**PERFORMANCE DATA SHEET**
**1818**

# *Monoclonal* anti-human perforin/FITC*

***mAb name/Clone:*** **delta G9**
***Isotype:*** Mouse IgG2b kappa
***Immunogen:*** Purified granules from human YT lymphoma cell line

**CATALOG#: 358-040**
**QUANTITY: 120 tests** **VOLUME IN VIAL: 0.2 ml**
**WORKING DILUTION: 1:50** (or use 1.6µl of concentrated stock per 5 x $10^5$-cell test)

**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 delta 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$^{o}$C***. Freeze/Thawing is not recommended. **Protect from light**.

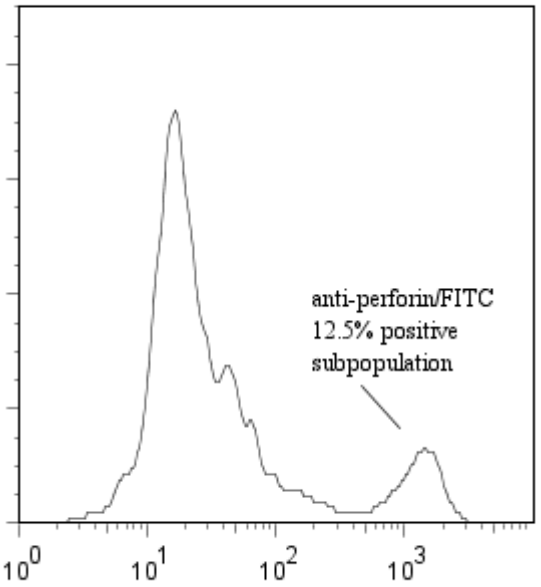
**PRODUCT STABILITY:** Product should retain activity for at least 12 months after shipping date when stored as recommended. Ship Date:_____

**BUFFER:** 50 mM Sodium Phosphate pH 7.5, 100 mM Potassium Chloride, 150mM NaCl, 5% Glycerol, 0.2% BSA, 0.04% $NaN_3$ (as a preservative).

**PRODUCTION:** Antibody from (low FBS containing) tissue culture supernatant was Protein A purified to 95% Immunoglobulin by SDS-PAGE, and reacted with FITC. Unconjugated FITC was removed from conjugate using a desalting column. Antibody conjugate is at a concentration of **0.1 mg/ml**.

**PERFORMANCE:** Ficoll prepared human **peripheral blood mononuclear cells** were stimulated 1 day by incubating 5 x $10^6$ cells/ml in RPMI 10% FBS media containing 5 µg/ml Phytohemagglutinin-P. Cells were then collected and fixed for 30 min with 2% buffered formaldehyde and washed two times with buffer. Cells were permeabolized using a buffer containing **0.3% Saponin**, washed and pre incubated 10 minutes with 20 µl human IgG at 250 µg/ml (to block non specific binding) and than incubated 45 minutes on ice with 80 µl of anti-perforin/FITC at a **1:50 dilution (2 µg/ml)**. Cells were washed three times, fixed and analyzed by FACS. A net **12.5%** sub population of the cells stained positive with a mean shift of **1.2** $\log_{10}$ fluorescent units when compared to a Mouse IgG2b/FITC negative control (Catalog #284-040) at a similar concentration. Binding was blocked when cells were pre incubated 10 minutes with 20 µl of 0.5 mg/ml anti-perforin antibody (Catalog #358-020).

**Binding of anti-perforin/FITC to stimulated, fixed-permeabolized human PBMC**




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

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