Technical Data Sheet

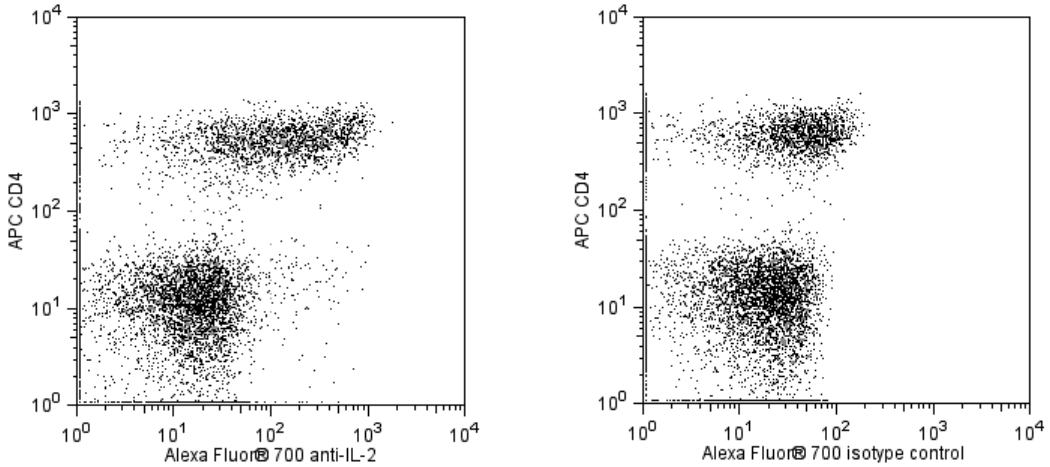
Alexa Fluor® 700 Rat Anti-Mouse IL-2

Product Information

Material Number:	**561287**
Alternate Name:	Il2; Interleukin-2; T-cell growth factor; TCGF
Size:	50 µg
Concentration:	0.2 mg/ml
Clone:	JES6-5H4
Immunogen:	Mouse IL-2 Recombinant Protein
Isotype:	Rat IgG2b
Reactivity:	QC Testing: Mouse
Storage Buffer:	Aqueous buffered solution containing protein stabilizer and ≤0.09% sodium azide.

Description

The JES6-5H4 monoclonal antibody specifically binds to mouse interleukin-2 (IL-2). IL-2 is a multifunctional cytokine that plays pivotal roles in immunity and tolerance. It is produced by activated T cells. IL-2 effects the activation, growth, proliferation and/or differentiation of various cell types including T and B lymphocytes and their precursors, LAK cells, NK cells, and monocytes/macrophages. IL-2 mediates its biological activities by binding to IL-2 receptor compleses. The intermediate affinity IL-2R is comprised of IL-2Rβ (CD122) and common gamma chain (γc; CD132) subunits whereas the high-affinity IL-2R is comprised of IL-2Rα (CD25), IL-2Rβ and γc subunits. The JES6-5H4 monoclonal antibody binds to IL-2 and neutralizes its biological activity.



Two-color flow cytometric analysis of IL-2 expressed by activated CD4-positive splenocytes. *Mouse BALB/c spleen cells were activated for 4 hours using Leukocyte Activation Cocktail, with BD GolgiPlug™ (Cat. No. 550583) that contains Phorbol 12-Myristate 13-Acetate (PMA), ionomycin and brefeldin A. The cells were then fixed and permeabilized using BD Cytofix™ Fixation Buffer (Cat. No. 554655) and BD Perm/Wash™ Buffer (Cat. No. 554723). The permeabilized cells were stained with APC Rat Anti-Mouse CD4 (Cat. No. 553051/561091) and either Alexa Fluor® 700 Rat Anti-Mouse IL-2 (Cat No. 561287, Left Panel) or Alexa Fluor® 700 Rat IgG2b Isotype Control (Cat No.557964, Right Panel). MiCK-1 Mouse Cytokine Positive Control Cells (Cat No. 554652) are prepared in a similar manner. These cells can be used as a positive control for cytokine flow cytometry experiments designed to characterize the nature of mouse IL-2-producing cells. Two-color flow cytometric dot plots showing the correlated expression of IL-2 (or Ig Isotype control staining) versus CD4 were derived from events with the forward and side light-scatter characteristics of intact lymphocytes. Flow cytometry was performed using a BD™ LSR II Flow Cytometer System.*

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Preparation and Storage

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

The antibody was conjugated to Alexa Fluor® 700 under optimum conditions, and unreacted Alexa Fluor® 700 was removed.

Store undiluted at 4°C and protected from prolonged exposure to light. Do not freeze.

Application Notes

Application

Intracellular staining (flow cytometry)	Routinely Tested

Recommended Assay Procedure:

Flow cytometry: The JES6-5H4 antibody is useful for immunofluorescent staining and flow cytometric analysis to identify and enumerate IL-2 producing cells within mixed cell populations. A useful control investigators may consider using for demonstrating specificity of staining, is to pre-block with one of the following reagents: (1) recombinant mouse IL-2 (Cat. No. 550069) or (2) unlabeled JES6-5H4 antibody (Cat. No. 554425), prior to staining.

Suggested Companion Products

Catalog Number	Name	Size	Clone
550583	Leukocyte Activation Cocktail, with BD GolgiPlug™	200 µl	(none)
554655	Fixation Buffer	100 ml	(none)
554723	Perm/Wash Buffer	100 ml	(none)
557964	Alexa Fluor® 700 Rat IgG2b, κ Isotype Control	0.1 mg	A95-1
554656	Stain Buffer (FBS)	500 ml	(none)
553051	APC Rat Anti-Mouse CD4	0.1 mg	RM4-5
561091	APC Rat Anti-Mouse CD4	25 µg	RM4-5

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. An isotype control should be used at the same concentration as the antibody of interest.
3. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
4. The Alexa Fluor®, Pacific Blue™, and Cascade Blue® dye antibody conjugates in this product are sold under license from Molecular Probes, Inc. for research use only, excluding use in combination with microarrays, or as analyte specific reagents. The Alexa Fluor® dyes (except for Alexa Fluor® 430), Pacific Blue™ dye, and Cascade Blue® dye are covered by pending and issued patents.
5. Alexa Fluor® 700 has an adsorption maximum of ~700nm and a peak fluorescence emission of ~720nm. Before staining cells with this reagent, please confirm that your flow cytometer is capable of exciting the fluorochrome and discriminating the resulting fluorescence.
6. Alexa Fluor® is a registered trademark of Molecular Probes, Inc., Eugene, OR.
7. 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.
8. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at www.bdbiosciences.com/colors.

References

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