsen, A. Yndestad, T. Ranheim, et al., NLRP3 inflammasome expression and activation in human atherosclerosis, J. Am. Heart Assoc. 5 (2016) e003031.
[36] H. Zhang, R. Fu, C. Guo, Y. Huang, H. Wang, S. Wang, et al., Anti-dsDNA antibodies bind to TLR4 and activate NLRP3 inflammasome in lupus monocytes/macrophages, J. Transl. Med. 14 (2016) 156.
[37] M.S. Shin, Y. Kang, N. Lee, E.R. Wahl, S.H. Kim, K.S. Kang, et al., Self double-stranded (ds)DNA induces IL-1beta production from human monocytes by activating NLRP3 inflammasome in the presence of anti-dsDNA antibodies, J. Immunol. 190 (2013) 1407–1415.
[38] M.S. Shin, Y. Kang, N. Lee, S.H. Kim, K.S. Kang, R. Lazova, et al., U1-small nuclear ribonucleoprotein activates the NLRP3 inflammasome in human monocytes, J. Immunol. 188 (2012) 4769–4775.
[39] J. Liu, C.C. Berthier, J.M. Kahlenberg, Enhanced inflammasome activity in systemic lupus erythematosus is mediated via type I interferon-induced up-regulation of interferon regulatory factor 1, Arthritis Rheum. 69 (2017) 1840–1849.
[40] Q. Yang, C. Yu, Z. Yang, Q. Wei, K. Mu, Y. Zhang, et al., Deregulated NLRP3 and NLRP1 inflammasomes and their correlations with disease activity in systemic lupus erythematosus, J. Rheumatol. 41 (2014) 444–452.
[41] C.Y. Wu, H.Y. Yang, T.C. Yao, S.H. Liu, J.L. Huang, Serum IL-18 as biomarker in predicting long-term renal outcome among pediatric-onset systemic lupus erythematosus patients, Medicine (Baltim.) 95 (2016) e5037.
[42] R. Panchanathan, X. Duan, H. Shen, V.A. Rathinam, L.D. Erickson, K.A. Fitzgerald, et al., Aim2 deficiency stimulates the expression of IFN-inducible Ifi202, a lupus susceptibility murine gene within the Nba2 autoimmune susceptibility locus, J. Immunol. 185 (2010) 7385–7393.
[43] R. Panchanathan, X. Duan, M. Arumugam, H. Shen, H. Liu, D. Choubey, Cell type and gender-dependent differential regulation of the p202 and Aim2 proteins: implications for the regulation of innate immune responses in SLE, Mol. Immunol. 49 (2011) 273–280.
[44] R. Panchanathan, H. Liu, Y.K. Leung, S.M. Ho, D. Choubey, Bisphenol A (BPA) stimulates the interferon signaling and activates the inflammasome activity in myeloid cells, Mol. Cell. Endocrinol. 415 (2015) 45–55.
[45] C.A. Yang, S.T. Huang, B.L. Chiang, Sex-dependent differential activation of NLRP3 and AIM2 inflammasomes in SLE macrophages, Rheumatology 54 (2015) 324–331.
[46] J. Zhao, H. Wang, C. Dai, H. Wang, H. Zhang, Y. Huang, et al., P2X7 blockade attenuates murine lupus nephritis by inhibiting activation of the NLRP3/ASC/caspase 1 pathway, Arthritis Rheum. 65 (2013) 3176–3185.
[47] J. Zhao, H. Zhang, Y. Huang, H. Wang, S. Wang, C. Zhao, et al., Bay11-7082 attenuates murine lupus nephritis via inhibiting NLRP3 inflammasome and NF-kappaB activation, Int. Immunopharmacol. 17 (2013) 116–122.
[48] M. Li, X. Shi, T. Qian, J. Li, Z. Tian, B. Ni, et al., A20 overexpression alleviates pristine-induced lupus nephritis by inhibiting the NF-kappaB and NLRP3 inflammasome activation in macrophages of mice, Int. J. Clin. Exp. Med. 8 (2015) 17430–17440.
[49] S.M. Ka, J.C. Lin, T.J. Lin, F.C. Liu, L.K. Chao, C.L. Ho, et al., Citral alleviates an

accelerated and severe lupus nephritis model by inhibiting the activation signal of NLRP3 inflammasome and enhancing Nrf2 activation, Arthritis Res. Ther. 17 (2015) 331.
[50] F.G. Zhu, W. Jiang, L. Bhagat, D. Wang, D. Yu, J.X. Tang, et al., A novel antagonist of Toll-like receptors 7, 8 and 9 suppresses lupus disease-associated parameters in NZBW/F1 mice, Autoimmunity 46 (2013) 419–428.
[51] P.Y. Tsai, S.M. Ka, J.M. Chang, H.C. Chen, H.A. Shui, C.Y. Li, et al., Epigallocatechin-3-gallate prevents lupus nephritis development in mice via enhancing the Nrf2 antioxidant pathway and inhibiting NLRP3 inflammasome activation, Free Radic. Biol. Med. 51 (2011) 744–754.
[52] J.M. Kahlenberg, C. Carmona-Rivera, C.K. Smith, M.J. Kaplan, Neutrophil extracellular trap-associated protein activation of the NLRP3 inflammasome is enhanced in lupus macrophages, J. Immunol. 190 (2013) 1217–1226.
[53] K.H. Lee, A. Kronbichler, D.D. Park, Y. Park, H. Moon, H. Kim, et al., Neutrophil extracellular traps (NETs) in autoimmune diseases: a comprehensive review, Autoimmun. Rev. 16 (2017) 1160–1173.
[54] H.H. Shen, Y.X. Yang, X. Meng, X.Y. Luo, X.M. Li, Z.W. Shuai, et al., NLRP3: a promising therapeutic target for autoimmune diseases, Autoimmun. Rev. 17 (2018) 694–702.
[55] Y. Zhang, Y. Zheng, H. Li, NLRP3 inflammasome plays an important role in the pathogenesis of collagen-induced arthritis, Mediat. Inflamm. 2016 (2016) 9656270.
[56] T.H. Shin, H.S. Kim, T.W. Kang, B.C. Lee, H.Y. Lee, Y.J. Kim, et al., Human umbilical cord blood-stem cells direct macrophage polarization and block inflammasome activation to alleviate rheumatoid arthritis, Cell Death Dis. 7 (2016) e2524.
[57] J. Rzepecka, M.A. Pineda, L. Al-Riyami, D.T. Rodgers, J.K. Huggan, F.E. Lumb, et al., Prophylactic and therapeutic treatment with a synthetic analogue of a parasitic worm product prevents experimental arthritis and inhibits IL-1beta production via NRF2-mediated counter-regulation of the inflammasome, J. Autoimmun. 60 (2015) 59–73.
[58] H. Ji, A. Pettit, K. Ohmura, A. Ortiz-Lopez, V. Duchatelle, C. Degott, et al., Critical roles for interleukin 1 and tumor necrosis factor alpha in antibody-induced arthritis, J. Exp. Med. 196 (2002) 77–85.
[59] X.F. Li, W.W. Shen, Y.Y. Sun, W.X. Li, Z.H. Sun, Y.H. Liu, et al., MicroRNA-20a negatively regulates expression of NLRP3-inflammasome by targeting TXNIP in adjuvant-induced arthritis fibroblast-like synoviocytes, Jt. Bone Spine 83 (2016) 695–700.
[60] C.J. Greenhill, G.W. Jones, M.A. Nowell, Z. Newton, A.K. Harvey, A.N. Moideen, et al., Interleukin-10 regulates the inflammasome-driven augmentation of inflammatory arthritis and joint destruction, Arthritis Res. Ther. 16 (2014) 419.
[61] L.A. Joosten, M.G. Netea, G. Fantuzzi, M.I. Koenders, M.M. Helsen, H. Sparrer, et al., Inflammatory arthritis in caspase 1 gene-deficient mice: contribution of proteinase 3 to caspase 1-independent production of bioactive interleukin-1beta, Arthritis Rheum. 60 (2009) 3651–3662.
[62] S.K. Ippagunta, D.D. Brand, J. Luo, K.L. Boyd, C. Calabrese, R. Stienstra, et al., Inflammasome-independent role of apoptosis-associated speck-like protein containing a CARD (ASC) in T cell priming is critical for collagen-induced arthritis, J. Biol. Chem. 285 (2010) 12454–12462.
[63] L. Vande Walle, N. Van Opdenbosch, P. Jacques, A. Fossoul, E. Verheugen, P. Vogel, et al., Negative regulation of the NLRP3 inflammasome by A20 protects against arthritis, Nature 512 (2014) 69–73.
[64] L. Kolly, M. Karababa, L.A. Joosten, S. Narayan, R. Salvi, V. Petrilli, et al., Inflammatory role of ASC in antigen-induced arthritis is independent of caspase-1, NALP-3, and IPAF, J. Immunol. 183 (2009) 4003–4012.
[65] S. Rosengren, H.M. Hoffman, W. Bugbee, D.L. Boyle, Expression and regulation of cryopyrin and related proteins in rheumatoid arthritis synovium, Ann. Rheum. Dis. 64 (2005) 708–714.
[66] P. Ruscitti, P. Cipriani, P. Di Benedetto, V. Liakouli, O. Berardicurti, F. Carubbi, et al., Monocytes from patients with rheumatoid arthritis and type 2 diabetes mellitus display an increased production of interleukin (IL)-1beta via the nucleotide-binding domain and leucine-rich repeat containing family pyrin 3(NLRP3)-inflammasome activation: a possible implication for therapeutic decision in these patients, Clin. Exp. Immunol. 182 (2015) 35–44.
[67] Q. Xie, M. Wei, B. Zhang, X. Kang, D. Liu, W. Zheng, et al., MicroRNA33 regulates the NLRP3 inflammasome signaling pathway in macrophages, Mol. Med. Rep. 17 (2018) 3318–3327.
[68] I. Lasithiotaki, I. Giannarakis, E. Tsitoura, K.D. Samara, G.A. Margaritopoulos, C. Choulaki, et al., NLRP3 inflammasome expression in idiopathic pulmonary fibrosis and rheumatoid lung, Eur. Respir. J. 47 (2016) 910–918.
[69] S. Nakamura, K. Suzuki, H. Iijima, Y. Hata, C.R. Lim, Y. Ishizawa, et al., Identification of baseline gene expression signatures predicting therapeutic responses to three biologic agents in rheumatoid arthritis: a retrospective observational study, Arthritis Res. Ther. 18 (2016) 159.
[70] C. Choulaki, G. Papadaki, A. Repa, E. Kampouraki, K. Kambas, K. Ritis, et al., Enhanced activity of NLRP3 inflammasome in peripheral blood cells of patients with active rheumatoid arthritis, Arthritis Res. Ther. 17 (2015) 257.
[71] T. Wang, C.L. Zhu, S. Wang, L.W. Mo, G.D. Yang, J. Hu, et al., Role of NLRP3 and NLRP1 inflammasomes signaling pathways in pathogenesis of rheumatoid arthritis, Asian Pac. J. Trop. Med. 7 (2014) 827–831.
[72] Z. Yang, J. Cao, C. Yu, Q. Yang, Y. Zhang, L. Han, Caspase-1 mediated interleukin-18 activation in neutrophils promotes the activity of rheumatoid arthritis in a NLRP3 inflammasome independent manner, Jt. Bone Spine 83 (2016) 282–289.
[73] S. Grandemange, E. Sanchez, P. Louis-Plence, F. Tran Mau-Them, D. Bessis, C. Coubes, et al., A new autoinflammatory and autoimmune syndrome associated with NLRP1 mutations: NAIAD (NLRP1-associated autoinflammation with arthritis and dyskeratosis), Ann. Rheum. Dis. 76 (2017) 1191–1198.
[74] A. Kastbom, D. Verma, P. Eriksson, T. Skogh, G. Wingren, P. Soderkvist, Genetic variation in proteins of the cryopyrin inflammasome influences susceptibility and severity of rheumatoid arthritis (the Swedish TIRA project), Rheumatology 47 (2008) 415–417.
[75] C. Addobbati, H.L.A. da Cruz, J.E. Adelino, A.L. Melo Tavares Ramos, T.S. Fragoso, A. Domingues, et al., Polymorphisms and expression of inflammasome genes are associated with the development and severity of rheumatoid arthritis in Brazilian patients, Inflamm. Res. 67 (2018) 255–264.
[76] J. Sui, H. Li, Y. Fang, Y. Liu, M. Li, B. Zhong, et al., NLRP1 gene polymorphism influences gene transcription and is a risk factor for rheumatoid arthritis in han Chinese, Arthritis Rheum. 64 (2012) 647–654.
[77] R.J. Mathews, J.I. Robinson, M. Battellino, C. Wong, J.C. Taylor, S. Eyre, et al., Evidence of NLRP3-inflammasome activation in rheumatoid arthritis (RA); genetic variants within the NLRP3-inflammasome complex in relation to susceptibility to RA and response to anti-TNF treatment, Ann. Rheum. Dis. 73 (2014) 1202–1210.
[78] B. Jenko, S. Praprotnik, M. Tomsic, V. Dolzan, NLRP3 and CARD8 polymorphisms influence higher disease activity in rheumatoid arthritis, J. Med. Biochem. 35 (2016) 319–323.
[79] J. Sode, U. Vogel, S. Bank, P.S. Andersen, M.K. Thomsen, M.L. Hetland, et al., Anti-TNF treatment response in rheumatoid arthritis patients is associated with genetic variation in the NLRP3-inflammasome, PLoS One 9 (2014) e100361.
[80] A. Kastbom, L. Arlestig, S. Rantapaa-Dahlqvist, Genetic variants of the NLRP3 inflammasome are associated with stroke in patients with rheumatoid arthritis, J. Rheumatol. 42 (2015) 1740–1745.
[81] A. Kastbom, M. Johansson, D. Verma, P. Soderkvist, S. Rantapaa-Dahlqvist, CARD8 p.C10X polymorphism is associated with inflammatory activity in early rheumatoid arthritis, Ann. Rheum. Dis. 69 (2010) 723–726.
[82] M. Ben Hamad, F. Cornelis, S. Marzouk, G. Chabchoub, Z. Bahloul, A. Rebai, et al., Association study of CARD8 (p.C10X) and NLRP3 (p.Q705K) variants with rheumatoid arthritis in French and Tunisian populations, Int. J. Immunogenet. 39 (2012) 131–136.
[83] M. Garcia-Bermudez, R. Lopez-Mejias, C. Gonzalez-Juanatey, A. Corrales, S. Castaneda, A.M. Ortiz, et al., CARD8 rs2043211 (p.C10X) polymorphism is not associated with disease susceptibility or cardiovascular events in Spanish rheumatoid arthritis patients, DNA Cell Biol. 32 (2013) 28–33.
[84] L. Zhu, J. Li, L. Guo, X. Yu, D. Wu, L. Luo, et al., Activation of NALP1 inflammasomes in rats with adjuvant arthritis; a novel therapeutic target of carboxyamidotriazole in a model of rheumatoid arthritis, Br. J. Pharmacol. 172 (2015) 3446–3459.
[85] L. Zhang, Y. Dong, F. Zou, M. Wu, C. Fan, Y. Ding, 11beta-Hydroxysteroid dehydrogenase 1 inhibition attenuates collagen-induced arthritis, Int. Immunopharmacol. 17 (2013) 489–494.
[86] F. Li, N. Guo, Y. Ma, B. Ning, Y. Wang, L. Kou, Inhibition of P2X4 suppresses joint inflammation and damage in collagen-induced arthritis, Inflammation 37 (2014) 146–153.
[87] H. Ding, G. Gao, L. Zhang, G. Shen, W. Sun, Z. Gu, et al., The protective effects of curculigoside A on adjuvant-induced arthritis by inhibiting NF-small ka, CyrillicB/NLRP3 activation in rats, Int. Immunopharmacol. 30 (2016) 43–49.
[88] M. Mertens, J.A. Singh, Anakinra for rheumatoid arthritis, Cochrane Database Syst. Rev. 1 (2009) CD005121.
[89] S.M. Nielsen, K. Zobbe, L.E. Kristensen, R. Christensen, Nutritional recommendations for gout: an update from clinical epidemiology, Autoimmun. Rev. 17 (2018) 1090–1096.
[90] K. Migita, T. Koga, K. Satomura, M. Izumi, T. Torigoshi, Y. Maeda, et al., Serum amyloid A triggers the mosodium urate -mediated mature interleukin-1beta production from human synovial fibroblasts, Arthritis Res. Ther. 14 (2012) R119.
[91] E.E. Mylona, M. Mouktaroudi, T.O. Crisan, S. Makri, A. Pistiki, M. Georgitsi, et al., Enhanced interleukin-1beta production of PBMCs from patients with gout after stimulation with Toll-like receptor-2 ligands and urate crystals, Arthritis Res. Ther. 14 (2012) R158.
[92] J. Nomura, A. So, M. Tamura, N. Busso, Intracellular ATP decrease mediates NLRP3 inflammasome activation upon nigericin and crystal stimulation, J. Immunol. 195 (2015) 5718–5724.
[93] N. Riteau, L. Baron, B. Villeret, N. Guillou, F. Savigny, B. Ryffel, et al., ATP release and purinergic signaling: a common pathway for particle-mediated inflammasome activation, Cell Death Dis. 3 (2012) e403.
[94] J. Xiao, X.L. Zhang, C. Fu, R. Han, W. Chen, Y. Lu, et al., Soluble uric acid increases NALP3 inflammasome and interleukin-1beta expression in human primary renal proximal tubule epithelial cells through the Toll-like receptor 4-mediated pathway, Int. J. Mol. Med. 35 (2015) 1347–1354.
[95] K. Yokose, S. Sato, T. Asano, M. Yashiro, H. Kobayashi, H. Watanabe, et al., TNF-alpha potentiates uric acid-induced interleukin-1beta (IL-1beta) secretion in human neutrophils, Mod. Rheumatol. 28 (2018) 513–517.
[96] J.J. Jhang, Y.T. Cheng, C.Y. Ho, G.C. Yen, Monosodium urate crystals trigger Nrf2- and heme oxygenase-1-dependent inflammation in THP-1 cells, Cell. Mol. Immunol. 12 (2015) 424–434.
[97] B.M. Alberts, J.S. Barber, S.M. Sacre, K.A. Davies, P. Ghezzi, L.M. Mullen, Precipitation of soluble uric acid is necessary for in vitro activation of the NLRP3 inflammasome in primary human monocytes, Mar 1. pii: jrheum 180855, J. Rheumatol. (2019), https://doi.org/10.3899/jrheum.180855 ([Epub ahead of print]).
[98] I. Allaeys, F. Marceau, P.E. Poubelle, NLRP3 promotes autophagy of urate crystals phagocytized by human osteoblasts, Arthritis Res. Ther. 15 (2013) R176.
[99] J.Y. Choe, H.Y. Jung, K.Y. Park, S.K. Kim, Enhanced p62 expression through impaired proteasomal degradation is involved in caspase-1 activation in

monosodium urate crystal-induced interleukin-1b expression, Rheumatology 53 (2014) 1043–1053.
[100] I. Galvao, A.C. Dias, L.D. Tavares, I.P. Rodrigues, C.M. Queiroz-Junior, V.V. Costa, et al., Macrophage migration inhibitory factor drives neutrophil accumulation by facilitating IL-1beta production in a murine model of acute gout, J. Leukoc. Biol. 99 (2016) 1035–1043.
[101] H.M. Hoffman, P. Scott, J.L. Mueller, A. Misaghi, S. Stevens, G.D. Yancopoulos, et al., Role of the leucine-rich repeat domain of cryopyrin/NALP3 in monosodium urate crystal-induced inflammation in mice, Arthritis Rheum. 62 (2010) 2170–2179.
[102] F. Martinon, V. Petrilli, A. Mayor, A. Tardivel, J. Tschopp, Gout-associated uric acid crystals activate the NALP3 inflammasome, Nature 440 (2006) 237–241.
[103] P. Scott, H. Ma, S. Viriyakosol, R. Terkeltaub, R. Liu-Bryan, Engagement of CD14 mediates the inflammatory potential of monosodium urate crystals, J. Immunol. 177 (2006) 6370–6378.
[104] C.S. Yang, J.J. Kim, T.S. Kim, P.Y. Lee, S.Y. Kim, H.M. Lee, et al., Small heterodimer partner interacts with NLRP3 and negatively regulates activation of the NLRP3 inflammasome, Nat. Commun. 6 (2015) 6115.
[105] Q.B. Zhang, Y.F. Qing, C.C. Yin, L. Zhou, X.S. Liu, Q.S. Mi, et al., Mice with miR-146a deficiency develop severe gouty arthritis via dysregulation of TRAF 6, IRAK 1 and NALP3 inflammasome, Arthritis Res. Ther. 20 (2018) 45.
[106] F.A. Amaral, V.V. Costa, L.D. Tavares, D. Sachs, F.M. Coelho, C.T. Fagundes, et al., NLRP3 inflammasome-mediated neutrophil recruitment and hypernociception depend on leukotriene B(4) in a murine model of gout, Arthritis Rheum. 64 (2012) 474–484.
[107] H.J. Khameneh, A.W. Ho, F. Laudisi, H. Derks, M. Kandasamy, B. Sivasankar, et al., C5a regulates IL-1beta production and leukocyte recruitment in a murine model of monosodium urate crystal-induced peritonitis, Front. Pharmacol. 8 (2017) 10.
[108] C. Schorn, B. Frey, K. Lauber, C. Janko, M. Strysio, H. Keppeler, et al., Sodium overload and water influx activate the NALP3 inflammasome, J. Biol. Chem. 286 (2011) 35–41.
[109] F.A. Amaral, L.F. Bastos, T.H. Oliveira, A.C. Dias, V.L. Oliveira, L.D. Tavares, et al., Transmembrane TNF-alpha is sufficient for articular inflammation and hypernociception in a mouse model of gout, Eur. J. Immunol. 46 (2016) 204–211.
[110] L.A. Joosten, M.G. Netea, E. Mylona, M.I. Koenders, R.K. Malireddi, M. Oosting, et al., Engagement of fatty acids with Toll-like receptor 2 drives interleukin-1beta production via the ASC/caspase 1 pathway in monosodium urate monohydrate crystal-induced gouty arthritis, Arthritis Rheum. 62 (2010) 3237–3248.
[111] O.M. Shaw, S. Steiger, X. Liu, J.A. Hamilton, J.L. Harper, Brief report: granulocyte-macrophage colony-stimulating factor drives monosodium urate monohydrate crystal-induced inflammatory macrophage differentiation and NLRP3 inflammasome up-regulation in an in vivo mouse model, Arthritis Rheum. 66 (2014) 2423–2428.
[112] M. Castelblanco, J. Lugrin, D. Ehirchiou, S. Nasi, I. Ishii, A. So, et al., Hydrogen sulfide inhibits NLRP3 inflammasome activation and reduces cytokine production both in vitro and in a mouse model of inflammation, J. Biol. Chem. 293 (2018) 2546–2557.
[113] A. Cumpelik, B. Ankli, D. Zecher, J.A. Schifferli, Neutrophil microvesicles resolve gout by inhibiting C5a-mediated priming of the inflammasome, Ann. Rheum. Dis. 75 (2016) 1236–1245.
[114] A.T. Vieira, l. Macia, I. Galvao, F.S. Martins, M.C. Canesso, F.A. Amaral, et al., A role for gut microbiota and the metabolite-sensing receptor GPR43 in a murine model of gout, Arthritis Rheum. 67 (2015) 1646–1656.
[115] W.C. Chang, Y.J. Jan Wu, W.H. Chung, Y.S. Lee, S.W. Chin, T.J. Chen, et al., Genetic variants of PPAR-gamma coactivator 1B augment NLRP3-mediated inflammation in gouty arthritis, Rheumatology 56 (2017) 457–466.
[116] A.L. Gosling, J. Boocock, N. Dalbeth, J. Harre Hindmarsh, L.K. Stamp, E.A. Stahl, et al., Mitochondrial genetic variation and gout in Maori and Pacific people living in Aotearoa New Zealand, Ann. Rheum. Dis. 77 (2018) 571–578.
[117] A. Karaarslan, S. Kobak, I. Kaya, N. Intepe, M. Orman, A. Berdeli, Prevalence and significance of MEFV gene mutations in patients with gouty arthritis, Rheumatol. Int. 36 (2016) 1585–1589.
[118] S.W. Lee, S.S. Lee, D.H. Oh, D.J. Park, H.S. Kim, J.R. Choi, et al., Genetic association for P2X7R rs3751142 and CARD8 rs2043211 polymorphisms for susceptibility of gout in Korean men: multi-center study, J. Korean Med. Sci. 31 (2016) 1566–1570.
[119] C. McKinney, L.K. Stamp, N. Dalbeth, R.K. Topless, R.O. Day, D.R. Kannangara, et al., Multiplicative interaction of functional inflammasome genetic variants in determining the risk of gout, Arthritis Res. Ther. 17 (2015) 288.
[120] H. Rasheed, C. McKinney, L.K. Stamp, N. Dalbeth, R.K. Topless, R. Day, et al., The toll-like receptor 4 (TLR4) variant rs2149356 and risk of gout in european and polynesian sample sets, PLoS One 11 (2016) e0147939.
[121] I. Sari, I. Simsek, Y. Tunca, B. Kisacik, H. Erdem, S. Pay, et al., Is there a relationship between gouty arthritis and Mediterranean fever gene mutations? Rev. Bras. Reumatol. 55 (2015) 325–329.
[122] N.G. Cavalcanti, C.D. Marques, T.U. Lins E Lins, M.C. Pereira, M.J. Rego, A.L. Duarte, et al., Cytokine profile in gout: inflammation driven by IL-6 and IL-18? Immunol, Invest 45 (2016) 383–395.
[123] J.Y. Choe, S.K. Kim, Clinical significance of serum NLRP3 levels in patients with chronic gouty arthritis, Jt. Bone Spine 85 (2018) 257–258.
[124] C.N. Son, S.Y. Bang, J.H. Kim, C.B. Choi, T.H. Kim, J.B. Jun, Caspase-1 level in synovial fluid is high in patients with spondyloarthropathy but not in patients with gout, J. Korean Med. Sci. 28 (2013) 1289–1292.
[125] G. Balasubramaniam, M. Almond, B. Dasgupta, Improved renal function in diabetic patients with acute gout treated with anakinra, Kidney Int. 88 (2015) 195–196.
[126] S. Ottaviani, A. Molto, H.K. Ea, S. Neveu, G. Gill, L. Brunier, et al., Efficacy of anakinra in gouty arthritis: a retrospective study of 40 cases, Arthritis Res. Ther. 15 (2013) R123.
[127] A. So, T. De Smedt, S. Revaz, J. Tschopp, A pilot study of IL-1 inhibition by anakinra in acute gout, Arthritis Res. Ther. 9 (2007) R28.
[128] R. Terkeltaub, J.S. Sundy, H.R. Schumacher, F. Murphy, S. Bookbinder, S. Biedermann, et al., The interleukin 1 inhibitor rilonacept in treatment of chronic gouty arthritis: results of a placebo-controlled, monosequence crossover, non-randomised, single-blind pilot study, Ann. Rheum. Dis. 68 (2009) 1613–1617.
[129] M.C. Cleophas, T.O. Crisan, H. Lemmers, H. Toenhake-Dijkstra, G. Fossati, T.L. Jansen, et al., Suppression of monosodium urate crystal-induced cytokine production by butyrate is mediated by the inhibition of class I histone deacetylases, Ann. Rheum. Dis. 75 (2016) 593–600.
[130] H.M. Doss, C. Dey, C. Sudandiradoss, M.K. Rasool, Targeting inflammatory mediators with ferulic acid, a dietary polyphenol, for the suppression of monosodium urate crystal-induced inflammation in rats, Life Sci. 148 (2016) 201–210.
[131] E.L. Goldberg, J.L. Asher, R.D. Molony, A.C. Shaw, C.J. Zeiss, C. Wang, et al., Beta-hydroxybutyrate deactivates neutrophil NLRP3 inflammasome to relieve gout flares, Cell Rep. 18 (2017) 2077–2087.
[132] A.J. Greaney, N.K. Maier, S.H. Leppla, M. Moayeri, Sulforaphane inhibits multiple inflammasomes through an Nrf2-independent mechanism, J. Leukoc. Biol. 99 (2016) 189–199.
[133] Y. Yuang, H. Jiang, Y. Chen, X. Wang, Y. Yang, J. Tao, et al., Tranilast directly targets NLRP3 to treat inflammasome-driven diseases, EMBO Mol. Med. 10 (2018) e8689.
[134] J.J. Jhang, C.C. Lu, G.C. Yen, Epigallocatechin gallate inhibits urate crystals-induced peritoneal inflammation in C57BL/6 mice, Mol. Nutr. Food Res. 60 (2016) 2297–2303.
[135] H. Jiang, H. He, Y. Chen, W. Huang, J. Cheng, J. Ye, et al., Identification of a selective and direct NLRP3 inhibitor to treat inflammatory disorders, J. Exp. Med. 214 (2017) 3219–3238.
[136] E.W. Orlowsky, T.V. Stabler, E. Montell, J. Verges, V.B. Kraus, Monosodium urate crystal induced macrophage inflammation is attenuated by chondroitin sulphate: pre-clinical model for gout prophylaxis? BMC Muscoskelet. Disord. 15 (2014) 318.
[137] H. Wei, C. Hu, J. Xie, C. Yang, Y. Zhao, Y. Guo, et al., Doliroside A attenuates monosodium urate crystals-induced inflammation by targeting NLRP3 inflammasome, Eur. J. Pharmacol. 740 (2014) 321–328.
[138] G. Yang, S.H. Yeon, H.E. Lee, H.C. Kang, Y.Y. Cho, H.S. Lee, et al., Suppression of NLRP3 inflammasome by oral treatment with sulforaphane alleviates acute gouty inflammation, Rheumatology 57 (2018) 727–736.
[139] E.H. Kim, M.J. Park, S. Park, E.S. Lee, Increased expression of the NLRP3 inflammasome components in patients with Behcet's disease, J. Inflamm. 12 (2015) 41.
[140] L. Liang, X. Tan, Q. Zhou, Y. Zhu, Y. Tian, H. Yu, et al., IL-1beta triggered by peptidoglycan and lipopolysaccharide through TLR2/4 and ROS-NLRP3 inflammasome-dependent pathways is involved in ocular Behcet's disease, Investig. Ophthalmol. Vis. Sci. 54 (2013) 402–414.
[141] F. Ture-Ozdemir, A. Tulunay, M.O. Elbasi, I. Tatli, A.M. Maurer, G. Mumcu, et al., Pro-inflammatory cytokine and caspase-1 responses to pattern recognition receptor activation of neutrophils and dendritic cells in Behcet's disease, Rheumatology 52 (2013) 800–805.
[142] L. Ortiz-Fernandez, J.R. Garcia-Lozano, M.A. Montes-Cano, M. Conde-Jaldon, N. Ortego-Centeno, F.J. Garcia-Hernandez, et al., Variants of the IFI16 gene affecting the levels of expression of mRNA are associated with susceptibility to Behcet disease, J. Rheumatol. 42 (2015) 695–701.
[143] S. Yüksel, E. Eren, G. Hatemi, A.C. Sahillioglu, Y. Gultekin, D. Demiroz, et al., Novel NLRP3/cryopyrin mutations and pro-inflammatory cytokine profiles in Behcet's syndrome patients, Int. Immunol. 26 (2014) 71–81.
[144] S. Burillo-Sanz, M.A. Montes-Cano, J.R. Garcia-Lozano, L. Ortiz-Fernandez, N. Ortego-Centeno, F.J. Garcia-Hernandez, et al., Mutational profile of rare variants in inflammasome-related genes in Behcet disease: a Next Generation Sequencing approach, Sci. Rep. 7 (2017) 8453.
[145] F. Alibaz-Oner, A.H. Sawalha, H. Direskeneli, Management of Behçet's disease, Curr. Opin. Rheumatol. 30 (2018) 238–242.
[146] C. Baldini, C. Rossi, F. Ferro, E. Santini, V. Seccia, V. Donati, et al., The P2X7 receptor-inflammasome complex has a role in modulating the inflammatory response in primary Sjogren's syndrome, J. Intern. Med. 274 (2013) 480–489.
[147] C. Baldini, E. Santini, C. Rossi, V. Donati, A. Solini, The P2X7 receptor-NLRP3 inflammasome complex predicts the development of non-Hodgkin's lymphoma in Sjogren's syndrome: a prospective, observational, single-centre study, J. Intern. Med. 282 (2017) 175–186.
[148] S.K. Kim, J.Y. Choe, G.H. Lee, Enhanced expression of NLRP3 inflammasome-related inflammation in peripheral blood mononuclear cells in Sjogren's syndrome, Clin. Chim. Acta 474 (2017) 147–154.
[149] A.G. Vakrakou, S. Boiu, P.D. Ziakas, E. Xingi, H. Boleti, M.N. Manoussakis, Systemic activation of NLRP3 inflammasome in patients with severe primary Sjogren's syndrome fueled by inflammagenic DNA accumulations, J. Autoimmun. 91 (2018) 23–33.
[150] M.G. Khalafalla, L.T. Woods, J.M. Camden, A.A. Khan, K.H. Limesand, M.J. Petris, et al., P2X7 receptor antagonism prevents IL-1beta release from salivary epithelial cells and reduces inflammation in a mouse model of autoimmune exocrinopathy, J. Biol. Chem. 292 (2017) 16626–16637.
[151] K.B. Norheim, E. Harboe, L.G. Gøransson, R. Omdal, Interleukin-1 inhibition and fatigue in primary Sjögren's syndrome–a double blind, randomised clinical trial, PLoS One 7 (2012) e30123.

ARTICLE IN PRESS



[152] G. Nocturne, D. Cornec, R. Seror, X. Mariette, Use of biologics in Sjögren's syndrome, Rheum. Dis. Clin. N. Am. 42 (2016) 407–417.
[153] A. Schreiber, F.C. Luft, R. Kettritz, Phagocyte NADPH oxidase restrains the inflammasome in ANCA-induced GN, J. Am. Soc. Nephrol. 26 (2015) 411–424.
[154] M. Tashiro, Y. Sasatomi, R. Watanabe, M. Watanabe, K. Miyake, Y. Abe, et al., IL-1beta promotes tubulointerstitial injury in MPO-ANCA-associated glomerulonephritis, Clin. Nephrol. 86 (2016) 190–199.
[155] X. He, H. Lu, S. Kang, J. Luan, Z. Liu, W. Yin, et al., MEFV E148Q polymorphism is associated with Henoch-Schonlein purpura in Chinese children, Pediatr. Nephrol. 25 (2010) 2077–2082.
[156] R. Cuchacovich, I. Garcia-Valladares, L.R. Espinoza, Pleiotropic effects of inflammasome modulation in chronic gout and associated comorbidities: potential therapeutic implications, Int. J. Clin. Rheumatol. 6 (2011) 669–676.