Datasheet for ABIN967653

anti-TNFRSF1A antibody

2 Images | 3 Publications

Go to Product page

Overview

Quantity:	0.5 mg
Target:	TNFRSF1A
Reactivity:	Mouse
Host:	Armenian Hamster
Clonality:	Monoclonal
Conjugate:	This TNFRSF1A antibody is un-conjugated
Application:	Western Blotting (WB), ELISA, Flow Cytometry (FACS), Immunoprecipitation (IP)

Product Details

Brand:	BD Pharmingen™
Immunogen:	Mouse TNFRI
Clone:	55R-286
Isotype:	IgG1 kappa
Characteristics:	1. Since applications vary, each investigator should titrate the reagent to obtain optimal results. 2. Please refer to us for technical protocols. 3. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.
Purification:	The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

Target Details

Target:	TNFRSF1A
Alternative Name:	CD120a (TNFRSF1A Products)
Background:	The 55R-286 antibody reacts with the extracellular region of the 55 kDa receptor for the mouse cytokines, Tumor Necrosis Factor (TNF) and lymphotoxin-alpha (LT-alpha). This receptor, referred to as the p55 or Type I Tumor Necrosis Factor Receptor (TNFRI) [aka, CD120a], is expressed by a variety of cell lines and normal cell types including T cells, monocytes, macrophages, and neutrophils. B cells express very low or undetectable levels of TNFRI whereas mature erythrocytes are uniformly negative for TNFRI expression. In addition, the 55R-286 antibody can bind to a soluble, truncated form of the mouse Type I TNFR that is shed by cells in response to certain stimuli, e.g., cells treated with LPS or TNF. The in vivo administration of nonblocking, nonagonistic 55R-286 antibody reportedly results in the linear accumulation of shed, soluble forms of the TNFRI in the circulation. The 55R-286 antibody does not recognize the 75 kD (p75) Type II TNFR (aka, CD120b). 55R-286 does not block the binding and does not neutralize the bioactivity of the TNF ligand on L929 target cell populations that express TNFRI. The immunogen used to generate the 55R-286 hybridoma was a purified, soluble extracellular domain of the mouse TNFRI. Synonyms: TNF Receptor, Type I/p55
Pathways:	NF-kappaB Signaling, Apoptosis, Caspase Cascade in Apoptosis, Hepatitis C, Ubiquitin Proteasome Pathway

Application Details

Application Notes:	1) ELISA Capture: The purified 55R-286 antibody (ABIN967653) is useful as a capture antibody for a sandwich ELISA for measuring mouse TNFRI protein levels. Purified 55R-286 antibody can be paired with the biotinylated 55R-170 hamster anti-mouse TNFRI as the detecting antibody. Purified 55R-286 antibody should be titrated 4 - 8 µg/ml to determine its optimal concentration for ELISA capture. Note: This ELISA antibody pair shows no cross-reactivity with other purified recombinant cytokines or receptors that were tested including: 1) Mouse IL-1beta, IL-2, IL-3, IL-4, IL-5, IL-6, IL-8, IL-9, IL-10, IL-12 p40, IL-12p70, IL-16, IL-17, GM-CSF, IFN-gamma, TNF, TNFRI, TNFRII, MCP-1, CRG-2, 2) Human IL-1alpha, IL-2, IL-3, IL-4, IL-5, IL-6, IL-7, IL-10, IL-12 p40, IL-12 p70, IL-13, IL-16, GM-CSF, G-CSF, TNF, LT-alpha, TNFRI, TNFRII, MCP-1, MCP-2, IFN-gamma, RANTES, MIG 3) Rat IL-1alpha, IL-2, IL-4, IL-6, IL-10, GM-CSF, IFN-gamma, MCP-1, TNF

Application Details

	2) Immunofluorescent Staining and Flow Cytometric Analysis: The purified form of 55R-286 (ABIN967653) can be used for the immunofluorescent staining (Less or equal than 1 µg antibody/10^6 cells) and flow cytometric analysis of normal mouse cells or cell lines to measure their expressed levels of TNFRI. An appropriate purified isotype control is clone A19-3. A three-layer staining protocol is recommended for maximizing the detection of TNFRI expressed by cells as detailed in the figure legend. Note: 55R-286 is a nonblocking antibody that can be used for the unobstructed immunofluorescent staining and flow cytometric analysis of cells in systems where ligands (eg, TNF) for TNF receptors are present. Based on our testing results (data not shown), the presence of exogenous recombinant mouse TNF at levels Less or equal than 1 µg per 10^6 cells was insufficient to inhibit the binding of 55R-286 to L929 cells that express p55 TNFR (at 0.25 µg antibody/10^6 cells). Please note also that as a consequence of in vivo or in vitro activation, cell surface TNFRI can either be shed by cells or transiently expressed at higher levels. As a result, cellular activation can affect the cells overall expressed level of surface TNFRI. Immunoprecipitation: The 55R-286 antibody has been reported to be useful for the immunoprecipitation of p55 TNFR from lysates of mouse Meth A fibrosarcoma cells. Please note that this application is not routinely tested. WB: The purified 55R-286 antibody has been found useful for Western blotting. Please note that this application is not routinely tested.
Restrictions:	For Research Use only

Handling

Format:	Liquid
Concentration:	0.5 mg/mL
Buffer:	Aqueous buffered solution containing ≤0.09 % sodium azide.
Preservative:	Sodium azide
Precaution of Use:	This product contains Sodium azide: a POISONOUS AND HAZARDOUS SUBSTANCE which

Handling

	should be handled by trained staff only.
Storage:	4 °C
Storage Comment:	Store undiluted at 4°C.

Publications

Product cited in:	Pinckard, Sheehan, Schreiber: "Ligand-induced formation of p55 and p75 tumor necrosis factor receptor heterocomplexes on intact cells." in: **The Journal of biological chemistry**, Vol. 272, Issue 16, pp. 10784-9, (1997) (PubMed). Pinckard, Sheehan, Arthur, Schreiber: "Constitutive shedding of both p55 and p75 murine TNF receptors in vivo." in: **Journal of immunology (Baltimore, Md. : 1950)**, Vol. 158, Issue 8, pp. 3869-73, (1997) (PubMed). Sheehan, Pinckard, Arthur, Dehner, Goeddel, Schreiber: "Monoclonal antibodies specific for murine p55 and p75 tumor necrosis factor receptors: identification of a novel in vivo role for p75." in: **The Journal of experimental medicine**, Vol. 181, Issue 2, pp. 607-17, (1995) (PubMed).

Images

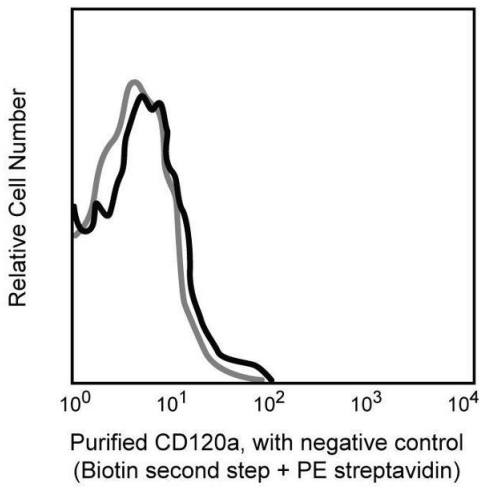
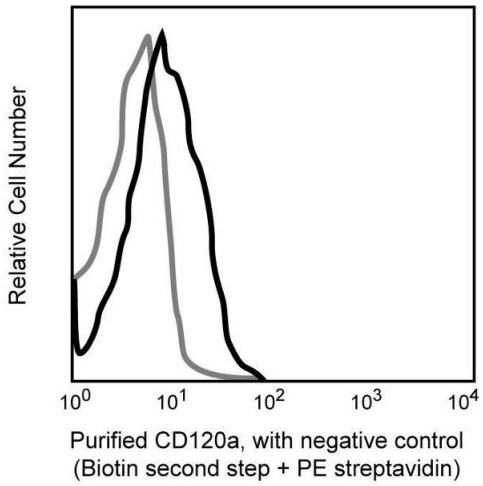


Image 1. Histogram: FITC-B220-positive



Flow Cytometry

Image 2. Overlapping Histograms Figure: Expression of cell surface TNFRI by BALB/c lymph node T cells. BALB/c lymph node cells were preincubated (~15 min., 4°C) with purified 2.4G2 antibody [rat anti-mouse CD16 (FcgIII)/CD32 (FcgII), 1 µg/10e6 cells] to block Fc receptor-mediated nonspecific staining by antibodies. The cells were incubated (30 min., 4° C) with purified 55R-286 antibody (0.5 µg mAb/10e6 cells, ABIN967653). The cells were washed and incubated (30 min., 4° C) with a biotin-conjugated cocktail of mouse anti-hamster antibodies (Clones G70-204 + G94-56, 0.5 µg mAb cocktail/10e6 cells). The cells were washed and incubated with R-PE-conjugated streptavidin (0.015 µg PE-SA/10e6 cells) and anti-CD45R/B220, FITC-RA3-6B2 (0.06 µg mAb/10e6 cells). Finally, the cells were washed in preparation for flow cytometric analysis with a BD FACScan™ Flow Cytometer. The immunofluorescent staining patterns for cells stained in the first stage with either purified 55R-286 antibody (black line histograms) or no antibody (background staining, gray line histograms) followed by the 2nd- and 3rd-layer reagents are shown. The histograms were generated from reanalyzed flow cytometric data files that were gated for FITC-B220-negative (first panel, mostly T cells) or FITC-B220-positive (second panel, mostly B cells) events with the light-scattering characteristics of lymphocytes.