

## DESCRIPTION

<b>Species Reactivity</b>	Human
<b>Specificity</b>	Detects human IL-33 in direct ELISAs and Western blots.
<b>Source</b>	Recombinant Monoclonal Goat IgG Clone # 40015C
<b>Purification</b>	Protein A or G purified from cell culture supernatant
<b>Immunogen</b>	<i>E. coli</i> -derived recombinant human IL-33 Ser112-Thr270 Accession # O95760
<b>Formulation</b>	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied either lyophilized or as a 0.2 µm filtered solution in PBS.

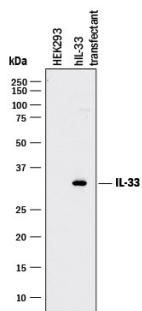
## APPLICATIONS

**Please Note:** Optimal dilutions should be determined by each laboratory for each application. [General Protocols](#) are available in the Technical Information section on our website.

	<b>Recommended Concentration</b>	<b>Sample</b>
<b>Western Blot</b>	1 µg/mL	See Below
<b>Immunohistochemistry</b>	0.1-25 µg/mL	See Below
<b>Intracellular Staining by Flow Cytometry</b>	0.25 µg/10 <sup>6</sup> cells	See Below
<b>Human IL-33 Sandwich Immunoassay</b>		<b>Reagent</b>
<b>ELISA Capture</b>	2-8 µg/mL	Human IL-33 Antibody (Catalog # <a href="#">MAB36253</a> )
<b>ELISA Detection</b>	0.1-0.4 µg/mL	Human IL-33 Biotinylated Antibody (Catalog # <a href="#">BAF3625</a> )
<b>Standard</b>		Recombinant Human IL-33 (Catalog # <a href="#">3625-IL</a> )

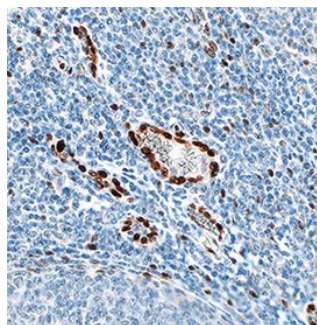
## DATA

### Western Blot



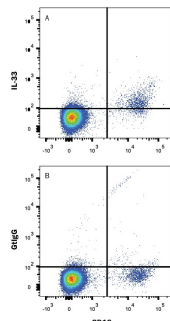
**Detection of Human IL-33 by Western Blot.** Western blot shows lysates of HEK293 human embryonic kidney cell line either mock transfected or transfected with human IL-33. PVDF membrane was probed with 1 µg/mL of Goat Anti-Human IL-33 Monoclonal Antibody (Catalog # [MAB36253](#)) followed by HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # [HAF017](#)). A specific band was detected for IL-33 at approximately 30 kDa (as indicated). This experiment was conducted under reducing conditions and using [Immunoblot Buffer Group 1](#).

### Immunohistochemistry



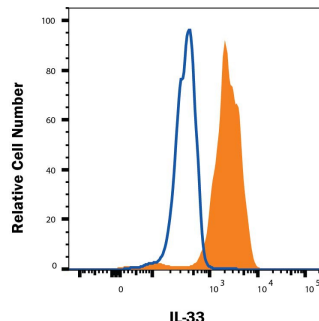
**IL-33 in Human Tonsil.** IL-33 was detected in immersion fixed paraffin-embedded sections of human tonsil using Goat Anti-Human IL-33 Monoclonal Antibody (Catalog # [MAB36253](#)) at 0.1 µg/mL overnight at 4 °C. Tissue was stained using the Anti-Goat HRP-DAB Cell & Tissue Staining Kit (brown; Catalog # [CTS008](#)) and counterstained with hematoxylin (blue). Specific staining was localized to nuclei in epithelial cells. View our protocol for [Chromogenic IHC Staining of Paraffin-embedded Tissue Sections](#).

### Intracellular Staining by Flow Cytometry



**Detection of IL-33 in Human Peripheral Blood Lymphocytes by Flow Cytometry.** Human peripheral blood lymphocytes were stained with (A) Goat Anti-Human IL-33 Monoclonal Antibody (Catalog # [MAB36253](#)) or (B) Goat IgG control antibody (Catalog # [AB-108-C](#)) followed by anti-Goat IgG PE-conjugated secondary antibody (Catalog # [F0107](#)) and Mouse anti-Human CD19 APC-conjugated Monoclonal Antibody (Catalog # [FAB4867A](#)). To facilitate intracellular staining, cells were fixed with Flow Cytometry Fixation Buffer (Catalog # [FC004](#)) and permeabilized with methanol. View our protocol for [Staining Intracellular Molecules](#).

### Intracellular Staining by Flow Cytometry



**Detection of IL-33 in HUVECs by Flow Cytometry.** HUVECs were stained with Goat Anti-Human IL-33 Monoclonal Antibody (Catalog # [MAB36253](#), filled histogram) or Goat IgG control antibody (Catalog # [AB-108-C](#), open histogram) followed by anti-Goat IgG PE-conjugated secondary antibody (Catalog # [F0107](#)). To facilitate intracellular staining, cells were fixed with Flow Cytometry Fixation Buffer (Catalog # [FC004](#)) and permeabilized with methanol. View our protocol for [Staining Intracellular Molecules](#).

#### PREPARATION AND STORAGE

<b>Reconstitution</b>	Reconstitute at 0.5 mg/mL in sterile PBS.
<b>Shipping</b>	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
<b>Stability &amp; Storage</b>	<p><b>Use a manual defrost freezer and avoid repeated freeze-thaw cycles.</b></p> <ul style="list-style-type: none"> <li>• 12 months from date of receipt, -20 to -70 °C as supplied.</li> <li>• 1 month, 2 to 8 °C under sterile conditions after reconstitution.</li> <li>• 6 months, -20 to -70 °C under sterile conditions after reconstitution.</li> </ul>

#### BACKGROUND

IL-33, also known as NF-HEV and DVS 27, is a 30 kDa proinflammatory protein that may also regulate gene transcription (1-3). DVS 27 was identified as a gene that is up-regulated in vasospastic cerebral arteries (1). NF-HEV was described as a nuclear factor that is preferentially expressed in the endothelial cells of high endothelial venules relative to endothelial cells from other tissues (2). IL-33 was identified based on sequence and structural homology with IL-1 family cytokines (3). DVS 27, NF-HEV, and IL-33 share 100% amino acid sequence identity. IL-33 is constitutively expressed in smooth muscle and airway epithelia. It is up-regulated in arterial smooth muscle, dermal fibroblasts, and keratinocytes following IL-1 $\alpha$  or IL-1 $\beta$  stimulation (1, 3). Similar to IL-1, IL-33 can be cleaved *in vitro* by caspase-1, generating an N-terminal fragment that is slightly shorter than the C-terminal fragment (3, 4). The N-terminal portion of full length IL-33 contains a predicted bipartite nuclear localization sequence and a homeodomain-like helix-turn-helix DNA binding domain. By immunofluorescence, full length IL-33 localizes to the nucleus in HUVECs and transfectants (2). The C-terminal fragment, corresponding to mature IL-33, binds and triggers signaling through mast cell IL-1 R4/ST2L, a longtime orphan receptor involved in the augmentation of Th2 cell responses (3, 5-7). A ternary signaling complex is formed by the subsequent association of IL-33 and ST2L with IL-1R AcP (8). Stimulation of Th2 polarized lymphocytes with mature IL-33 *in vitro* induces IL-5 and IL-13 secretion (3). *In vivo* administration of mature IL-33 promotes increased production of IL-5, IL-13, IgE, and IgA, as well as splenomegaly and inflammatory infiltration of mucosal tissues (3). Full length and mature human IL-33 share 52-58% aa sequence identity with mouse and rat IL-33. Human IL-33 shares less than 20% aa sequence identity with other IL-1 family proteins.

#### References:

1. Onda, H. *et al.* (1999) J. Cereb. Blood Flow Metab. **19**:1279.
2. Baekkevold, E.S. *et al.* (2003) Am. J. Pathol. **163**:69.
3. Schmitz, J. *et al.* (2005) Immunity **23**:479.
4. Black, R.A. *et al.* (1989) J. Biol. Chem. **264**:5323.
5. Xu, D. *et al.* (1998) J. Exp. Med. **187**:787.
6. Lohning, M. *et al.* (1998) Proc. Natl. Acad. Sci. USA **95**:6930.
7. Dinarello, C.A. (2005) Immunity **23**:461.
8. Chackerian, A.A. *et al.* (2007) J. Immunol. **179**:2551.