Technical Data Sheet

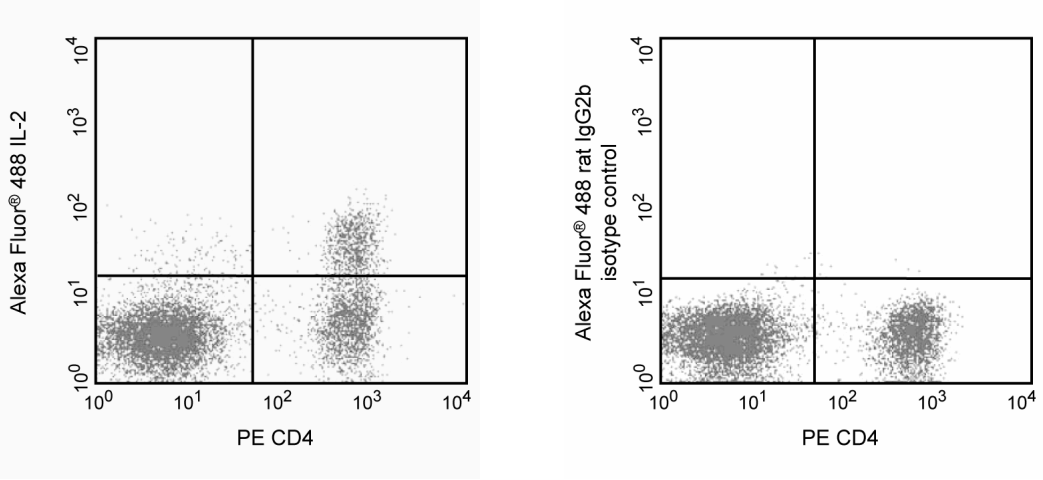
Alexa Fluor® 488 Rat IgG2b, κ Isotype Control

Product Information

Material Number:	**557726**
Size:	0.1 mg
Concentration:	0.2 mg/ml
Clone:	A95-1
Immunogen:	TNP-Keyhole Limpet Hemocyanin
Isotype:	Rat (LOU) IgG2b, κ
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

Description

The A95-1 antibody has unknown specificity. Trinitrophenal (TNP), the immunogen, is a hapten that is not expressed on human, mouse, rat, or non-human primate cells. The A95-1 immunoglobulin was selected as an isotype control following screening for low background on a variety of mouse and human tissues.



Expression of IL-2 by stimulated CD4+ and CD4- BALB/c spleen cells. *Splenocytes from BALB/c mice were stimulated for 4 hours with PMA (5 ng/ml, Sigma, Cat. No. P-8139) and Ionomycin (500 ng, Sigma, P-8139) in the presence of BD GolgiPlug™ (Cat. No. 555029). Cells were harvested, fixed, permeabilized, and stained with PE-conjugated rat anti-mouse CD4 (PE-RM4-5, Cat. No. 553048) and either rat anti-mouse IL-2 antibody (Alexa Fluor® 488-JES6-5H4, Cat. No. 557725), (left panel) or immunoglobulin isotype control (Alexa Fluor® 488-A95-1, Cat. No. 557726), (right panel) by using Pharmingen's staining protocol. Dot plots were derived from gated events with the forward and side light scatter characteristics of lymphocytes. The quadrant markers for the bivariate dot plots were set based on the autofluorescence and isotype controls.*

Preparation and Storage

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

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

The antibody was conjugated to Alexa Fluor® 488 under optimum conditions, and unreacted Alexa Fluor® 488 was removed.

Application Notes

Application

Intracellular staining (flow cytometry)	Routinely Tested
Isotype control	Routinely Tested

Suggested Companion Products

Catalog Number	Name	Size	Clone
555029	Protein Transport Inhibitor (Containing Brefeldin A)	1 mL	(none)
557725	Alexa Fluor® 488 Rat Anti-Mouse IL-2	0.1 mg	JES6-5H4
553048	PE Rat Anti-Mouse CD4	0.1 mg	RM4-5
554656	Stain Buffer (FBS)	500 mL	(none)
554657	Stain Buffer (BSA)	500 mL	(none)

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Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Alexa Fluor® 488 fluorochrome emission is collected at the same instrument settings as for fluorescein isothiocyanate (FITC).
3. Alexa Fluor® is a registered trademark of Molecular Probes, Inc., Eugene, OR.
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. 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.
6. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
7. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

References

Prussin C, Metcalfe DD. Detection of intracytoplasmic cytokine using flow cytometry and directly conjugated anti-cytokine antibodies. *J Immunol Methods.* 1995; 188(1):117-128. (Methodology)

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