

## Technical Data Sheet

## PE Rat Anti-Mouse CD126

## Product Information

<b>Material Number:</b>	<b>554462</b>
<b>Alternate Name:</b>	IL-6 Receptor $\alpha$ chain
<b>Size:</b>	0.2 mg
<b>Concentration:</b>	0.2 mg/ml
<b>Clone:</b>	D7715A7
<b>Immunogen:</b>	OKT4 hybridoma cells
<b>Isotype:</b>	Rat IgG2b, $\kappa$
<b>Reactivity:</b>	QC Testing: Mouse
<b>Storage Buffer:</b>	Aqueous buffered solution containing $\leq 0.09\%$ 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

## Application

Flow cytometry	Routinely Tested
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## 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  $\mu$ g of the purified anti-CD32/CD16 (anti-Fc $\gamma$ II/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  $\mu$ g 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

Catalog Number	Name	Size	Clone
553142	Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block™)	0.5 mg	2.4G2
553989	PE Rat IgG2b, $\kappa$ Isotype Control	0.1 mg	A9S-1
554656	Stain Buffer (FBS)	500 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/pharmingen/protocols](http://www.bdbiosciences.com/pharmingen/protocols) for technical protocols.
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.bdbiosciences.com/colors](http://www.bdbiosciences.com/colors).
5. An isotype control should be used at the same concentration as the antibody of interest.

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## References

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Coulie PG, Vink A, Van Snick J. A monoclonal antibody specific for the murine IL-6-receptor inhibits the growth of a mouse plasmacytoma in vivo. *Curr Top Microbiol Immunol.* 1990; 166:43-46. (Clone-specific)

Vink A, Coulie P, Warnier G, Renauld JC, Stevens M, Donckers D, Van Snick J. Mouse plasmacytoma growth in vivo: enhancement by interleukin 6 (IL-6) and inhibition by antibodies directed against IL-6 or its receptor. *J Exp Med.* 1990; 172(3):997-1000. (Biology)