Technical Data Sheet

PE Rat Anti-Mouse CD126

Product Information

Material Number: 554462
Alternate Name: IL-6 Receptor α chain
Size: 0.2 mg
Concentration: 0.2 mg/ml
Clone: D7715A7
Immunogen: OKT4 hybridoma cells
Isotype: Rat IgG2b, κ
Reactivity: QC Testing: Mouse
Storage Buffer: Aqueous buffered solution containing ≤0.09% sodium azide.

Description
The D7715A7 antibody reacts with the mouse IL-6 receptor. The immunogen used to generate the D7715A7 hybridoma was OKT4 hybridoma cells. The D7715A7 hybridoma was selected based on its capacity to produce antibody that inhibited IL-6 binding to 2F4 cells (mouse B-cell hybridoma expressing high levels of IL-6 receptors). The binding of purified D7715A7 to B9 cells (mouse B-cell hybridoma expressing high levels of IL-6 receptors) is inhibited by recombinant mouse IL-6. This antibody has been reported to inhibit the in vitro and in vivo growth of the IL-6-dependent plasmacytoma line, T1033C2.

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

Application Notes

Recommended Assay Procedure:
The PE conjugated D7715A7 antibody can be used for the immunofluorescent staining and flow cytometric analyses of mouse peripheral blood leukocytes and cell lines which express IL-6 receptors.

1. To help block nonspecific staining due to Fc receptors, preincubate ~1 million cells with 1 µg of the purified anti-CD32/CD16 (anti-FcγII/III receptor) antibody (clone 2.4G2, Fc Block™, Cat. No. 553142/553143) for 30 minutes at 4°C.

2. Incubate the cells with 0.12 - 2.0 µg of PE conjugated D7715A7 antibody (Cat. No. 554462) at 4°C for 45 minutes. Wash cells three times with staining medium containing sodium azide (e.g., Dulbecco’s PBS or tissue culture medium [without phenol red] with 0.09% sodium azide and 1% heat-inactivated FCS). We encourage investigators to titrate the D7715A7 antibody up to saturating levels for optimal performance, minimizing the risk for dim staining.

3. Resuspend cells in staining medium and analyze by flow cytometry using appropriate specificity and compensation controls. Using this method, positive staining was seen with the B9 cell line but not with the mouse MC/9 mast cell line and C20.4 CD4+ T cell line.

Suggested Companion Products

<table>
<thead>
<tr>
<th>Catalog Number</th>
<th>Name</th>
<th>Size</th>
<th>Clone</th>
</tr>
</thead>
<tbody>
<tr>
<td>553142</td>
<td>Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block™)</td>
<td>0.5 mg</td>
<td>2.4G2</td>
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<tr>
<td>553989</td>
<td>PE Rat IgG2b, κ Isotype Control</td>
<td>0.1 mg</td>
<td>A95-1</td>
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<tr>
<td>554656</td>
<td>Stain Buffer (FBS)</td>
<td>500 ml</td>
<td>(none)</td>
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Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
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. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at www.bd biosciences.com/colors.
5. An isotype control should be used at the same concentration as the antibody of interest.

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References
Zola H. Detection of receptors for cytokines and growth factors. The Immunologist. 1994:47. (Biology)