

Datasheet for 210-401-323S**IL-18 Antibody****Overview**

Description:	Anti-IL-18 (RABBIT) Antibody - 210-401-323S
Item No.:	210-401-323S
Size:	25 µL
Applications:	IF, IHC, WB
Reactivity:	Mouse
Host Species:	Rabbit

Product Details**Background:**

Interleukin-18 (IL-18) is a member of the IL-1 cytokine family and was initially identified as an Interferon- γ (IFN- γ) 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- γ 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- γ produced by T cells as measured in tissue culture supernatants.

Synonyms:	rabbit anti-IL-18 antibody, rabbit anti-interleukin-18 antibody, Ibctadekin antibody, IFN gamma inducing factor antibody, IGIF antibody, IL 1 gamma antibody, IL 18 antibody, IL 1g antibody, IL-1F4, IL 18, Interleukin 18, IL18, Interleukin18, IL1 F4, IL1F4, Ms IL-18, mouse IL18
Host Species:	Rabbit
Clonality:	Polyclonal

Format: IgG

Target Details

Gene Name:	Il18
Reactivity:	Mouse
Immunogen Type:	Recombinant Protein
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.
Purity/Specificity:	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.
Relevant Links:	• GenelD - 16173 • NCBI - P70380.2 • UniProtKB - P70380

Application Details

Tested Applications:	IF, IHC
Suggested Applications:	WB (Based on references)
Application Note:	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.
Assay Dilutions:	All assays should be optimized by the user. Recommended dilutions (if any) may be listed below.
ELISA:	1:1,000 - 1:5,000
IF:	1:50-1:200
IHC:	User Optimized
Neutralization:	User Optimized
WB:	1:500 - 1:2,000

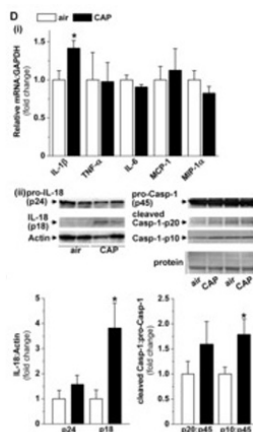
Formulation

Physical State:	Liquid (sterile filtered)
Concentration:	1.0 mg/ml by UV absorbance at 280 nm
Buffer:	0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2
Preservative:	None
Stabilizer:	None

Shipping & Handling

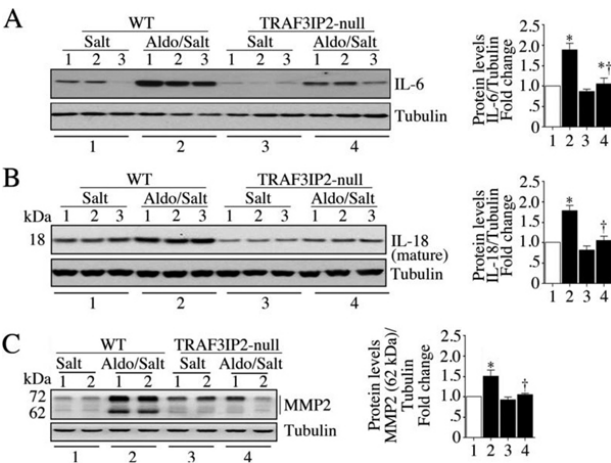
Shipping Condition:	Dry Ice
Storage Condition:	Store vial at -20° C or below prior to opening. This vial contains a relatively low volume of reagent (25 µL). To minimize loss of volume dilute 1:10 by adding 225 µL of the buffer stated above directly to the vial. Recap, mix thoroughly and briefly centrifuge to collect the volume at the bottom of the vial. Use this intermediate dilution when calculating final dilutions as recommended below. Store the vial at -20°C or below after dilution. Avoid cycles of freezing and thawing.
Expiration:	Expiration date is one (1) year from date of receipt.

Images



Western Blot

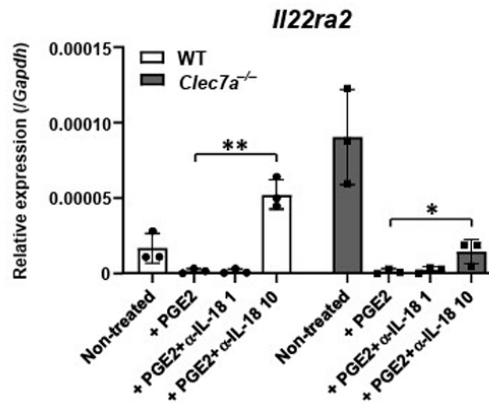
CAP exposure induces vascular oxidative stress and inflammation. A: correlation between CAP-induced vascular insulin resistance and EPC levels. Data are shown as discrete points (mean in % of control) for the densitometric Western blot analysis of aortic insulin-induced Akt phosphorylation vs. the abundance of blood or bone marrow endothelial progenitor cells (EPCs) measured in air or CAP-exposed mice since August 2010. The curve is a linear fit [$y=A+B \cdot x$] between the CAP-induced vascular insulin resistance and the depletion of circulating and the retention of bone marrow EPCs. CAP-induced aortic oxidative stress (C) and inflammation (D) was analyzed in aortas isolated from mice exposed for 9 days to air or CAP as indicated in the experimental protocol (B). The mRNA levels of antioxidant defense genes (Ci) and pro-inflammatory genes (Di) were measured by RT-PCR and abundance of protein-HNE adducts (Cii), and the activation of IL-18 and Casp-1 (Dii) were analyzed by Western blot. Data are means $\pm$ SE normalized to the air control (* $P < 0.05$, air vs. CAP, $n = 5$). Fig 4. PMID: 27016579



Western Blot

Effect of aldosterone treatment on cardiac IL-6, IL-18 and MMP2 expression. (A) Immunoblot analysis of IL-6. Densitometric analysis of the immunoreactive bands is shown on the right. (B). Immunoblot analysis of IL-18. Densitometric analysis of the immunoreactive bands is shown on the right. (C). Immunoblot analysis of MMP2. Densitometric analysis of the immunoreactive bands is shown on the right. Figure 6. PMID: 27040306

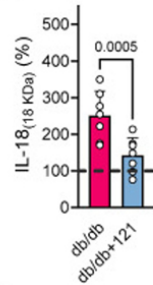
i



A



D



Figure

(i) CD11b⁺ and CD11c⁺ cells isolated from the colon of ApcMin/+ and ApcMin/+Clec7a^{-/-} mice were treated with 10 μM PGE2 for 20h in the presence of anti-IL-18 antibody (p/n 210-401-323) atv (1 or 10 μg/ml), and Il22ra2 expression was determined by qPCR (n=3 biologically independent samples/group; **P=0.0031, *P=0.0264). SF5. PMID: 36932082

Western Blot

Liver inflammatory markers in db/db mice fed control diet or diet with CMS121. Representative blot images

(A). Vertical lines indicate non-adjacent lanes from the same blot, and dashed lines adjacent groups.

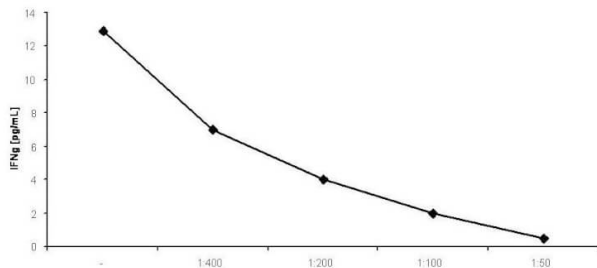
(D) Respective quantification of blots: IL-18. Data are presented as mean ± SD (n = 6–8), and p-values are indicated for the untreated control db/db mice compared to the CMS121-treated mice. Values were normalized to WT group (dashed line), and data are presented as percentages of WT values. One-way ANOVA was used to detect mean changes, and differences between db/db and db/db + 121 were evaluated by the Holm–Sidak post-hoc test. Fig 4.

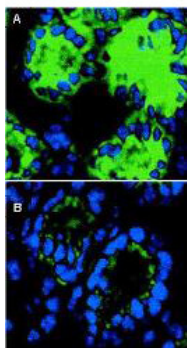
PMID: 37047807

Neutralization

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.

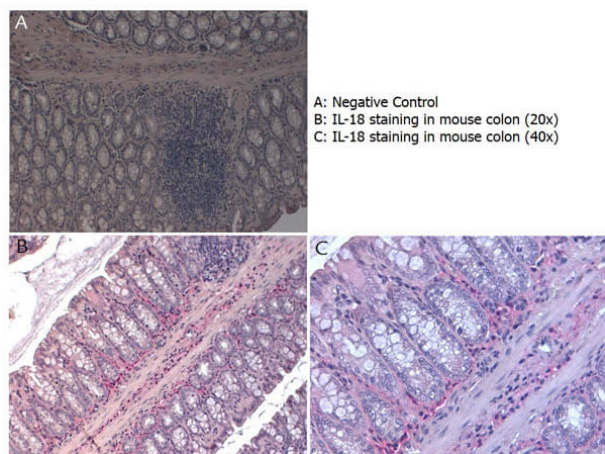
Neutralising effect of anti-IL-18 antibody on LPS induced IFNγ production in murine splenocytes





Immunofluorescence Microscopy

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 μ m) 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 Rockland's rabbit anti-Mouse IL-18 antibody or 1 μ g/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 μ g/100 ml bisbenzimidazole. 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.



Immunohistochemistry

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 μ g/ml for 60 minutes. The reaction was developed using MACH 4 universal AP polymer detection system and visualized with WARP RED.

References

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