## PERFORMANCE DATA SHEET

2011

## Ancell

## Monoclonal anti-human CD274 (B7-H1, PD-L1)/FITC\*

*mAb name/Clone:* ANC6H1 *Isotype:* Mouse IgG1κ

Immunogen: Recombinant human CD274

CATALOG#: 274-040 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:\_\_\_\_\_\_

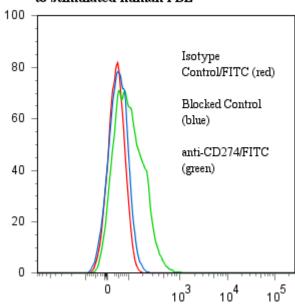
**BUFFER:** 50 mM Sodium Phosphate pH 7.5, 100 mM Potassium Chloride, 150mM NaCl, 5% Glycerol, 0.2% BSA, 0.04% NaN<sub>3</sub> (as a preservative).

**PRODUCTION:** Protein A purified antibody from tissue culture supernatant was reacted with FITC. Unconjugated FITC was separated from antibody/FITC conjugate by desalting column. The antibody/FITC conjugate is at approximately **0.5 mg/ml** with a Fluorescein/IgG molar ratio of 10.5.

**PERFORMANCE:** Ficoll prepared human peripheral blood mononuclear cells (PBMC) were stimulated overnight in culture with 5 μg/ml PHA. Five x  $10^5$  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/FITC at a **1:50** dilution (10 μg/ml). They were washed three times, fixed and analyzed by FACS. Cells stained positive with a mean shift of **0.5**  $\log_{10}$  fluorescent units when compared to a Mouse IgG1/FITC negative control (Catalog #278-040) 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.

## Binding of anti-CD274(B7-HL)/FITC to stimulated human PBL



Ancell Corporation P.O. Box 87 Bayport, MN 55003-0087 USA Phone: Toll free 800-374-9523 or 651-439-0835 Fax: 651-439-1940