



## CELL LAB Rat Anti-Mouse CD25/IL-2R $\alpha$

Cat. No.	Form	Quantity
732095	Purified (UNLB) Antibody	0.5 mg
732096	Fluorescein (FITC) Conjugate	0.5 mg
733281	Biotin (BIOT) Conjugate	0.5 mg
732097	Phycoerythrin (PE) Conjugate	0.1 mg
733282	Phycoerythrin (PE) Conjugate	0.2 mg
732098	Allophycocyanin (APC) Conjugate	0.1 mg
733283	Spectral Red™ (SPRD) Conjugate	0.1 mg

### For Laboratory Use Only

#### DESCRIPTION

**Clone:** 3C7  
**Isotype:** Rat IgG2b $\kappa$   
**Specificity:** Low affinity  $\alpha$  chain IL-2 receptor, Mr 55 kDa (IL-2R $\alpha$ )

The IL-2 receptor (IL-2R) exists in three alternative forms made up from the individual components of CD25, CD122, and CD132. CD25 represents the low affinity  $\alpha$  chain of the IL-2R, a type I transmembrane glycoprotein containing two CCP domains. It is rich in O-linked carbohydrates and has a short cytoplasmic tail.<sup>14</sup> CD25 is expressed on activated T cells, B cells, NK cells and monocytes of all mouse strains tested.<sup>2,3,15</sup> Expression of CD25 on activated T lymphocytes is transitory and endogenously regulated.<sup>4</sup> CD25 is expressed on precursor B cells in bone marrow. Its expression is initiated by functional rearrangement and expression of IgM heavy chain genes and is down-regulated when immature B cells mature and express IgD.<sup>5</sup> It is expressed at a higher level on CD4<sup>+</sup>CD8<sup>+</sup> thymocytes.<sup>5,6</sup> It is also expressed on cultured epidermal Langerhans cells.<sup>7</sup> The biochemical features of murine CD25 have been characterized in detail.<sup>8-10</sup> The 3C7 monoclonal antibody reacts with an epitope of CD25 which is distinct from that recognized by clone 7D4.<sup>2,11,12</sup> 3C7, used in combination with 7D4 in culture, results in higher levels of inhibition of proliferation driven by IL-2 and generation of alloreactive CTL than either of these monoclonal antibodies alone.<sup>2,13</sup>

#### APPLICATIONS

- Identification and enumeration of CD25<sup>+</sup> cells by flow cytometry<sup>1-4</sup>
- Immunoprecipitation<sup>1,2,8,10</sup>
- Inhibition of IL-2 or mitogen-induced T cell proliferation<sup>13</sup>
- *In vitro* blocking of IL-2 binding<sup>2,12</sup>

#### CHARACTERIZATION

To ensure lot-to-lot consistency, each batch of product is tested to conform with characteristics of a standard reference reagent using immunofluorescence staining and flow cytometry.

#### WORKING DILUTIONS

**Flow Cytometry:**

FITC conjugate	$\leq 3 \mu\text{g}/10^6$ cells
BIOT conjugate	$\leq 3 \mu\text{g}/10^6$ cells
PE conjugate	$\leq 0.3 \mu\text{g}/10^6$ cells
APC conjugate	$\leq 0.3 \mu\text{g}/10^6$ cells
SPRD conjugate	$\leq 0.3 \mu\text{g}/10^6$ cells

**Other Applications:** Since applications vary, determine the optimum working dilution of the product that is appropriate for your specific needs.

### **HANDLING AND STORAGE**

- The purified (UNLB) antibody is supplied as 0.5 mg of purified immunoglobulin in 1.0 mL of 100 mM borate buffered saline, pH 8.0. No preservatives or amine-containing buffer salts added.
- The fluorescein (FITC) conjugate is supplied as 0.5 mg in 1.0 mL of PBS/NaN<sub>3</sub>
- The phycoerythrin (PE) conjugates are supplied as 0.1 mg in 1.0 mL or 0.2 mg in 2.0 mL of PBS/NaN<sub>3</sub> and a stabilizing agent.
- The allophycocyanin (APC) conjugate is supplied as 0.1 mg in 1.0 mL of PBS/NaN<sub>3</sub> and a stabilizing agent.
- The Spectral Red (SPRD) conjugate is supplied as 0.1 mg in 1.0 mL of PBS/NaN<sub>3</sub> and a stabilizing agent.
- Protect fluorochrome-conjugated forms from light. Do not freeze.
- Reagent is stable until the expiration date on the vial when stored at 2-8°C.

### **STATEMENT OF WARNINGS**

1. Specimens, samples and all material coming in contact with them should be handled as if capable of transmitting infection and disposed of with proper precautions.
2. Never pipet by mouth and avoid contact of samples with skin and mucous membranes.
3. Do not use reagent beyond the expiration date on the vial label.
4. Minimize exposure of reagent to light during storage or incubation.
5. Avoid microbial contamination of reagent or erroneous results may occur.
6. Use Good Laboratory Practice (GLP) when handling this reagent.
7. Harmful if swallowed.
8. After contact with skin, wash immediately with plenty of water.
9. Contains sodium azide. Sodium azide under acidic conditions yields hydrazoic acid, an extremely toxic compound. Azide compounds should be flushed with running water while being discarded. These precautions are recommended to avoid deposits in metal piping in which explosive conditions can develop. If skin or eye contact occurs, immediately wash excessively with water.

### **TRADEMARKS**

The Beckman Coulter logo is a trademark of Beckman Coulter, Inc.

Spectral Red is a trademark of Southern Biotechnology Associates, Inc.

Cy5 is a trademark of GE Healthcare, Inc.

For additional information or if damaged product is received, contact your local Beckman Coulter Representative.

Spectral Red is a PE/Cy<sup>TM</sup>5 tandem conjugate. Cy5 is for non-commercial research use only, not for therapeutic or in vivo applications. Other use needs license from Amersham Biosciences Corp., under U.S. Patent Nos. 4,981,977 and 5,268,486 and other patents pending. This material (or portions of this material) is subject to proprietary rights of Amersham Biosciences Corp. and Carnegie Mellon University and made and sold under license from Amersham Biosciences Corp. This product is licensed for sale only for research. It is not licensed for any other use. There is no implied license hereunder for any commercial use. Commercial use shall include: 1) sale, lease, license or other transfer of the material or any material derived or produced from it 2) sale, lease, license or other grant of rights to use this material or any material derived or produced from it 3) use of this material to perform services for a fee for third parties. If you require a commercial license to use this material and do not have one, return this material, unopened to Beckman Coulter, Inc. 11800 SW 147 Ave. Miami, FL 33196, USA and any money paid for the material will be refunded.

## REFERENCES

1. Malek TR, Robb RJ and Shevach EM. 1983. Identification and initial characterization of a rat monoclonal antibody reactive with the murine interleukin 2 receptor-ligand complex. *Proc Natl Acad Sci U S A*, 80:5694-5698.
2. Ortega G, Robb RJ, Shevach EM and Malek TR. 1984. The murine IL 2 receptor. I. Monoclonal antibodies that define distinct functional epitopes on activated T cells and react with activated B cells. *J Immunol*, 133:1970-1975.
3. Malek TR, Schmidt JA and Shevach EM. 1985. The murine IL 2 receptor. III. Cellular requirements for the induction of IL 2 receptor expression on T cell subpopulations. *J Immunol*, 134:2405-2413.
4. Andrew ME, Churilla AM, Malek TR, Braciale VL and Braciale TJ. 1985. Activation of virus specific CTL clones: antigen-dependent regulation of interleukin 2 receptor expression. *J Immunol*, 134:920-925.
5. Chen J, Ma A, Young F and Alt FW. 1994. IL-2 receptor alpha chain expression during early B lymphocyte differentiation. *Int Immunol*, 6:1265-1268.
6. Habu S, Okumura K, Diamantstein T and Shevach EM. 1985. Expression of interleukin 2 receptor on murine fetal thymocytes. *Eur J Immunol*, 15:456-460.
7. Steiner G, Tschachler E, Tani M, Malek TR, Shevach EM, Holter W, Knapp W, Wolff K and Stingl G. 1986. Interleukin 2 receptors on cultured murine epidermal Langerhans cells. *J Immunol*, 137:155-159.
8. Malek TR and Korty PE. 1986. The murine interleukin 2 receptor. IV. Biochemical characterization. *J Immunol*, 136:4092-4098.
9. Malek TR, Ashwell JD, Germain RN, Shevach EM and Miller J. 1986. The murine interleukin-2 receptor: biochemical structure and regulation of expression. *Immunol Rev*, 92:81-101.
10. Saragovi H and Malek TR. 1988. Direct identification of the murine IL-2 receptor p55-p75 heterodimer in the absence of IL-2. *J Immunol*, 141:476-482.
11. Lorenzo F, Jaulin C, Vita N, Froussard P, Ferrara P, Jankovic DL and Theze J. 1991. Structure-function study of the p55 subunit of murine IL-2 receptor by epitope mapping. *J Immunol*, 147:2970-2977.
12. Moreau JL, Nabholz M, Diamantstein T, Malek T, Shevach E and Theze J. 1987. Monoclonal antibodies identify three epitope clusters on the mouse p55 subunit of the interleukin 2 receptor: relationship to the interleukin 2-binding site. *Eur J Immunol*, 17:929-935.
13. Malek TR, Ortega G, Jakway JP, Chan C and Shevach EM. 1984. The murine IL 2 receptor. II. Monoclonal anti-IL 2 receptor antibodies as specific inhibitors of T cell function in vitro. *J Immunol*, 133:1976-1982.
14. Leonard WJ, Depper JM, Crabtree GR, Rudikoff S, Pumphrey J, Robb RJ, Kronke M, Svetlik PB, Peffer NJ, Waldmann TA. 1984. Molecular cloning and expression of cDNAs for the human interleukin-2 receptor. *Nature*, 311:626-631.
15. Rolink A, Grawunder U, Winkler TH, Karasuyama H and Melchers F. 1994. IL-2 receptor alpha chain (CD25, TAC) expression defines a crucial stage in pre-B cell development. *Int Immunol*, 6:1257-1264.



Manufactured for:  
Beckman Coulter, Inc.  
4300 N. Harbor Blvd.  
Fullerton, CA 92835  
[www.beckmancoulter.com](http://www.beckmancoulter.com)

Printed in USA  
Made in USA

© 2005 Beckman Coulter, Inc.  
All Rights Reserved.

PN 733926-A