

IOTest CD28-APC-Alexa Fluor 700

PN B12696 – 0.5 mL – Liquid – 10 µL/test – Clone CD28.2

Analyte Specific Reagent.

Analytical and performance characteristics are not established.

SPECIFICITY

The CD28 antigen, (Tp44), is a homodimeric, disulfide-linked, type 1 transmembrane protein; the monomer is 202 amino acid-long, with a molecular weight of 44 kDa (1, 2). The extracellular region, homologous to an Ig V-like domain, shares significant amino acid sequence with the CD152 antigen (CTLA-4) (3).

The CD28 antigen is involved in the interaction of T lymphocytes with professional antigen-presenting cells (APCs), through its counter-receptors, B7-1/BB-1 (CD80) and B7-2/B70 (CD86). It provides a major co-stimulatory signal for T cell activation, proliferation and lymphokine production. The CD28 family of receptors (CD28, CTLA-4, ICOS, PD-1 and BTLA) plays a critical role in controlling the adaptive immune response. The CD28 receptor can enhance T cell antigen receptor (TCR) signals, as well as deliver independent signals. Although the signals through CD28 are crucial for the initial co-stimulation of interleukin-2 (IL-2) production (4), a TCR-independent CD28 signal leads to the selective transcription of survival, but not proliferative genes (5).

The cytoplasmic region of CD28 can associate with the PI3-kinase (2), the GRB-2/SOS complex, and the T cell-specific protein-tyrosine kinase ITK (6, 7). The pYMN motif of the intracellular domain of CD28 binds to the SH2 domains of PI3-kinase and GRB-2.

CD28 is expressed on T cells, on plasma cells and thymocytes (1, 8).

The CD28.2 monoclonal antibody has been assigned to the CD28 cluster of differentiation at the 5th International Workshop on Human Leucocyte Differentiation Antigens in Boston, U.S.A., in 1993 (1).

REAGENT

IOTest CD28-APC-Alexa Fluor 700
Conjugated antibody
PN B12696 - 0.5 mL - Liquid - 10 µL/test

Clone	CD28.2
Isotype	IgG1, Mouse
Immunogen	Transfected murine cell line
Hybridoma	X63 x (balb/c x C3H)
Source	Ascites fluid or supernatant of in vitro cultured hybridoma cells.
Purification	Affinity chromatography
Conjugation	Allophycocyanin-Alexa Fluor 700 (APC-Alexa Fluor)
Molar Ratio	APC-Alexa Fluor / Ig : 0.5 - 1.5
Fluorescence	Excites at 633/638 nm Emits at 720 nm

REAGENT CONTENTS

This antibody is provided in phosphate-buffered saline, containing 0.1% sodium azide and 2 mg/mL bovine serum albumin.

STATEMENTS OF WARNING

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 considered potentially infectious 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. Avoid microbial contamination of reagents or incorrect results might occur.
7. Use good laboratory practices when handling this reagent.

PRECAUTIONS

Due to the tandem structure of the fluorochrome, APC-Alexa Fluor 700 also emits light at 660 nm. This secondary emission peak varies from lot-to-lot of APC-Alexa Fluor 700. Therefore, for multi-color analysis, the compensation matrix should be carefully checked when changing the lot of a APC-Alexa Fluor 700-conjugate.

Weak non-specific binding on a lymphocyte subpopulation may occur on some donors with APC-Alexa Fluor 700 conjugates.

STORAGE AND HANDLING CONDITIONS AND STABILITY

This reagent is stable up to the expiration date when stored at 2 – 8°C. Do not freeze. No reconstitution is necessary. This monoclonal antibody may be used directly from the vial. Bring reagent to 18 – 25°C prior to use.

SELECTED RESEARCH REFERENCES

1. 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, Vol. 1. Schlossman, S.F., et al., Eds., Oxford University Press, 360-370.
2. Ghiotto-Ragueneau, M., Battifora, M., Truneh, A., Waterfield, M.D., Olive, D., "Comparison of CD28-B7.1 and B7.2 functional interaction in resting human T cells: phosphatidylinositol 3-kinase association to CD28 and cytokine production", 1996, Eur. J. Immunol., 26, 34-41.
3. Buonavista, N., "Molecular linkage of the human CTLA4 and CD28 Ig-superfamily genes in yeast artificial chromosomes", 1992, Genomics, 13, 856-861.
4. Sharpe, A.H., Freeman, G.J., "The B7-CD28 superfamily", 2002, Nature Rev. Immunol., 2, 116-126.
5. Marinari, B., Costanzo, A., Marzano, V., Piccolella, E., Tuosto, L., "CD28 delivers a unique signal leading to the selective recruitment of RelA and p52 NF-κB subunits on IL-8 and Bcl-xL gene promoters", 2004, Proc. Natl. Acad. Sci. USA, 101, 6098-6103.
6. June, C.H., Bluestone, J.A., Nadler, L.M., Thompson, C.B., "The B7 and CD28 receptor families", 1994, Immunol. Today, 15, 321-31.
7. Raab, M., Cai, Y., Bunnell, S.C., Heyeck, S.D., Berg, L.J., Rudd, C.E., "p⁵⁶Lck and p⁵⁹Fyn regulate CD28 binding to phosphatidylinositol 3-kinase, growth factor receptor-bound protein GRB-2, and T cell-specific protein-tyrosine kinase ITK: implications for T-cell costimulation", 1994, Proc. Natl. Acad. Sci. USA, 92, 8891-8895.
8. Bani, L., David, D., Moreau, J.L., Cayota, A., Nakarai, T., Ritz, J., Thèze, J., "Expression of the IL-2 receptor γ subunit in resting human CD4 T lymphocytes: mRNA is constitutively transcribed and the protein stored as an intracellular component", 1997, Int. Immunol., 4, 9, 573-580.

TRADEMARKS

Beckman Coulter logo and IOTest, are trademarks of Beckman Coulter; Beckman Coulter logo, IOTest are registered in the USPTO and SIPO.

Alexa Fluor is a trademark of Molecular Probes, Inc.

IOTest CD28-APC-Alexa Fluor 700

PN B12696 – 0.5 mL – Liquid – 10 µL/test – Clone CD28.2

MANUFACTURED BY :
IMMUNOTECH SAS
a Beckman Coulter Company
130, avenue de Lattre de Tassigny
B.P. 177 - 13276 Marseille Cedex 9
France

For additional information in the USA, call
800-526-7694.
Outside the USA, contact your local Beckman
Coulter representative.

www.beckmancoulter.com

Printed in France.
Made in France.

©2011 Beckman Coulter, Inc.