

Interleukin-33

Interleukin-33 [1,2] (IL-33; HF-NEV [3]; IL-1F11 [4]), a member of the IL-1 family of cytokines, is expressed by many cell types following pro-inflammatory stimulation and is thought to be released on cell lysis. The predicted 270-amino acid human pro-IL-33 has a calculated molecular mass of 30 kDa. Whether pro-IL-33 needs to be cleaved to be active and whether caspase-1 is required is still a matter of controversy. However, it has been reported that IL-33 is biologically active in a caspase-1 independent manner [5]. The heterodimeric IL-33 receptor complex consists of ST2 [6] (interleukin-1 receptor-like 1 [7]; IL1RL1; DER4 [8]; Fit-1 [9]; T1 [10]) and IL-1 receptor accessory protein [11] (IL-1RAP) and mediates signaling through the TIR domain of IL-1RAP. Binding of IL-33 to its receptor results in the recruitment of MyD88, IRAK1, and IRAK4 to the receptor complex, activating NF- κ B, I κ B α and numerous MAPKs. Cell-specific variations in the IL-33 induced signaling pathway have been described, such as the requirement of TRAF6 for IRAK recruitment in fibroblasts. In addition, IL-33 may function as a transcriptional repressor mediated by the N-terminal homeobox domain [3]. Soluble ST2 can bind IL-33 directly and acts as a decoy receptor. Taken together IL-33 may be a dual function cytokine with both extracellular and intracellular signaling, a property it shares with IL-1 α . IL-33 affects the various cell types that express membrane ST2. ST2 is selectively expressed by Th2 but not Th1 cells. IL-33 is described as a modulator of inflammation, mediating Th2 immune responses. Administration of purified IL-33 *in vitro* and *in vivo* induces Th2-associated cytokines such as IL-5 and IL-13 and reduces production of IFN- γ from Th1 cells. IL-33 is a potent inducer of pro-inflammatory cytokines and chemokines.

For a latest review summarizing IL-33 signaling and cellular targets see: *Disease-associated functions of IL-33: the new kid in the IL-1 family*: F. Y. Liew, et al.; Nat. Rev. Immunology **10**, 103 (2010).

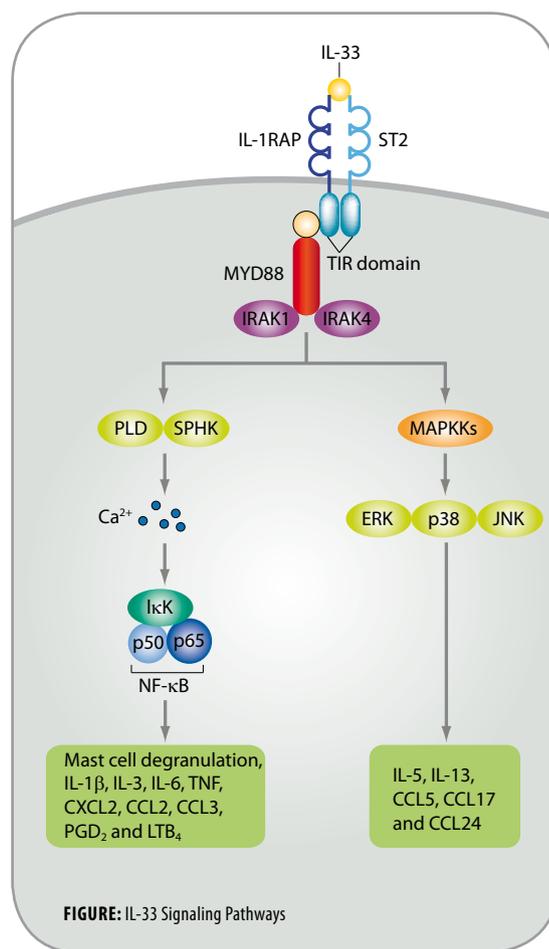


FIGURE: IL-33 Signaling Pathways

The Role of IL-33 in Disease

The ability of IL-33 to target numerous immune cell types, like Th2-like cells, mast cells and B1 cells, and to induce cytokine and chemokine production underlines its potential in influencing the outcome of a wide range of diseases.

Arthritis

IL-33 has recently been found to be involved in the pathogenesis of chronic inflammatory arthritis like its other family members, IL-1 and IL-18. IL-33 and its receptor, ST2, are increased in the synovia of patients with rheumatoid arthritis [12,13]. In a mouse model of collagen-induced arthritis (CIA), blocking the function of IL-33 results in a decreased disease severity [13]. IL-33 exacerbates autoantibody-induced arthritis (AIA) in a mast-cell dependent signaling pathway [14]. In summary, locally produced IL-33 contributes to the pathogenesis of joint inflammation and destruction [13]. Therefore, blocking IL-33 signaling may offer a novel strategy against rheumatoid arthritis.

Only recently, it has been shown that none of healthy individuals yielded detectable ranges of serum IL-33 recovery, whereas serum IL-33 could be detected in rheumatoid arthritis patients. These data clearly indicate that IL-33 serum levels may be an important biomarker for human rheumatoid arthritis.

Asthma

In clinical and experimental asthma, IL-33 levels are elevated compared to healthy individuals or wild-type mice [16,17, 18]. IL-33 administrated to the mouse lung induces airway hyperresponsiveness (AHR) and eosinophilic inflammation [16,19]. Administrated IL-33 not only induces features of asthma in animals, but can also exacerbate experimental asthma [20]. Depending on the model, the disease can be attenuated by blocking IL-33 or ST2 [20,21,22,23]. In summary, during the development or exacerbation of certain types of airway inflammation, IL-33 likely exhibits a critical role.

Atopic Allergy and Anaphylaxis

Elevated levels of IgE antibodies are characteristic for anaphylactic shock and anaphylaxis. IL-33 was shown to activate and directly induce degranulation of IgE-sensitized mast cells [24]. This may have clinical importance, as human serum levels of IL-33 are increased in atopic patients during anaphylactic shock, and in inflamed skin tissue of atopic dermatitis patients [24]. Thus, IL-33 could be a therapeutic target for anaphylaxis.

Cardiovascular Disease/Atherosclerosis

A possible association between IL-33 and cardiovascular disease has been highlighted by the finding that soluble ST2 levels are increased in the blood of mice following myocardial infarction [25]. IL-33 and ST2 are expressed by vascular endothelial and vascular smooth muscle cells of the heart and aorta in mice and humans [26]. IL-33-treated mice produces significantly higher levels of antibodies specific for oxidized low-density lipoprotein (oxLDL), which are atheroprotective, in an IL-5-dependent manner [26]. IL-33 thus provides a new therapeutic approach for the treatment or prevention of atherosclerotic vascular diseases.

Nervous System Diseases

Various cells and tissues in the central nervous system express IL-33 and ST2 [27,28]. IL-33 may have a pathogenic role in inflammatory diseases of the CNS, such as experimental autoimmune encephalomyelitis (EAE). In the brain of patients with Alzheimer's disease IL-33 expression is decreased compared with controls, eventually due to the downregulation of β -amyloid peptide secretion by IL-33 [29]. Furthermore, IL-33 is linked to inflammatory pain in the peripheral nervous system [30].

Sepsis

During infection by bacteria, sepsis can develop when the host response fails to contain the infection. This leads to widespread inflammation and organ failure. Treatments against sepsis have limited success. IL-33 has been shown to attenuate sepsis by increasing neutrophil influx to the site of infection [31]. It prevents the down-regulation of CXCR2 and chemotaxis in circulating neutrophils. Patients with severe sepsis have reduced levels of IL-33.

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IL-33 Proteins

IL-33 (human) (rec.)

AG-40B-0038-C010		10 µg
AG-40B-0038-5010	MultiPack	5 x 10 µg
AG-40B-0038AA-C500	BULK	500 µg

Produced in *E. coli*. Untagged human IL-33 (aa 112-270). **SPECIFICITY:** Binds to human ST2. **BIOLOGICAL ACTIVITY:** Activates the human ST2-dependent NF-κB pathway. Does not activate mouse ST2-dependent NF-κB.

LIT: Epithelial-derived IL-33 and its receptor ST2 are dysregulated in ulcerative colitis and in experimental Th1/Th2 driven enteritis: L. Pastorelli, et al; PNAS 107, 8017 (2010)

IL-33 (mouse) (rec.)

AG-40B-0041-C010		10 µg
AG-40B-0041-5050	MultiPack	5 x 10 µg
AG-40B-0041AA-C500	BULK	500 µg

Produced in *E. coli*. Untagged mouse IL-33 (aa 109-266). **SPECIFICITY:** Binds to mouse and human ST2. **BIOLOGICAL ACTIVITY:** Activates the mouse and human ST2-dependent NF-κB pathway.

LIT: Epithelial-derived IL-33 and its receptor ST2 are dysregulated in ulcerative colitis and in experimental Th1/Th2 driven enteritis: L. Pastorelli, et al; PNAS 107, 8017 (2010)

IL-33 (human) (rec.)

AG-40A-0054-C010		10 µg
AG-40A-0054-C050		50 µg

Produced in HEK 293 cells. Recombinant human IL-33 (aa 112-270) fused at the C-terminus to a FLAG®-tag. **SPECIFICITY:** Binds to human ST2. **BIOLOGICAL ACTIVITY:** Activates the human ST2-dependent NF-κB pathway.

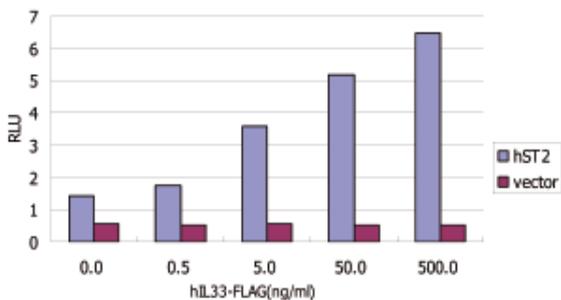


FIGURE: Activation of a human ST2-dependent NF-κB pathway. HEK 293 cells were transiently co-transfected with 500 ng of human ST2-containing vector (or empty vector as a negative control), a vector containing NF-κB-driven firefly luciferase, and a vector containing Renilla luciferase as an internal control in a 12-well plate. At 40hr after transfection, cells were treated with IL33 (human) (rec.) (Prod. No. AG-40A-0054) at the concentration as indicated for 8hr, followed by measurement of a dual luciferase assay.

IL-33 (human) (homeodomain-like) (rec.) (His)

AG-40A-0162-C010		10 µg
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Produced in *E. coli*. Recombinant homeodomain of human IL-33 (aa 1-111) fused at the C-terminus to a His-tag.

IL-33 (human) (rec.) (His)

AG-40A-0042-C010		10 µg
AG-40A-0042-C050		50 µg

Produced in *E. coli*. Recombinant human IL-33 (aa 112-270) fused at the C-terminus to a His-tag. **SPECIFICITY:** Binds to human ST2. **BIOLOGICAL ACTIVITY:** Activates the human ST2-dependent NF-κB pathway.

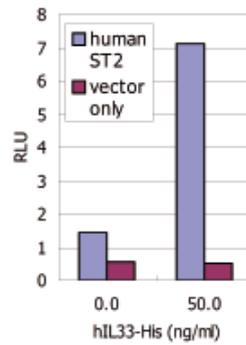


FIGURE: Activation of a human ST2-dependent NF-κB pathway. HEK 293 cells were transiently co-transfected with 500 ng of human ST2-containing vector (or empty vector as a negative control), a vector containing NF-κB-driven firefly luciferase, and a vector containing Renilla luciferase as an internal control in 12-well plate. At 40hr after transfection, cells were treated with IL33 (human) (rec.) (His) (Prod. No. AG-40A-0042) at the concentration as indicated for 8hr, followed by measurement of a dual luciferase assay.

IL-33 (mouse) (rec.) (His)

AG-40A-0053-C010		10 µg
AG-40A-0053-C050		50 µg

Produced in *E. coli*. Recombinant mouse IL-33 (aa 109-266) fused at the C-terminus to a His-tag. **SPECIFICITY:** Binds to human ST2. **BIOLOGICAL ACTIVITY:** Activates the human ST2-dependent NF-κB pathway.

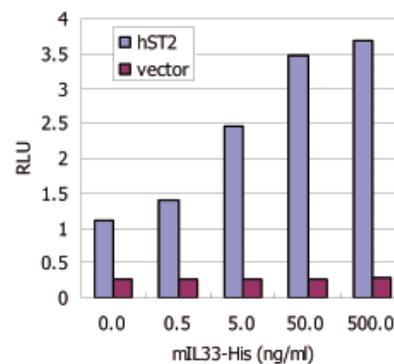


FIGURE: Activation of a human ST2-dependent NF-κB pathway. HEK 293 cells were transiently co-transfected with 500 ng of human ST2-containing vector (or empty vector as a negative control), a vector containing NF-κB-driven firefly luciferase, and a vector containing Renilla luciferase as an internal control in a 12-well plate. At 40hr after transfection, cells were treated with IL33 (mouse) (rec.) (His) (Prod. No. AG-40A-0053) at the concentration as indicated for 24hr, followed by measurement of a dual luciferase assay.

IL-33 Antibodies

The Standard!

anti-IL-33 (human), mAb (IL33305B)

AG-20A-0041-C050 50 µg
AG-20A-0041-C100 100 µg

CLONE: IL33305B. **ISOTYPE:** Mouse IgG2a. **IMMUNOGEN:** Recombinant human IL-33. **SPECIFICITY:** Recognizes human IL-33. Does not cross-react with mouse IL-33. **APPLICATION:** IHC, IP, WB.

LIT: Inhibition of Interleukin-33 Signaling Attenuates the Severity of Experimental Arthritis: G. Palmer, et al.; *Arthritis Rheum.* 60, 738 (2009) ■ Interleukin-33 Is Biologically Active Independently of Caspase-1 Cleavage: D. Talbot-Ayer, et al.; *JBC* 284, 19420 (2009)

FIGURE: Immunohistochemical staining of human IL-33 with anti-IL-33 (human), mAb (IL33305B) (Prod. No. AG-20A-0041) in human tissue (1:100 dilution). **A.** Immunoperoxidase staining of formalin-fixed, paraffin-embedded human heart showing cytoplasmic staining (100X, Brown color). **B.** Immunohistochemical staining with isotype control IgG2a in human heart (negative control).



anti-IL-33, mAb (IL33068A)

AG-20A-0042-C050 50 µg
AG-20A-0042-C100 100 µg

CLONE: IL33068A. **ISOTYPE:** Mouse IgG2b. **IMMUNOGEN:** Recombinant human IL-33. **SPECIFICITY:** Recognizes human IL-33. Weakly cross-reacts with mouse IL-33. **APPLICATION:** WB.

anti-IL-33, mAb (IL33026B)

AG-20A-0043-C050 50 µg
AG-20A-0043-C100 100 µg

CLONE: IL33026B. **ISOTYPE:** Mouse IgG1. **IMMUNOGEN:** Recombinant human IL-33. **SPECIFICITY:** Recognizes human and mouse IL-33. **APPLICATION:** IP, WB.

anti-IL-33 (human), pAb

AG-25A-0045-C100 100 µg
AG-25A-0045B-C050 Biotin 50 µg

From rabbit. **IMMUNOGEN:** Recombinant human IL-33. **SPECIFICITY:** Recognizes human IL-33. **APPLICATION:** WB.

anti-IL-33 (mouse), pAb

AG-25A-0047-C100 100 µg

From rabbit. **IMMUNOGEN:** Recombinant mouse IL-33. **SPECIFICITY:** Recognizes mouse IL-33. Does not cross-react with human IL-33. **APPLICATION:** WB.

Pro-IL-33 (human) (rec.) (His)

AG-40A-0165-C010

10 µg

Produced in *E. coli*. Recombinant human pro-IL-33 (aa 1-270) fused at the C-terminus to a His-tag.

Coming soon!

ST2 Proteins & Antibodies

ST2, Soluble (human) (rec.)

AG-40A-0062-C050 50 µg
Produced in HEK 293 cells. Recombinant soluble ST2 (aa 1-328) fused at the C-terminus to a FLAG®-tag.

anti-ST2 (human), mAb (ST33868)

AG-20A-0044-C050 50 µg
AG-20A-0044-C100 100 µg

CLONE: ST33868. **ISOTYPE:** Mouse IgG1. **IMMUNOGEN:** Recombinant soluble human ST2. **SPECIFICITY:** Recognizes human ST2. **APPLICATION:** WB.

anti-ST2 (human), pAb

AG-25A-0058-C100 100 µg

From rabbit. **IMMUNOGEN:** Recombinant soluble human ST2. **SPECIFICITY:** Recognizes human ST2. **APPLICATION:** WB.

ST2 (human):Fc (human), Soluble (rec.)

AG-40A-0059-C050 50 µg

Produced in HEK 293 cells. Recombinant soluble ST2 (aa 1-328) fused at the C-terminus to the Fc portion of human IgG1. **BIOLOGICAL ACTIVITY:** Interacts with human IL-33.

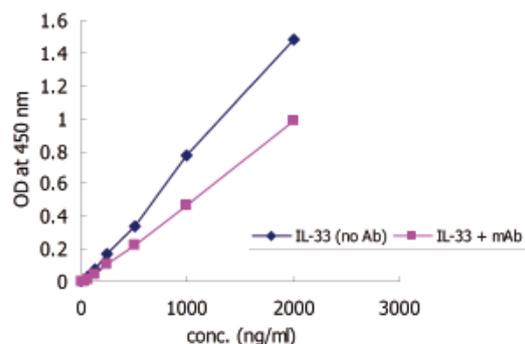


FIGURE Specific binding of IL-33 (human) (rec.) (Prod. No. AG-A40-0054) to ST2 (human):Fc (human), Soluble (rec.) (Prod. No. AG-40A-0059) *in vitro*. An indirect competitive ELISA was performed as follows; 1 coat microtiter plate wells with hST2-Fc (10 µg/ml); 2) add varying concentrations of hIL-33 with or without a hIL-33 mAb to the wells followed by washing; 3) add anti-FLAG HRP conjugated (1:2,000) to an enzyme; 4) After adding the TMB solution, incubate at RT in the dark for 10 to 45 minutes. Immediately read the plate at 450 nm.

NEW High-Sensitivity IL-33 (human) ELISA Kit

AG-45A-0033EK-KI01 96 wells
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AG-45A-0033PP-KI01 5 x 96 wells

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RANGE: 0.016 to 1 ng/ml
DETECTION TYPE: Colorimetric
ASSAY TYPE: Sandwich

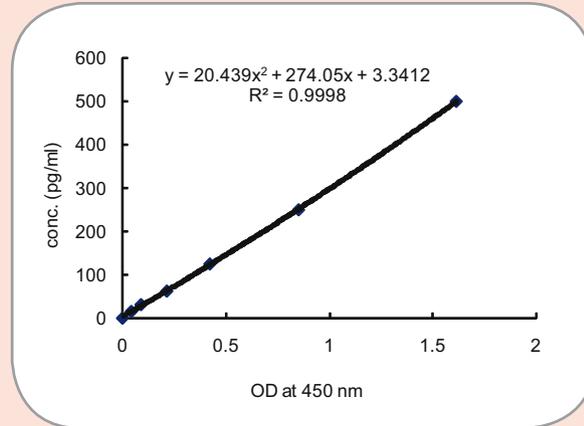
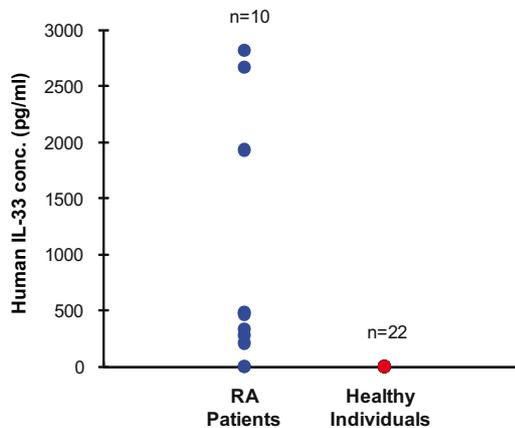


FIGURE: Mean values of the human IL-33 concentrations from 10 RA (rheumatoid arthritis) patients and 22 healthy individuals, detected with IL-33 (human) ELISA Kit (Prod. No. AG-45A-0033EK).

None of the healthy sera yielded detectable ranges of serum IL-33 recovery, whereas 7 out of the 10 RA sera showed a significant range of serum IL-33 recovery. The data clearly show that IL-33 may be an important biomarker for human rheumatoid arthritis.



This ELISA Kit is for the quantitative determination of IL-33 in human serum, plasma or cell culture supernatants.

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