

MONOCLONAL ANTIBODY

# PE labeled CD9

Code No.	Clone	Subclass	Quantity
D252-5	10H6	Mouse IgG1 $\kappa$	1 mL (50 tests)

**BACKGROUND:** CD9 is expressed on platelets, eosinophils, basophils, pre-B cells, activated T cells and neural cell lines. It belongs to a member of the tetraspanin transmembrane-protein (TM4) superfamily, which includes CD37, CD53, CD63, CD81, CD82, CD151 and CD231. Several members of this family, including CD9, form noncovalent associations with integrins, particularly  $\beta$ 1 integrins (CD29). Tetraspanins are involved in membrane events such as sperm-oocyte fusion, myoblast fusion, mononuclear phagocyte fusion, osteoclastogenesis, and paranodal junction formation in the peripheral nervous system. In addition, tetraspanins are implicated in viral processes such as CD63 in HIV infection, CD81 in hepatitis C virus infection, CD82 in cell-to-cell human T cell leukemia virus type I (HTLV-1) spreading, and CD9 in feline immunodeficiency virus (FIV) and canine distemper virus spreading.

**SOURCE:** This antibody was purified from hybridoma (clone 10H6) supernatant using protein A agarose. This hybridoma was established by fusion of mouse myeloma cell P3U1 with Balb/c mouse splenocyte immunized with human prostate carcinoma cell line (PC3).

**FORMULATION:** 50 tests in 1 mL volume of PBS containing 1% BSA and 0.09%  $\text{NaN}_3$ .

\*Azide may react with copper or lead in plumbing system to form explosive metal azides. Therefore, always flush plenty of water when disposing materials containing azide into drain.

**STORAGE:** This antibody solution is stable for one year from the date of purchase when stored at 4°C.

**REACTIVITY:** This antibody reacts with CD9 antigen on Flow cytometry.

**APPLICATION:**

Flow cytometry; 20  $\mu$ L (ready for use)

\*Please refer to the data sheet (MBL code no. D252-3) for other applications.

Detailed procedure is provided in the following **PROTOCOL.**

**INTENDED USE:**

For Research Use Only. Not for use in diagnostic procedures.

**SPECIES CROSS REACTIVITY:**

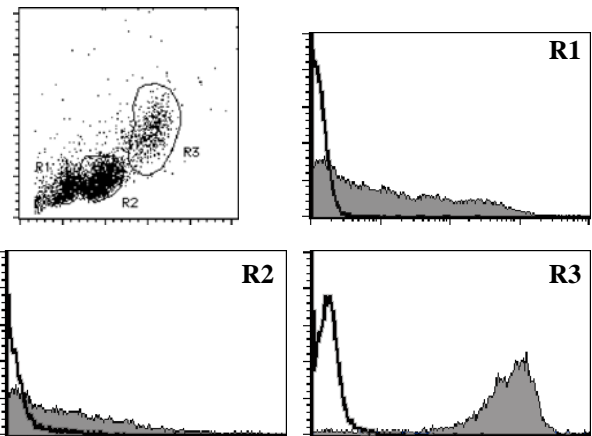
Species	Human	Mouse	Rat
Cells	HL-60, lymphocyte, granulocyte, monocyte	Not Tested	Not Tested
Reactivity on FCM	+		

**REFERENCE:**

1) Boucheix, C., *et al.*, *J. Biol. Chem.* **266**, 117-122 (1991)

**RELATED PRODUCTS:**

- D252-3 CD9 (10H6)
- D131-3 Mouse CD9 (JF9)
- D131-4 FITC labeled Mouse CD9 (JF9)
- D050-3 CD29/ $\beta$ 1-Integrin (AG89)
- D050-5 PE labeled CD29/ $\beta$ 1-Integrin (AG89)
- D263-3 Mouse CD63 (R5G2.1)
- M075-5 PE labeled Mouse IgG1 Isotype control (2E12)



**Flow cytometric analysis of CD9 expression on human lymphocyte (R1), monocyte (R2) and granulocyte (R3) using D252-5.** Open histograms indicate the reaction of isotypic control to the cells. Shaded histograms indicate the reaction of D252-5 to the cells.

**PROTOCOLS:**

**Flow cytometric analysis for floating cells**

We usually use Fisher tubes or equivalents as reaction tubes for all steps described below.

- 1) Wash the cells 2 times with washing buffer [PBS containing 2% fetal calf serum (FCS) and 0.1% NaN<sub>3</sub>].
- 2) Resuspend the cells with washing buffer (5 x 10<sup>6</sup> cells/mL).
- 3) Add 50 µL of the cell suspension into each tube, and centrifuge at 500 x g for 1 minute at room temperature (20~25°C). Remove supernatant by careful aspiration.
- 4) Add 20 µL of Clear Back (human Fc receptor blocking reagent, MBL; code no. MTG-001) to the cell pellet after tapping. Mix well and incubate for 5 minutes at room temperature.
- 5) Add 20 µL of the primary antibody. Mix well and incubate for 30 minutes at room temperature.
- 6) Add 1 mL of the washing buffer followed by centrifugation at 500 x g for 1 minute at room temperature. Remove supernatant by careful aspiration.
- 7) Resuspend the cells with 500 µL of the washing buffer and analyze by a flow cytometer.