



Product Datasheet Mouse IL-27/p28 Antibody (orb345244)

Catalog Number	orb345244
Description	IL-27/P28 antibody
Species/Host	Rat
Reactivity	Mouse
Conjugation	Unconjugated
Tested Applications	ELISA, FC, Multiplex Assay, WB
Immunogen	This Protein A purified monoclonal antibody was produced in rats by repeated immunizations with mature length recombinant mouse p28 protein (produced in E.coli) followed by hybridoma development.
Preservatives	0.01% (w/v) Sodium Azide
Form/Appearance	Liquid (sterile filtered)
Concentration	1.0 mg/mL
Storage	Store vial at -20° C or below prior to opening. This vial contains a relatively low volume of reagent (25 μ L). To minimize loss of volume dilute 1:10 by adding 225 μ L 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.
Note	For research use only

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Application notes	IL-27 is expressed in activated antigen presenting cells including monocytes, endothelial cells, and dendritic cells, for example mouse CD4 splenocytes. This purified antibody has been tested for use in Flow Cytometry, ELISA and western blotting. Specific conditions for reactivity should be optimized by the end user. Expect a band approximately 26,411 Da in size corresponding to the mature mouse p28 protein, a non-glycosylated polypeptide chain consisting of amino acids, by western blotting in appropriate cell lysate or extract.
Isotype	lgG2a
Clonality	Monoclonal
Clone Number	3H12.F10
Antibody Type	Primary Antibody
Purity	Anti-IL-27 / p28 antibody is purified by a multi-step process which includes delipidation, salt fractionation and ion exchange chromatography followed by extensive dialysis against the buffer stated above. This antibody is specific for mouse. Cross-reactivity with IL-27 from other sources has not been determined.
Uniprot ID	Q8K3I6
NCBI	NP_663611.1
Dilution Range	ELISA: 1:10,000, FC: 1 ug/mL, WB: 1:1000
Expiration Date	12 months from date of receipt.

72 kD 55 kD 36 kD 28 kD Anti-IL-27/p28 antibody in western blot shows detection of recombinant mouse IL-27/p28. Recombinant protein (0.1 µg) was loaded on to an SDS-PAGE gel, and after separation, transferred to nitrocellulose. The expected band is approximately 26 kDa in size. The membrane was blocked with 1% BSA in TBST for 30 min at RT, followed by incubation with Biorbyt's Anti-IL-27/p28 antibody diluted 1:1000 in 1% BSA in TBST overnight at 4°C. After washes, the blot was reacted with secondary antibody HRP Goat anti-Rat IgG antibody (p/n orb347790) diluted 1:40000 in blocking buffer (p/n orb348637) for 30 min at RT.

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Mouse peritoneal macrophages were grown in culture for 24 hours, stimulated with 10 ng/mL IFN gamma and 1 ug/ml LPS for 14 hours and incubated for 4 hours with Bredfeldin A. Cells were harvested, washed, aliquoted 1x106 cells per sample, and fixed and permeabilized according to a standard protocol. Samples were stained with biotinylated primary anti-mouse p28 antibody at (0.1-10 ug/ml primary antibody alongside negative controls of unstimulated cells and isotype controls. Cells were stained with 0.25 ug/ml rat anti-mouse CD107b conjugated Alexa Fluor 647 and PHYCOERYTHRIN Conjugated secondary at 1:100 and analyzed by flow cytometry. Stimulated cells showed increase PE staining (horizontal axis) when compared with unstimulated cells.

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