

## Technical Data Sheet

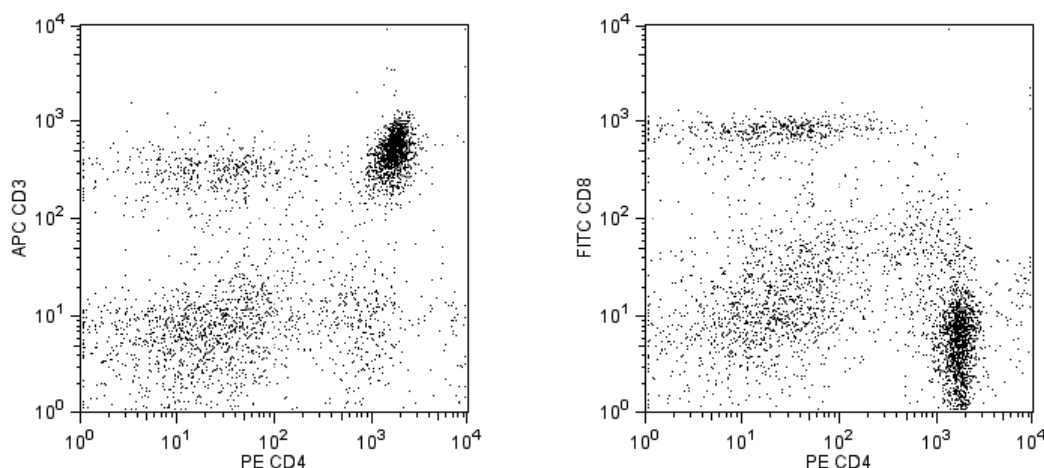
## Rat T Lymphocyte Cocktail

## Product Information

<b>Material Number:</b>	<b>558493</b>
<b>Size:</b>	50 Tests
<b>Vol. per Test:</b>	20 µl
<b>RRID:</b>	AB_771205
<b>Storage Buffer:</b>	Aqueous buffered solution containing BSA and ≤0.09% sodium azide.
<b>Component:</b>	<b>51-540-22074</b>
<b>Description:</b>	FITC anti-Rat CD8a
<b>Clone Name:</b>	OX-8
<b>Isotype:</b>	Mouse (BALB/c) IgG1, κ
<b>Component:</b>	<b>51-590-22739</b>
<b>Description:</b>	APC anti-Rat CD3
<b>Clone Name:</b>	1F4
<b>Isotype:</b>	Mouse (BALB/c) IgM, κ
<b>Component:</b>	<b>51-550-22025</b>
<b>Description:</b>	PE anti-Rat CD4
<b>Clone Name:</b>	OX-35
<b>Isotype:</b>	Mouse (BALB/c) IgG2a, κ

## Description

The Rat T Lymphocyte Cocktail is a three-color reagent cocktail designed to identify rat T lymphocytes by direct immunofluorescence staining with flow cytometric analysis. The OX-35 antibody reacts with the CD4 antigen on most thymocytes, a subpopulation of mature T lymphocytes (ie, MHC class II-restricted T cells, including most T helper cells), monocytes, macrophages, and some dendritic cells (1,2). The OX-8 antibody reacts with the hinge-like membrane-proximal domain of the 32-kDa alpha chain of the CD8 differentiation antigen (1,4). The CD8α and β chains (CD8a and CD8b, respectively) form a heterodimer on the surface of most thymocytes and a subpopulation of mature T-lymphocytes (ie, MHC class I-restricted T cells, including most T suppressor/cytotoxic cells). The 1F4 antibody reacts with the T-cell receptor-associated CD3 cell-surface antigen found on thymocytes and peripheral T lymphocytes (1,3).



**Three-color analysis of the expression of CD3, CD4, and CD8 on rat splenocytes.** A single-cell suspension of Lewis splenocytes was stained with either Isotype Control Cocktail B (Cat. no. 558508; data not shown) or Rat T Lymphocyte Cocktail (Cat. no. 558493). The figure on the left represents the CD3 and CD4 profile gating on whole spleen. The figure on the right represents the CD4 and CD8 profile gating on whole spleen. Flow cytometry was performed on a BD FACSCalibur™.

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558493 Rev. 2



## 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 the dye under optimum conditions and unconjugated antibody and free dye were removed.

## Application Notes

### Application

Flow cytometry	Tested During Development
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### Recommended Assay Procedure:

BD® CompBeads can be used as surrogates to assess fluorescence spillover (compensation). When fluorochrome conjugated antibodies are bound to BD® CompBeads, they have spectral properties very similar to cells. However, for some fluorochromes there can be small differences in spectral emissions compared to cells, resulting in spillover values that differ when compared to biological controls. It is strongly recommended that when using a reagent for the first time, users compare the spillover on cell and BD® CompBeads to ensure that BD® CompBeads are appropriate for your specific cellular application.

### Product Notices

1. Please refer to [www.bdbiosciences.com/us/s/resources](http://www.bdbiosciences.com/us/s/resources) for technical protocols.
2. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
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.
4. Please refer to <http://regdocs.bd.com> to access safety data sheets (SDS).
5. This APC-conjugated reagent can be used in any flow cytometer equipped with a dye, HeNe, or red diode laser.
6. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at [www.bdbiosciences.com/colors](http://www.bdbiosciences.com/colors).
7. An isotype control should be used at the same concentration as the antibody of interest.

### References

Brideau RJ, Carter PB, McMaster WR, Mason DW, Williams AF. Two subsets of rat T lymphocytes defined with monoclonal antibodies. *Eur J Immunol.* 1980; 10:609-615. (Clone-specific)

Jefferies WA, Green JR, Williams AF. Authentic T helper CD4 (W3/25) antigen on rat peritoneal macrophages. *J Exp Med.* 1985; 162(1):117-127. (Immunogen: Flow cytometry, Functional assay, Immunoaffinity chromatography, Immunoprecipitation, Inhibition)

Tanaka T, Masuko T, Yagita H, Tamura T, Hashimoto Y. Characterization of a CD3-like rat T cell surface antigen recognized by a monoclonal antibody. *J Immunol.* 1989; 142(8):2791-2795. (Clone-specific)

Torres-Nagel N, Kraus E, Brown MH, et al. Differential thymus dependence of rat CD8 isoform expression. *Eur J Immunol.* 1992; 22(11):2841-2848. (Biology)