PERFORMANCE DATA SHEET

1818

Monoclonal anti-human TNF alpha*

mAb name/Clone: J1D9
Isotype: Mouse IgG1

Immunogen: Recombinant TNF alpha

CATALOG#: 398-020 QUANTITY: 100 μg

CONCENTRATION: 1.0 mg/ml

INFORMATION: Human tumor necrosis factor alpha (TNF alpha) is a proinflammatory cytokine expressed mainly by activated monocytes and macrophages. TNF alpha exerts a key role with regard to pathogenesis of many infectious and inflammatory diseases. Antibody J1D9 neutralizes TNF alpha biological activities (2). *References:* 1) P. Vassalli, (1992) Annu Rev Immunol 10: 411-452. 2) P.J. McLaughlin, et al, (1992) Anticancer Res 12: 1243-1246. 3) M. Pasparakis, et al, (1996) Cytokine and Growth Factor Reviews 7: 223-229. 4) A. Eigler, et al, (1997) Immunology Today 18: 487-492.

STORAGE CONDITIONS: *Store at 2 - 5^oC*. Freeze/Thawing is 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, 0.5 mg/ml Gentamicin Sulfate (as a preservative).

PRODUCTION: Antibody was Protein A purified from (low FBS containing) tissue culture supernatant. Purity was >95% Immunoglobulin by SDS-PAGE with less than 1% Bovine Immunoglobulin.

PERFORMANCE: Ficoll prepared human peripheral blood mononuclear cells were stimulated by culturing 6 hours in the presence of 50 ng/ml PMA, 1 µM Ionomycin and 1 µg/ml Brefeldin A. 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 pre incubated 5 minutes with 20 µl of 250 µg/ml human IgG (to block non specific binding) after which they were incubated 45 minutes on ice with 80 μl of anti-TNFα antibody at 5 μg/ml. Cells were washed twice and incubated with 20 reagent Goat anti-Mouse IgG/FITC (Catalog #232-011), after which they were washed three times, fixed and analyzed by FACS. A 20% sub population of the cells stained positive with a mean shift of 0.89 log₁₀ fluorescent units when compared to a Mouse IgG1 negative control (Catalog #278-010) at a similar concentration.

*This Product is intended for Laboratory Research use only.

Binding of anti-TNF alpha mAb +GAM/FITC to fixed permeabolized stimulated human PBMC

