PERFORMANCE DATA SHEET

2440

Monoclonal anti-human CD279(PD-1)/Biotin*



mAb name/Clone: ANC4H6 *Isotype:* Murine IgG1κ

Immunogen: Recombinant human CD279

CATALOG#: 279-030 QUANTITY: 100 μg

CONCENTRATION: 1.0 mg/ml

INFORMATION: Human CD279 (PD-1, Programmed death 1) is a 55 kd Ig superfamily member with sililarity to CD28 and CD152(CTLA-4). It is expressed on activated T and B and myeloid cells, and engagement by its ligands PD-L1 (CD274, B7-H1) or PD-L2 (B7-DC) can inhibit proliferation and cytokine expression. In mice blockade of PD-1 ligand interaction has been used to augment T cell anti cancer responses (3,4,5).

Anti-CD279 antibody clone ANC4H6 binds to CD279 present on PHA activated PBMC in FACS, and binds to recombinant CD279 in EIA. Pre incubation of recombinant CD279-coated plates with ANC4H6 blocks binding of biotinylated recombinant CD273 in EIA.

References: 1) Ishida et al. (1992) *EMBO J* **11**(11):3887-95. 2) Carreno BM, M Collins, et al. (2001) *Annu Rev Immunol* **20**:29. 3) MA Curran, Allison JP, et al. (2010) *PNAS* **107**(9): 4275-80. 4) Mangsbo SM, TH Totterman, et al. (2010) *J Immunotherapy* **33**(3):225. 5) Hirano F, L Chen, et al. (2005) *Canc Res* **65**: 1089.

STORAGE CONDITIONS: *Store at 2 - 5^oC*. Freeze/thawing not recommended.

PRODUCT STABILITY: Product should retain activity for at least 12 months after shipping date when stored as recommended. Ship Date:_____

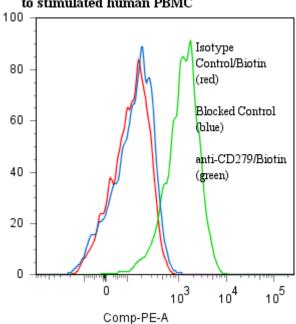
BUFFER: 50 mM Sodium Phosphate pH 7.5, 100 mM Potassium Chloride, 150mM NaCl, 5% Glycerol, 0.2% BSA, 0.04% NaN₃ (as a preservative).

PRODUCTION: Antibody from (low FBS containing) tissue culture supernatant was Protein A purified to 95% Immunoglobulin by SDS-PAGE, and reacted with NHS-Biotin.

Unconjugated Biotin was removed from conjugate using a desalting column.

PERFORMANCE: Ficoll prepared human peripheral blood mononuclear cells (**PBMC**) were stimulated 5 days in culture with 5 μg/ml Phytohemagglutanin (PHA). Five x 10^5 cells per tube were pre incubated 10 minutes with 250 μg/ml human IgG (to block nonspecific binding) after which they were incubated 45 minutes on ice with 80 μl of product at a concentration of **10** μg/ml. Cells were then washed twice and incubated 45 minutes with 20 μl of 2^0 reagent Streptavidin/R-PE (Catalog #253-050), after which they were washed three times, fixed and analyzed by FACS. Cells stained positive with a mean shift of **1.04** log₁₀ fluorescent units when compared to a Mouse IgG1/Biotin negative control (Catalog #278-030) at a similar concentration. Binding was blocked when reagent was pre incubated 10 minutes with a ten-fold excess of recombinant CD279-muIg.

Binding of anti-CD279(PD-1)/Biotin +SA/PE to stimulated human PBMC



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