PERFORMANCE DATA SHEET ¹⁹³¹ *Monoclonal* anti-mouse CD3/APC*



mAb name/Clone: 145-2C11 *Isotype:* IgG *Immunogen:* Mouse BM10-37 T cell membrane prep

CATALOG#: 703-060 QUANTITY: 120 tests WORKING DILUTION: 1:50 (or use 1.6µl of concentrated stock per 5 x 10⁵-cell test)

INFORMATION: Antibody 145-2C11binds to an extracellular epitope of the 25 kd CD3e molecule present on murine T cells (1). CD3 molecules associate with T cell Receptors and provide intracellular signaling mechanism. Cross linking Cell surface CD3 with soluble or immobilized 145-2C11 can activate this mechanism(2). *References:* 1) O Leo, J A Bluestone, et al. (1987) *PNAS USA* **84**(5): 1374-1378. 2) L E Samelson, J A Bluestone, et al. (1987) *J Immunol* **139**(8): 2708-2714.

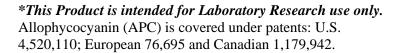
STORAGE CONDITIONS: Store at 2 - 5°C. Do not freeze! Protect from light.

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

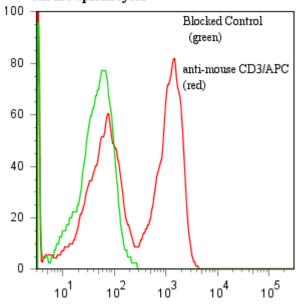
BUFFER: 50 mM Sodium Phosphate pH 7.5, 500 mM Potassium Chloride, 150mM NaCl, 15% Glycerol, 0.2% BSA, 0.04% NaN₃ (as a preservative).

PRODUCTION: Protein G purified antibody from tissue culture supernatant was conjugated to Allophycocycanin through a sulfo-ester linkage. Unconjugated antibody was removed using size exclusion chromatography. Antibody conjugate is at **250** μ g/ml with an A₆₅₀/A₂₈₀ ratio of 2.26.

PERFORMANCE: Reagent was tested for binding to ACK lysed murine splenocytes in FACS. Five x 10^5 **splenocytes** per tube were washed and pre incubated with 20 µl of 300 µg/ml Mouse IgG (to block non specific binding), after which they were incubated 45 minutes on ice with 80 µl of anti-Mouse CD3/APC at a **1:50** dilution (5 µg/ml). They were then washed three times, fixed and analyzed by FACS. A 44.4% sub population of the cells stained positive with a mean shift of **1.35** log_{10} fluorescent units when compared to background. Binding was blocked when cells were pre incubated 10 minutes with 20 µl of 0.5 mg/ml anti-mouse CD3 antibody (Catalog #703-020).



Binding of anti-CD3/APC to RBC-lysed Mouse splenocytes



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