PERFORMANCE DATA SHEET
3039

Monoclonal anti-human IFNγ/R-PE*

mAb name/Clone: **ANC2E11**
Isotype: Mouse IgG1κ
Immunogen: Recombinant human IFN-gamma

CATALOG#: 247-050
QUANTITY: 120 tests **VOLUME IN VIAL: 0.2 ml**
WORKING DILUTION: 1:50 (or use 1.6µl of concentrated stock per 5 x 10^5-cell test)

INFORMATION: The cytokine human interferon-gamma (IFN-gamma) is a glycosylated protein that exists as a homodimer of 34 kd in its biologically active form. IFN-gamma is expressed upon activation by TH1 cells $CD8^+$ cells and NK cells (2). Secreted IFN-gamma is a potent inhibitor of *in vitro* hematopoiesis and appears to serve as an important regulator of lymphocyte and macrophage function by exerting antiproliferative effects (1). Antibody ANC2E11 binds to recombinant and cellular IFN-gamma in EIA and Flow cytometry.
References: 1) H.A. Young & K.J. Hardy (1995) J Leuk Biol 58:373-381. 2) A. Bilkiau (1996) Adv Immunol 62:61-130. 3) T. Sato, et al (1997) Blood 90:4749-4758.

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

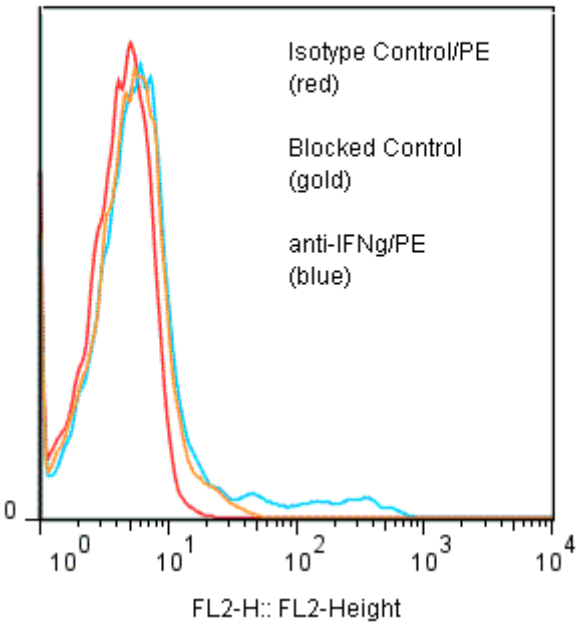
PRODUCT STABILITY: Product should retain activity for at least 12 months after shipping date when stored as recommended. Ship Date:_____

BUFFER: 50 mM Sodium Phosphate pH 7.5, 500 mM Potassium Chloride, 150mM NaCl, 15% Glycerol, 0.2% BSA, 0.04% NaN_3 (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 **250 µg/ml** with an A_{565}/A_{280} ratio of 3.69.

PERFORMANCE: Ficoll prepared human **peripheral blood lymphocytes** were stimulated 4 hours in the presence of 50 ng/ml PMA, 2 µM ionomicin and 5 µM monensin. Cells were then harvested, washed and fixed for 30 min with 2% buffered formaldehyde after which they were washed two times. Fixed cells were permeabolized 10 minutes in a buffer containing **0.3% Saponin**. Subsequent reagent incubations and washes were done using this buffer. Five x 10^5 cells per tube were preincubated 5 minutes with 20 µl of 250 µg/ml human IgG (to block nonspecific binding), after which they were incubated 45 minutes on ice with 80 µl of anti-IFNγ/FITC at a **1:50** dilution factor (5µg/ml). Cells were washed three times, fixed and analyzed by FACS. A net **13%** sub population of the cells stained positive with a mean shift of **1.1** $\log_{10}$ fluorescent units when compared to a Mouse IgG1/R-PE negative control (Catalog # 278-050). Binding was blocked when cells were pre incubated 10 minutes with 20 µl of 0.5 mg/ml anti-IFNγ antibody (Catalog #247-020).

Binding of anti-IFNg/PE to fixed permeabolized stimulated human PBL



Research use only. Not for use in Diagnostic procedures.

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