Technical Data Sheet

# APC Rat Anti-Human IL-10

## Product Information

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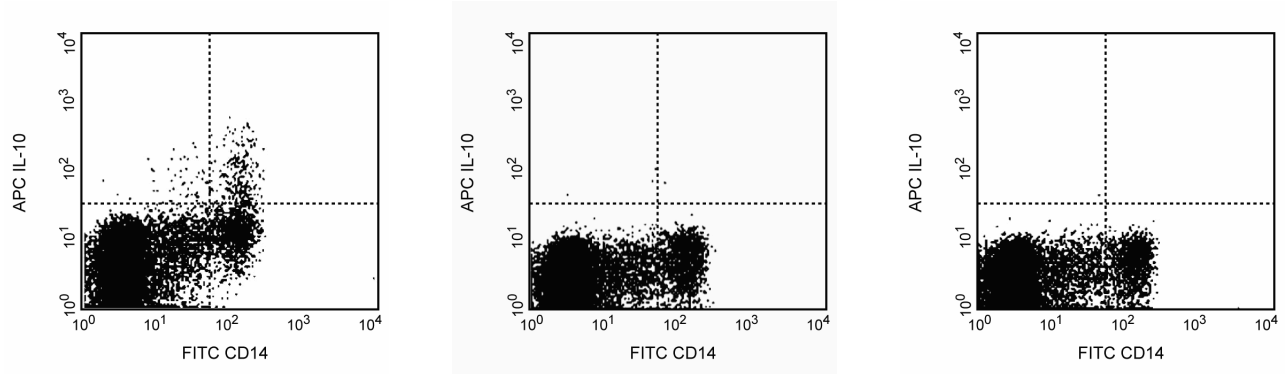
| **Material Number:** | **562036** |
| **Size:** | 25 µg |
| **Concentration:** | 0.2 mg/ml |
| **Clone:** | JES3-19F1 |
| **Immunogen:** | Recombinant Human IL-10 |
| **Isotype:** | Rat IgG2a |
| **Reactivity:** | QC Testing: Human<br>Tested in Development: Viral |
| **Storage Buffer:** | Aqueous buffered solution containing ≤0.09% sodium azide. |

## Description

The JES3-19F1 antibody reacts with human interleukin-10 (IL-10). The immunogen used to generate the JES3-19F1 hybridoma was recombinant human IL-10 expressed in COS cells. This is a neutralizing antibody. This antibody also reacts with viral IL-10.




***Expression of IL-10 by stimulated CD14+ human monocytes.*** *Human PBMC were stimulated for 24 hours with LPS (1 µg/ml final concentration) in the presence of BD GolgiStop™ (2 µM final concentration; Cat. No. 554724). The PBMC were harvested, stained with FITC-mouse anti-human CD14 antibody (Cat. No. 557153), fixed, permeabilized, and subsequently stained with 0.007 µg of APC Rat anti-Human IL-10 antibody following the BD Biosciences staining protocol (left panel). The data reflects gating on monocytes, based on forward and side scatter. To demonstrate specificity of staining, the binding of APC-JES3-19F1 was blocked by the preincubation of the conjugated antibody with recombinant human IL-10 (0.25 µg, Cat. No. 554611; middle panel), and by preincubation of the fixed/permeabilized cells with Purified Rat anti-Human IL-10 (10 µg, Cat. No. 554705; right panel). The quadrant markers for the bivariate dot plots were set based on the autofluorescence control, and verified with the recombinant cytokine blocking (middle panel) and unlabelled antibody (right panel) blocking specificity 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 APC under optimum conditions, and unconjugated antibody and free APC were removed.

## Application Notes

**Application**

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| Intracellular staining (flow cytometry) | Routinely Tested |

**Recommended Assay Procedure:**

**Immunofluorescent Staining and Flow Cytometric Analysis:** The JES3-19F1 antibody is useful for immunofluorescent staining and flow cytometric analysis to identify and enumerate IL-10 producing cells within mixed cell populations. For optimal immunofluorescent staining and flow cytometric analysis, this anti-cytokine antibody should be titrated (≤ 0.5 µg mAb/million cells). For specific methodology, please visit our web site, www.bdbiosciences.com, and go to the protocol section or the chapter on intracellular staining in the Immune Function Handbook. In addition, staining techniques and use of blocking controls are described in detail by C. Prussin and D. Metcalfe.

**BD Biosciences**

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## Suggested Companion Products

| Catalog Number | Name | Size | Clone |
|---|---|---|---|
| 555062 | HiCK-2 Human Cytokine Positive Control Cells | 1.0 ml | (none) |
| 554705 | Purified Rat Anti-Human IL-10 | 0.5 mg | JES3-19F1 |
| 554690 | APC Rat IgG2a κ Isotype Control | 0.1 mg | R35-95 |
| 554715 | BD Cytofix/Cytoperm Plus Kit (with BD GolgiStop) | 250 tests | (none) |
| 557153 | FITC Mouse Anti-Human CD14 | 50 tests | M5E2 |
| 554611 | Recombinant Human IL-10 | 5 µg | (none) |

## Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Please refer to www.bdbiosciences.com/pharmingen/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.
5. This APC-conjugated reagent can be used in any flow cytometer equipped with a dye, HeNe, or red diode laser.
6. An isotype control should be used at the same concentration as the antibody of interest.

## References


Andersson EC, Christensen JP, Marker O, Thomsen AR. Changes in cell adhesion molecule expression on T cells associated with systemic virus infection. *Immunology.* 1994; 152(3):1237-1245. (Clone-specific)

D'Andrea A, Aste-Amezaga M, Valiante NM, Ma X, Kubin M, Trinchieri G. Interleukin 10 (IL-10) inhibits human lymphocyte interferon gamma-production by suppressing natural killer cell stimulatory factor/IL-12 synthesis in accessory cells. *J Exp Med.* 1993; 178(3):1041-1048. (Clone-specific: Neutralization)

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: Blocking)