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antibody and ELISA experts

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Anti-Mouse IL-18 Antibody

Catalog Number: A00124

Introduction

CD3epsilon is a 20kD chain, which together with CD3lambda, CD3delta, and CD3zeta, and a T cell receptor (alpha/beta or gamma/) form the CD3/T-cell receptor complex. It is a specific marker for T lymphocytes, NK T cells, and some thymocytes. Crosslinking of TCR initiates an intracellular signaling cascade resulting in cellular activation and proliferation. The OKT3 antibody has been reported to have potent immunosuppressive properties in vivo and has been proved effective in the treatment of renal, heart, and liver allograft rejection.

This antibody is routinely tested by flow cytometric analysis. Flow cytometry and other applications were tested during antibody development or are reported in the literature.

Application Information

Each lot of this antibody has been quality control tested by flow cytometric analysis of human PBMCs. For flow cytometric staining, the recommended use of this antibody is ≤ 0.5µg per 1x106 cells in 100µl of staining volume followed by a secondary florescent conjugated anti-mouse antibody. However, it is strongly suggested that the antibody reactivity be empirically titrated for optimal performance in the application of interest.

About IL18

Interleukin-18 (IL-18) is a member of the IL-1 cytokine family and was initially identified as an Interferon-g (IFN-g) inducing factor (IGIF). The IL-18 gene was originally cloned from liver cells and has since been shown to be produced by activated monocytes/ macrophages, Kupffer cells, keratinocytes, glucocorticoid-secreting adrenal cortex cells, osteoblasts and dendritic cells. IL-18 is a 24 kDa, non-glycosylated polypeptide that lacks a classical signal sequence and possesses a structure recognizably similar to IL-1. IL-18 is synthesized as a bio-inactive propeptide that undergoes proteolytic cleavage by either ICE (interleukin-1 beta converting enzyme) or another caspase to generate a mature, bioactive, 18 kDa molecule. In both the mature and propeptide forms, IL-18 shows 64% aa sequence identity from mouse to human. IL-18 does not appear to show any primary sequence similarity to any other known cytokines. Rat IL-18 has also been isolated, and found to be 194 aa in length with a 91% aa sequence identity to mouse IL-18. Human IL-18 has been found to induce the production of IFN-g and GM-CSF while inhibiting the production of IL-10 by PBMC. With respect to human T cells, IL-18 enhances Th1 cytokine production and stimulates cell proliferation via an IL-2-dependent pathway. Human IL-18 can also inhibit the synthesis of IgE by B cells. Thus, IL-18 plays an important role in immunological and inflammatory reactions. Currently, the bioactivity of human IL-18 is often determined by its capacity to augment the levels of IFN-g produced by T cells as measured in tissue culture supernatants.

Overview

Product Name	Anti-Mouse IL-18 Antibody
Reactive Species	Mouse
Description	Boster Bio Anti-Mouse IL-18 Antibody (Catalog # A00124). Tested in IF, IHC applications. This antibody reacts with Mouse.
Conjugate	Biotin

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Application	ELISA, IF, IHC, WB
Clonality	Polyclonal
Formulation	0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2
Storage Instructions	Store vial at -20°C prior to opening. Aliquot contents and freeze at -20°C or below for extended storage. Avoid cycles of freezing and thawing. Centrifuge product if not completely clear after standing at room temperature. This product is stable for several weeks at 4°C as an undiluted liquid. Dilute only prior to immediate use. Expiration date is one (1) year from date of opening. (Ship on dry ice.)
Host	Rabbit
Uniprot ID	P70380

Technical Details

Immunogen	The whole rabbit serum used to produce this IgG fraction antibody was prepared by repeated immunizations with native 157 aa mouse IL-18 produced in E.coli.
Predicted Reactive Species	Mouse, Rat
Cross Reactivity	beta-Actin antibody is human, mouse, rat, rabbit, chicken, zebrafish and drosophila reactive.
Isotype	IgG
Form	Liquid (sterile filtered)
Concentration	1.0 mg/mL by UV absorbance at 280 nm
Purification	This is an IgG preparation of whole rabbit serum purified by protein A chromatography using a low endotoxin methodology. This antibody is primarily directed against mature 18,000 MW mouse IL-18 and is useful in determining its presence in various assays. This antibody will also recognize the 24,000 inactive precursor form of mouse IL-18. In general, this antibody also detects rat IL-18 in the same formats using similar dilutions. A control of similarly diluted LOW ENDOTOXIN CONTROL RABBIT IgG (code # 011-001-297) is recommended.
Suggested Dilutions	ELISA: 1:1,000 - 1:5,000 IHC: User optimized IF Microscopy: 1:50-1:200 WB: 1:500 - 1:2,000 Neutralization: User optimized Anti-Mouse IL-18 has been tested in immunohistochemistry and immunofluorescence and is suitable for use in neutralizations, ELISA, and immunoblotting. Although untested, this reagent may be useful for radioimmunoassays, flow cytometry and immunoprecipitation. It recognizes the 18,000 MW mature (active) IL-18. Reactivity in other immunoassays is unknown.

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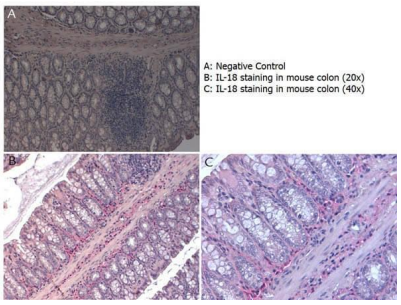
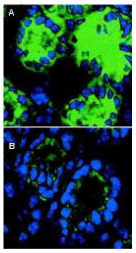
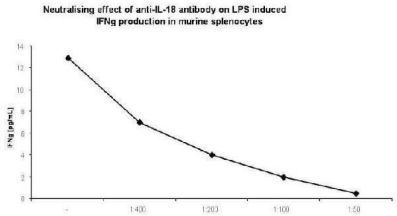
Anti-Mouse IL-18 Antibody (A00124) Images



Immunohistochemistry with Rabbit anti-Mouse IL-18 antibody showing IL-18 staining in inflammatory cells of the mucous corium of mouse colon at 20x and 40x. Slide A is a negative control. Slides B and C show staining. Formalin fixed/paraffin embedded sections were subjected to heat induced epitope retrieval (HIER) at pH 6.2 and then incubated with mouse anti-IL-18 antibody at 4.0 µg/ml for 60 minutes. The reaction was developed using MACH 4 universal AP polymer detection system and visualized with WARP RED.



Immunofluorescence microscopy of IL-18 in mouse colon sections. The transversing portion of the large intestine from DSS-exposed (Panel A) and -unexposed mice (Panel B) was excised, rinsed in PBS, and frozen on isopentane cooled with liquid nitrogen. Frozen sections (5 µm) were cut on a Leica CM 1850 cryostat. The slides were fixed for 10 min in 4% paraformaldehyde, air-dried, and incubated for 20 min in PBS supplemented with 10% normal goat serum. Sections were incubated in a 1:50 dilution of Boster's rabbit anti-Mouse IL-18 antibody or 1 µg/ml nonimmune rabbit IgG (not shown) as negative control. The antibodies were diluted in PBS containing 1% bovine serum albumin. After an overnight incubation at 4°C, the sections were washed three times with 0.5% bovine serum albumin in PBS. The sections were then incubated with a secondary goat anti-rabbit antibody conjugated to Alexa488 (Molecular Probes) for 60 min at room temperature in the dark. Nuclei were counterstained blue using 1 µg/100 ml bisbenzimide. After staining, sections were washed and examined with the Leica DM RXA confocal laser scanning system and analyzed. Similar staining will occur with other systems.



In-vitro neutralization. Spleens were aseptically removed and cell suspensions were prepared. Cells were washed twice and resuspended in RPMI supplemented with 10% FBS. For cytokine measurement, spleen cells were cultured at 5 mln/mL in 24-well, flat-bottom culture plates in the presence of several dilutions of rabbit anti-murine IL-18 antibody (1:400; 1:200; 1:100; 1:50) and 100 ng/mL of LPS (a phenol-extracted preparation from Escherichia coli 055:B5, Sigma Chemical Co). Cultures were incubated at 37°C in a humidified atmosphere with 5% CO2. At the end of the incubation period, cultures were frozen at -70°C and subjected to 3 freeze-thaw cycles to obtain total cytokine levels. Before assaying, samples were centrifuged for 10 minutes at 10,000g to remove debris.

2 Publications Citing This Product

1. PubMed ID: 16052682, Yang Yj, Shen Y, Chen Sh, Ge Xr. World J Gastroenterol. 2005 Aug 7;11(29):4524-9. Role Of Interleukin 18 In Acute Lung Inflammation Induced By Gut Ischemia Reperfusion.

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2. PubMed ID: 25955000 , High and fluctuating glucose levels increase the expression and secretion of interleukin‐18 in mouse peritoneal macrophages

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