

Product datasheet for **AM26651AF-N**

IL18 (37-194) Mouse Monoclonal Antibody [Clone ID: 21A12]

Product data:

Product Type:	Primary Antibodies
Clone Name:	21A12
Applications:	WB
Recommended Dilution:	Western blot: 1 µg/ml for chemiluminescence detection system. For details see protocol below.
Reactivity:	Mouse, Rat
Host:	Mouse
Isotype:	IgG2b
Clonality:	Monoclonal
Immunogen:	Mature rat IL-18 fusion protein corresponding to 37-194 aa
Specificity:	This antibody reacts with IL-18.
Formulation:	PBS containing 50% glycerol, pH 7.2. No preservative is contained. State: Azide Free State: Liquid Ig fraction
Concentration:	lot specific
Purification:	Protein A agarose
Conjugation:	Unconjugated
Storage:	Upon receipt, store (in aliquots) at -20 °C. Avoid repeated freezing and thawing.
Stability:	Shelf life: One year from despatch.
Gene Name:	interleukin 18
Database Link:	Entrez Gene 29197 Rat P97636



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Background:

Interleukin 18 (IL-18) is an 18-kDa cytokine which identified as a costimulatory factor for production of interferon- γ (IFN- γ) in response to toxic shock and shares functional similarities with IL-12. IL-18 is synthesized as a precursor 24-kDa molecule without a signal peptide and must be cleaved to produce an active molecule. IL-1 converting enzyme (ICE, Caspase-1) cleaves pro-IL-18 at aspartic acid in the P1 position, producing the mature, bioactive peptide that is readily released from the cells. It is reported that IL-18 is produced from Kupffer cells, activated macrophages, keratinocytes, intestinal epithelial cells, osteoblasts, adrenal cortex cells and murine diencephalon. IFN- γ is produced by activated T or NK cells and plays critical roles in the defense against microbial pathogens. IFN- γ activates macrophages and enhances NK activity and B cell maturation, proliferation and Ig secretion. IFN- γ also induces expression of MHC class I and II antigens and inhibits osteoclast activation. IL-18 acts on T helper type-1 (Th1) T cells and in combination with IL-12 strongly induces them to produce IFN- γ . Pleiotropic effects of IL-18 have also been reported, such as, enhancement production of IFN- γ and GM-CSF in peripheral blood mononuclear cells, production of Th1 cytokines, IL-2, GM-CSF and IFN- γ in T cells, enhancement of Fas ligand expression by Th1 cells.

Synonyms:

IL-18, IGIF, IL1F4, Iboctadekin, IL1 gamma, IL-1 gamma

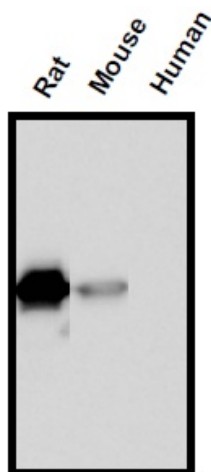
Note: This product was originally produced by MBL International.

Protocol:

SDS-PAGE & Western Blotting

- 1) Mix the sample with equal volume of Laemmli's sample buffer. Boil the samples for 3 minutes and centrifuge.
- 2) Load the sample per lane in a 1 mm thick SDS-polyacrylamide gel for electrophoresis.
- 3) Blot the protein to a polyvinylidene difluoride (PVDF) membrane at 1 mA/cm² for 1 hour in a semi-dry transfer system (Transfer Buffer: 25 mM Tris, 190 mM glycine, 20% MeOH). See the manufacture's manual for precise transfer procedure.
- 4) To reduce nonspecific binding, soak the membrane in 5% skimmed milk (in PBS, pH 7.2) for 1 hour at room temperature, or overnight at 4°C.
- 5) Incubate the membrane with primary antibody diluted with PBS, pH 7.2 containing 1% skimmed milk as suggest in the APPLICATIONS for 1 hour at room temperature. (The concentration of antibody will depend on condition.)
- 6) Wash the membrane with PBS-T [0.05% Tween-20 in PBS] (5 minutes x 6 times).
- 7) Incubate the membrane with the 1:10,000 HRP-conjugated anti-mouse IgG diluted with 1% skimmed milk (in PBS, pH 7.2) for 1 hour at room temperature.
- 8) Wash the membrane with PBS-T (5 minutes x 6 times).
- 9) Wipe excess buffer on the membrane, then incubate it with appropriate chemiluminescence reagent for 1 minute.
- 10) Remove extra reagent from the membrane by dabbing with paper towel, and seal it in plastic wrap.
- 11) Expose to an X-ray film in a dark room for 1 minute.
- 12) Develop the film as usual. The condition for exposure and development may vary. (Positive control for Western blotting; transfectant)

Product images:



Western blot analysis of mature rat IL-18 expression in culture supernatant of rat, mouse and human IL-18 transfected 293T cells using AM26651AF-N.