

PERFORMANCE DATA SHEET

1818

*Monoclonal anti-human Perforin/Biotin**

mAb name/Clone: δ G9

Isotype: Mouse IgG2b

Immunogen: Purified granules from human YT lymphoma cell line

CATALOG#: 358-030

QUANTITY: 100 μ g

CONCENTRATION: 0.9 mg/ml

INFORMATION: Human perforin (cytolysin) is believed to be one of the major effector molecules used by cytotoxic T cells and NK cells to mediate targeted cell lysis. Perforin expression is constitutive on NK cells, but increases in resting CD8⁺ cytotoxic cells upon activation. Antibody δ G9 recognizes the human perforin molecule of 70 kd.

References: M.J. Smyth, et al, (1990) J Exp Med **171**: 1269-1281. A. Hameed, et al, Am J Pathol (1992) **140**: 1025-1030. P.L. Jose, et al, (1994) J Immunol **148**: 3354-3360. B.C. Schlesinger & L. Cheng, Immunol (1994) **81**: 291-295. C.C. Liu, et al, (1995) Immunol Today **16**: 194-201. A.H. Hombach, H Abken, et al. (2006) J Immunol **177**: 5668-5675.

STORAGE CONDITIONS: *Store at 2 - 5°C.* 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, 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% mouse immunoglobulin by SDS-PAGE (<1% bovine immunoglobulin), and reacted with NHS-Biotin. Unconjugated Biotin was removed from conjugate using a desalting column.

PERFORMANCE: Ficoll prepared human **peripheral blood mononuclear cells** were stimulated by incubating 1 day in culture at 5 x 10⁶ cells/ml with 10 ng/ml **PMA** and 2 μ M **Ionomycin**. Cells were harvested and then fixed for 30 min with 2% buffered formaldehyde after which they were washed two times. Subsequent reagent incubations and washes were done using a buffer containing **0.3% Saponin** to permeabilize cells. Five x 10⁵ cells per tube were washed and incubated 45 minutes on ice with 80 μ l of anti-perforin/Biotin at 2 μ g/ml. They were then washed twice and incubated with 50 μ l of 2^o reagent Streptavidin R-Phycoerythrin (Catalog #253-050), after which they were washed three times, fixed and analyzed by FACS. A net **4%** sub population of the cells stained positive with a mean shift of **1.1** log₁₀ fluorescent units. Binding was blocked when cells were pre incubated with 20 ul of 0.5 mg/ml anti-perforin mAb.

**This Product is intended for Laboratory Research use only.*

Binding of anti-Perforin/Biotin +SA/PE to fixed stimulated PBMC

