



**For Research Use Only.**  
**Not for use in diagnostic procedures.**

### SPECIFICITY

CD28 is a T-cell surface homodimeric molecule that belongs to the Ig superfamily.<sup>1</sup> It is composed of disulfide-linked chains with a molecular weight of 44 kDa.<sup>2</sup>

CD28 is involved in cell adhesion T-B through the binding to its ligand B7/BB1 that is also the ligand for CTLA-4. CD28-B7 interaction is an important cosignal that allows T-cell proliferation and lymphokine production. CD28 is associated to a PI3 kinase activity.<sup>2,4</sup>

The majority of CD4<sup>+</sup> T-cells and 50% of CD8<sup>+</sup> T-cells express CD28.<sup>3</sup> The CD28.2 monoclonal antibody induces T-cell proliferation in costimulation with CD2 monoclonal antibodies.<sup>2</sup> It also inhibits the CD4<sup>+</sup> proliferation in the allogeneic T-cell response.<sup>2</sup> This antibody has been studied at the 5th International Workshop on Human Leucocyte Differentiation Antigens.<sup>2</sup>

### REAGENTS

IOtest CD28-ECD Conjugated Antibodies  
PN 6607111 - 100 tests - 10 µL/test

**CLONE:** CD28.2

**ISOTYPE:** IgG1

**IMMUNOGEN:** Transfected murine cell line

**HYBRIDOMA:** Myeloma X63 Ag8.653 x BALB/c spleen cells

**SOURCE:** Ascites fluid

**PURIFICATION:** Ion exchange or affinity chromatography

**CONJUGATION:** ECD is conjugated at a Molar Ratio ECD/Ig: 0.5-1.5  
Excitation wavelength at 486-575 nm  
Emission wavelength at 610-635 nm

**BUFFER:** 2 mg/mL bovine serum albumin in phosphate-buffered saline containing 0.1% sodium azide.

### STATEMENT OF WARNINGS

1. This reagent contains 0.1% sodium azide. Sodium azide under acid 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, wash excessively with water.
2. 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.
3. Never pipet by mouth and avoid contact of samples with skin and mucous membranes.

4. Do not use antibody beyond the expiration date on the label.
5. Do not expose reagents to strong light during storage or incubation.
6. Use Good Laboratory Practices (GLP) when handling reagent.
7. Harmful if swallowed.
8. After contact with skin, wash immediately with plenty of water.

### STORAGE CONDITIONS AND STABILITY

This reagent is stable up to the expiration date when stored at 2-8°C. Do not freeze. Minimize exposure to light.

### REAGENT PREPARATION

No reconstitution is necessary. This monoclonal antibody may be used directly from the vial. Bring reagent to 18-25°C prior to use.

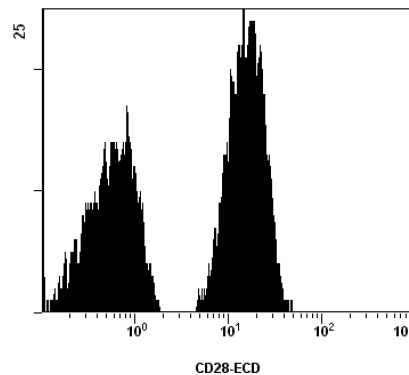
### PROCEDURE

This reagent is designed for flow cytometry. Assay volume: 10 µL per 5 x 10<sup>5</sup> cells in one test, or per 100 µL whole blood. A wash is required to yield optimal results.

### EXAMPLE DATA

The histogram shown is a monoparametric representation (Count versus Fluorescence Intensity) of lysed normal whole blood sample stained with CD28-ECD monoclonal antibody (PN 6607111) and gated on lymphocytes.

**Figure 1:**  
**Acquisition with a COULTER® EPICS® XL™/XL-MCL™ flow cytometer. Analysis with EXPO™32 ADC software.**



### SELECTED RESEARCH REFERENCES

1. McMichael, A.J. and Gotch, F.M., "T-cell antigens: new previously defined clusters", 1987, in *Leucocyte Typing III, White Cell Differentiation Antigens*, A.J. McMichael et al., Eds., Oxford University Press, p. 30-62.
2. Olive D., Cerdan C., Costello R., Sielleur I., Ragueneau M., Pages F., Klasen S. Nunès J., Imbert J., "CD28 and CTLA-4 cluster report", 1995, in *Leucocyte Typing V, White Cell Differentiation Antigens*, S. F. Schlossman et al., Eds., Oxford University Press, p. 360-370.
3. Tan, R., Teh, S.J., Ledbetter, J.A., Linsley, P., Teh, H.S. "B7 costimulates proliferation of CD4<sup>+</sup> T lymphocytes but is not required for the deletion of immature CD4<sup>+</sup>8<sup>+</sup> Thymocytes", 1992, *J. Immunol.*, 149, 3217-3224.
4. Nunès J., Klasen S., Ragueneau M., Pavon C., Couez D., Mawas C., Bagnasco M., Olive D., "CD28 mAbs with distinct binding properties differ in their ability to induce T cell activation: analysis of early and late activation events", 1993, *International Immunology*, 5, 3, 311-315.

### TRADEMARKS

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