MONOCLONAL ANTIBODY

Anti-IL-18 (Human) mAb

Code No.  
Clone  
Subclass  
Quantity  
Concentration
D043-3  
25-2G  
Mouse IgG1 κ  
100 μL  
1 mg/mL

BACKGROUND: Interleukin 18 (IL-18) is an 18 kDa cytokine which identified as a costimulator 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, enhances NK activity and B cell maturation, proliferation and Ig secretion, induces MHC class I and II antigens, and inhibits osteoclast activation. IL-18 acts on Th1 helper type T cells and in combination with IL-12 strongly induces them to produce IFN-γ. Pleiotropic effects of IL-18 has 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.

SOURCE: This antibody was purified from mouse ascites fluid using protein A agarose. This hybridoma was established by fusion of mouse myeloma cell SP2/0 with Balb/c mouse splenocyte immunized with recombinant human IL-18.

FORMULATION: 100 μg IgG in 100 μL volume of PBS containing 50% glycerol, pH 7.2. No preservative is contained.

STORAGE: This antibody solution is stable for one year from the date of purchase when stored at -20°C.

REACTIVITY: This antibody reacts with human IL-18 on Western blotting.

SPECIES CROSS REACTIVITY:

<table>
<thead>
<tr>
<th>Species</th>
<th>Human</th>
<th>Mouse</th>
<th>Rat</th>
</tr>
</thead>
<tbody>
<tr>
<td>Samples</td>
<td>Recombinant</td>
<td>Recombinant</td>
<td>Not tested</td>
</tr>
<tr>
<td>Reactivity on WB</td>
<td>+</td>
<td>-</td>
<td></td>
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</tbody>
</table>

APPLICATIONS:
Western blotting: 1 μg/mL for chemiluminescence detection system
Immunoprecipitation: Not tested
Immunocytochemistry: Not tested
Immunohistochemistry: Not tested

*It is reported that this monoclonal antibody can be used in Immunohistochemistry for paraffin section in the reference number 3-5, and for frozen section in the reference number 4.

Flow cytometry: Not tested

Detailed procedure is provided in the following PROTOCOL.

INTENDED USE:
For Research Use Only. Not for use in diagnostic procedures.

REFERENCES:
6) Takiyama, Y., et al., Thyroid 12, 935-943 (2002) [IHC]

Clone 25-2G is used in reference number 1) - 8).

![Western blot analysis of Human IL-18 expression in recombinant Human IL-18 using D043-3](image-url)
PROTOCOL: SDS-PAGE & Western Blotting

1) Boil the samples for 2 minutes and centrifuge. Load 10 μL of sample per lane on a 1-mm-thick SDS-polyacrylamide gel and carry out electrophoresis.

2) 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 manufacturer's manual for precise transfer procedure.

3) To reduce nonspecific binding, soak the membrane in 10% skimmed milk (in PBS, pH 7.2) for 1 hour at room temperature, or overnight at 4°C.

4) Incubate the membrane with primary antibody diluted with 1% skimmed milk (in PBS, pH 7.2) as suggested in the APPLICATIONS for 1 hour at room temperature. (The concentration of antibody will depend on the conditions.)

5) Wash the membrane with PBS-T [0.05% Tween-20 in PBS] (5 minutes x 3 times).

6) Incubate the membrane with 1:10,000 of Anti-IgG (Mouse) pAb-HRP (MBL; code no. 330) diluted with 1% skimmed milk (in PBS, pH 7.2) for 1 hour at room temperature.

7) Wash the membrane with PBS-T (10 minutes x 3 times).

8) Wipe excess buffer on the membrane, then incubate it with appropriate chemiluminescence reagent for 1 minute. Remove extra reagent from the membrane by dabbing with paper towel, and seal it in plastic wrap.

9) Expose to an X-ray film in a dark room for 1 minute. Develop the film as usual. The condition for exposure and development may vary.

(Positive control for Western blotting; Recombinant)

RELATED PRODUCTS:

Antibodies
D044-3 Anti-IL-18 (Human) mAb (125-2H)
D045-3 Anti-IL-18 (Human) mAb (159-12B)
D045-6 Anti-IL-18 (Human) mAb-Biotin (159-12B)
PM014 Anti-IL-18 (Human) pAb
D046-3 Anti-IL-18 (Mouse) mAb (39-3F)
D047-3 Anti-IL-18 (Mouse) mAb (74)
D048-3 Anti-IL-18 (Mouse) mAb (93-10C)
D048-6 Anti-IL-18 (Mouse) mAb-Biotin (93-10C)
M157-3 Anti-IL-18 (Rat) mAb (21A12)
M158-3 Anti-IL-18 (Rat) mAb (91D8)

ELISA Kits
7620 Human IL-18 ELISA Kit
7625 Mouse IL-18 ELISA Kit

Recombinant Proteins
B001-5 Recombinant Human IL-18
B003-5 Recombinant Human IL-18 (without BSA)
B002-5 Recombinant Mouse IL-18
B004-5 Recombinant Mouse IL-18 (without BSA)