

DESCRIPTION

Species Reactivity	Mouse
Specificity	Detects mouse IL-33 in direct ELISAs.
Source	Monoclonal Rat IgG _{2A} Clone # 396118
Purification	Protein A or G purified from hybridoma culture supernatant
Immunogen	<i>E. coli</i> -derived recombinant mouse IL-33 Ser109-Ile266 Accession # Q8BVZ5
Formulation	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.

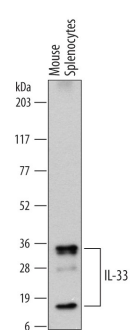
APPLICATIONS

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.

	Recommended Concentration	Sample
Western Blot	0.5 µg/mL	See Below
Immunocytochemistry	8-25 µg/mL	See Below
Intracellular Staining by Flow Cytometry	0.25 µg/10 ⁶ cells	See Below
CyTOF-ready	Ready to be labeled using established conjugation methods. No BSA or other carrier proteins that could interfere with conjugation.	

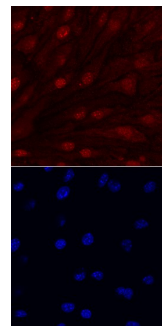
DATA

Western Blot



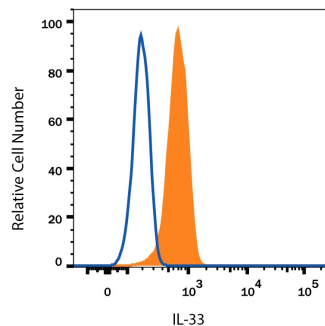
Detection of Mouse IL-33 by Western Blot. Western blot shows lysates of mouse splenocytes. PVDF membrane was probed with 0.5 µg/mL of Rat Anti-Mouse IL-33 Monoclonal Antibody (Catalog # MAB3626) followed by HRP-conjugated Anti-Rat IgG Secondary Antibody (Catalog # HAF005). Specific bands were detected for IL-33 at approximately 18 and 35 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Immunocytochemistry



IL-33 in bEnd.3 Mouse Cell Line. IL-33 was detected in immersion fixed bEnd.3 mouse endothelioma cell line using Rat Anti-Mouse IL-33 Monoclonal Antibody (Catalog # MAB3626) at 10 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Rat IgG Secondary Antibody (red, upper panel, Catalog # NL013) and counterstained with DAPI (blue, lower panel). View our protocol for [Fluorescent ICC Staining of Cells on Coverslips](#).

Intracellular Staining by Flow Cytometry



Detection of IL-33 in bEnd.3 Mouse Cell Line by Flow Cytometry. bEnd.3 mouse endothelioma cell line was stained with Rat Anti-Mouse IL-33 Monoclonal Antibody (Catalog # MAB3626, filled histogram) or isotype control antibody (Catalog # MAB006, open histogram), followed by Phycoerythrin-conjugated Anti-Rat IgG Secondary Antibody (Catalog # F0105B). To facilitate intracellular staining, cells were fixed with Flow Cytometry Fixation Buffer (Catalog # FC004) and permeabilized with Flow Cytometry Permeabilization/Wash Buffer I (Catalog # FC005). View our protocol for [Staining Intracellular Molecules](#).

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.5 mg/mL in sterile PBS.
Shipping	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
Stability & Storage	Use a manual defrost freezer and avoid repeated freeze-thaw cycles. <ul style="list-style-type: none"> • 12 months from date of receipt, -20 to -70 °C as supplied. • 1 month, 2 to 8 °C under sterile conditions after reconstitution. • 6 months, -20 to -70 °C under sterile conditions after reconstitution.

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 upregulated 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 upregulated in arterial smooth muscle, dermal fibroblasts, and keratinocytes following IL-1 α or IL-1 β 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 mouse IL-33 share approximately 55% and 90% amino acid (aa) sequence identity with human and rat IL-33, respectively. Mouse IL-33 shares less than 25% 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.