

**PERFORMANCE DATA SHEET**  
2011

**Monoclonal anti-human CD274 (B7-H1, PD-L1)/R-PE\***

**mAb name/Clone:** ANC6H1  
**Isotype:** Mouse IgG1κ  
**Immunogen:** Recombinant human CD274

**CATALOG#:** 274-050

**QUANTITY:** 120 tests

**VOLUME IN VIAL:** 0.2 ml

**WORKING DILUTION:** 1:50 (or use 1.6µl of concentrated stock per 5 x 10<sup>5</sup>-cell test)

**INFORMATION:** CD274 (B7-H1, PD-L1, Programmed Death Ligand) is a member of the B7 family and is expressed on a variety of tissues including lymphoid cells. It plays an important role in regulation of T cell activation, and is involved in progression of cancer, arthritis and HIV infection (3). CD274 binding to its receptor CD279 (PD-1) on activated T cells can decrease proliferation. Conversely, ligation of CD279 on primed T cells can stimulate IL-10 production. High levels of CD274 present in Renal cell carcinoma is associated with poor prognosis (1). Tumor expressed CD274 can increase apoptosis of tumor specific T cells resulting in better tumor cell survival (2). Gamma Interferon and PHA can up regulate CD274 expression on T cells. Antibody ANC6H1 binds to recombinant CD274 and native cell surface CD274 expressed on stimulated PBMC.

**REFERENCES:** 1)Cancer Research (April 2006) 66:3381. 2)J Molecular Medicine (Feb 2004) 81(5):281. 3)Int J Hematol (Nov 2003) 78(4):321.

**STORAGE CONDITIONS:** Store at 2 - 5°C. Do not freeze! Protect from light.

**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, 500 mM Potassium Chloride, 150mM NaCl, 15% Glycerol, 0.2% BSA, 0.04% NaN<sub>3</sub> (as a preservative).

**PRODUCTION:** Protein A purified antibody from tissue culture supernatant was conjugated to R-Phycoerythrin through a sulfo-ester linkage. Unconjugated antibody was removed using size exclusion chromatography. Antibody conjugate is at 1 mg/ml with an A<sub>565</sub>/A<sub>280</sub> ratio of 3.04.

**PERFORMANCE:** Ficoll prepared human peripheral blood mononuclear cells (PBMC) were stimulated overnight in culture with 5 µg/ml PHA. Five x 10<sup>5</sup> cells per tube were washed and pre incubated 5 minutes with 20µl of 250µg/ml human IgG (to block non specific binding), after which they were incubated 45 minutes on ice with 80 µl of anti-human CD274 at a dilution factor of 1:50 (20 µg/ml). They were washed three times, fixed and analyzed by FACS. Cells stained positive with a mean shift of 0.67 log<sub>10</sub> fluorescent units when compared to a Mouse IgG1/R-PE negative control (Catalog #278-050) at a similar concentration. Binding was blocked when cells were pre incubated 10 minutes with 20 µl of 0.5 mg/ml anti-CD274 antibody (Catalog #274-020).

*\*This Product is intended for Laboratory Research use only.* R-Phycoerythrin (R-PE) is covered under patents: U.S. 4,520,110; European 76,695 and Canadian 1,179,942.

