Immunity
Review
CellPress

Proteolytic Processing of Interleukin-1 Family Cytokines: Variations on a Common Theme

Inna S. Afonina,[1,2] Christina Müller,[1,2] Seamus J. Martin,[3] and Rudi Beyaert[1,2,*]
[1]Unit of Molecular Signal Transduction in Inflammation, Inflammation Research Center, VIB, Technologiepark 927, 9052 Ghent, Belgium
[2]Department of Biomedical Molecular Biology, Ghent University, Technologiepark 927, 9052 Ghent, Belgium
[3]Molecular Cell Biology Laboratory, Department of Genetics, The Smurfit Institute, Trinity College, Dublin 2, Ireland
*Correspondence: rudi.beyaert@irc.vib-ugent.be
http://dx.doi.org/10.1016/j.immuni.2015.06.003

Members of the extended interleukin-1 (IL-1) cytokine family, such as IL-1, IL-18, IL-33, and IL-36, play a pivotal role in the initiation and amplification of immune responses. However, deregulated production and/or activation of these cytokines can lead to the development of multiple inflammatory disorders. IL-1 family members share a broadly similar domain organization and receptor signaling pathways. Another striking similarity between IL-1 family members is the requirement for proteolytic processing in order to unlock their full biological potential. Although much emphasis has been put on the role of caspase-1, another emerging theme is the involvement of neutrophil- and mast cell-derived proteases in IL-1 family cytokine processing. Elucidating the regulation of IL-1 family members by proteolytic processing is of great interest for understanding inflammation and immunity. Here, we review the identity of the proteases involved in the proteolytic processing of IL-1 family cytokines and the therapeutic implications in inflammatory disease.

Introduction

Interleukins are a group of secreted proteins that function as cytokines to mediate inter-cellular communication during immune responses. In this review, we concentrate on a particular group of interleukins, the interleukin (IL)-1 family, which is comprised of 11 members (Table 1). Some family members are pro-inflammatory (IL-1α, IL-1β, IL-18, IL-33, IL-36α, IL-36β, IL-36γ), although IL-37 is thought to play an anti-inflammatory role. Other members of the extended IL-1 family are anti-inflammatory (IL-1RA, IL-36RA, IL-38; not discussed in the present review) by acting as receptor antagonists, thereby setting a threshold for the activities of the pro-inflammatory family members. IL-1 family cytokines play a central role in mediating immune responses due to their broad spectrum of biological functions and the diverse range of cells they target. Therefore, the activity of these potent cytokines needs to be tightly regulated and this is achieved in several ways: by controlling the production and maturation of cytokine precursors, through synthesis of cytokine receptor antagonists (e.g., IL-1RA and IL-36RA) or decoy receptors, and by intracellular signaling inhibitors. As a testament to the importance of these cytokines, deregulated activity of several IL-1 family members is a major cause of inflammatory and autoimmune disorders. Thus, multiple members of the IL-1 family, as well as their activating proteases, represent attractive targets for therapeutic manipulation (Dinarello, 2012).

Members of the IL-1 family engage similar receptors (Table 1) and initiate similar signaling pathways culminating in the expression of a diverse array of pro-inflammatory cytokines, chemokines, and other factors. For more details on their function in innate and adaptive immunity, we refer to a recent review (Garlanda et al., 2013). IL-1 family members also share a common structure and are mostly synthesized as precursor proteins with little or no bioactivity, requiring limited proteolytic processing to unlock their full biological potential (see below). This is an important regulatory mechanism that maintains potent inflammatory mediators in an inactive state, thereby minimizing the potential for erroneous activation of pro-inflammatory signaling. In the case of IL-1α and IL-1β, proteolytic maturation leads to conformational changes that convert them from a proteinase K-sensitive to a proteinase K-insensitive form, likely representing the transition from conformational states that are incompetent for receptor binding to states that are competent (Hazuda et al., 1991). Interestingly, most proteolytic events that activate IL-1α and IL-1β take place within a limited region at the N terminus (Figure 1; Afonina et al., 2011; Hazuda et al., 1991). This suggests that the N-terminal region within members of the extended IL-1 family acts as a regulatory switch that keeps these cytokines inactive, possibly by obscuring the receptor binding site. In this review we summarize current knowledge on the processing of several IL-1 cytokine family members by different proteases and highlight important similarities and differences.

Veteran Members of the IL-1 Family: IL-1α and IL-1β

IL-1 is the prototypic and undoubtedly the most extensively studied cytokine of the IL-1 family, and it is implicated in a wide range of inflammatory diseases, including rheumatoid arthritis, osteoarthritis, gout, periodic fever, and type II diabetes (Dinarello, 2009). IL-1 affects cells of the innate and adaptive immune system and exerts a wide range of biological activities including promoting fever, vasodilation, hematopoiesis, lymphocyte activation, leukocyte attraction and extravasation, angiogenesis, and antibody synthesis (Garlanda et al., 2013). This diverse spectrum of IL-1-mediated activities is shared by two separately encoded proteins, IL-1α and IL-1β, which surprisingly have only 26% amino acid homology (Auron et al., 1984; March et al., 1985). Nevertheless, the two proteins activate the same cell surface receptor and have a practically identical range of biological activities. Interestingly, IL-1β but not IL-1α deficiency has been shown to result in impaired febrile and acute-phase inflammatory responses in a turpentine-induced model of local

Table 1. Overview of IL-1 Family Members, Their (Co)-receptors, Inhibitory Ligands plus Receptors, and Main Functions

Cytokine	Alternative Name	Receptor	Co-receptor	Inhibitory Ligands and Receptors	Main Function
IL-1α	IL-1F1	IL-1R1	IL-1RAcP	IL-1RA, IL-1R2	pro-inflammatory
IL-1β	IL-1F2	IL-1R1	IL-1RAcP	IL-1RA, IL-1R2	pro-inflammatory
IL-18	IL-1F4	IL-18Rα	IL-18Rβ	IL-18BP	pro-inflammatory
IL-33	IL-1F11	ST2 (IL-1RL1)	IL-1RAcP	sST2	pro-inflammatory
IL-36α IL-36β IL-36γ	IL-1F6 IL-1F8 IL-1F9	IL-36R (IL-1Rrp2)	IL-1RAcP	IL-36RA	pro-inflammatory
IL-37	IL-1F7	IL-18Rα	SIGIRR (TIR8, IL-1R8)	unknown	anti-inflammatory
IL-38	IL-1F10	IL-36R (IL-1Rrp2)	–	unknown	anti-inflammatory
IL-1RA	IL-1F3	IL-1R1	–	NA	IL-1R antagonist
IL-36RA	IL-1F5	IL-36R	–	NA	IL-36R antagonist

Abbreviation is as follows: NA, not applicable.

inflammation and tissue injury, while displaying normal susceptibility to LPS challenge (Horai et al., 1998; Zheng et al., 1995). On the other hand, IL-1α has been uniquely implicated in initiating sterile inflammation in response to dying cells by inducing neutrophil recruitment, whereas IL-1β has been shown to regulate recruitment of macrophages only in a later phase (Rider et al., 2011). Collectively, these data imply that, despite triggering the same receptor and signaling pathways, IL-1α and IL-1β are not completely redundant, which might reflect differences in their localization, expression, and mechanisms of release in different cell types.

IL-1β

Caspase-1

IL-1β is expressed by hematopoietic cells as a 31-kDa precursor, mostly in response to inflammatory stimuli. However, full-length pro-IL-1β cannot bind the IL-1R and first needs to be converted into its mature 17-kDa form (Mosley et al., 1987b). An IL-1β-converting enzyme was initially described in monocytes and referred to as ICE, which was later re-named caspase-1 (Kostura et al., 1989; Thornberry et al., 1992). Caspase-1 is the founding member of the caspase (Cysteine aspartate-specific protease) family, members of which play crucial roles in inflammation and immunity (Creagh et al., 2003; Martin et al., 2012). Caspase-1 processes IL-1β at two distinct sites, producing a minor 26-kDa product, the function of which is unknown, and a mature 17-kDa form (Figure 1; Howard et al., 1991). Mice lacking caspase-1 are reported to be highly resistant to lipopolysaccharide (LPS)-induced endotoxic shock and to have decreased serum amounts of IL-1β and other inflammatory cytokines (Kuida et al., 1995; Li et al., 1995), indicating that caspase-1 is a key protease in IL-1β processing. However, it has since become clear that, due to the very close chromosomal location of the *Casp1* and *Casp11* genes, the caspase-1-deficient animals used in these early studies in fact lack both caspase-1 and caspase-11 (caspase-11 is the murine homolog of human caspase-4 and caspase-5) (Kayagaki et al., 2011). This raises questions about previous conclusions based on experiments with these mice. Cells from double $Casp1^{-/-}Casp11^{-/-}$ mice reconstituted with a *Casp11* transgene still do not process and secrete IL-1β (and IL-18) (Kayagaki et al., 2011), confirming the essential role of caspase-1 in IL-1β processing. Moreover, in contrast to caspase-1, recombinant caspase-11 processes IL-1β poorly (Kang et al., 2000). However, the above-mentioned transgenic mice are sensitive to LPS-induced lethal septic shock, whereas $Casp1^{-/-}$ $Casp11^{-/-}$ as well as $Casp11^{-/-}$ mice are resistant, arguing that caspase-11 rather than caspase-1 is the dominant mediator of LPS-induced lethal shock.

Similar to pro-IL-1β, caspase-1 is also present in cells as an inactive precursor that needs to be activated via auto-proteolytic processing, adding another level of complexity to the regulation of IL-1β activity. Caspase-1 activation involves the formation of the inflammasome, a cytosolic multi-protein signaling complex that contains caspase-1 and a nucleotide-binding oligomerization domain-like receptor (NLR) that senses pathogen-associated molecular patterns (PAMPs) such as bacterial flagellin or host-derived danger-associated molecular pattern (DAMP) molecules such as uric acid crystals (Lamkanfi and Dixit, 2014). Different inflammasomes have been identified and are defined by the NLR protein that they contain. The NLRP3 (also known as NALP3) inflammasome is the most studied and its activation in macrophages can be achieved with a plethora of PAMPs and DAMPs. Genetic studies indicate that mutations in *NLRP3* are linked with the development of auto-inflammatory diseases (Aganna et al., 2002; Gattorno et al., 2007). It is now generally accepted that NLRP3-mediated activation and release of IL-1β requires two distinct signals: the first signal can be triggered by various PAMPs after Toll-like receptor (TLR) activation which induces the NF-κB-dependent synthesis of NALP3 and pro-IL-1β. The second signal is provided by the activation of the inflammasome and caspase-1 leading to IL-1β processing (Figure 2). This two-tiered regulation minimizes the unwanted and potentially detrimental activation of IL-1β. Importantly, another outcome of caspase-1 activation is pyroptosis, a form of cell death that is characterized by cell swelling and membrane rupture (Kayagaki et al., 2011). Cell lysis allows the release of not only mature IL-1β but also pro-IL-1β, which thus becomes accessible to proteolysis by extracellular proteases.

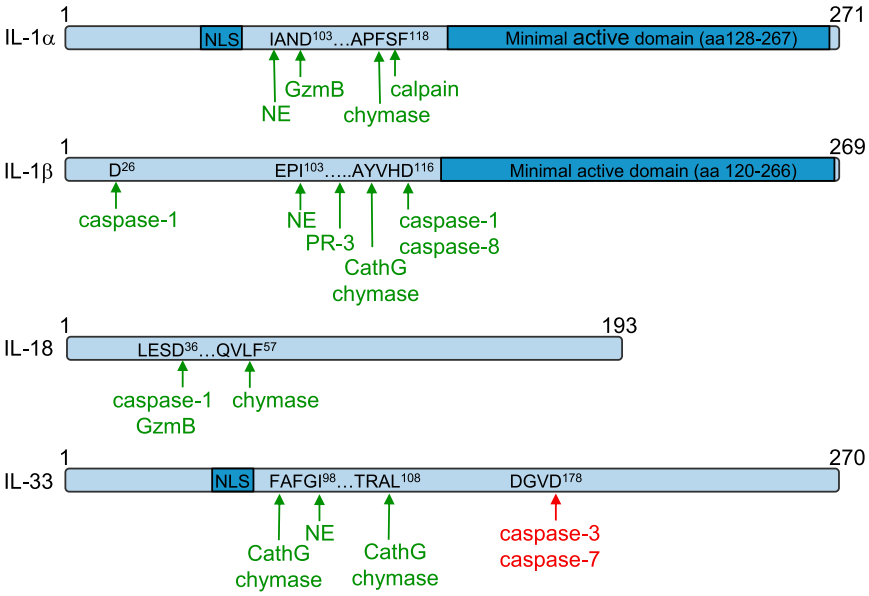

Figure 1. Overview of Proteolytic Events Known to Regulate the Biological Activity of Specific IL-1 Family Cytokines

Schematic representation of IL-1α, IL-1β, IL-18, and IL-33 proteins showing the cleavage sites of different proteases (NE, neutrophil elastase; GzmB, granzyme B; CathG, cathepsin G; PR-3, proteinase-3). Cleavage resulting in increased biological activity is indicated in green, and cleavage resulting in decreased biological activity is indicated in red. Numbering is according to the primary amino acid sequence of the human proteins. The minimal active domain in IL-1α and IL-1β (according to Mosley et al., 1987a) and nuclear localization signal (NLS) are also indicated.

Proteases Derived from Neutrophils and Mast Cells

Joosten et al. (2009) have demonstrated that disease progression in mice with neutrophil-dominated forms of arthritis is comparable in *Casp1*$^{-/-}$ and wild-type mice, suggesting that other enzymes might have an important role in regulating the activity of IL-1β. Neutrophils contain a battery of proteases that, upon release into the extracellular space, promote viral and bacterial killing, modulate cell proliferation and migration, and regulate production and activity of cytokines and chemokines (reviewed in Heutinck et al., 2010). Neutrophil elastase and cathepsin G have been shown to process IL-1β just a few amino acids upstream of the caspase-1 cleavage site (Figure 1; Black et al., 1988; Hazuda et al., 1990). Proteolysis at these sites activate IL-1β, but the resultant activity is much weaker compared to activation by caspase-1 (Black et al., 1988; Hazuda et al., 1990). Similarly, proteinase-3 (PR-3) has been shown to process and activate IL-1β in vitro, but again less efficiently than caspase-1 (Coeshott et al., 1999; Li et al., 1995). In addition to neutrophils, mast cell-derived proteases are also able to activate IL-1β. In this context, the chymotrypsin-like serine protease chymase has been shown to convert pro-IL-1β into a biologically active form that is three amino acids longer at its N terminus than the mature protein generated by caspase-1 (Figure 1; Mizutani et al., 1991). However, the relative activity of mast cell chymase-processed IL-1β and caspase-1-processed IL-1β has not been determined.

It is likely that in certain inflammatory conditions characterized by the release of pro-IL-1β in the extracellular milieu, neutrophil- and mast cell-derived proteases play an important role in IL-1β activation. Notably, LPS-treated peritoneal macrophages have been shown to release pro-IL-1β in response to necrotic stimuli or mechanical stress (Hogquist et al., 1991), making it accessible to extracellular proteases. Furthermore, neutrophil- and mast cell-derived proteases might act secondary to caspase-1 activation and pro-IL-1β release during pyroptosis (Figure 2). In this way, extracellular proteases would help to enhance and amplify the initial caspase-1-mediated inflammatory response.

Caspase-8

Caspase-8 is most well known for its role in the transmission of pro-apoptotic signals downstream of death-domain-containing members of the tumor necrosis factor (TNF) receptor family, such as Fas and TRAIL (van Raam and Salvesen, 2012). However, accumulating evidence also points to caspase-8 involvement in a non-canonical, caspase-1-independent IL-1β processing pathway (Figure 2). Thus, cellular stress (inhibition of protein translation, chemotherapeutics, or endoplasmic reticulum stress) enables macrophages and dendritic cells to produce mature IL-1β in response to TLR3 or TLR4 stimulation through a caspase-8-dependent and caspase-1-independent pathway (Antonopoulos et al., 2013; Maelfait et al., 2008; Shenderov et al., 2014). Furthermore, stimulation of dectin-1 (a macrophage-specific C-type lectin receptor) or Fas receptors, as well as downregulation of inhibitor of apoptosis (IAP) proteins, have been independently shown to lead to caspase-8 activation and caspase-1-independent maturation of proIL-1β (Bossaller et al., 2012; Gringhuis et al., 2012; Petrella et al., 2012). It is still unclear whether the role for caspase-8 in pro-IL-β processing reflects a direct effect of caspase-8 on IL-1β or the caspase-8-mediated activation of another protease that cleaves IL-1β. However, the observation that recombinant caspase-8 can cleave pro-IL-1β in vitro at exactly the same site (Asp116) as caspase-1 makes a direct effect in cells very likely (Figure 1; Maelfait et al., 2008).

IL-1α

IL-1α is constitutively expressed as a 31-kDa precursor by epithelial cells, endothelial cells, and keratinocytes. However, unlike IL-1β, IL-1α exhibits a basal amount of activity in its immature unprocessed form. IL-1α is released from damaged cells, and for a long time research on this cytokine was dominated by the idea that IL-1α does not require proteolytic processing to exhibit bioactivity. Therefore, the importance of proteolysis in the regulation of IL-1α activity has been underappreciated. This paradigm was fuelled by the work of Mosley and colleagues, who demonstrated that both full-length and processed IL-1α can bind the IL-1R and are biologically active (Mosley et al., 1987b). This view persisted despite a parallel report by the same group (Mosley et al., 1987a) comparing a panel of N-terminally truncated IL-1α constructs which indicated that the specific

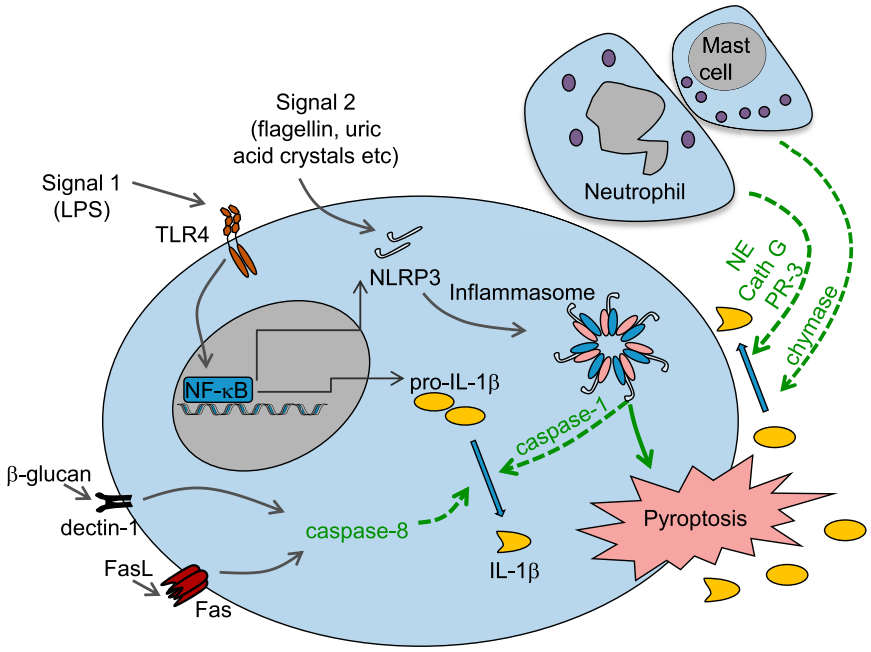

Figure 2. Caspase-1-Dependent and -Independent IL-1β Processing

Activation of caspase-1 requires two signals. Signal 1 involves the NF-κB-dependent expression of NLRP3 and pro-IL-1β, which can be provided for example by LPS triggering of TLR4. Signal 2 involves the formation of the inflammasome and caspase-1 activation that is triggered upon sensing of specific PAMPs (e.g., flagellin) or DAMPs (e.g., uric acid crystals) by NLRs such as NLRP3. Activated caspase-1 then processes pro-IL-1β into its mature form. Additionally, activation of caspase-1 can lead to pyroptotic cell death, facilitating the release of both pro- and mature forms of IL-1β into the extracellular space, where proteases derived from neutrophils and mast cells can also cleave and activate pro-IL-1β, thus further amplifying the inflammatory response. Under certain conditions, caspase-8 activation in response to dectin-1 or Fas triggering can mediate caspase-1- and inflammasome-independent IL-1β processing.

bioactivity and receptor binding affinity of mature IL-1α is several fold higher than the unprocessed form. In agreement with the latter observation, it has been recently demonstrated that cleavage of IL-1α by a number of inflammatory proteases, including elastase, granzyme B, and mast cell chymase, results in a dramatic increase in IL-1α bioactivity (Afonina et al., 2011). Thus, similar to IL-1β, the biological activity of IL-1α is also regulated via proteolytic processing.

Calpain

Calpain belongs to a family of proteases involved in cellular proliferation, differentiation, and apoptosis. Calcium-induced processing of IL-1α by calpain was first reported by two groups in the early 1990s (Figure 1; Carruth et al., 1991; Kobayashi et al., 1990). However, these reports did not explore the functional consequences of calpain-mediated IL-1α proteolysis. Formal comparison of the bioactivity of the full-length and calpain-cleaved forms of IL-1α has shown that proteolysis of IL-1α by calpain results in a several-fold increase in bioactivity (Afonina et al., 2011). This can be explained by the different receptor binding affinities of full-length and mature IL-1α, with the former having nearly 50-fold lower affinity for IL-1R (Zheng et al., 2013). Collectively, these data suggest that calpain might play a role as a regulator of inflammatory processes through converting IL-1α into its more potent form (Figure 3).

Granzyme B

Granzyme B, a major pro-apoptotic serine protease that is contained within the lytic granules of cytotoxic lymphocytes and natural killer (NK) cells, was also found to process and activate IL-1α (Figure 1; Afonina et al., 2011). IL-1α is readily processed by granzyme B both intra- and extracellularly during NK cell-dependent cytotoxicity (Figure 3). Although the classical view of cytotoxic granule proteases (granzymes) depicts these solely as apoptosis-inducing proteases that destroy virus-infected or transformed cells (Cullen and Martin, 2008), growing evidence suggests that granzymes also participate in the modulation of inflammatory responses (Afonina et al., 2010). Thus, elevated amounts of granzyme B have been detected in plasma samples and synovial fluid from patients with rheumatoid arthritis and in bronchoalveolar lavage from patients with allergic asthma. Moreover, granzyme B expression is detected in a wide range of non-cytotoxic cells including keratinocytes, mast cells, and B cells (reviewed in Afonina et al., 2010), which equips these cells with the machinery to amplify an immune response via proteolytic processing of inflammatory cytokines. Strikingly, granzyme B-deficient mice are more resistant to LPS lethality than wild-type mice, suggesting that granzyme B might also contribute to immune signaling in response to inflammatory stimuli (Metkar et al., 2008). Furthermore, when compared to wild-type animals, mice lacking granzyme B are less effective in generating antibody responses to ovalbumin in combination with IL-1α as an adjuvant, possibly because *Gzmb*$^{-/-}$ mice cannot process IL-1α to unlock its full biological potential (Afonina et al., 2011). These data also suggest that adjuvant activity of IL-1α can be further boosted via proteolytic processing by extracellular proteases present at the inflammatory site, such as granzyme B or neutrophil-derived proteases.

Proteases Derived from Neutrophils and Mast Cells

Similar to IL-1β, IL-1α is also processed and activated by neutrophil-derived proteases and mast cell chymase with an efficiency that is comparable to its processing by calpain or granzyme B (Figure 1; Afonina et al., 2011). Persistent inflammatory conditions, such as cystic fibrosis, are characterized by elevated neutrophil infiltration of tissues, accompanied by increased amounts of extracellular neutrophil proteases, predominantly elastase (Hayes et al., 2011). Of note, IL-1α processing activity is readily detected in the bronchoalveolar lavage fluids from patients with cystic fibrosis or bronchiectasis (Afonina et al., 2011). In addition to neutrophils, emerging evidence points to an important role for mast cells in pulmonary fibrosis (Andersson et al., 2011; Overed-Sayer et al., 2013). Importantly, *Pseudomonas aeruginosa*, a common respiratory pathogen in cystic fibrosis,

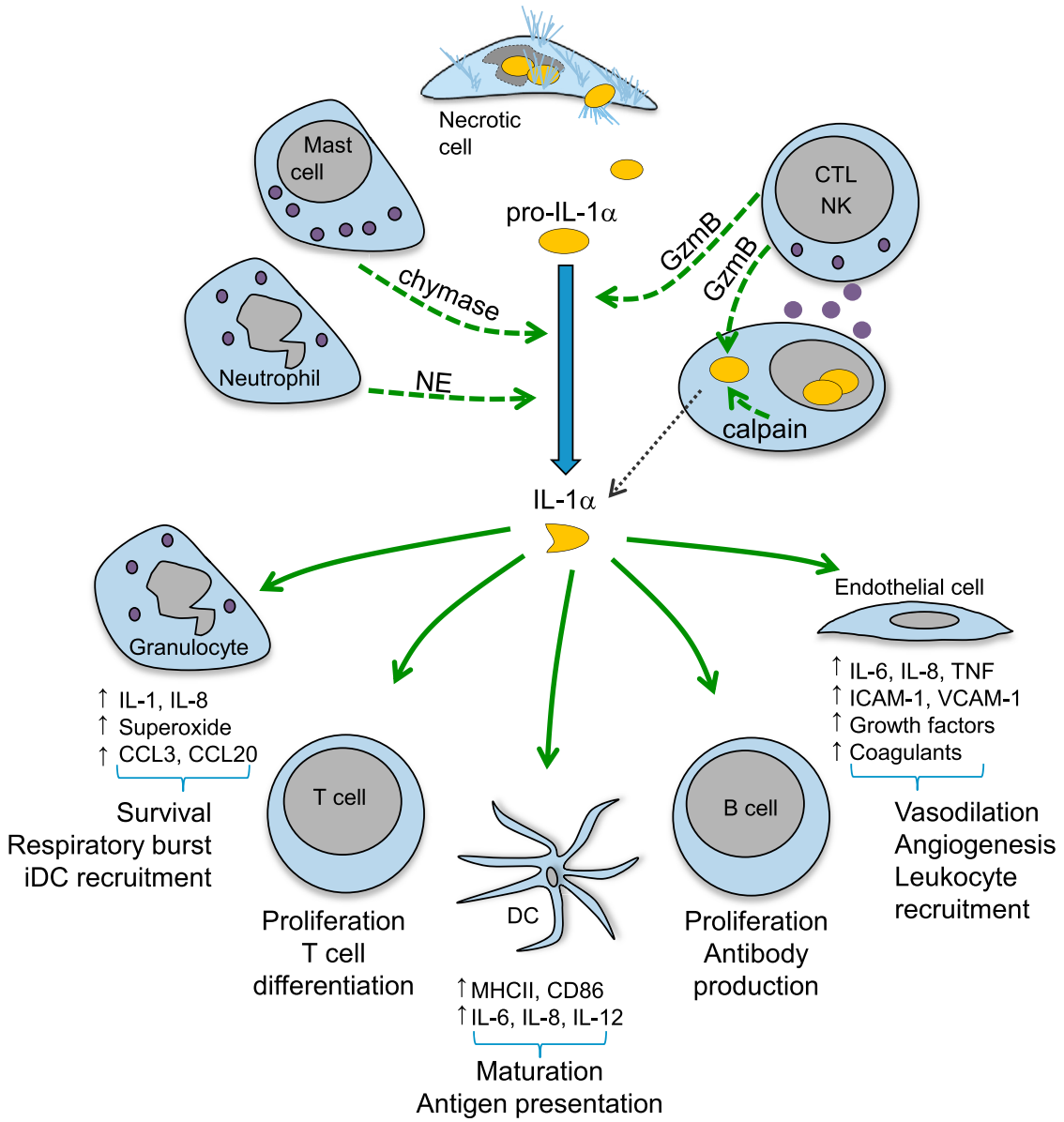

Figure 3. Processing and Biological Functions of IL-1α

Necrotic cells release pro-IL-1α into the extracellular milieu, where it is processed and activated by granzyme B (GzmB), neutrophil elastase (NE), or mast cell chymase. Intracellularly, pro-IL-1α can be cleaved by calcium-activated calpain or by granzyme B during CTL attack. Although pro-IL-1α is active, cleavage dramatically enhances its bioactivity. IL-1α subsequently targets a wide range of cells initiating both innate and adaptive immune responses.

has been reported to upregulate IL-1α and IL-1β in mast cells, further facilitating neutrophil transendothelial migration (Lin et al., 2002). IL-1α has also been suggested to initiate sterile inflammation and mediate early neutrophil recruitment (Chen et al., 2007; Rider et al., 2011). Collectively, these data suggest that IL-1-mediated neutrophil recruitment creates an amplification loop where neutrophil-derived proteases can further enhance the inflammatory response via proteolytic activation of IL-1α (Figure 3).

Although IL-1α and IL-1β bind to the same receptor and exhibit a similar range of biological effects, IL-1α has some unique properties that might be affected through proteolysis. First, although IL-1α is rarely secreted from cells, a membrane-anchored form of this cytokine has been reported to induce gene expression in neighboring cells via a juxtacrine mechanism (Kurt-Jones et al., 1985, 1987; Sasu et al., 2001). Niki et al. (2004) have demonstrated that membrane-bound IL-1α contributes to cartilage destruction and arthritis development in IL-1α transgenic mice. Interestingly, mild trypsin or plasmin digestion was shown to remove IL-1α from the cell surface without destroying its bioactivity (Matsushima et al., 1986; Niki et al., 2004). These data suggest that proteolytic processing of membrane IL-1α by proteases prevalent in the extracellular milieu of an arthritic joint, such as neutrophil proteases or granzymes, might facilitate

the transition from localized IL-1 signaling to systemic and can actually protect from cartilage destruction. Additionally, IL-1α possesses an N-terminal nuclear localization signal (NLS) that directs this cytokine to the nucleus, where it might regulate the activity of transcription factors in a receptor-independent manner (Werman et al., 2004; Wessendorf et al., 1993). Moreover, apoptotic cells retain IL-1α within their nuclei, whereas necrotic cells release IL-1α into the extracellular space (Cohen et al., 2010). Thus, cleavage of IL-1α by proteases liberated during necrosis might abolish its nuclear retention through removal of the NLS, thereby facilitating IL-1α release during necrosis but retention during apoptosis.

IL-18

IL-18 is primarily a T helper 1 (Th1) cell-oriented cytokine, which contributes to immune responses by inducing interferon (IFN)-γ production, Th1 cell proliferation, and activation of NK cells (Okamura et al., 1995; Ushio et al., 1996). However, depending on the context and interaction with other cytokines, IL-18 can display various inflammatory activities typical of the other IL-1 family members (Novick et al., 2013). This two-sided nature of IL-18 makes it a double-edged sword in cancer: whereas Th1 cell-promoting activities of IL-18 ensure anti-tumor immune responses, the pro-inflammatory functions of IL-18 might facilitate tumor progression and angiogenesis (Fabbi et al., 2015). Deletion of *Il18* in mice results in obesity, insulin resistance, atherosclerosis, and hyperglycemia, which together resemble the features of metabolic syndrome in humans (Netea et al., 2006). Abnormal IL-18 function is also involved in autoimmune and inflammatory diseases such as psoriasis and inflammatory bowel disease (reviewed in Dinarello et al., 2003).

IL-18 is constitutively expressed by most cells in healthy humans and animals as an intracellular inactive precursor of 24 kDa. Caspase-1 cleaves pro-IL-18 after Asp36 into an active molecule of 17.2 kDa that is secreted from cells (Figure 1; Ghayur et al., 1997; Gu et al., 1997). To date, caspase-1 is the only protease that has been convincingly demonstrated to convert IL-18 into its active form. However, in some cases caspase-1-independent processing of IL-18 can be observed. For example, Fas stimulation results in the release of biologically active IL-18 from caspase-1-deficient murine macrophages (Tsutsui et al., 1999). This phenomenon is sensitive to caspase inhibitors, pointing to the involvement of another member of the caspase family. It was recently shown that caspase-8-deficient macrophages do not secrete IL-18 (or IL-1β) in response to Fas stimulation (Bossaller et al., 2012), so caspase-8 is a likely candidate IL-18-processing enzyme.

Keratinocytes constitutively express and release pro-IL-18 (Companjen et al., 2000). IL-18 has been implicated in several inflammatory skin conditions, but its activation mechanism remains unclear. Notably, IL-18 can be processed by mast cell chymase, which cleaves pro-IL-18 after Phe57, downstream of the caspase-1 cleavage site (Figure 1; Omoto et al., 2006). However, the chymase-induced IL-18 cleavage product has only 20% of the activity of mature IL-18 generated by caspase-1. Notably, the caspase-1-generated IL-18 product is resistant to subsequent chymase-mediated cleavage, suggesting that IL-18 undergoes a conformational change upon processing by caspase-1, which is in line with other findings in the case of IL-1β (Hazuda et al., 1991). Omoto and colleagues have also reported granzyme B-mediated activation of recombinant IL-18 and increased IL-18 release upon incubation of IL-18-expressing HaCaT keratinocytes with $CD8^+$ T cells (Akeda et al., 2014; Omoto et al., 2010). Interestingly, keratinocytes have been shown to express granzyme B in response to UV radiation, suggesting a non-cytotoxic function for granzyme B in these cells (Hernandez-Pigeon et al., 2006, 2007). Additionally, IL-18 processing and release has been reported to be induced upon co-incubation of IFN-γ-primed oral epithelial cells with neutrophil-derived proteinase-3 and LPS, in the absence of caspase-1. However, evidence for direct processing of IL-18 by proteinase-3 has not been demonstrated (Sugawara et al., 2001). Collectively, these data suggest that proteases such as mast cell chymase and granzyme B released from resident skin cells can contribute to skin inflammation via activation of IL-18.

IL-33

IL-33 is a more recently identified member of the IL-1 cytokine family. Initial studies showed that injection of recombinant IL-33 into mice leads to splenomegaly, eosinophilia, production of Th2 cell-associated cytokines (IL-4, IL-5, and IL-13), and increased serum amounts of IgE and IgA (Schmitz et al., 2005). IL-33 acts on a wide range of innate immune cells, inducing the production of Th2 cell-type cytokines and chemokines, maturation of mast cells, survival and expansion of eosinophils, and M2-polarization of macrophages (Figure 4; Oboki et al., 2010). IL-33 has also been shown to activate innate lymphoid cells group 2 (ILC2) in combination with IL-7 or thymic stromal lymphopoeitin (TSLP) (Halim et al., 2012). This has a crucial effect on allergic airway responses ensuring ILC2 expansion and migration, release of IL-5 and IL-13, followed by eosinophil recruitment, mucus production, and airway hyper-reactivity. Moreover, IL-33 and ILC2-derived IL-13 promote activation and migration of dendritic cells (DCs), which then present antigen to naive $CD4^+$ T cells and mediate their differentiation into Th2 cells (Besnard et al., 2011; Halim et al., 2014). Thus, IL-33 affects cells of the innate and adaptive immune system and plays a central role in mediating type 2 inflammatory conditions. As a result, IL-33 plays a major role in a broad range of diseases, including rheumatoid arthritis, atherosclerosis, cardiovascular disease, inflammatory bowel disease, asthma, and chronic obstructive pulmonary disease (Garlanda et al., 2013; Pei et al., 2014).

IL-33 engages a structurally similar receptor complex (consisting of ST2 and IL-1RAcP) and largely overlapping signaling pathways to other IL-1 family members (Garlanda et al., 2013). It is expressed as a 31-kDa precursor protein by multiple cell types, including endothelial cells, keratinocytes, fibroblasts, and leukocytes, either constitutively or in response to different stimuli (Schmitz et al., 2005). Similar to IL-1α, IL-33 is predominantly released from cells upon cell damage and acts as an alarmin that initiates inflammatory responses in the context of cell death (Cayrol and Girard, 2009; Lüthi et al., 2009). IL-33 also possesses an NLS within its N terminus and is, therefore, normally tethered in the nucleus. Whether IL-33 has a nuclear function is still unclear, but it has been proposed that nuclear IL-33 is a chromatin-associated factor and transcriptional repressor.

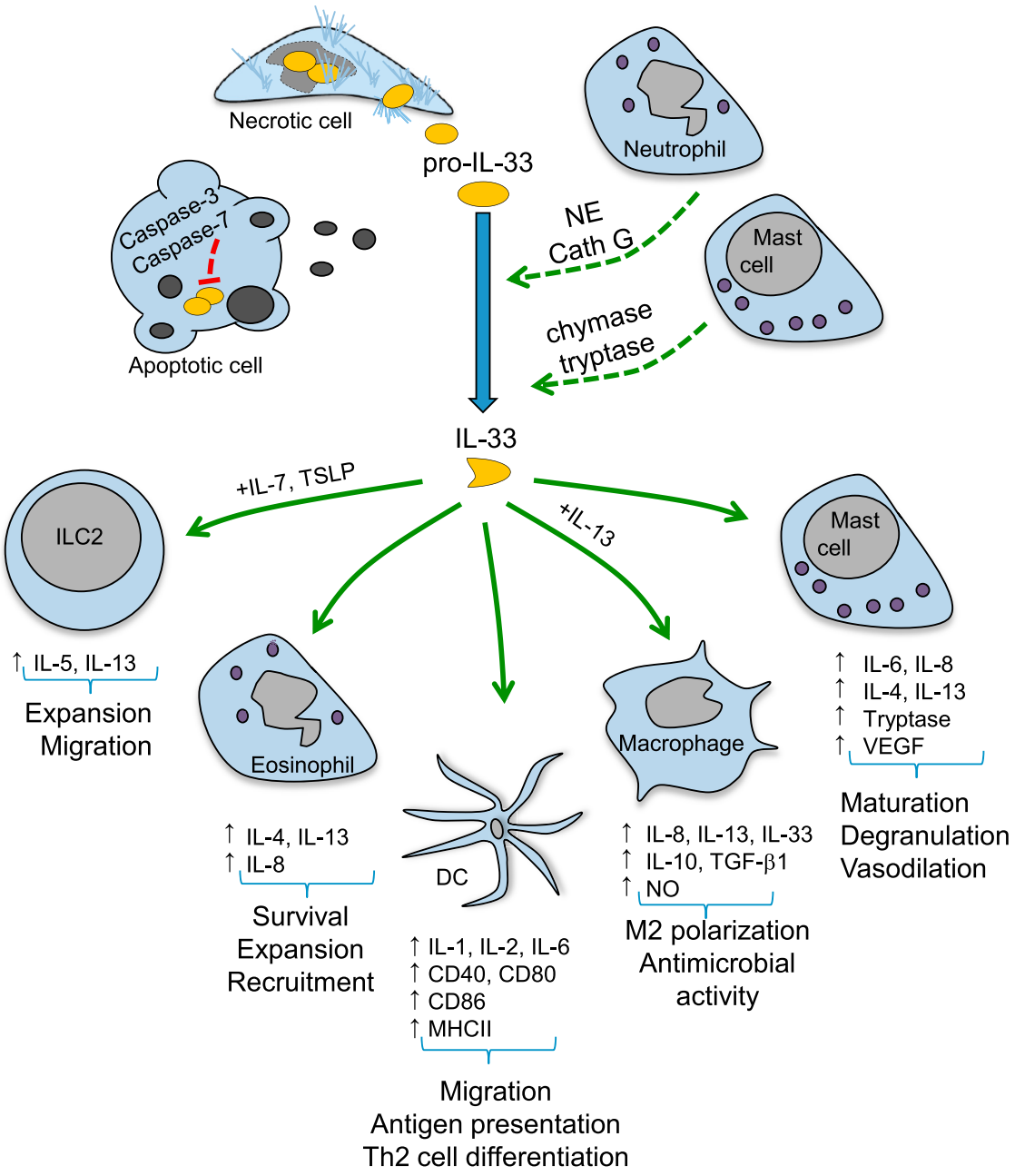

Figure 4. Processing and Biological Functions of IL-33
Similar to IL-1α, active pro-IL-33 is released by necrotic cells. On the other hand, caspase-3 and -7 inactivate IL-33 via proteolytic cleavage in apoptotic cells, thereby silencing its alarmin function. Pro-IL-33 that is released can be processed by extracellular neutrophil- or mast cell-derived proteases, which strongly enhances its activity. IL-33 targets different cell types and mainly drives Th2-cell-mediated immune responses, characterized by mast cell activation, eosinophil recruitment, and ILC2 expansion.

Chromatin association is mediated by an evolutionarily conserved homeodomain-like helix-turn-helix motif within the IL-33 N-terminal part (Carriere et al., 2007; Roussel et al., 2008). This domain also associates with the p65 NF-κB subunit, preventing its binding to DNA and dampening NF-κB-dependent gene expression (Ali et al., 2011). Alternatively and similar to IL-1α, nuclear localization of IL-33 has been proposed as a mechanism to prevent its constitutive release from cells and to limit its pro-inflammatory potential. This is supported by the detection of increased amounts of serum IL-33 in mice expressing a genetically targeted allele of IL-33 in which the whole N-terminal chromatin-binding domain (containing also the NLS) has been replaced by the coding region of the DsRed fluorescent protein. As a consequence, these animals manifest widespread non-resolving inflammation and premature death (Bessa et al., 2014).

Caspase-1, -3, and -7

Initial reports have proposed that IL-33 is activated through caspase-1-mediated processing (Schmitz et al., 2005). This view was largely based on artificial truncation of IL-33 at a proposed caspase-1 processing site, which is, however, not conserved between the human and murine forms of this cytokine (Schmitz et al., 2005). Subsequent studies have demonstrated that neither recombinant nor endogenous caspase-1 can cleave IL-33 at concentrations that are sufficient to completely process IL-1β (Lüthi et al., 2009). Instead, it has been reported that IL-33 is cleaved after Asp178 by caspase-3 and -7 in vitro and in apoptotic cells, leading to its inactivation (Lüthi et al., 2009). Importantly, the latter study also demonstrated that, similar to IL-1α, IL-33 displays basal activity and does not strictly require maturation in order to bind and activate the IL-33 receptor. These findings have been independently confirmed by several parallel reports (Ali et al., 2010; Cayrol and Girard, 2009; Talabot-Ayer et al., 2009). It is now generally accepted that full-length IL-33 is biologically active upon release from damaged cells, but can undergo processing by certain neutrophil-derived proteases (see below) to amplify its activity. Processing of IL-33 by pro-apoptotic caspases might represent a mechanism to inactivate the pro-inflammatory activities of IL-33 during apoptosis (Martin et al., 2012). This is in contrast to necrotic caspase-independent cell death, which results in the release of active full-length IL-33 and activation of an inflammatory immune response (Figure 4).

Proteases Derived from Neutrophils and Mast Cells

It has been recently shown that the neutrophil-derived proteases cathepsin G and elastase cleave full-length IL-33 at different sites within the N terminus, leading to a 10-fold increase in IL-33 activity (Figure 1; Lefrançais et al., 2012). Importantly, processing of extracellular IL-33 has been detected in bronchoalveolar lavage fluid from mice with neutrophil-dominated acute lung injury, suggesting a physiological role for neutrophil-derived proteases in the regulation of IL-33 activity (Lefrançais et al., 2012). Another neutrophil protease, PR-3, also processes IL-33 at multiple sites (Bae et al., 2012). However, only brief incubation (5 min) of IL-33 with PR-3 produces biologically active fragments, whereas prolonged exposure results in IL-33 degradation, suggesting that PR-3 might actually dampen the inflammatory response by inactivating IL-33 (Bae et al., 2012). Once again, these data implicate neutrophil-derived proteases in the amplification of the biological potency of IL-1 family members (Figure 4).

Recently, mast cell chymase has also been shown to process IL-33 (Lefrançais et al., 2012; Roy et al., 2014; Waern et al., 2013). The first two studies suggest that incubation of recombinant IL-33 with mast cell chymase results in its degradation, but bioactivity of the cleaved IL-33 was not assessed in these studies. In contrast, Lefrancais and colleagues demonstrated that mast cell chymase and tryptase cleave IL-33 at the N terminus and that the corresponding in vitro translated mature forms of IL-33 exhibit increased ability to activate ILC2 ex vivo. Further studies are clearly required to resolve these discrepancies. Of interest, mast cell chymase-deficient mice show elevated airway and inflammatory responses to house dust mite extracts, suggesting a protective and regulatory role of mast cell chymase and possibly chymase-mediated IL-33 processing during sterile inflammation (Waern et al., 2013).

IL-36

Members of the IL-36 subfamily (IL-36α, IL-36β, and IL-36γ) are encoded by separate genes on the IL-1 locus and share 36%–46% sequence identity (Smith et al., 2000). The IL-36 cytokines have attracted much interest due to accumulating evidence that they play an important initiating role in psoriasis (Johnston et al., 2011; Marrakchi et al., 2011). Mice transgenic for IL-36α transiently exhibit features of severe skin inflammation that are exacerbated through deletion of a competitive antagonist of IL-36 receptor signaling, known as IL-36RA, or through irritation of the skin with chemical agents (Blumberg et al., 2007). Moreover, hypomorphic mutations in *IL1f5* (the gene encoding IL-36RA) are linked to generalized pustular psoriasis in humans, a highly severe and life-threatening form of this disease (Marrakchi et al., 2011; Sugiura et al., 2014).

IL-36 cytokines are predominantly expressed in skin and act in an autocrine fashion on skin-resident fibroblasts and keratinocytes. However, high amounts of recombinant unprocessed IL-36 are required to elicit a biological response in vitro (Towne et al., 2011). Taking into account that proteolytic processing is required for activation of other IL-1 family members, it was not entirely unexpected that artificial removal of N-terminal amino acids from IL-36 proteins results in a more than 1,000-fold increase in bioactivity (Towne et al., 2011). However, it is not known how endogenous IL-36 undergoes maturation to its bioactive form, and the identity of the protease(s) involved remains unknown. The N termini of IL-36 cytokines lack caspase-cleavage motifs, making a role for caspases rather unlikely. Considering the accumulating evidence that IL-36 plays a key role in psoriasis, identifying the IL-36-converting enzyme(s) is of great interest.

IL-37

IL-37 has recently been implicated as an important cytokine in autoimmunity and inflammation. Elevated amounts of IL-37 have been found in cases of rheumatoid arthritis, Crohn's disease, psoriasis, and lupus and during *Mycobaterium avium* infection (Clavel et al., 2013). Mouse IL-37 has not been reported, but human IL-37 is active on mouse cells. Five isoforms (IL-37a–e, of which IL-37b is the most studied) are generated through alternative splicing and have distinct expression profiles (Clavel et al., 2013). IL-37 expression is induced by TGF-β, as well as by IL-1β and TLR ligands (Nold et al., 2010). Unlike other members of the IL-1 family, IL-37 appears to be an anti-inflammatory cytokine. Human *Il37* transgenic mice show a decreased response to LPS-induced shock and other inflammatory stimuli (McNamee et al., 2011; Nold et al., 2010). It has been proposed that IL-37 acts by dampening LPS- and IL-1β-induced inflammatory responses or by enhancing the immunosuppressive and anti-inflammatory properties of TGF-β (Grimsby et al., 2004; Nold et al., 2010). IL-37 can bind the IL-18 receptor α chain and further requires the orphan IL-1 receptor family protein IL-1R8 (also known as TIR8 or SIGIRR) for its anti-inflammatory activity (Li et al., 2015; Lunding et al., 2015; Nold-Petry et al., 2015; Pan et al., 2001). Additionally, IL-37 has been shown to translocate to the nucleus upon LPS stimulation, where it inhibits the expression of pro-inflammatory cytokines (Bulau et al., 2014; Sharma et al., 2008).

Similar to other IL-1 family members, all IL-37 variants lack a typical signal peptide. Some in vitro evidence suggests that

caspase-1 and, to a lesser extent, caspase-4 are capable of cleaving IL-37 after Asp20 (Kumar et al., 2002; Sharma et al., 2008). However, although overexpressed human IL-37 is processed in LPS-stimulated mouse macrophages, cleavage of IL-37 by endogenous caspase-1 has not been demonstrated unequivocally (Bulau et al., 2014; Sharma et al., 2008). Moreover, mutation of the proposed caspase-1 cleavage site has only a marginal effect on the LPS-induced processing of IL-37, suggesting the involvement of other proteases (Bulau et al., 2014). Interestingly, mutation of the putative caspase-1 cleavage site significantly impairs translocation of IL-37 to the nucleus, which correlates with this mutant's inability to inhibit LPS-induced IL-6 production upon transfection into bone-marrow-derived macrophages (Bulau et al., 2014). Moreover, the anti-inflammatory function of IL-37 is lost in the absence of Asc or NLRP3, known components of the caspase-1-activating inflammasome, further pointing to a possible role for caspase-1 in the nuclear translocation of IL-37 (Bulau et al., 2014). However, it is important to stress that all these studies were performed with human IL-37 in mouse cells, which might be prone to artifacts, illustrating the need for further investigation. To date, only one IL-37 isoform (IL-37b) has been studied in detail. The above-mentioned putative caspase-1 cleavage site is conserved among four of the five isoforms, but it is missing in IL-37a. Compared to the other isoforms, IL-37a lacks 20 N-terminal amino acids. It will therefore be interesting to test whether the pro-form of IL-37a is active, because this could reveal a role for alternative splicing in the regulation of IL-37 activity.

Therapeutic Implications

Members of the IL-1 family are prominent players in different inflammatory diseases, such as rheumatoid arthritis, psoriasis, diabetes, autoinflammatory syndromes, and asthma. Therefore, products that prevent their biological activity are likely to have considerable therapeutic utility. Various strategies that are already in use or under investigation include targeting of cytokine receptors via soluble receptor antagonists (Anakinra, a recombinant version of the IL-1 receptor antagonist IL-1RA) or antagonistic antibodies, neutralization of cytokines via soluble decoy receptors (the IL-1 inhibitor Rilonacept, also known as IL-1 Trap, and the soluble IL-33 receptor sST2) or neutralizing antibodies, and inhibition of cytokine maturation via protease inhibitors (reviewed in Dinarello et al., 2012). Although Anakinra has shown much promise in patients with different hereditary and common autoinflammatory disorders (Dinarello et al., 2012; Gabay et al., 2011), it is rapidly metabolized by the kidneys and daily injections of Anakinra are required to sustain its therapeutic effect, which is accompanied by side effects, such as pain and discomfort. Currently, there is a clear need for alternative strategies.

Because of its essential role in the proteolytic activation of IL-1β, caspase-1 has been originally considered a very attractive therapeutic target. However, clinical trials with caspase-1 inhibitors have been disappointing. One peptidomimetic caspase-1 inhibitor, pralnacasan (VX-740), has shown much promise in preclinical studies on mice with collagen-induced arthritis (Rudolphi et al., 2003). Subsequently, this inhibitor has demonstrated anti-inflammatory potential in phase II trials in patients with rheumatoid arthritis, was well tolerated, and caused minimum side effects (reviewed in MacKenzie et al., 2010). However, pralnacasan has been discontinued due to potential liver toxicity demonstrated in animal trials. Another peptidomimetic caspase-1 inhibitor, VX-765, has shown much promise in phase I trials in patients with rheumatoid arthritis and osteoarthritis, as well as in phase II trials in patients with psoriasis (MacKenzie et al., 2010). However, no recent developments of VX-765 or other caspase-1 inhibitors for the treatment of inflammatory disorders have been reported.

The potential clinical relevance of caspase-1-independent proteolytic maturation of IL-1 family members has been demonstrated in different mouse models. For example, Guma et al. (2009) have shown that small molecule inhibitors of elastase or chymase decrease experimental arthritis in caspase-1-deficient mice, while an elastase inhibitor reduces IL-1-dependent neutrophil infiltration in monosodium urate-induced peritonitis. Likewise, high doses of a calpain inhibitor alleviate clinical symptoms in a mouse model of antibody-induced arthritis, presumably due to its ability to block the production of mature IL-1α (Yoshifuji et al., 2005).

For the successful clinical development of IL-1 maturation inhibitors, it will be important to discriminate between disease conditions in which IL-1 activation relies on caspase-1 versus those that are dependent on other proteases, such as neutrophil-derived enzymes. For example, the presence of mutations in *NLRP3*, leading to increased inflammasome activation, that are associated with the development of CAPS (cryopyrin-associated periodic syndromes), are indicative for the use of inflammasome inhibitors in these patients (Aganna et al., 2002; Agostini et al., 2004). Notably, a new NLRP3 inflammasome-specific inhibitor MCC950 has recently been shown to inhibit caspase-1 activation and IL-1β maturation in cells isolated from CAPS patients in response to LPS ex vivo (Coll et al., 2015). Moreover, MCC950 is highly effective in a mouse model of CAPS and attenuates disease severity in a mouse model of multiple sclerosis. Importantly, inhibiting caspase-1 activation will also prevent pyroptosis and release of DAMPs, such as IL-1α and IL-33, further dampening the immune response. On the other hand, neutrophil-dominated conditions like arthritis, gout, and pulmonary inflammatory diseases would benefit more from the use of neutrophil protease inhibitors. However, inhibition of neutrophil proteases should be considered with great caution, because these proteases regulate several cytokines and are instrumental for the host defense against invading pathogens. Also, because multiple functionally divergent proteases can activate the same cytokine, inhibiting a single enzyme might not be sufficient to completely prevent cytokine activation. Therefore, combined treatment with multiple inhibitors that target different proteases might be the way to go. Moreover, this might allow the use of lower doses that only partially inhibit each protease, still allowing the cleavage of other substrates and therefore reducing the chance for possible side effects.

An interesting safety latch against IL-1α activation was recently described by Zheng et al. (2013). In some cells, IL-1α is constitutively bound to an intracellular IL-1 receptor-2 (IL-1R2), which prevents calpain and other inflammatory proteases from cleaving and activating IL-1α. Upon initiation of necrosis, IL-1R2 is degraded and IL-1α becomes available to proteases (Zheng et al., 2013). It will be worthwhile to investigate

whether reagents can be developed that, similarly to IL-1R2, mask the protease cleavage region in IL-1α or other IL-1 family members and prevent their proteolytic activation while avoiding the inhibition of proteases and their vital functionality in host defense.

Future Perspectives

Although the proteolytic maturation of several IL-1 family members is well characterized, many questions remain. For example, how does proteolytic cleavage activate these cytokines? Does this affect protein stability, protein conformation, or interactions with other proteins or receptors? Available evidence suggests that processing of specific IL-1 family cytokines can lead to conformational changes that affect their susceptibility to proteolytic degradation (Hazuda et al., 1991; Lüthi et al., 2009). These changes most likely reflect significant internal structural reorganization that influence receptor binding but might also impact on the half-lives of these cytokines. In the case of IL-1α, it is likely that its long N-terminal pro-peptide blocks receptor binding and acts as a switch to permit rapid toggling of IL-1α activity (Afonina et al., 2011). To delineate the role of the N-termini of all IL-1 family members, it would be interesting to solve and compare the crystal structures of the full-length and mature forms. However, due to difficulties in obtaining large quantities of precursor interleukins, only the crystal structures of mature or truncated forms are available (Liu et al., 2013; Priestle et al., 1988). Several protocols describing the purification of full-length IL-1α have been published in recent years (Afonina et al., 2011; Zheng et al., 2013), which might help efforts to determine the crystal structure of full-length IL-1α or other IL-1 family proteins.

Most members of the IL-1 family can be cleaved by multiple proteases. However, the relative contribution of each protease in vivo remains unclear. Processing of a specific IL-1 family member might be tissue dependent and might vary in different disease states. Studies on protease-deficient mice have already contributed important knowledge (Guma et al., 2009; Joosten et al., 2009). The generation and use of genetically targeted mice expressing catalytically inactive proteases or uncleavable mutants of IL-1 family cytokines, combined with specific inflammatory disease models, are expected to further increase our understanding of the biological effects of IL-1 family cytokine processing and the role of specific proteases. In addition, new and rapidly developing proteomic approaches might reveal novel cytokine activating proteases that are more easy to target therapeutically than those currently known.

IL-1 family members have also been implicated in tumor development. For example, IL-1 enhances angiogenesis and in this way might facilitate tumor invasiveness and metastasis (Voronov et al., 2014). IL-18 has a dual role in cancer, promoting antitumor activity of cytotoxic T lymphocytes (CTLs) and NK cells, while in some conditions enhancing tumor progression, evasion, and angiogenesis (Fabbi et al., 2015). One could speculate that granzyme B-mediated activation of IL-1α and IL-18 initially plays a protective role by stimulating cytotoxic cells, while neutrophil infiltration in the tumor microenvironment might promote angiogenesis and tumorigenesis via proteolytic activation of IL-1 family members by neutrophil-derived proteases. Thus, it might be worthwhile to explore the idea of targeting proteases of tumor-associated neutrophils as a means of antitumor therapy.

A major unresolved question regarding the IL-1 family is the identity of the proteases that process some of the most recently identified members of the IL-1 family, i.e., IL-36 and IL-37. Given their proven role in the regulation of other IL-1 family members, neutrophil-derived or mast cell-derived proteases need to be explored as potential proteases involved in the processing of IL-36 and IL-37. Because of the clear evidence that IL-36 is a major player in certain forms of psoriasis, knowledge of the proteases involved in the activation of IL-36 is extremely interesting for the development of novel therapeutic approaches.

In conclusion, accumulating evidence obtained in the last few years indicates that proteolytic processing is a common theme in the regulation of the activity of IL-1 family cytokines. Although caspases have received most interest until recently, it is now clear that other proteases including some well-known neutrophil- and mast-cell derived proteases might be equally important. The identification of additional proteases involved in the fine-tuning of IL-1 family cytokines and the elucidation of their role in inflammation and immunity is likely to keep the scientific community engaged for many years to come and can be expected to reveal innovative therapeutic targets.

ACKNOWLEDGMENTS

Research in the lab of R.B. is supported by grants from the following sources: Fund for Scientific Research Flanders (FWO) (grant numbers G028712N, G046612N, G016413N, G027413N, G090914N), Belgian Foundation Against Cancer, Agency for Innovation by Science and Technology, Interuniversity Attraction Poles (grant number P7/32), Belgian Science Policy (Belgian Coordinated Collections of Microorganisms [BCCM]/LMBP), and Ghent University (Hercules, Concerted Research Actions [GOA] [grant number BOF13-GOA-005], Group-ID Multidisciplinary research partnership). I.S.A. is supported by a postdoctoral fellowship and a research grant from FWO. C.M. is supported by a doctoral fellowship from the IWT.

REFERENCES

Afonina, I.S., Cullen, S.P., and Martin, S.J. (2010). Cytotoxic and non-cytotoxic roles of the CTL/NK protease granzyme B. Immunol. Rev. *235*, 105–116.

Afonina, I.S., Tynan, G.A., Logue, S.E., Cullen, S.P., Bots, M., Lüthi, A.U., Reeves, E.P., McElvaney, N.G., Medema, J.P., Lavelle, E.C., and Martin, S.J. (2011). Granzyme B-dependent proteolysis acts as a switch to enhance the proinflammatory activity of IL-1α. Mol. Cell *44*, 265–278.

Aganna, E., Martinon, F., Hawkins, P.N., Ross, J.B., Swan, D.C., Booth, D.R., Lachmann, H.J., Bybee, A., Gaudet, R., Woo, P., et al. (2002). Association of mutations in the NALP3/CIAS1/PYPAF1 gene with a broad phenotype including recurrent fever, cold sensitivity, sensorineural deafness, and AA amyloidosis. Arthritis Rheum. *46*, 2445–2452.

Agostini, L., Martinon, F., Burns, K., McDermott, M.F., Hawkins, P.N., and Tschopp, J. (2004). NALP3 forms an IL-1beta-processing inflammasome with increased activity in Muckle-Wells autoinflammatory disorder. Immunity *20*, 319–325.

Akeda, T., Yamanaka, K., Tsuda, K., Omoto, Y., Gabazza, E.C., and Mizutani, H. (2014). CD8+ T cell granzyme B activates keratinocyte endogenous IL-18. Arch. Dermatol. Res. *306*, 125–130.

Ali, S., Nguyen, D.Q., Falk, W., and Martin, M.U. (2010). Caspase 3 inactivates biologically active full length interleukin-33 as a classical cytokine but does not prohibit nuclear translocation. Biochem. Biophys. Res. Commun. *391*, 1512–1516.

Ali, S., Mohs, A., Thomas, M., Klare, J., Ross, R., Schmitz, M.L., and Martin, M.U. (2011). The dual function cytokine IL-33 interacts with the transcription factor NF-κB to dampen NF-κB-stimulated gene transcription. J. Immunol. *187*, 1609–1616.

Andersson, C.K., Andersson-Sjöland, A., Mori, M., Hallgren, O., Pardo, A., Eriksson, L., Bjermer, L., Löfdahl, C.G., Selman, M., Westergren-Thorsson, G., and Erjefält, J.S. (2011). Activated MCTC mast cells infiltrate diseased lung areas in cystic fibrosis and idiopathic pulmonary fibrosis. Respir. Res. *12*, 139.

Antonopoulos, C., El Sanadi, C., Kaiser, W.J., Mocarski, E.S., and Dubyak, G.R. (2013). Proapoptotic chemotherapeutic drugs induce noncanonical processing and release of IL-1β via caspase-8 in dendritic cells. J. Immunol. *191*, 4789–4803.

Auron, P.E., Webb, A.C., Rosenwasser, L.J., Mucci, S.F., Rich, A., Wolff, S.M., and Dinarello, C.A. (1984). Nucleotide sequence of human monocyte interleukin 1 precursor cDNA. Proc. Natl. Acad. Sci. USA *81*, 7907–7911.

Bae, S., Kang, T., Hong, J., Lee, S., Choi, J., Jhun, H., Kwak, A., Hong, K., Kim, E., Jo, S., and Kim, S. (2012). Contradictory functions (activation/termination) of neutrophil proteinase 3 enzyme (PR3) in interleukin-33 biological activity. J. Biol. Chem. *287*, 8205–8213.

Besnard, A.G., Togbe, D., Guillou, N., Erard, F., Quesniaux, V., and Ryffel, B. (2011). IL-33-activated dendritic cells are critical for allergic airway inflammation. Eur. J. Immunol. *41*, 1675–1686.

Bessa, J., Meyer, C.A., de Vera Mudry, M.C., Schlicht, S., Smith, S.H., Iglesias, A., and Cote-Sierra, J. (2014). Altered subcellular localization of IL-33 leads to non-resolving lethal inflammation. J. Autoimmun. *55*, 33–41.

Black, R.A., Kronheim, S.R., Cantrell, M., Deeley, M.C., March, C.J., Prickett, K.S., Wignall, J., Conlon, P.J., Cosman, D., Hopp, T.P., et al. (1988). Generation of biologically active interleukin-1 beta by proteolytic cleavage of the inactive precursor. J. Biol. Chem. *263*, 9437–9442.

Blumberg, H., Dinh, H., Trueblood, E.S., Pretorius, J., Kugler, D., Weng, N., Kanaly, S.T., Towne, J.E., Willis, C.R., Kuechle, M.K., et al. (2007). Opposing activities of two novel members of the IL-1 ligand family regulate skin inflammation. J. Exp. Med. *204*, 2603–2614.

Bossaller, L., Chiang, P.I., Schmidt-Lauber, C., Ganesan, S., Kaiser, W.J., Rathinam, V.A., Mocarski, E.S., Subramanian, D., Green, D.R., Silverman, N., et al. (2012). Cutting edge: FAS (CD95) mediates noncanonical IL-1β and IL-18 maturation via caspase-8 in an RIP3-independent manner. J. Immunol. *189*, 5508–5512.

Bulau, A.M., Nold, M.F., Li, S., Nold-Petry, C.A., Fink, M., Mansell, A., Schwerd, T., Hong, J., Rubartelli, A., Dinarello, C.A., and Bufler, P. (2014). Role of caspase-1 in nuclear translocation of IL-37, release of the cytokine, and IL-37 inhibition of innate immune responses. Proc. Natl. Acad. Sci. USA *111*, 2650–2655.

Carriere, V., Roussel, L., Ortega, N., Lacorre, D.A., Americh, L., Aguilar, L., Bouche, G., and Girard, J.P. (2007). IL-33, the IL-1-like cytokine ligand for ST2 receptor, is a chromatin-associated nuclear factor in vivo. Proc. Natl. Acad. Sci. USA *104*, 282–287.

Carruth, L.M., Demczuk, S., and Mizel, S.B. (1991). Involvement of a calpain-like protease in the processing of the murine interleukin 1 alpha precursor. J. Biol. Chem. *266*, 12162–12167.

Cayrol, C., and Girard, J.P. (2009). The IL-1-like cytokine IL-33 is inactivated after maturation by caspase-1. Proc. Natl. Acad. Sci. USA *106*, 9021–9026.

Chen, C.J., Kono, H., Golenbock, D., Reed, G., Akira, S., and Rock, K.L. (2007). Identification of a key pathway required for the sterile inflammatory response triggered by dying cells. Nat. Med. *13*, 851–856.

Clavel, G., Thiolat, A., and Boissier, M.C. (2013). Interleukin newcomers creating new numbers in rheumatology: IL-34 to IL-38. Joint Bone Spine *80*, 449–453.

Coeshott, C., Ohnemus, C., Pilyavskaya, A., Ross, S., Wieczorek, M., Kroona, H., Leimer, A.H., and Cheronis, J. (1999). Converting enzyme-independent release of tumor necrosis factor alpha and IL-1beta from a stimulated human monocytic cell line in the presence of activated neutrophils or purified proteinase 3. Proc. Natl. Acad. Sci. USA *96*, 6261–6266.

Cohen, I., Rider, P., Carmi, Y., Braiman, A., Dotan, S., White, M.R., Voronov, E., Martin, M.U., Dinarello, C.A., and Apte, R.N. (2010). Differential release of chromatin-bound IL-1alpha discriminates between necrotic and apoptotic cell death by the ability to induce sterile inflammation. Proc. Natl. Acad. Sci. USA *107*, 2574–2579.

Coll, R.C., Robertson, A.A., Chae, J.J., Higgins, S.C., Muñoz-Planillo, R., Inserra, M.C., Vetter, I., Dungan, L.S., Monks, B.G., Stutz, A., et al. (2015). A small-molecule inhibitor of the NLRP3 inflammasome for the treatment of inflammatory diseases. Nat. Med. *21*, 248–255.

Companjen, A.R., van der Velden, V.H., Vooys, A., Debets, R., Benner, R., and Prens, E.P. (2000). Human keratinocytes are major producers of IL-18: predominant expression of the unprocessed form. Eur. Cytokine Netw. *11*, 383–390.

Creagh, E.M., Conroy, H., and Martin, S.J. (2003). Caspase-activation pathways in apoptosis and immunity. Immunol. Rev. *193*, 10–21.

Cullen, S.P., and Martin, S.J. (2008). Mechanisms of granule-dependent killing. Cell Death Differ. *15*, 251–262.

Dinarello, C.A. (2009). Immunological and inflammatory functions of the interleukin-1 family. Annu. Rev. Immunol. *27*, 519–550.

Dinarello, C.A. (2012). Keep up the heat on IL-1. Blood *120*, 2538–2539.

Dinarello, C.A., Novick, D., Rubinstein, M., and Lonnemann, G. (2003). Interleukin 18 and interleukin 18 binding protein: possible role in immunosuppression of chronic renal failure. Blood Purif. *21*, 258–270.

Dinarello, C.A., Simon, A., and van der Meer, J.W. (2012). Treating inflammation by blocking interleukin-1 in a broad spectrum of diseases. Nat. Rev. Drug Discov. *11*, 633–652.

Fabbi, M., Carbotti, G., and Ferrini, S. (2015). Context-dependent role of IL-18 in cancer biology and counter-regulation by IL-18BP. J. Leukoc. Biol. *97*, 665–675.

Gabay, E., Wolf, G., Shavit, Y., Yirmiya, R., and Tal, M. (2011). Chronic blockade of interleukin-1 (IL-1) prevents and attenuates neuropathic pain behavior and spontaneous ectopic neuronal activity following nerve injury. Eur. J. Pain *15*, 242–248.

Garlanda, C., Dinarello, C.A., and Mantovani, A. (2013). The interleukin-1 family: back to the future. Immunity *39*, 1003–1018.

Gattorno, M., Tassi, S., Carta, S., Delfino, L., Ferlito, F., Pelagatti, M.A., D'Osualdo, A., Buoncompagni, A., Alpigiani, M.G., Alessio, M., et al. (2007). Pattern of interleukin-1beta secretion in response to lipopolysaccharide and ATP before and after interleukin-1 blockade in patients with CIAS1 mutations. Arthritis Rheum. *56*, 3138–3148.

Ghayur, T., Banerjee, S., Hugunin, M., Butler, D., Herzog, L., Carter, A., Quintal, L., Sekut, L., Talanian, R., Paskind, M., et al. (1997). Caspase-1 processes IFN-gamma-inducing factor and regulates LPS-induced IFN-gamma production. Nature *386*, 619–623.

Grimsby, S., Jaensson, H., Dubrovska, A., Lomnytska, M., Hellman, U., and Souchelnytskyi, S. (2004). Proteomics-based identification of proteins interacting with Smad3: SREBP-2 forms a complex with Smad3 and inhibits its transcriptional activity. FEBS Lett. *577*, 93–100.

Gringhuis, S.I., Kaptein, T.M., Wevers, B.A., Theelen, B., van der Vlist, M., Boekhout, T., and Geijtenbeek, T.B. (2012). Dectin-1 is an extracellular pathogen sensor for the induction and processing of IL-1β via a noncanonical caspase-8 inflammasome. Nat. Immunol. *13*, 246–254.

Gu, Y., Kuida, K., Tsutsui, H., Ku, G., Hsiao, K., Fleming, M.A., Hayashi, N., Higashino, K., Okamura, H., Nakanishi, K., et al. (1997). Activation of interferon-gamma inducing factor mediated by interleukin-1beta converting enzyme. Science *275*, 206–209.

Guma, M., Ronacher, L., Liu-Bryan, R., Takai, S., Karin, M., and Corr, M. (2009). Caspase 1-independent activation of interleukin-1beta in neutrophil-predominant inflammation. Arthritis Rheum. *60*, 3642–3650.

Halim, T.Y., Krauss, R.H., Sun, A.C., and Takei, F. (2012). Lung natural helper cells are a critical source of Th2 cell-type cytokines in protease allergen-induced airway inflammation. Immunity *36*, 451–463.

Halim, T.Y., Steer, C.A., Mathä, L., Gold, M.J., Martinez-Gonzalez, I., McNagny, K.M., McKenzie, A.N., and Takei, F. (2014). Group 2 innate lymphoid cells are critical for the initiation of adaptive T helper 2 cell-mediated allergic lung inflammation. Immunity *40*, 425–435.

Hayes, E., Pohl, K., McElvaney, N.G., and Reeves, E.P. (2011). The cystic fibrosis neutrophil: a specialized yet potentially defective cell. Arch. Immunol. Ther. Exp. (Warsz.) *59*, 97–112.

Hazuda, D.J., Strickler, J., Kueppers, F., Simon, P.L., and Young, P.R. (1990). Processing of precursor interleukin 1 beta and inflammatory disease. J. Biol. Chem. *265*, 6318–6322.

Hazuda, D.J., Strickler, J., Simon, P., and Young, P.R. (1991). Structure-function mapping of interleukin 1 precursors. Cleavage leads to a conformational change in the mature protein. J. Biol. Chem. *266*, 7081–7086.

Hernandez-Pigeon, H., Jean, C., Charruyer, A., Haure, M.J., Titeux, M., Tonasso, L., Quillet-Mary, A., Baudouin, C., Charveron, M., and Laurent, G. (2006). Human keratinocytes acquire cellular cytotoxicity under UV-B irradiation. Implication of granzyme B and perforin. J. Biol. Chem. *281*, 13525–13532.

Hernandez-Pigeon, H., Jean, C., Charruyer, A., Haure, M.J., Baudouin, C., Charveron, M., Quillet-Mary, A., and Laurent, G. (2007). UVA induces granzyme B in human keratinocytes through MIF: implication in extracellular matrix remodeling. J. Biol. Chem. *282*, 8157–8164.

Heutinck, K.M., ten Berge, I.J., Hack, C.E., Hamann, J., and Rowshani, A.T. (2010). Serine proteases of the human immune system in health and disease. Mol. Immunol. *47*, 1943–1955.

Hogquist, K.A., Nett, M.A., Unanue, E.R., and Chaplin, D.D. (1991). Interleukin 1 is processed and released during apoptosis. Proc. Natl. Acad. Sci. USA *88*, 8485–8489.

Horai, R., Asano, M., Sudo, K., Kanuka, H., Suzuki, M., Nishihara, M., Takahashi, M., and Iwakura, Y. (1998). Production of mice deficient in genes for interleukin (IL)-1alpha, IL-1beta, IL-1alpha/beta, and IL-1 receptor antagonist shows that IL-1beta is crucial in turpentine-induced fever development and glucocorticoid secretion. J. Exp. Med. *187*, 1463–1475.

Howard, A.D., Chartrain, N., Ding, G.F., Kostura, M.J., Limjuco, G., Schmidt, J.A., and Tocci, M.J. (1991). Probing the role of interleukin-1 beta convertase in interleukin-1 beta secretion. Agents Actions Suppl. *35*, 77–83.

Johnston, A., Xing, X., Guzman, A.M., Riblett, M., Loyd, C.M., Ward, N.L., Wohn, C., Prens, E.P., Wang, F., Maier, L.E., et al. (2011). IL-1F5, -F6, -F8, and -F9: a novel IL-1 family signaling system that is active in psoriasis and promotes keratinocyte antimicrobial peptide expression. J. Immunol. *186*, 2613–2622.

Joosten, L.A., Netea, M.G., Fantuzzi, G., Koenders, M.I., Helsen, M.M., Sparrer, H., Pham, C.T., van der Meer, J.W., Dinarello, C.A., and van den Berg, W.B. (2009). Inflammatory arthritis in caspase 1 gene-deficient mice: contribution of proteinase 3 to caspase 1-independent production of bioactive interleukin-1beta. Arthritis Rheum. *60*, 3651–3662.

Kang, S.J., Wang, S., Hara, H., Peterson, E.P., Namura, S., Amin-Hanjani, S., Huang, Z., Srinivasan, A., Tomaselli, K.J., Thornberry, N.A., et al. (2000). Dual role of caspase-11 in mediating activation of caspase-1 and caspase-3 under pathological conditions. J. Cell Biol. *149*, 613–622.

Kayagaki, N., Warming, S., Lamkanfi, M., Vande Walle, L., Louie, S., Dong, J., Newton, K., Qu, Y., Liu, J., Heldens, S., et al. (2011). Non-canonical inflammasome activation targets caspase-11. Nature *479*, 117–121.

Kobayashi, Y., Yamamoto, K., Saido, T., Kawasaki, H., Oppenheim, J.J., and Matsushima, K. (1990). Identification of calcium-activated neutral protease as a processing enzyme of human interleukin 1 alpha. Proc. Natl. Acad. Sci. USA *87*, 5548–5552.

Kostura, M.J., Tocci, M.J., Limjuco, G., Chin, J., Cameron, P., Hillman, A.G., Chartrain, N.A., and Schmidt, J.A. (1989). Identification of a monocyte specific pre-interleukin 1 beta convertase activity. Proc. Natl. Acad. Sci. USA *86*, 5227–5231.

Kuida, K., Lippke, J.A., Ku, G., Harding, M.W., Livingston, D.J., Su, M.S., and Flavell, R.A. (1995). Altered cytokine export and apoptosis in mice deficient in interleukin-1 beta converting enzyme. Science *267*, 2000–2003.

Kumar, S., Hanning, C.R., Brigham-Burke, M.R., Rieman, D.J., Lehr, R., Khandekar, S., Kirkpatrick, R.B., Scott, G.F., Lee, J.C., Lynch, F.J., et al. (2002). Interleukin-1F7B (IL-1H4/IL-1F7) is processed by caspase-1 and mature IL-1F7B binds to the IL-18 receptor but does not induce IFN-gamma production. Cytokine *18*, 61–71.

Kurt-Jones, E.A., Beller, D.I., Mizel, S.B., and Unanue, E.R. (1985). Identification of a membrane-associated interleukin 1 in macrophages. Proc. Natl. Acad. Sci. USA *82*, 1204–1208.

Kurt-Jones, E.A., Fiers, W., and Pober, J.S. (1987). Membrane interleukin 1 induction on human endothelial cells and dermal fibroblasts. J. Immunol. *139*, 2317–2324.

Lamkanfi, M., and Dixit, V.M. (2014). Mechanisms and functions of inflammasomes. Cell *157*, 1013–1022.

Lefrançais, E., Roga, S., Gautier, V., Gonzalez-de-Peredo, A., Monsarrat, B., Girard, J.P., and Cayrol, C. (2012). IL-33 is processed into mature bioactive forms by neutrophil elastase and cathepsin G. Proc. Natl. Acad. Sci. USA *109*, 1673–1678.

Li, P., Allen, H., Banerjee, S., Franklin, S., Herzog, L., Johnston, C., McDowell, J., Paskind, M., Rodman, L., Salfeld, J., et al. (1995). Mice deficient in IL-1 beta-converting enzyme are defective in production of mature IL-1 beta and resistant to endotoxic shock. Cell *80*, 401–411.

Li, S., Neff, C.P., Barber, K., Hong, J., Luo, Y., Azam, T., Palmer, B.E., Fujita, M., Garlanda, C., Mantovani, A., et al. (2015). Extracellular forms of IL-37 inhibit innate inflammation in vitro and in vivo but require the IL-1 family decoy receptor IL-1R8. Proc. Natl. Acad. Sci. USA *112*, 2497–2502.

Lin, T.J., Garduno, R., Boudreau, R.T., and Issekutz, A.C. (2002). *Pseudomonas aeruginosa* activates human mast cells to induce neutrophil transendothelial migration via mast cell-derived IL-1 alpha and beta. J. Immunol. *169*, 4522–4530.

Liu, X., Hammel, M., He, Y., Tainer, J.A., Jeng, U.S., Zhang, L., Wang, S., and Wang, X. (2013). Structural insights into the interaction of IL-33 with its receptors. Proc. Natl. Acad. Sci. USA *110*, 14918–14923.

Lunding, L., Webering, S., Vock, C., Schröder, A., Raedler, D., Schaub, B., Fehrenbach, H., and Wegmann, M. (2015). IL-37 requires IL-18Rα and SIGIRR/IL-1R8 to diminish allergic airway inflammation in mice. Allergy *70*, 366–373.

Lüthi, A.U., Cullen, S.P., McNeela, E.A., Duriez, P.J., Afonina, I.S., Sheridan, C., Brumatti, G., Taylor, R.C., Kersse, K., Vandenabeele, P., et al. (2009). Suppression of interleukin-33 bioactivity through proteolysis by apoptotic caspases. Immunity *31*, 84–98.

MacKenzie, S.H., Schipper, J.L., and Clark, A.C. (2010). The potential for caspases in drug discovery. Curr. Opin. Drug Discov. Devel. *13*, 568–576.

Maelfait, J., Vercammen, E., Janssens, S., Schotte, P., Haegman, M., Magez, S., and Beyaert, R. (2008). Stimulation of Toll-like receptor 3 and 4 induces interleukin-1beta maturation by caspase-8. J. Exp. Med. *205*, 1967–1973.

March, C.J., Mosley, B., Larsen, A., Cerretti, D.P., Braedt, G., Price, V., Gillis, S., Henney, C.S., Kronheim, S.R., Grabstein, K., et al. (1985). Cloning, sequence and expression of two distinct human interleukin-1 complementary DNAs. Nature *315*, 641–647.

Marrakchi, S., Guigue, P., Renshaw, B.R., Puel, A., Pei, X.Y., Fraitag, S., Zribi, J., Bal, E., Cluzeau, C., Chrabieh, M., et al. (2011). Interleukin-36-receptor antagonist deficiency and generalized pustular psoriasis. N. Engl. J. Med. *365*, 620–628.

Martin, S.J., Henry, C.M., and Cullen, S.P. (2012). A perspective on mammalian caspases as positive and negative regulators of inflammation. Mol. Cell *46*, 387–397.

Matsushima, K., Taguchi, M., Kovacs, E.J., Young, H.A., and Oppenheim, J.J. (1986). Intracellular localization of human monocyte associated interleukin 1 (IL 1) activity and release of biologically active IL 1 from monocytes by trypsin and plasmin. J. Immunol. *136*, 2883–2891.

McNamee, E.N., Masterson, J.C., Jedlicka, P., McManus, M., Grenz, A., Collins, C.B., Nold, M.F., Nold-Petry, C., Bufler, P., Dinarello, C.A., and Rivera-Nieves, J. (2011). Interleukin 37 expression protects mice from colitis. Proc. Natl. Acad. Sci. USA *108*, 16711–16716.

Metkar, S.S., Menaa, C., Pardo, J., Wang, B., Wallich, R., Freudenberg, M., Kim, S., Raja, S.M., Shi, L., Simon, M.M., and Froelich, C.J. (2008). Human and mouse granzyme A induce a proinflammatory cytokine response. Immunity *29*, 720–733.

Mizutani, H., Schechter, N., Lazarus, G., Black, R.A., and Kupper, T.S. (1991). Rapid and specific conversion of precursor interleukin 1 beta (IL-1

beta) to an active IL-1 species by human mast cell chymase. J. Exp. Med. *174*, 821–825.

Mosley, B., Dower, S.K., Gillis, S., and Cosman, D. (1987a). Determination of the minimum polypeptide lengths of the functionally active sites of human interleukins 1 alpha and 1 beta. Proc. Natl. Acad. Sci. USA *84*, 4572–4576.

Mosley, B., Urdal, D.L., Prickett, K.S., Larsen, A., Cosman, D., Conlon, P.J., Gillis, S., and Dower, S.K. (1987b). The interleukin-1 receptor binds the human interleukin-1 alpha precursor but not the interleukin-1 beta precursor. J. Biol. Chem. *262*, 2941–2944.

Netea, M.G., Joosten, L.A., Lewis, E., Jensen, D.R., Voshol, P.J., Kullberg, B.J., Tack, C.J., van Krieken, H., Kim, S.H., Stalenhoef, A.F., et al. (2006). Deficiency of interleukin-18 in mice leads to hyperphagia, obesity and insulin resistance. Nat. Med. *12*, 650–656.

Niki, Y., Yamada, H., Kikuchi, T., Toyama, Y., Matsumoto, H., Fujikawa, K., and Tada, N. (2004). Membrane-associated IL-1 contributes to chronic synovitis and cartilage destruction in human IL-1 alpha transgenic mice. J. Immunol. *172*, 577–584.

Nold, M.F., Nold-Petry, C.A., Zepp, J.A., Palmer, B.E., Bufler, P., and Dinarello, C.A. (2010). IL-37 is a fundamental inhibitor of innate immunity. Nat. Immunol. *11*, 1014–1022.

Nold-Petry, C.A., Lo, C.Y., Rudloff, I., Elgass, K.D., Li, S., Gantier, M.P., Lotz-Havla, A.S., Gersting, S.W., Cho, S.X., Lao, J.C., et al. (2015). IL-37 requires the receptors IL-18Rα and IL-1R8 (SIGIRR) to carry out its multifaceted anti-inflammatory program upon innate signal transduction. Nat. Immunol. *16*, 354–365.

Novick, D., Kim, S., Kaplanski, G., and Dinarello, C.A. (2013). Interleukin-18, more than a Th1 cytokine. Semin. Immunol. *25*, 439–448.

Oboki, K., Ohno, T., Kajiwara, N., Saito, H., and Nakae, S. (2010). IL-33 and IL-33 receptors in host defense and diseases. Allergol. Int. *59*, 143–160.

Okamura, H., Nagata, K., Komatsu, T., Tanimoto, T., Nukata, Y., Tanabe, F., Akita, K., Torigoe, K., Okura, T., Fukuda, S., et al. (1995). A novel costimulatory factor for gamma interferon induction found in the livers of mice causes endotoxic shock. Infect. Immun. *63*, 3966–3972.

Omoto, Y., Tokime, K., Yamanaka, K., Habe, K., Morioka, T., Kurokawa, I., Tsutsui, H., Yamanishi, K., Nakanishi, K., and Mizutani, H. (2006). Human mast cell chymase cleaves pro-IL-18 and generates a novel and biologically active IL-18 fragment. J. Immunol. *177*, 8315–8319.

Omoto, Y., Yamanaka, K., Tokime, K., Kitano, S., Kakeda, M., Akeda, T., Kurokawa, I., Gabazza, E.C., Tsutsui, H., Katayama, N., et al. (2010). Granzyme B is a novel interleukin-18 converting enzyme. J. Dermatol. Sci. *59*, 129–135.

Overed-Sayer, C., Rapley, L., Mustelin, T., and Clarke, D.L. (2013). Are mast cells instrumental for fibrotic diseases? Front. Pharmacol. *4*, 174.

Pan, G., Risser, P., Mao, W., Baldwin, D.T., Zhong, A.W., Filvaroff, E., Yansura, D., Lewis, L., Eigenbrot, C., Henzel, W.J., and Vandlen, R. (2001). IL-1H, an interleukin 1-related protein that binds IL-18 receptor/IL-1Rrp. Cytokine *13*, 1–7.

Pei, C., Barbour, M., Fairlie-Clarke, K.J., Allan, D., Mu, R., and Jiang, H.R. (2014). Emerging role of interleukin-33 in autoimmune diseases. Immunology *141*, 9–17.

Petrella, B.L., Armstrong, D.A., and Vincenti, M.P. (2012). Interleukin-1 beta and transforming growth factor-beta 3 cooperate to activate matrix metalloproteinase expression and invasiveness in A549 lung adenocarcinoma cells. Cancer Lett. *325*, 220–226.

Priestle, J.P., Schär, H.P., and Grütter, M.G. (1988). Crystal structure of the cytokine interleukin-1 beta. EMBO J. *7*, 339–343.

Rider, P., Carmi, Y., Guttman, O., Braiman, A., Cohen, I., Voronov, E., White, M.R., Dinarello, C.A., and Apte, R.N. (2011). IL-1α and IL-1β recruit different myeloid cells and promote different stages of sterile inflammation. J. Immunol. *187*, 4835–4843.

Roussel, L., Erard, M., Cayrol, C., and Girard, J.P. (2008). Molecular mimicry between IL-33 and KSHV for attachment to chromatin through the H2A-H2B acidic pocket. EMBO Rep. *9*, 1006–1012.

Roy, A., Ganesh, G., Sippola, H., Bolin, S., Sawesi, O., Dagälv, A., Schlenner, S.M., Feyerabend, T., Rodewald, H.R., Kjellén, L., et al. (2014). Mast cell chymase degrades the alarmins heat shock protein 70, biglycan, HMGB1, and interleukin-33 (IL-33) and limits danger-induced inflammation. J. Biol. Chem. *289*, 237–250.

Rudolphi, K., Gerwin, N., Verzijl, N., van der Kraan, P., and van den Berg, W. (2003). Pralnacasan, an inhibitor of interleukin-1beta converting enzyme, reduces joint damage in two murine models of osteoarthritis. Osteoarthritis Cartilage *11*, 738–746.

Sasu, S., Cooper, A.L., and Beasley, D. (2001). Juxtacrine effects of IL-1 alpha precursor promote iNOS expression in vascular smooth muscle cells. Am. J. Physiol. Heart Circ. Physiol. *280*, H1615–H1623.

Schmitz, J., Owyang, A., Oldham, E., Song, Y., Murphy, E., McClanahan, T.K., Zurawski, G., Moshrefi, M., Qin, J., Li, X., et al. (2005). IL-33, an interleukin-1-like cytokine that signals via the IL-1 receptor-related protein ST2 and induces T helper type 2-associated cytokines. Immunity *23*, 479–490.

Sharma, S., Kulk, N., Nold, M.F., Gräf, R., Kim, S.H., Reinhardt, D., Dinarello, C.A., and Bufler, P. (2008). The IL-1 family member 7b translocates to the nucleus and down-regulates proinflammatory cytokines. J. Immunol. *180*, 5477–5482.

Shenderov, K., Riteau, N., Yip, R., Mayer-Barber, K.D., Oland, S., Hieny, S., Fitzgerald, P., Oberst, A., Dillon, C.P., Green, D.R., et al. (2014). Cutting edge: Endoplasmic reticulum stress licenses macrophages to produce mature IL-1β in response to TLR4 stimulation through a caspase-8- and TRIF-dependent pathway. J. Immunol. *192*, 2029–2033.

Smith, D.E., Renshaw, B.R., Ketchem, R.R., Kubin, M., Garka, K.E., and Sims, J.E. (2000). Four new members expand the interleukin-1 superfamily. J. Biol. Chem. *275*, 1169–1175.

Sugawara, S., Uehara, A., Nochi, T., Yamaguchi, T., Ueda, H., Sugiyama, A., Hanzawa, K., Kumagai, K., Okamura, H., and Takada, H. (2001). Neutrophil proteinase 3-mediated induction of bioactive IL-18 secretion by human oral epithelial cells. J. Immunol. *167*, 6568–6575.

Sugiura, K., Shoda, Y., and Akiyama, M. (2014). Generalized pustular psoriasis triggered by amoxicillin in monozygotic twins with compound heterozygous IL36RN mutations: comment on the article by Navarini et al. J. Invest. Dermatol. *134*, 578–579.

Talabot-Ayer, D., Lamacchia, C., Gabay, C., and Palmer, G. (2009). Interleukin-33 is biologically active independently of caspase-1 cleavage. J. Biol. Chem. *284*, 19420–19426.

Thornberry, N.A., Bull, H.G., Calaycay, J.R., Chapman, K.T., Howard, A.D., Kostura, M.J., Miller, D.K., Molineaux, S.M., Weidner, J.R., Aunins, J., et al. (1992). A novel heterodimeric cysteine protease is required for interleukin-1 beta processing in monocytes. Nature *356*, 768–774.

Towne, J.E., Renshaw, B.R., Douangpanya, J., Lipsky, B.P., Shen, M., Gabel, C.A., and Sims, J.E. (2011). Interleukin-36 (IL-36) ligands require processing for full agonist (IL-36α, IL-36β, and IL-36γ) or antagonist (IL-36Ra) activity. J. Biol. Chem. *286*, 42594–42602.

Tsutsui, H., Kayagaki, N., Kuida, K., Nakano, H., Hayashi, N., Takeda, K., Matsui, K., Kashiwamura, S., Hada, T., Akira, S., et al. (1999). Caspase-1-independent, Fas/Fas ligand-mediated IL-18 secretion from macrophages causes acute liver injury in mice. Immunity *11*, 359–367.

Ushio, S., Namba, M., Okura, T., Hattori, K., Nukada, Y., Akita, K., Tanabe, F., Konishi, K., Micallef, M., Fujii, M., et al. (1996). Cloning of the cDNA for human IFN-gamma-inducing factor, expression in Escherichia coli, and studies on the biologic activities of the protein. J. Immunol. *156*, 4274–4279.

van Raam, B.J., and Salvesen, G.S. (2012). Proliferative versus apoptotic functions of caspase-8 Hetero or homo: the caspase-8 dimer controls cell fate. Biochim. Biophys. Acta *1824*, 113–122.

Voronov, E., Carmi, Y., and Apte, R.N. (2014). The role IL-1 in tumor-mediated angiogenesis. Front. Physiol. *5*, 114.

Waern, I., Lundequist, A., Pejler, G., and Wernersson, S. (2013). Mast cell chymase modulates IL-33 levels and controls allergic sensitization in dust-mite induced airway inflammation. Mucosal Immunol. *6*, 911–920.

Werman, A., Werman-Venkert, R., White, R., Lee, J.K., Werman, B., Krelin, Y., Voronov, E., Dinarello, C.A., and Apte, R.N. (2004). The precursor form of IL-1alpha is an intracrine proinflammatory activator of transcription. Proc. Natl. Acad. Sci. USA *101*, 2434–2439.

Wessendorf, J.H., Garfinkel, S., Zhan, X., Brown, S., and Maciag, T. (1993). Identification of a nuclear localization sequence within the structure of the human interleukin-1 alpha precursor. J. Biol. Chem. *268*, 22100–22104.

Yoshifuji, H., Umehara, H., Maruyama, H., Itoh, M., Tanaka, M., Kawabata, D., Fujii, T., and Mimori, T. (2005). Amelioration of experimental arthritis by a calpain-inhibitory compound: regulation of cytokine production by E-64-d in vivo and in vitro. Int. Immunol. *17*, 1327–1336.

Zheng, H., Fletcher, D., Kozak, W., Jiang, M., Hofmann, K.J., Conn, C.A., Soszynski, D., Grabiec, C., Trumbauer, M.E., Shaw, A., et al. (1995). Resistance to fever induction and impaired acute-phase response in interleukin-1 beta-deficient mice. Immunity *3*, 9–19.

Zheng, Y., Humphry, M., Maguire, J.J., Bennett, M.R., and Clarke, M.C. (2013). Intracellular interleukin-1 receptor 2 binding prevents cleavage and activity of interleukin-1α, controlling necrosis-induced sterile inflammation. Immunity *38*, 285–295.