

Technical Data Sheet

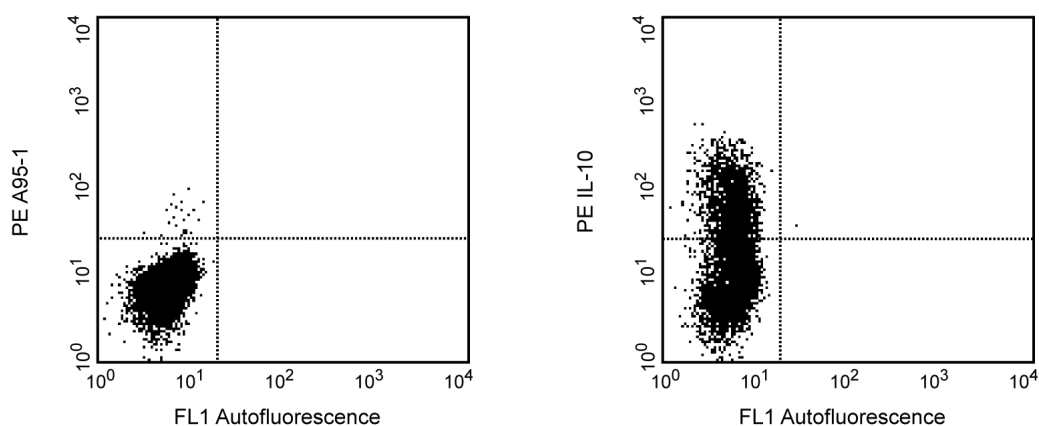
PE Rat IgG2b, κ Isotype Control

Product Information

Material Number:	556925
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 $\leq 0.09\%$ 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-10 by MiCK-2 Cytokine Positive Control Cells. MiCK-2 cells (Cat. No. 554653) were permeabilized, and subsequently stained with 0.12 μg of PE-conjugated rat anti-mouse IL-10 antibody (PE-JES5-16E3, Cat. No. 554467; right panel) or 0.12 μg of PE-A95-1 rat IgG2b isotype control immunoglobulin (Cat. No. 556925; left panel) by using the BD Pharmingen staining protocol. The quadrant markers for the bivariate dot plots were set based on autofluorescence control.

Preparation and Storage

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

The antibody was conjugated with R-PE under optimum conditions, and unconjugated antibody and free PE were removed.

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

Application Notes

Application

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

Recommended Assay Procedure:

Immunofluorescent Staining and Flow Cytometric Analysis: The PE-conjugated A95-1 immunoglobulin (Cat. No. 556925) is a suitable rat IgG2b, κ isotype control for assessing the level of background staining on paraformaldehyde-fixed/saponin-permeabilized mouse and human cells for flow cytometric analysis (see right panel). The intracellular cytokine staining technique and use of blocking controls are described in detail by C. Prussin and D. Metcalfe.

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Suggested Companion Products

<u>Catalog Number</u>	<u>Name</u>	<u>Size</u>	<u>Clone</u>
554467	PE Rat Anti-Mouse IL-10	0.1 mg	JES5-16E3
554653	MiCK-2 Mouse Cytokine Positive Control Cells	1.0 ml	(none)

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Please refer to www.bdbiosciences.com/pharming/en/protocols for technical protocols.
3. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at www.bdbiosciences.com/colors.
4. 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.

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)