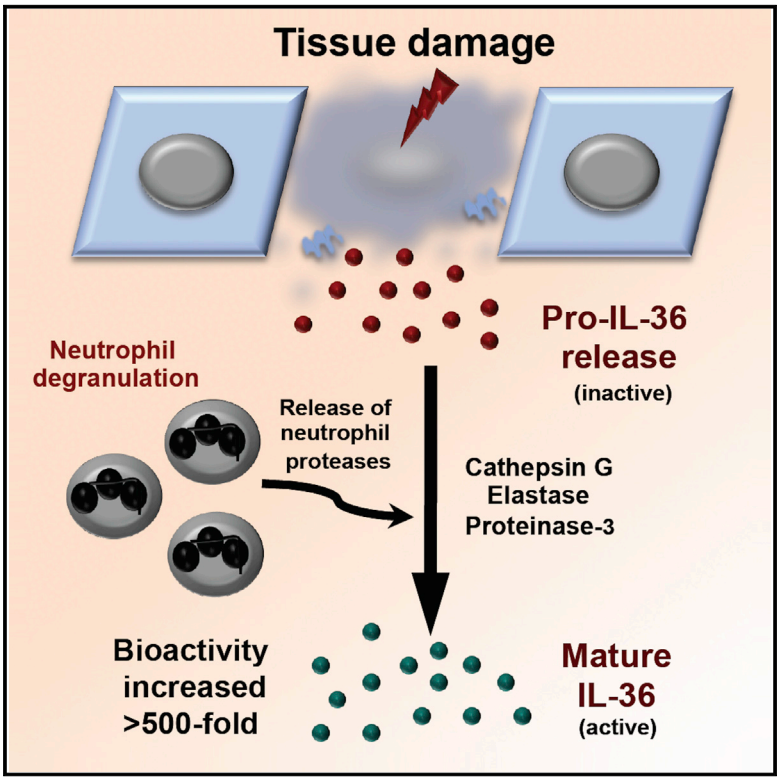
Article

Cell Reports

Neutrophil-Derived Proteases Escalate Inflammation through Activation of IL-36 Family Cytokines

Graphical Abstract



Highlights

- Neutrophils can escalate inflammation via processing of extracellular cytokines
- Activated neutrophils liberate proteases that can process IL-36 family cytokines
- IL-36α, β, and γ are activated differentially by cathepsin G, elastase, or proteinase-3
- Cathepsin G activity is elevated in human psoriatic lesions

Authors

Conor M. Henry, Graeme P. Sullivan, Danielle M. Clancy, Inna S. Afonina, Dagmar Kulms, Seamus J. Martin

Correspondence

martinsj@tcd.ie

In Brief

IL-36 cytokines require proteolytic processing for activation, but how this is achieved is unknown. Here, Henry et al. show that proteases liberated from activated neutrophils—cathepsin G, elastase, and proteinase-3—differentially process and activate all three IL-36 family members. Therefore, neutrophil-derived proteases can escalate inflammation through processing of extracellular cytokines.

Henry et al., 2016, Cell Reports *14*, 708–722
February 2, 2016
http://dx.doi.org/10.1016/j.celrep.2015.12.072

OPEN ACCESS
CellPress

Cell Reports
Article

Neutrophil-Derived Proteases Escalate Inflammation through Activation of IL-36 Family Cytokines

Conor M. Henry,[1] Graeme P. Sullivan,[1] Danielle M. Clancy,[1] Inna S. Afonina,[1,3] Dagmar Kulms,[2] and Seamus J. Martin[1,*]
[1]Molecular Cell Biology Laboratory, Department of Genetics, The Smurfit Institute, Trinity College, Dublin 2, Ireland
[2]Experimental Dermatology, Department of Dermatology, Dresden University of Technology, 01307 Dresden, Germany
[3]Present address: Unit of Molecular Signal Transduction in Inflammation, VIB Inflammation Research Center, Department for Biomedical Molecular Biology, University of Gent, Technologiepark 927, 9052 Gent, Belgium
*Correspondence: martinsj@tcd.ie
http://dx.doi.org/10.1016/j.celrep.2015.12.072

SUMMARY

Recent evidence has strongly implicated the IL-1 family cytokines IL-36α, IL-36β, and IL-36γ as key initiators of skin inflammation. Similar to the other members of the IL-1 family, IL-36 cytokines are expressed as inactive precursors and require proteolytic processing for activation; however, the responsible proteases are unknown. Here, we show that IL-36α, IL-36β, and IL-36γ are activated differentially by the neutrophil granule-derived proteases cathepsin G, elastase, and proteinase-3, increasing their biological activity ~500-fold. Active IL-36 promoted a strong pro-inflammatory signature in primary keratinocytes and was sufficient to perturb skin differentiation in a reconstituted 3D human skin model, producing features resembling psoriasis. Furthermore, skin eluates from psoriasis patients displayed significantly elevated cathepsin G-like activity that was sufficient to activate IL-36β. These data identify neutrophil granule proteases as potent IL-36-activating enzymes, adding to our understanding of how neutrophils escalate inflammatory reactions. Inhibition of neutrophil-derived proteases may therefore have therapeutic benefits in psoriasis.

INTRODUCTION

Interleukin 1 (IL-1) family cytokines, which include the recently described IL-36α, β, and γ proteins, play major roles as initiators of inflammation and are frequently among the first cytokines produced in response to infection or injury (Kono and Rock, 2008; Sims and Smith, 2010; Afonina et al., 2015). IL-1 family cytokines are capable of triggering complex cascades of additional cytokine production from diverse cell types, such as resident tissue macrophages and dendritic cells as well as keratinocytes and endothelial cells lining local blood vessels (Towne et al., 2004; Dinarello, 2009; Vigne et al., 2011, 2012; Milovanovic et al., 2012). IL-36α, IL-36β, and IL-36γ are encoded by distinct genes, and evidence is accumulating rapidly to suggest that these cytokines play a key role in skin inflammation, particularly in psoriasis (Blumberg et al., 2007, 2010; Johnston et al., 2011; Marrakchi et al., 2011; Tortola et al., 2012; Towne and Sims, 2012; Farooq et al., 2013; Kanazawa et al., 2013).

Individuals that carry hypomorphic mutations in the IL-36 receptor antagonist (IL-36RA) display a severe and highly debilitating form of psoriasis called generalized pustular psoriasis (Marrakchi et al., 2011; Farooq et al., 2013; Kanazawa et al., 2013). This suggests that deregulated IL-36 cytokine signaling is sufficient to drive aggressive skin inflammation and also that IL-36 is an important barrier cytokine. Analysis of IL-36 mRNA expression in skin biopsies from individuals with the most common form of psoriasis, psoriasis vulgaris, found dramatically elevated expression (100-fold) of all three IL-36 transcripts compared with non-lesional skin from the same individuals or non-affected controls (Blumberg et al., 2010; Johnston et al., 2011). Consistent with the idea that elevated IL-36 activity is an initiating event in psoriasis, transgenic expression of IL-36α in the mouse leads to a psoriasis-like condition at birth that can be exacerbated further with the skin irritant phorbol acetate (Blumberg et al., 2007). Moreover, application of a Toll receptor agonist (imiquimod) to the skin of humans and mice can provoke psoriasis outbreaks (Wu et al., 2004; Tortola et al., 2012) that are increased in severity in *IL-36RA*$^{-/-}$ mice (Tortola et al., 2012). Furthermore, imiquimod-induced psoriasis in mouse models is abolished completely on an *IL-36R*$^{-/-}$ background (Tortola et al., 2012). Finally, transplantation of human psoriatic lesions onto immunodeficient (severe combined immunodeficiency) mice produces a psoriasis-like condition that is greatly improved through blocking the IL-36 receptor (Blumberg et al., 2010).

IL-36α, IL-36β, and IL-36γ are all generated as leaderless cytokines that lack biological activity (Towne et al., 2011). Therefore, proteolytic processing of IL-36 cytokines is required to unleash their pro-inflammatory activity, similar to other members of the IL-1 family, such as IL-1β and IL-18 (Afonina et al., 2015). Sims and colleagues (Towne et al., 2011) have shown that removal of a small number of residues from the N termini of IL-36α, IL-36β, and IL-36γ increases their biological activity by more than 10,000-fold. Because IL-36 cytokines appear to play a key role as initiators of inflammation in the skin barrier, inhibitors of IL-36 proteolytic activation may have considerable potential for the treatment of inflammatory skin conditions.